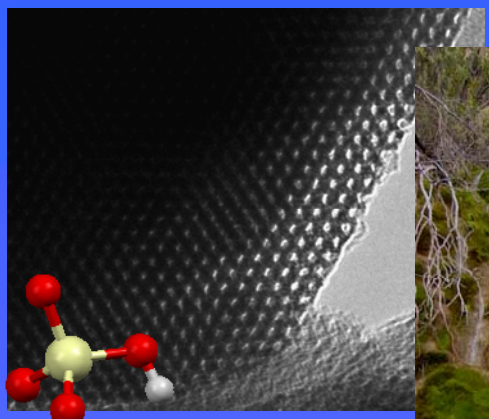
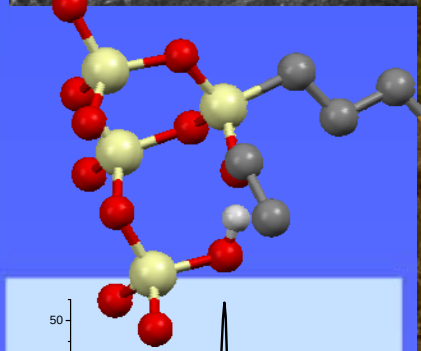
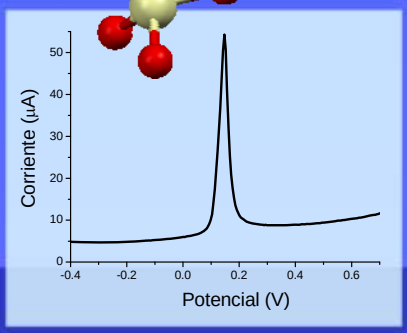
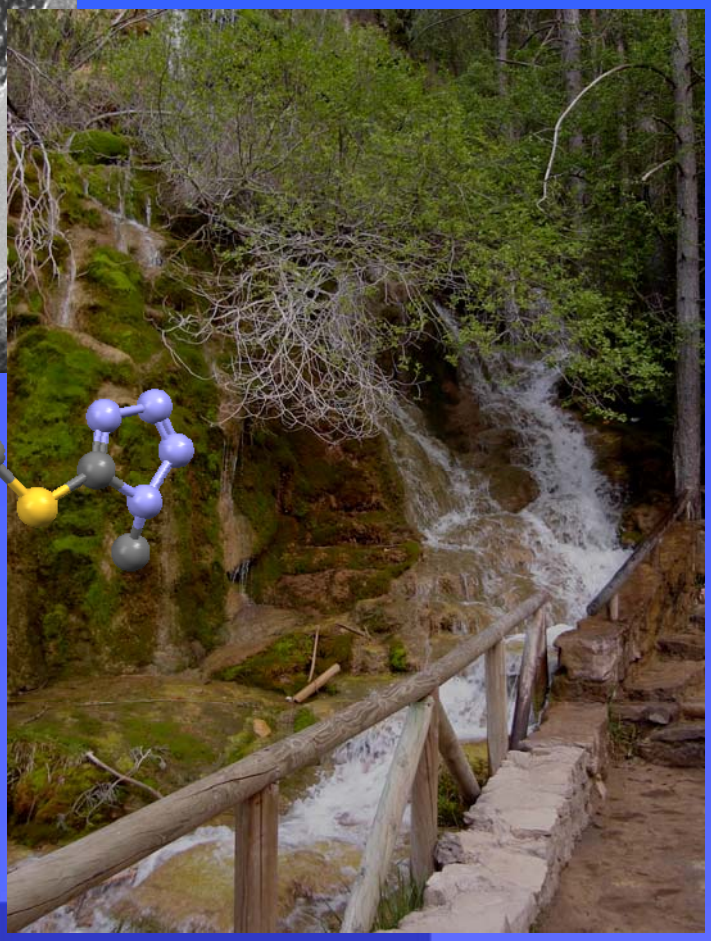
Tesis Doctoral

APLICACIÓN DE MATERIALES HÍBRIDOS PARA LA DETERMINACIÓN ELECTROQUÍMICA DE METALES PESADOS EN AGUAS









ALFREDO SÁNCHEZ SÁNCHEZ

Universidad Rey Juan Carlos

Septiembre 2011

TESIS DOCTORAL

APLICACIÓN DE MATERIALES HÍBRIDOS PARA LA DETERMINACIÓN ELECTROQUÍMICA DE METALES PESADOS EN AGUAS

Departamento de Química Inorgánica y Analítica
UNIVERSIDAD REY JUAN CARLOS

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Septiembre 2011

Dra. Isabel Sierra Alonso, Profesora Titular de Química Analítica del Departamento de Química Inorgánica y Analítica de la Universidad Rey Juan Carlos, campus de Móstoles (Madrid), y **Dra. Isabel del Hierro Morales**, Profesora Titular de Química Inorgánica del Departamento de Química Inorgánica y Analítica de la Universidad Rey Juan Carlos, campus de Móstoles (Madrid),

CERTIFICAN:

Que el presente trabajo titulado "Aplicación de materiales híbridos para la determinación electroquímica de metales pesados en aguas" el cual constituye la memoria que, bajo su dirección, presenta **D. Alfredo Sánchez Sánchez** para optar al Grado de *Doctor por la Universidad Rey Juan Carlos*, con mención de *Doctor Europeo*, ha sido realizado bajo su supervisión en el Departamento de Química Inorgánica y Analítica de la Universidad Rey Juan Carlos, así como durante cuatro meses en el Laboratoire de Chimie Physique et Microbiologie pour l´Environnement de la Universidad Henri Poincaré de Nancy (Francia), cumpliendo todos los requisitos necesarios.

Y para que así conste, expiden y firman el presente certificado, en Móstoles a 21 de junio de 2011.

Fdo. Isabel Sierra Alonso Fdo. Isabel del Hierro Morales

A mis padres y hermana

A Cristina

Agradecimientos:

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PRESENTACIÓN

La presente tesis se muestra como un compendio de artículos de investigación publicados en revistas científicas internacionales o enviados para su publicación, en los cuales la temática común es el desarrollo sílices mesoporosas funcionalizadas con el ligando orgánico 5-mercapto-1-metiltetrazol (MTTZ) para la determinación de metales pesados en medios acuosos.

En el primer capítulo se recoge una introducción teórica, se presentan los objetivos del trabajo, se muestra la metodología seguida y, por último, se recogen las conclusiones más relevantes obtenidas del trabajo realizado.

En el capítulo II se presenta la síntesis de un nuevo material híbrido formado a partir de la sílice mesoporosa tipo MSU-2 y el ligando MTTZ, comprobándose su capacidad para la adsorción de metales pesados, así como su selectividad hacia diferentes metales.

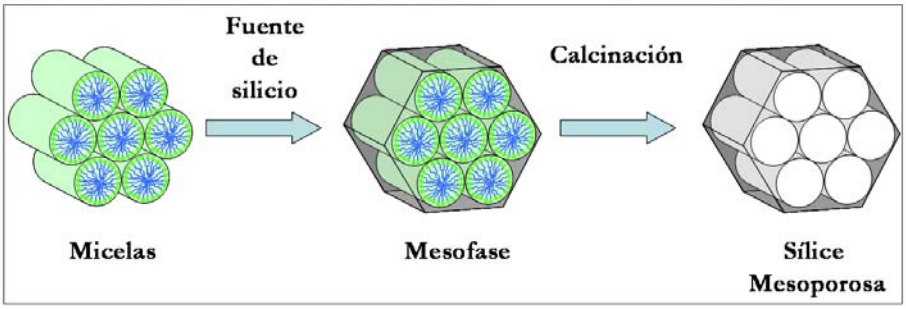
El capítulo III se centra en la determinación de Pb(II) en medios acuosos mediante el empleo de un electrodo de pasta de carbono modificado con sílice mesoporosa funcionalizada, optimizando las variables que afectan a la medida, tanto en la etapa de preconcentración como en la de electrólisis y redisolución. Por último, el método desarrollado se aplica a la determinación de Pb(II) en muestras de agua de grifo y subterránea.

En el capítulo IV se presenta un método para la determinación de Pb(II) en medios acuosos mediante el empleo de electrodos serigrafiados de carbono modificados con sílice mesoporosa híbrida, utilizando un potenciostato portátil, optimizándose las variables que afectan a la medida. El método desarrollado se aplica a la determinación de este metal en agua de grifo, de río y subterránea.

El capítulo V presenta la determinación de Hg(II) en medios acuosos mediante el empleo de un electrodo de pasta de carbono modificado con sílice mesoporosa funcionarizada, optimizándose las variables que afectan a la medida. El método desarrollado se aplica a la determinación de este metal en agua de grifo y subterránea.

Por último, el capítulo VI muestra el trabajo desarrollado en la Universidad de Nancy (Francia) en el que se llevó a cabo la preparación de un electrodo consistente en una película de sílice mesoporosa con el ligando MTTZ depositada sobre un electrodo de grafito vitrificado. El electrodo desarrollado se aplica a la determinación de Hg(II) en medios acuosos, optimizándose las variables que afectan a la medida.

RESUMEN


Fuente
de
silicio
Calcinación
Micelas
Mesofase
Sílice
Mesoporosa

1.-INTRODUCCIÓN

El agua es uno de los recursos naturales más importantes para los seres humanos y la vida en general, ya que cabe señalar que todas las formas de vida requieren un aporte importante de agua y que casi todas las funciones de nutrición y excreción en los seres vivos se basan en el agua.

El agua es un recurso natural renovable, ya que sigue un ciclo que hace que se mantenga constante (Figura 1), y abundante en términos globales, ya que aproximadamente el 72% de la superficie terrestre se encuentra cubierta por agua, el problema es que solo el 3% del agua del planeta es agua dulce, del cual una gran parte se encuentra en forma de hielo en los glaciares o como aguas subterráneas, quedando únicamente como agua dulce superficial y disponible directamente (ríos y lagos) un 0,014%.

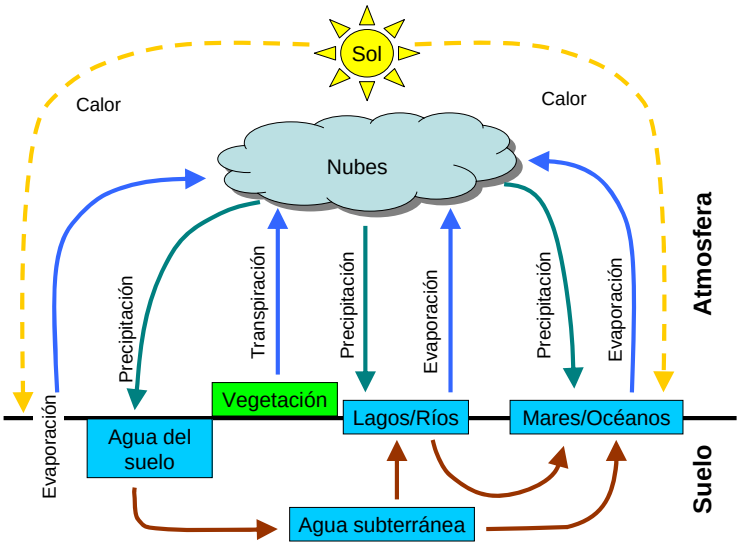


Figura 1. Ciclo del agua simplificado.

Los usos del agua son tan amplios que engloban desde necesidades básicas como calmar la sed, la preparación de alimentos, la higiene, fuente de alimentos tanto en agricultura y ganadería como en el sector pesquero, como otras también importantes como la producción de energía, el uso industrial, el transporte e incluso el ocio. A nivel mundial, los usos que se le dan al agua son en agricultura un 66%, en la industria un 24%, en consumo urbano un 8% y para otros fines (recreativos,

ganadería,…) un 2%. Como se ha comentado anteriormente, la cantidad de agua dulce directamente aprovechable es un porcentaje minúsculo de la totalidad del agua en el planeta, aunque se encuentra en constante renovación; además se trata de un recurso mal repartido, y aunque hoy en día existe tecnología suficiente para poder explotar las aguas subterráneas así como para transformar el agua salada en agua dulce, la calidad y abundancia del agua determina el desarrollo socio-económico de la región.

Así como el agua determina el desarrollo socio-económico de la región, la industrialización, el crecimiento demográfico, etc., todos los usos que el hombre da al agua provocan la aparición de sustancias tóxicas y contaminantes en las corrientes de aguas (aguas residuales). En los últimos años se ha tomado conciencia del grave problema que provoca tanto la escasez de agua como su contaminación. Estos contaminantes generalmente se degradan en las estaciones de depuración de aguas residuales, bien mediante tratamientos físicos, químicos o biológicos. En este aspecto los metales pesados presentan una problemática especial debido a que no son fáciles de eliminar y su toxicidad se da incluso a concentraciones muy reducidas.

A lo largo de la historia, la calidad del agua potable ha sido un factor determinante del bienestar humano. Las enfermedades propagadas por aguas contaminadas han causado y siguen causando grandes problemas a las personas que se ven obligadas a usarlas, tanto para el consumo directo como para el cultivo de alimentos. La contaminación de las aguas naturales tanto por contaminantes químicos como biológicos, es un problema a nivel mundial, no importa el nivel de desarrollo del país sino que todos sufren de una u otra forma esta contaminación. El agua contaminada causa la muerte de 2,2 millones de personas al año, por lo que la calidad de este recurso es una característica muy a tener en cuenta [1-8].

En la actualidad, uno de los retos desde el punto de vista ambiental es desarrollar y mejorar las técnicas analíticas para poder obtener información fiable, lo suficientemente precisa y a tiempo real de los parámetros de calidad de las aguas, de forma que la respuesta ante cualquier peligro sea lo más rápida posible y minimizar así los potenciales efectos en la salud human y el medioambiente [9].

Unos de los contaminantes ambientales más importantes son los metales pesados. Una de las características y uno de los principales problemas que presentan en cuanto a su control y cuantificación es la baja concentración en la que se suelen encontrar y que permite la ley establecida para aguas de consumo, que marca como valores paramétricos niveles de partes por billón para todos los casos. Estas concentraciones no son fácilmente cuantificables y para muchos de los casos las técnicas analíticas disponibles no consiguen llegar a límites de detección adecuados. Además, la creciente preocupación por el medioambiente hace que las leyes sean cada vez más estrictas y que los valores permitidos sean cada vez más bajos. Por todo ello, es necesario el desarrollo e implantación de técnicas capaces de adaptarse a esta demanda cada vez más restrictiva [9, 10].

1.1.-Metales pesados

La designación de metal pesado no está muy bien definida, pudiéndose encontrar diversos criterios para clasificar este tipo de contaminantes, empleando el criterio de densidad se puede considerar como metal pesado aquellos metales con una densidad superior a 4,5 g/cm^3, mientras que si tenemos en cuenta peso atómico se denomina metal pesado a aquellos elementos químicos que poseen un peso atómico comprendido aproximadamente entre el del cromo (51,99) y el del plomo (207,19).

Los metales pesados se pueden englobar en dos grupos diferentes en relación a su posible función biológica, pudiéndose diferenciar en oligoelementos o tóxicos. Los que presentan carácter de micronutrientes u oligoelementos son metales esenciales para la vida, ya que desarrollan una función importante en el organismo, el caso más claro es el del hierro, fundamental para la formación de la hemoglobina de la sangre, otros ejemplos son As, B, Co, Cr, Mo, Mn, Ni, Se y Zn. Por el contrario, otros de los metales pesados no son esenciales y presentan efectos tóxicos sobre el organismo. Incluso los metales esenciales, cuando sobrepasan las concentraciones requeridas por el organismo, pueden tener efectos tóxicos. Los metales pesados tóxicos son principalmente Hg, Cd, Pb, Sb y Bi, tratándose de metales que han acompañado al hombre desde la más remota antigüedad. Los envenenamientos por arsénico, plomo y mercurio poseen un carácter legendario.

La toxicidad que presentan estos metales no esenciales radica, generalmente, en que presentan actividad cancerígena y mutagénica, envenenamiento, así como en la reducción de la actividad de alguno de los órganos vitales. Bioquímicamente, el mecanismo de su acción tóxica proviene de la fuerte afinidad de los cationes de estos metales por el azufre. Por esta razón, los metales ingeridos se enlazan fácilmente a los grupos tiol (-SH) presentes en las enzimas, provocando una inhibición de la actividad enzimática y afectando a la salud humana.

La persistencia en el ambiente de los metales ocasiona una problemática especial. A diferencia de los contaminantes orgánicos, los metales no pueden degradarse ni biológicamente ni químicamente en la naturaleza, son elementos químicos que el hombre no crea ni destruye. La estabilidad de los metales permite también que sean transportados a distancias considerables, tanto por aire como por agua. El papel que el hombre desempeña en la presencia ambiental de los metales es introducir en el medio ambiente estos elementos como consecuencia de distintas actividades antropogénicas. Los compuestos que contienen metales pueden alterarse, pero los elementos metálicos indeseables aún permanecen. En algunos casos tales alteraciones desembocan en especies que contienen el metal y son más tóxicas aún que las iniciales.

Los metales pesados presentan una cualidad especial debido a que no son biodegradables, esta es su capacidad de bioacumulación en los organismos vivos. La importancia de este fenómeno es debida a que en muchos casos lo que hace tóxicos a los metales pesados no son, en general, sus características esenciales, sino las concentraciones en las que pueden presentarse.

La mayor parte de los metales pesados son de interés industrial y económico, lo que hace que las principales fuentes antropogénicas sean las actividades de minería, fundición y refinado, que implican arrancar los metales de sus menas en los depósitos subterráneos y, posteriormente, fundirlos y refinarlos hasta obtenerlos y convertirlos en bienes de consumo que después de usados se desechan. Durante estas operaciones se liberan metales al medio ambiente; también se emiten metales a la atmósfera por combustión de combustibles fósiles, que en un periodo más o menos largo acabarán depositándose en la superficie terrestre.

En el transcurso de su vida útil los metales están sujetos a corrosión y desgaste, lo que produce las pérdidas al ambiente. La mayoría de los contaminantes metálicos terminan por alcanzar las aguas superficiales y subálveas. Los residuos de muchas operaciones industriales que contienen metales se encuentran en forma líquida y alcanzan rápidamente las aguas naturales. Además, se liberan al aire cantidades significativas de muchos metales que son de nuevo transportados por la lluvia a la superficie terrestre. La escorrentía agrícola, los residuos mineros y el alcantarillado doméstico contribuyen también a las concentraciones de metales que se encuentran en las aguas naturales.

Los casos conocidos de desastres provocados por los metales pesados son amplios, y engloban desde la posible caída del imperio romano debido a la intoxicación crónica por plomo procedente de las fabricación del edulcorante *sapa*, obtenido por ebullición de mosto o vino en recipientes de plomo, la muerte de casi 200 personas y 400 afectados por la enfermedad de Itai-Itai, en años posteriores a la II Guerra Mundial, debido a la contaminación con cadmio procedente de la actividad minera en los cultivos de arroz en el distrito de Toyama en Japón, la muerte de 280 personas y más de 5.000 afectados en el norte de Irak por el consumo de harinas procedentes de semillas tratadas con fungicidas mercuriados destinadas inicialmente a la siembra o la enfermedad de Minamata, responsable de la muerte de 111 personas y más de 400 afectados entre 1.953 y 1.965 (cerca de 10.000 casos diagnosticados hasta 2.001) producida por la alimentación basada en pescado y marisco de la zona de Minamata (Japón) contaminado con metilmercurio procedente de la actividad minera nuevamente [1, 2, 5].

Los metales pesados se encuentran presentes en los sistemas acuosos a muy baja concentración, por lo que se denominan elementos traza, pero a pesar de esta baja concentración, y debido a su poder de bioacumulación, es muy importante poder

conocer y controlar de forma rigurosa los niveles en los que se encuentran estos contaminantes en los medios acuosos, principalmente en las aguas de consumo.

1.1.1.-El Plomo

El plomo (Pb) es un elemento químico de número atómico 82 y peso atómico 207,19. Se clasifica dentro de los llamados metales pesados con una densidad relativa de 11,4 g/mL. La capacidad de combinarse con los átomos de otros elementos viene determinada por los estados de oxidación 2 y 4 (Pb(II) y Pb(IV), siendo el estado de oxidación dos el más estable y por tanto, el que se presenta en mayor porcentaje en la naturaleza. Su punto de ebullición, se observa a 1.725 ºC y su punto de fusión a 328 ºC. Es un metal blando, maleable, dúctil, pobre conductor de electricidad y muy resistente a la corrosión.

En la naturaleza el plomo no se presenta habitualmente en su estado elemental sino que forma parte de minerales como la galena (PbS), la anglesita ($PbSO_4$) y la cerusita ($PbCO_3$), entre otros. Existen así mismo numerosos compuestos inorgánicos en cuyas estructuras está presente el plomo, ejemplo de estos compuestos son los acetatos, bromatos y oxalatos.

El plomo metálico es poco resistente al ataque de los ácidos sulfúrico y clorhídrico y se disuelve con lentitud en ácido nítrico. En condiciones normales el plomo metálico es pobremente soluble en agua [11].

1.1.1.1.-Origen de la contaminación por Plomo

La distribución del plomo en la corteza terrestre es muy abundante ya que se encuentra formando parte de numerosas estructuras. Sin embargo, la problemática de su aumento y acumulación en el medio ambiente es una consecuencia directa de los múltiples usos industriales del elemento, ya que el hombre ha hecho uso de este metal desde hace unos 6.000 años, lo que ha dado lugar a la aparición de un ciclo no natural del plomo. Debido a este hecho, el plomo se distribuye por suelo, agua y atmósfera y en ocasiones lo hace en concentraciones superiores a las recomendables.

En la actualidad, aproximadamente el 35% del plomo se utiliza para fabricar baterías para automóviles, el segundo uso del plomo, por su importancia cuantitativa, es su incorporación al cristal de monitores de ordenador y pantallas de televisión, con la función de absorber radiaciones perjudiciales. El amplio espectro de usos del plomo en la industria abarca además la fabricación de pinturas, barnices para cerámicas, tetraetilplomo para elementos de la construcción, diversos cosméticos, la fabricación de tuberías de plomo y como constituyente de combustibles entre otros muchos usos. Estas dos últimas aplicaciones son las causas más comunes de contaminación de aguas por plomo, así como las baterías de automóviles gastadas [5].

Otra de las fuentes que se suele quedar en el olvido es la actividad de caza y pesca, se estima que, en España, la caza y el tiro son responsables de dispersar anualmente cerca de 6.000 toneladas en el ambiente (60.000 toneladas anuales en

EE.UU.), y la pesca deja en las zonas acuáticas unas 100 toneladas más cada año. Si tenemos en cuenta estos valores a nivel global y los comparamos con los 300 millones de toneladas que se estima que los humanos hemos extraído a lo largo de la historia, podemos ver como una cantidad importante de este plomo se encuentra como contaminante en el medioambiente [12].

1.1.1.2.-El Plomo en disolución

En condiciones normales el plomo metálico no reacciona con el agua. Sin embargo, cuando se pone en contacto con aire húmedo, la reactividad con el agua aumenta. En la superficie del metal se forma una pequeña capa de óxido de plomo (PbO); en presencia de oxígeno y agua, el plomo metálico se convierte en hidróxido de plomo ($Pb(OH)_2$):

$$2Pb(s) + O_2(g) + 2H_2O(l) \rightarrow 2\,Pb(OH)_2(s)$$

lo que hace que este pueda entrar en el agua potable a través de la corrosión de las tuberías de plomo muy usadas a nivel doméstico hasta el año 1980.

En sistemas acuáticos contaminados el plomo presente tiende casi en su totalidad a sedimentar y acumularse en el fondo en forma de sulfuro, carbonato y fosfato. Sin embargo, en ocasiones una fracción menor del elemento permanece disuelta en el agua, esto viene marcado por el pH del medio.

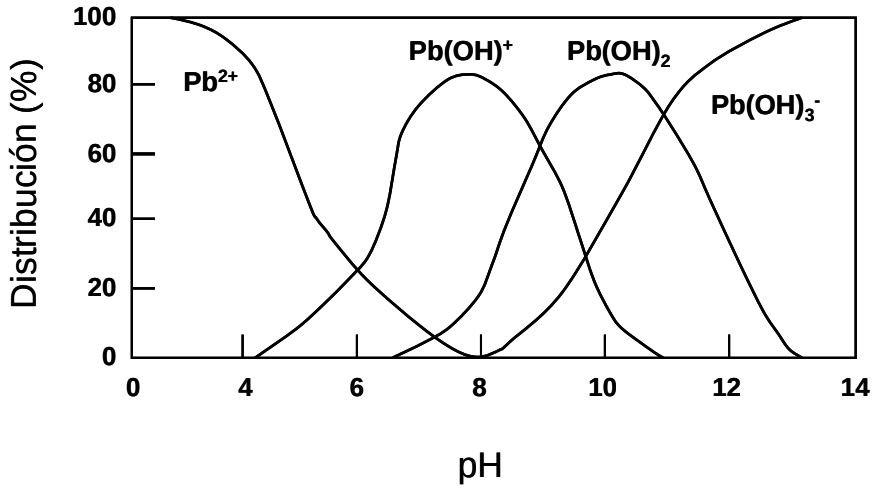


Figura 2. Especiación del plomo en disolución en función del pH.

1.1.1.3.-Toxicidad del Plomo

El plomo y los compuestos de plomo son generalmente contaminantes tóxicos. Las sales de plomo II y los compuestos orgánicos del plomo son dañinos desde un punto de vista toxicológico. La EPA, Agencia de Control Ambiental Americana, ha clasificado al plomo como probable carcinógeno en el hombre (grupo B2) [13] y la Agencia Internacional para la Investigación del Cáncer lo clasifica como posible carcinógeno en el hombre (grupo 2B) [14].

El plomo disuelto en los medios acuáticos, una vez asimilado por un organismo, se acumula en determinados órganos o tejidos. Se conoce, desde hace tiempo, el peligro de bioacumulación del plomo, derivado de la dificultad de excreción del mismo. Un estudio que ilustra este hecho es el realizado por Anderson en 1978 [15]. Anderson expuso a cangrejos (Orconectes virilis) a varias disoluciones de acetato de plomo en concentraciones crecientes. Tras un período de exposición de 40 días, se observó un aumento marcado del contenido de plomo en agallas y exoesqueleto. Dicho aumento fue directamente proporcional a la concentración y el tiempo de exposición.

El plomo es un metal neurotóxico que puede ser absorbido fácilmente por el cuerpo humano, en primer lugar a través de estómago y los pulmones. Los efectos tóxicos del plomo en humanos, especialmente niños han sido bien documentados. Los estudios sugieren que los daños neurológicos en niños pueden ocurrir a niveles de plomo en sangre tan bajos como 10 μg/dL [16].

El plomo limita la síntesis clorofílica de las plantas. No obstante las plantas pueden absorber del suelo altos niveles de plomo, hasta 500 ppm. Mediante la absorción por parte de las plantas, el plomo también puede ser introducido en la cadena alimenticia. Consecuentemente, la aplicación de pesticidas de plomo está prohibida en la mayor parte de los países. En el agua residual, el plomo proviene principalmente de las calles, tubería y suelos, debido a que se utilizó en fontanería y todavía está presente en construcciones antiguas.

La persistencia y bioacumulación del plomo en organismos induce una vía de intoxicación a través de la cadena trófica. La máxima concentración se alcanza en los huesos, que actúan como lugares de almacenamiento de este elemento, los órganos en los que aparece en mayor medida son el hígado, los riñones y el sistema nervioso central. Esta intoxicación presenta diversos efectos, como nauseas, dolores abdominales, vómitos, parestesia, dolor y debilidad muscular, fatiga, espasmos, cambios enzimáticos en la sangre, anemia, hiperactividad y desórdenes neurológicos; los efectos en los niños son más graves que en adultos, siendo los síntomas más característicos el letargo, vómitos, irritabilidad, pérdida del apetito y mareos, que avanza hasta desembocar en ataxia y reducción de la consciencia, y pueden provocar finalmente coma y la muerte. La intoxicación por plomo afecta además al desarrollo del feto [5, 11, 17].

Un estudio llevado a cabo por Gil y col. en 2006 [18] tuvo como objetivo aportar datos de base sobre los niveles de plomo, entre otros metales pesados, en diferentes especies de moluscos de importancia económica y alimentaria, los cuales fueron recolectados entre marzo y mayo de 1.994 en diferentes puntos de la línea costera en la Patagonia argentina. La concentración de plomo fluctuó entre 1,64 μg/g y 6,74 μg/g sobre materia seca. El riesgo que significa para la salud humana, se determina comparando la ingesta estimada del tóxico, con los niveles seguros recomendados. Estas recomendaciones se basan en el concepto de Ingesta Semanal Tolerable Provisional (ISTP) que tiene un valor para el plomo de 25 μg/kg [19]. El envenenamiento por plomo puede ocurrir en población susceptible como bebes y niños o debido a la exposición ocupacional en trabajadores.

Los criterios sanitarios que rigen la calidad del agua de consumo humano en España vienen definidos dentro del REAL DECRETO 140/2.003, de 7 de febrero [10], por el que se establece que entre las fechas 01/01/2.004 a 31/12/2.013 el valor paramétrico del plomo disminuya de 50 μg/L a 25 μg/L. A partir del 01/01/2.014 se endurecerá la norma permitiendo un valor paramétrico de 10 μg/L. Este último valor es el máximo límite tolerable que establece la WHO para 2010 [20, 21].

1.1.2.-El Mercurio

El mercurio (Hg) es un elemento químico de número atómico 80 y peso atómico 200,59. Se clasifica dentro de los llamados metales pesados con una densidad relativa de 13,55 g/mL. Es el único metal líquido a temperatura ambiente, ya que presenta una temperatura de fusión de -38,8 ºC, siendo además un elemento muy volátil, con una presión de vapor a 20 ºC de $1{,}22 \cdot 10^{-3}$ mm. La capacidad de combinarse con los átomos de otros elementos viene determinada por los estados de oxidación 1 y 2 (Hg(I) y Hg(II)), y a pesar de tratarse de un metal noble, se encuentra estable en la naturaleza presente en cualquiera de sus tres estados de oxidación. Se alea fácilmente con otros metales como el oro y la plata formando amalgamas. Además de las distintas formas inorgánicas, generalmente unido a oxigeno, cloro y azufre, el mercurio se presenta también formando parte de compuestos orgánicos entre los que destaca el metilmercurio por su toxicidad. Es un metal pobre conductor del calor pero muy buen conductor de la electricidad [11].

El mineral más importante del mercurio es el cinabrio (HgS), encontrándose sus mayores reservas mineras en Almadén, Ciudad Real, de donde se han extraído unas 200.000 toneladas de mercurio desde hace 2.500 años. El mercurio se extrae fácilmente de la mena, que al calentarla en el aire a temperaturas superiores a los 540 ºC desprende vapor de mercurio que posteriormente se condensa para obtener el metal líquido [22].

1.1.2.1.-Origen de la contaminación por Mercurio

La distribución del mercurio en la corteza terrestre es abundante, encontrándose tanto en estado elemental como combinado en forma de sales inorgánicas como en compuestos organometálicos. Se trata de un metal que se emplea desde la antigüedad más remota, la muestra de mercurio líquido más antigua se encontró en una tumba egipcia en Kurna y data de 1.600 a.C. La problemática de su aumento y acumulación en el medio ambiente es una consecuencia directa de los múltiples usos industriales del elemento, debido a este hecho, el mercurio se distribuye por suelo, agua y atmósfera y entra en la cadena trófica a través de los organismos acuáticos principalmente.

En la actualidad, los usos más importantes del mercurio se encuentran en la industria cloro-alcalina, la fabricación de pilas, lámparas fluorescentes y material para empastes dentales (sustituido en los países desarrollados). Otra de las aplicaciones más importantes hasta el momento ha sido su empleo en la extracción de oro y plata, gracias a la capacidad del mercurio de amalgamar estos metales y la facilidad que presenta para separarse posteriormente por calentamiento. Esta aplicación junto con la de la industria cloro-alcalina son las que más problemas medioambientales han causado, y han sido la razón de numerosas intoxicaciones con fatales consecuencias. Pero esta no es la única aplicación del mercurio responsable de victimas mortales, ya que también ha causado desgracias como fungicida. Otras aplicaciones que cabe destacar del mercurio son su empleo en termómetros, para la confección de espejos, como detonante, en motores de turbinas e incluso en medicina como antiséptico.

A pesar de su gran variedad de aplicaciones, las principales fuentes de liberación de mercurio elemental al medioambiente son procesos naturales de volatilización a partir de depósitos minerales y volcanes principalmente, se calcula que estos procesos liberan entre 25.000 y 150.000 toneladas anuales de mercurio a la atmósfera, el cual pasa al agua y el suelo posteriormente [5].

1.1.2.2.-El Mercurio en disolución

El mercurio en el medioambiente sigue un ciclo complejo que engloba sus tres estados de oxidación, siendo el más escaso de ellos el mercurio (I), y se extiende por aire, agua y suelo (Figura 3), interviniendo en dicho ciclo procesos químicos y biológicos [5]. En el medio acuoso coexisten diversas especies, desde mercurio elemental, sales inorgánicas de mercurio que presentan cierta solubilidad, hasta metilmercurio formado por procesos metabólicos de distintas bacterias o por procesos puramente químicos. Las formas orgánicas de mercurio, principalmente el metilmercurio son muy asimilables por los organismos vivos, por lo que una vez producido entra en la cadena trófica sufriendo un proceso de biomagnificación a lo largo de ella.

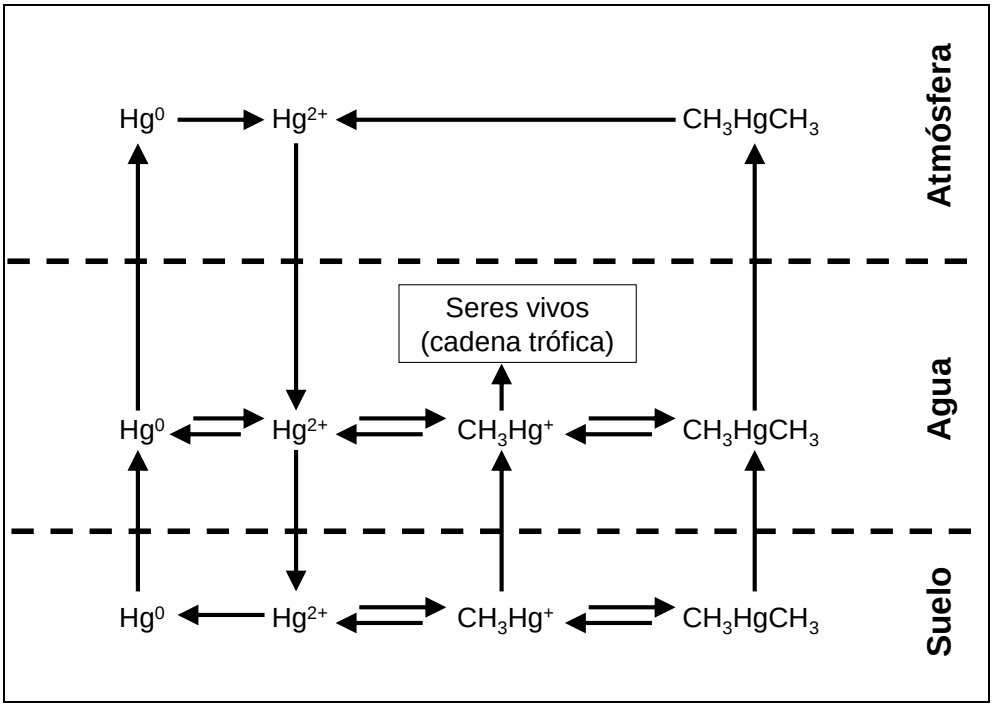


Figura 3. Ciclo biogeoquímico del mercurio.

1.1.2.3.-Toxicidad del Mercurio

El mercurio es uno de los mejores ejemplos de la multiplicidad de efectos tóxicos de los metales dependiendo de la forma química en que se encuentre. Las principales formas en las que presenta una importante toxicidad son en estado elemental y como compuestos organomercuriales, de los que cabe destacar el metilmercurio.

La principal vía de exposición al mercurio elemental es por inhalación de sus vapores, provocando desordenes neurológicos y de comportamiento como temblores labilidad emocional, insomnio, pérdida de la memoria, cambios en el sistema neuromuscular y dolores de cabeza. Se han observado efectos en el riñón y la tiroides y las altas exposiciones han ocasionado mortalidad. Alrededor del 80% del vapor de mercurio inhalado es absorbido por los tejidos pulmonares, se disuelve en la sangre, y es transportado al cerebro, en donde produce daños irreversibles al sistema nervioso central, también es oxidado por los glóbulos rojos de la sangre acumulándose en sus formas oxidadas en los riñones. Por otra parte la absorción intestinal de mercurio elemental es baja y puede oxidarse en los tejidos corporales a la forma divalente inorgánica [23].

En cuanto al metilmercurio, este se absorbe de forma muy eficaz tras su ingestión, gracias a su carácter lipofílico se produce la absorción de más del 90% del metilmercurio ingerido, además presenta una fuerte tendencia a la bioconcentración en la cadena trófica. Este compuesto se acumula en el cerebro, especialmente en la corteza cerebral. Los principales efectos tóxicos en adultos son neurológicos, mientras que la exposición a este compuesto durante el embarazo provoca la aparición de lesiones congénitas del sistema nervioso causando la disrupción de la arquitectura del cerebro. La neurotoxicidad del metilmercurio se manifiesta en temblores y pérdida de sensibilidad en dedos de extremidades superiores e inferiores, ataxia, neurastenia, pérdida de visión y audición, espasmos y finalmente, coma y muerte [5].

El mercurio inorgánico es poco asimilable por ingestión, del orden del 15% se consigue asimilar y se concentra principalmente en los riñones. Su forma más común es el cloruro de mercurio(II) y sus efectos son la ulceración corrosiva del sistema digestivo, causando hemorragias y necrosis en el tubo digestivo, incluyendo la insuficiencia renal generalizada, pudiendo ser los efectos fatales en dosis elevadas [5].

Los niños son más susceptibles al mercurio que los adultos, en ellos puede producir daños en el sistema nervioso y digestivo y lesiones en el riñón. En mujeres embarazadas puede pasar de la madre al feto, en donde puede acumularse y producir daño cerebral, retardo mental, ceguera, incapacidad para hablar, etc. también puede transmitirse al niño a través de la leche materna [24].

La Agencia de Protección Ambiental Americana (EPA) y la Agencia Internacional para la Investigación del Cáncer (IARC) han determinado que el cloruro de mercurio y el metilmercurio son posiblemente carcinógenos en seres humanos (grupo 2B), así mismo concluyen que el mercurio metálico y los compuestos orgánicos de mercurio no son clasificables en cuanto a carcinogenicidad para los seres humanos (grupo 3).

El Real decreto 140/2003 del Ministerio de Sanidad y Consumo del 7 de febrero [10] establece como límite máximo permitido de mercurio en aguas para el consumo 1,0 μg/L (1ppb). Igualmente la EPA estableció normas sobre la calidad del agua respecto a las concentraciones máximas de mercurio en medios acuosos, con una concentración de 12 partes por billón (ppb) como valor máximo en los ecosistemas de agua dulce, 25 ppb en agua salobre y 2 ppb en agua potable, así mismo la Ley Nacional de Tóxicos de la EPA recomienda que la concentración segura de mercurio sea de tan solo 1,3 ppb. Normalmente, las concentraciones menores a esta norma representan el total de mercurio disuelto (formas inorgánicas mas formas orgánicas) en el agua de mar. Las aguas superficiales de algunas partes del mar contienen concentraciones inferiores a 0,05 ppb, y las capas intermedias del agua contienen hasta 2 ppb (Skoog, 2008). La Administración Americana de Alimentos y Fármacos (FDA) ha establecido un nivel permisible máximo de 1 ppm de metilmercurio en pescados y mariscos [24].

1.2.-Métodos de análisis de metales pesados en aguas

Desde el punto de vista de la Química Analítica surgen dos problemas iniciales en la determinación de metales pesados en muestras acuosas, primero la baja concentración de los metales pesados en las muestras, que se encuentran por debajo del límite de detección de muchas técnicas instrumentales y segundo la necesidad de adaptación a los requerimientos de las nuevas normativas que restringen cada vez más los valores paramétricos requeridos.

En la actualidad se dispone de varias técnicas que pueden utilizarse para determinar cuantitativa y/o cualitativamente la presencia de trazas o ultratrazas de elementos metálicos. Entre éstas destacan las técnicas espectroscópicas, como la espectroscopia de absorción atómica (AAS) y la espectroscopia de emisión atómica por atomización en plasma acoplado por inducción (ICP) principalmente, y las técnicas voltamperométricas. La elección de la técnica analítica a emplear va a depender de los niveles de concentración en los que nos movamos, de la matriz que acompaña al analito y de la disponibilidad económica principalmente.

1.2.1.-Técnicas espectroscópicas

El término *técnicas espectroscópicas* engloba un conjunto de técnicas que estudian la interacción de la radiación electromagnética con la materia. Cuando se suministra energía por medio de la radiación electromagnética a un átomo o molécula, parte de esa energía es absorbida pasando los electrones a un nivel excitado, esa energía equivale exactamente a la diferencia entre los niveles energéticos. Por el contrario, cuando los electrones que se encuentran en un nivel excitado caen a otro de menor energía, se produce la emisión de energía correspondiente exactamente al salto de nivel que se produce. La espectrometría y los métodos espectrométricos se refieren a la medida de esta intensidad de radiación que el analito absorbe o emite, relacionándolo directamente con su concentración [25].

Los métodos analíticos que comúnmente se aplican en la determinación de metales pesados, son la espectroscopia de absorción atómica (AAS) con atomización de la muestra por llama (FAAS), o atomización electrotérmica a través de un horno de grafito (GFAAS) o espectroscopía de fluorescencia atómica de vapor frío (CVAFS), la emisión atómica con plasma acoplado por inducción (ICP-AES) y la espectrometría de masas con plasma acoplado por inducción (ICP-MS).

En el caso de espectroscopía de absorción atómica con atomización por llama podemos hablar de una técnica simple, robusta, relativamente libre de interferencias y que permite alcanzar límites de detección próximos a las ppb. En el caso de ser necesario alcanzar límites de detección inferiores se puede realizar la atomización en horno de grafito (GFAAS), ya que esta técnica presenta ventajas como una mayor

selectividad y sensibilidad, aunque conlleva un mayor coste económico. La técnica de absorción atómica de vapor frío es un método de atomización aplicable solo a la determinación de mercurio porque es el único elemento metálico que tiene una presión de vapor considerable a temperatura ambiente. Varios estudios han demostrado la eficacia de la espectroscopía de absorción atómica en el análisis de trazas, aunque por lo general, es necesaria una etapa de preconcentración y/o separación de la matriz original previa al análisis con el fin de asegurar una eficaz detección [26].

En el caso de la utilización de la espectrometría de emisión atómica con plasma acoplado por inducción (ICP-AES), se alcanzan temperaturas muy elevadas, en torno a los 10.000 K lo que aporta a la técnica una mayor sensibilidad y selectividad, puesto que conlleva una atomización más completa con la presencia de menos impurezas. Una de las variaciones que se pueden llevar a cabo sobre la técnica de ICP es acoplar esta técnica a espectrometría de masas (ICP-MS). Debido a las mejoras que presentan estas técnicas se consiguen unos rangos lineales más amplios y unos límites de detección más bajos e incluso la posibilidad de llevar a cabo un análisis multicomponente. Sin embargo, son técnicas de elevado coste, ICP-MS representa un coste aun mayor que ICP-AES, y son técnicas de una gran complejidad instrumental por lo que además es necesario personal cualificado para su uso [13].

1.2.2.-Técnicas voltamperométricas

La voltamperometría es una técnica electroanalítica en la cual la información sobre el analito se deriva de la medición de la corriente originada por una reacción de transferencia de electrones en la superficie de un electrodo en función del potencial aplicado en condiciones que favorezcan la polarización del mismo (un electrodo polarizado es aquel al que se le ha aplicado un voltaje superior al pronosticado por la ecuación de Nernst para que ocurra oxidación o reducción). Para ello se aplica un programa de potencial en función del tiempo y se obtiene un gráfico de corriente-potencial. En general, los electrodos de trabajo en voltamperometría son relativamente pequeños con un área superficial de unos pocos milímetros cuadrados para favorecer la polarización.

Los componentes básicos empleados en la técnica constan de una celda electroquímica compuesta por tres electrodos y un electrolito no reactivo o electrolito soporte. Uno de los tres electrodos es el de trabajo, en el que ocurre la reacción de interés y que se comporta como transductor de la información química del sistema en señal eléctrica. El segundo electrodo es el de referencia, generalmente de calomel saturado o de plata-cloruro de plata (Ag/AgCl), cuyo potencial permanece constante durante el análisis y con respecto al cual se compara el potencial del electrodo de trabajo. El tercero es un contraelectrodo o electrodo auxiliar, que a menudo es una espiral de alambre de platino o grafito, que conduce la electricidad desde la fuente de la señal a través de la solución hasta el electrodo de trabajo. La resistencia eléctrica del

circuito de control que contiene el electrodo de referencia es tan grande que prácticamente no pasa corriente por él. Por tanto, toda la corriente de la fuente circula del contraelectrodo al electrodo de trabajo. El potenciostato es el dispositivo electrónico que controla el potencial aplicado al electrodo de trabajo con respecto al electrodo de referencia y además mide la corriente que circula en el electrodo de trabajo. Se trata de técnicas sencillas con instrumentación fácil de manejar e incluso de tamaño reducido lo que hace que se trate de una técnica apropiada para realizar medidas *in situ* y que mejore los costes frente a técnicas más complejas como las espectroscopías.

La primera técnica voltamperométrica utilizada fue la polarografía, desarrollada por el químico Jaroslav Heyrovsky a principios de los años veinte, quien años más tarde fue galardonado con el premio Nobel de Química por el descubrimiento y desarrollo de ésta técnica analítica. La polarografía difiere de otros tipos de voltametría en que el electrodo de trabajo es un electrodo de gota de mercurio y se trabaja en condiciones dinámicas, así mismo cuando el electrodo de trabajo es sólido o bien de mercurio pero la superficie de trabajo permanece constante a lo largo de todo el proceso redox, se conoce como voltamperometría [27].

Hasta hace unos años la polarografía era una importante herramienta que utilizaban los químicos para identificar iones inorgánicos y algunas especies orgánicas en soluciones acuosas. Sin embargo a finales de los años cincuenta, muchas de estas aplicaciones analíticas fueron sustituidas por diversos métodos espectroscópicos y la polarografía dejo de ser un método importante de análisis, excepto en algunas aplicaciones especiales, como la determinación de oxígeno molecular en solución. Entre las limitaciones más reseñables de la polarografía están la incapacidad de trabajar a potenciales positivos debido a la facilidad con que se oxida el mercurio y la formación de amalgamas entre el mercurio y el metal a determinar, lo que conlleva a un tratamiento más exhaustivo de los residuos generados, ya que presentan la importante toxicidad del mercurio. Además, los electrodos de gota de mercurio son mecánicamente inestables durante varias etapas del procedimiento de ensayo y presentan problemas relacionados con la alta toxicidad que genera el mercurio, su adecuado manejo y posterior eliminación [28].

A mediados de los años setenta se desarrollaron algunas modificaciones importantes de las técnicas voltamétricas clásicas que aumentaron de manera significativa la sensibilidad y selectividad del método. Aunque la importancia de la polarografía ha declinado, la voltametría y la amperometría en electrodos de trabajo diferentes al electrodo de gota de mercurio han aumentado a una gran velocidad. Sin embargo, la sensibilidad, fiabilidad, y competitividad en costos de estos electrodos todavía no han alcanzado el nivel de los electrodos a base de mercurio. En consecuencia, en los últimos años ha aumentado el interés por el desarrollo y la aplicación de nuevos electrodos sólidos, libres de mercurio, como los preparados con pasta de carbono modificados químicamente, sobre todo por su capacidad para

aumentar la sensibilidad y la selectividad de las diversas técnicas electroanalíticas [29-31].

La evolución que han llevado estos electrodos sólidos libres de mercurio ha sido inicialmente la incorporación de grupos funcionales al electrodo en forma de ligandos con el fin de aportar selectividad hacia determinados analitos, pero la problemática que presentan dichos electrodos es que al ponerlos en contacto con la matriz o con el electrolito soporte se produce la liberación del ligando desde el electrodo al medio, por lo que para evitar ese fenómeno se comenzó a usar soportes sólidos sobre los que se une el ligando de forma que se mantiene la selectividad y se aumenta la estabilidad del electrodo modificado. Con este fin se proponen las superficies silíceas como una alternativa interesante debido a la posibilidad de anclar fácilmente el ligando deseado sobre dicha superficie y una gran estabilidad química térmica y mecánica aportada por la estructura inorgánica. En las últimas dos décadas, con el descubrimiento de las sílices mesoporosas los avances en este campo han ido encaminados al empleo de sílices mesoporosas funcionalizadas con ligandos orgánicos como modificadores de distintos electrodos sólidos [29, 32].

1.2.2.1.-Voltamperometría de redisolución

La voltamperometría de redisolución incluye una etapa de adsorción previa al análisis puramente electroquímico, en dicha etapa el analito se adsorbe sobre el electrodo de trabajo, normalmente a partir de una disolución agitada y sin aplicar potencial (circuito abierto). Después de un tiempo perfectamente controlado se suspende la agitación y se lleva a cabo la medida electroquímica, en esta etapa el analito previamente adsorbido en el electrodo de trabajo se redisuelve, en función de cómo se comporte en la etapa de redisolución la voltamperometría será anódica o catódica.

Como resultado de la etapa de adsorción, los métodos de redisolución poseen los límites de detección más bajos de todos los procedimientos voltamétricos y son técnicas de gran importancia en el análisis de trazas, permitiendo la determinación de cantidades muy pequeñas de analito con una exactitud razonable. Estos límites de detección se pueden mejorar aumentando el tiempo de la etapa de adsorción, ya que la cantidad de analito depositado en el electrodo de trabajo es proporcional al tiempo de adsorción. Además, el hecho de llevar a cabo las dos etapas físicamente en medios distintos permite realizar una adsorción selectiva del analito en su matriz de forma que al pasar el electrodo a un medio limpio en el que se lleva a cabo la electrólisis y posterior redisolución reducimos las interferencias presentes. Por todo ello el rango de aplicación de esta técnica se ubica entre las ppm y fracciones de ppb mediante métodos rápidos, sencillos, en donde la preparación de la muestra es mínima y su sensibilidad y selectividad son excelentes para obtener resultados reproducibles [33].

1.3.-Sílices mesoporosas

Desde que los investigadores de la Mobil Research and Development Corporation sintetizaron y caracterizaron por primera vez la MCM-41 en 1992 [34, 35], una sílice mesoporosa con una distribución uniforme hexagonal de los poros, las sílices mesoporosas han tenido un gran auge desde el punto de vista de la investigación debido a sus buenas propiedades, desarrollándose distintas sílices mesoporosas como las SBA, FDU, HMS, MSU, FSM, etc.

Lo que hace que estos materiales sean interesantes es que se trata de estructuras porosas, de alta superficie específica, con una estrecha distribución de poro, con alta estabilidad térmica, química y mecánica, que pueden presentar una amplia variedad en el tamaño de poro, entre 20 y 100 Å normalmente (pudiéndose alcanzar poros con diámetro superior a 300 Å), presentan una superficie susceptible a funcionalizarse mediante una unión fuerte y estable entre el soporte inorgánico y el compuesto orgánico, y una vez funcionalizados los grupos funcionales se dispersan de forma homogénea por todo el sólido, lo que hace que la accesibilidad a estos grupos sea buena. Otra característica interesante es la gran versatilidad de procesos y reactivos que existen para obtener materiales silíceos mesoporosas híbridos, pudiéndose obtener mediante procesos sol-gel en una etapa o mediante un anclaje post-síntesis del grupo funcional [32].

Gracias a todas las propiedades que presentan las sílices mesoporosas, en los últimos años se han empleado para numerosos fines, con unos resultados muy interesantes. Los usos que se dan generalmente a estos materiales son:

- Tamices moleculares, incluso para separar proteínas gracias a su gran tamaño de poro.
- Soportes, con numerosas aplicaciones.
- Absorbentes.
- Fases estacionarias en cromatografía.
- Moldes para otros materiales.
- Catálisis.
- Adsorción.
- Almacenamiento de gases.
- Liberación controlada de fármacos.

1.3.1.-Empleo de sílices mesoporosas en la determinación de metales pesados

Desde que se dieron a conocer las sílices mesoporosas en 1992, su aplicación en la determinación de metales ha sido ampliamente investigada, desarrollándose para tal fin numerosos materiales híbridos gracias a la incorporación de diversos ligandos

orgánicos anclados a la superficie de las sílices que presentan selectividad hacia la adsorción de metales pesados debido a la presencia de átomos dadores de electrones como oxígeno, nitrógeno, azufre y fósforo.

Gracias a las características que presentan estos materiales híbridos basados en sílices mesoporosas funcionalizadas como una gran resistencia mecánica, selectividad en función del ligando orgánico anclado, una buena velocidad de adsorción de metales pesados y con una gran capacidad de adsorción, estabilidad química, fácil regeneración del material permitiendo tanto la reutilización del propio material como la recuperación de los metales, etc. este tipo de materiales se ha usado extensamente en la determinación de metales pesados [2].

El empleo de sílices mesoporosas en la determinación de metales pesados, generalmente llevado a cabo en muestras acuosas debido a su importancia medioambiental, se ha desarrollado generalmente en dos vías, una primera vía en la que estos materiales se han preparado para una etapa de preconcentración previa al análisis, generalmente espectroscópico, con el fin de reducir los niveles de detección de la técnica empleada y una segunda vía en la que se han modificado diversos electrodos con sílices mesoporosas funcionalizadas aplicándose posteriormente en la determinación electroquímica de metales pesados mediante medidas voltamperométricas de redisolución principalmente.

1.3.1.1.-Preconcentración de metales pesados

En el caso de los metales pesados, los límites admisibles que marca la ley para aguas de consumo son valores muy bajos de concentraciones, todos ellos por debajo de las ppm [10], por lo que en muchas ocasiones sucede que las técnicas de las que se dispone no consiguen medir las concentraciones del metal a determinar, o también puede suceder que dicho metal se encuentra en una matriz que presenta interferencias en la técnica de análisis que se va a emplear. En estos casos se puede solucionar el problema recurriendo a una etapa previa de extracción. Si lo que pretendemos es simplemente eliminar interferencias, esto se lleva a cabo realizando una extracción selectiva del analito y una posterior recuperación de este llevándolo a un volumen final igual al inicial. En el caso en que se requiere de una preconcentración, lo que se hace tras la extracción es una recuperación del analito con un volumen inferior al inicial [36].

En este contexto se puede hablar de la importancia que representan las sílices mesoporosas funcionalizadas como fases sólidas para la extracción de metales pesados de matrices acuosas, en este caso se conoce como *extracción en fase sólida*. El empleo de este tipo de sílices como fase extractora de metales como etapa de preconcentración previa a la cuantificación del analito mediante diversas técnicas analíticas se encuentra ampliamente documentado en la bibliografía con buenos resultados.

Algunos estudios desarrollados con sílices mesoporosas modificadas para la preconcentración o inmovilización de metales pesados son los realizados por Brown y col. [37, 38] empleando HMS 9.0 y MSU-2 funcionalizadas con 3-mercaptopropilo para distintos iones metálicos, Fryxell y col. [39] utilizando sílice mesoporosa modificada con grupos amino para la adsorción de cromo, Hossain y Mercier [40] con HMS funcionalizada con grupos etilendiamina para adsorber cobre, níquel y cinc, Yoshitake y col. [41] empleando mono-, di- y triaminosilanos para retener cromo y arsénico, Bois y col. [42] con MCM-41 y SBA-15 funcionalizadas con grupos mercapto y amino para adsorber cobre, níquel, cromo y cobalto, Sayari y col. [43] empleando diversas sílices mesoporosas funcionalizadas con grupos N,N-dimetildecilamina para la adsorción de mercurio, cromatos y arseniatos, Yang y col. [44] usando 3-aminopropilo en sílices mesoporosas para adsorber distintos iones metálicos, Jiang y col. [45] utilizando poliamidoamina y EDTA-poliamidoamina en SBA-15 para la adsorción de cobre, níquel, plomo y cinc, Bruzzoniti y col. [46] llevando a cabo estudios con SBA-15 modificada con grupos butíricos para la retención de plata, cadmio, hierro, cobre y cinc, Jamali y col. [47] usando tiofeno-2-carbaldehído anclado en sílice mesoporosa para preconcentrar paladio.

En los últimos años, nuestro grupo también ha llevado a cabo una amplia investigación en este campo, en la que cabe destacar los resultados obtenidos empleando el ligando mercaptobenzotiazol soportado en SBA-15 y MCM-41 para la adsorción de mercurio [48], mercaptotiazolina anclado en SBA-15 y MCM-41 también para adsorber mercurio [49], mercaptopirimidina soportada nuevamente sobre SBA-15 y MCM-41 para adsorber mercurio, cadmio y cromo [50-52], así como los obtenidos empleando el ligando orgánico 5-mercapto-1-metiltetrazol anclado en MCM-41, HMS, MSU-2 y SBA-15 para la adsorción de plomo y cinc [53, 58].

1.3.1.2.-Determinación electroquímica de metales pesados

El empleo en electrodos es la aplicación más directa que se puede dar a las sílices mesoporosas para la determinación de metales pesados. El uso de estos materiales en electroquímica se ha convertido en el centro de un gran interés en los últimos 15 años. Además de las numerosas características que presentan estos materiales, las cuales se han comentado anteriormente, la principal razón por la que los materiales híbridos basados en sílices mesoporosas resultan tan interesantes para la electroquímica es que se trata de sólidos robustos porosos con espacios abiertos distribuidos uniformemente en forma de una estructura tridimensional, que presenta un alto numero de grupos organofuncionales unidos mediante enlaces covalentes a las paredes de los mesoporos.

El alto grado de funcionalización resulta especialmente importante al aplicar una técnica voltamperométrica de redisolución, ya que potencia la adsorción de los analitos sobre la superficie del electrodo, produciéndose una preconcentración que mejora la sensibilidad de la medida, así como nos permite mejorar la selectividad de dicha etapa mediante la selección del grupo funcional, induciendo a un

reconocimiento preferente o específico del analito. Por otro lado, un diámetro de poro grande mejora el proceso de difusión del analito hacia los grupos funcionales del electrodo, paso que resulta limitante en muchas determinaciones, facilitando así la acumulación del analito.

La utilización de las sílices mesoporosas híbridas como parte del electrodo de trabajo para el análisis electroquímico de metales pesados se lleva a cabo fundamentalmente de dos formas, un primer método ampliamente descrito en bibliografía consistente en la incorporación de las sílices mesoporosas en matrices de pasta de carbono, mediante una dispersión física de la sílice junto a una mezcla de grafito y un aglutinante. Esto representa una vía conveniente de modificación de electrodos, sencilla, y que ofrece la ventaja de permitir el uso de materiales más sofisticados o más desarrollados, que requieren condiciones de síntesis más complejas o más agresivas. El segundo método consiste en la producción de una película del material mesoporoso híbrido sobre la superficie de un electrodo inerte mediante una síntesis por co-condensación a través de un proceso sol-gel, la problemática de este método viene de la necesidad de eliminar el reactivo plantilla usado para dar forma a los poros y de que no es posible trabajar en condiciones agresivas [32].

Debido a todas las posibilidades que nos ofrecen estos materiales y las ventajas que presentan como la no toxicidad, reusabilidad, etc. frente al clásico electrodo de mercurio, los estudios que existen actualmente son muy numerosos, algunos de ellos son los descritos por el grupo de Walcarius, que lleva desde el año 1998 desarrollando electrodos modificados con sílice mesoporosa (MCM-41 y SBA-15) pura y también funcionalizada con distintos ligandos orgánicos tales como grupos aminopropilo, mercaptopropilo, sulfónicos, etc. para el análisis principalmente de mercurio, plomo y cobre [59-68], el grupo de Yantasee también lleva varios años trabajando con electrodos modificados con sílice mesoporosa MCM-41 funcionalizada con distintos ligandos orgánicos (mercaptopropilo, grupos fosfónico, glicinilurea, salicilamida) auto-ensamblados en forma de monocapa sobre la sílice (SAMMS) para determinar especialmente mercurio, plomo, cadmio y cobre , incluso en muestras de agua [28, 29, 69-74]. El grupo de Cesarino trabaja desde hace unos años incorporando SBA-15 funcionalizada con el ligando 2-benzotiazoltiol a electrodos de pasta de carbono para analizar mercurio, plomo, cadmio y cobre en distintas muestras naturales [75-79]. El grupo de Javanbakht ha empleado electrodos de pasta de carbono modificados con sílices mesoporosas (SBA-15 y LUS-1) funcionalizadas con grupos tiourea para determinar plata y mercurio en muestras naturales [80, 81]. Más recientemente, los grupos de Xia y de Popa han empleado como sílice mesoporosa una HMS, los primeros funcionalizándola con quercetina para determinar plomo, cadmio y cobre [82] y los segundos con acetilacetona para determinar cadmio y cobre [83, 84].

Como se puede ver, el empleo de este tipo de materiales en la determinación electroquímica de metales pesados representa un importante campo en la

investigación actual, desarrollándose cada vez materiales más sofisticados, que aportan una mayor selectividad obteniéndose como resultado unos niveles de detección cada vez más bajos.

2.-OBJETIVOS

Los objetivos principales de la presente tesis han sido los siguientes:

La preparación y caracterización de un nuevo material híbrido, obtenido a partir de sílice mesoporosa tipo MSU-2 funcionalizada con el ligando orgánico 5-mercapto-1-metiltetrazol (MTTZ), con elevada capacidad para la adsorción selectiva de metales pesados en medios acuosos.

La preparación de diversos sensores electroquímicos basados en el empleo de sílices mesoporosas funcionalizadas con el ligando orgánico 5-mercapto-1-metiltetrazol

El desarrollo de métodos analíticos para la determinación de Pb(II) y Hg(II) en medios acuosos mediante la técnica de voltamperometría de redisolución anódica, utilizando los sensores previamente preparados y optimizando de las variables que afectan a la medida (etapas de preconcentración, electrólisis y redisolución).

La validación de los métodos desarrollados y su aplicación a la determinación de Pb(II) y Hg(II) en distintas muestras de agua contaminadas (agua corriente, agua de río y agua subterránea).

3.-METODOLOGÍA

Todos los materiales y métodos se encuentran detallados, con sus respectivas referencias bibliográficas, en cada uno de los capítulos. A continuación, se muestra un resumen de la metodología empleada en la realización de la tesis.

3.1.-Preparación de disoluciones patrón y electrolitos

Todas las disoluciones se prepararon a diario con agua ultrapura obtenida mediante el sistema Millipore Milli-Q (Waters, EE. UU.). Todo el material de vidrio y polietileno utilizado en la preparación de disoluciones, el tratamiento y el almacenamiento de muestras, se lavó con una disolución acuosa de ácido nítrico (50%), enjuagándose bien posteriormente con agua ultrapura.

Las disoluciones patrón de Pb(II) y Hg(II) se prepararon a partir de disoluciones patrón con calidad ICP con una concentración de 1,000 g/L ± 0,5% y ± 0,2% respectivamente, diluyéndose directamente con agua ultrapura.

3.2.-Muestras de aguas

Las muestras de aguas empleadas en los distintos análisis llevados a cabo en el presente trabajo han sido: agua corriente del grifo recogida en el laboratorio (Canal de Isabel II, Móstoles), agua de río, recogida del río Alberche (Toledo) y agua subterránea recogida en un pozo en Escalona (Toledo).

Las muestras se filtraron a vacío con una membrana de 0,45 μm (Millipore) para eliminar las partículas en suspensión y posteriormente se almacenaron a -26 ºC en botellas de polietileno hasta su utilización. No se realizó ningún tratamiento más a las muestras.

Para determinar los metales pesados en dichas muestras (Pb(II) y Hg(II)) se trabajó mediante la técnica de adición estándar para compensar los efectos de matriz, para lo que además se doparon las muestras con cantidades conocidas de los metales. En el caso de las medidas con plomo, las muestras de aguas se doparon hasta una concentración de 10 ppb. Por el contrario, para el mercurio las muestras se doparon hasta una concentración de 30 ppb.

3.3.-Procedimiento experimental

El análisis mediante voltamperometría de redisolución anódica adsortiva (Ad-ASV) consiste, como se ha indicado anteriormente, en una etapa de preconcentración, en la que se acumula físicamente el catión metálico en la superficie del electrodo, seguida de una etapa de electrólisis, o proceso de electrodeposición, y una etapa de redisolución anódica que corresponde al paso de medida o cuantificación. Antes de iniciar una nueva medida es necesario realizar un último paso de regeneración del electrodo que se realiza de manera similar a la etapa de medida.

Durante el paso de preconcentración, el electrodo se sumerge en una disolución del catión metálico a analizar y se mantiene en agitación constante durante un tiempo determinado. Tras esta etapa, el electrodo se extrae de la disolución y se procede a su lavado con agua ultrapura. Seguidamente se introduce en la celda electroquímica, junto con el electrodo de referencia y el auxiliar, que contiene el medio de electrolisis. A continuación, se aplica un potencial negativo, suficiente para la reducción del metal (-1,2 V para Pb(II) y -0,8 ó -0,4 V para Hg(II)) sobre el electrodo de trabajo que se mantiene durante un tiempo determinado, llamado tiempo de electrólisis. El potencial aplicado provoca que los cationes metálicos adsorbidos en el electrodo de trabajo se reduzcan a metal en estado elemental. Por último, se realiza la medida en la misma celda mediante una variación de potencial hacia valores positivos, hasta 0,4 V para el plomo o 0,8 V para el mercurio, produciéndose la oxidación del metal. Por lo tanto, el metal depositado en el electrodo se redisolverá en el medio acuoso volviendo a su estado inicial.

La representación gráfica de la intensidad de la corriente generada frente al potencial aplicado recibe el nombre de voltamperograma (Figura 4) donde se observa un pico correspondiente al analito determinado cuya área es directamente proporcional a la concentración.

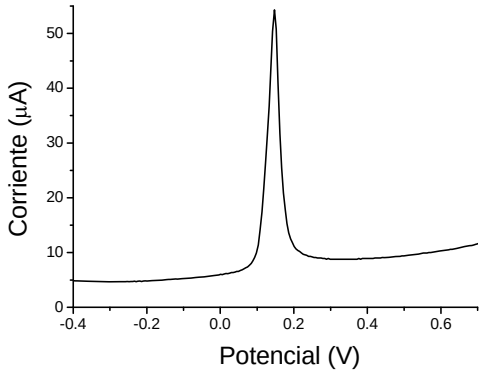


Figura 4. Voltamperograma.

3.4.-Optimización del procedimiento experimental

3.4.1.-Composición del electrodo

En el caso de los electrodos de pasta de carbono modificados con sílice mesoporosa funcionalizada, se optimiza la mezcla de los tres componentes que lo forman, la pasta de carbono, sílice mesoporosa modificada con el ligando MTTZ y el aceite mineral. Una vez bien homogenizada la mezcla de pasta de carbono y sílice mesoporosa se añade el aceite y se deja en agitación hasta conseguir una mezcla completamente homogénea. De estas mezclas con distinta composición se evalúa la compactación en el soporte del electrodo, la estabilidad al sumergirla en las disoluciones con agitación y la señal que dan frente a una determinada concentración de analito.

En el caso de las películas de sílice, lo que se varía es la composición del sol inicial en cuanto a la relación entre el ligando MTTZ y la fuente de silicio empleada (tetraetilortosilicato), de forma que se compara para cada caso la señal obtenida frente a una determinada concentración de analito.

3.4.2.-Parámetros operativos

Se ha estudiado el efecto de las distintas variables que afectan a las etapas de la medida sobre la señal obtenida en el análisis.

3.4.2.1.-Factores que afectan a la etapa de preconcentración

Tiempo de preconcentración

El tiempo de preconcentración incide directamente en la cantidad de metal en estado catiónico que se adsorbe en la superficie del electrodo, por lo que es una variable importante a optimizar. La optimización del tiempo de preconcentración se lleva a cabo en condiciones de circuito abierto (sin aplicar potencial) y en agitación constante de la disolución de metal, sumergiendo el electrodo en dicha disolución durante periodos perfectamente medidos.

pH de la disolución acuosa de metal

La formación del complejo entre el catión metálico en disolución y el ligando MTTZ se ve influenciada directamente por el pH de la disolución del medio. El objetivo de este ensayo es determinar que pH facilita la unión entre el metal oxidado y el ligando, mejorando así la señal voltamperométrica. Para realizar este experimento se utilizaron disoluciones acuosas de metal ajustando el pH con hidróxido sódico y ácido nítrico, manteniendo como tiempo de preconcentración de 5 minutos.

En el caso de las muestras de aguas no se lleva a cabo el ajuste de pH debido a que al tratarse de matrices complejas, pueden aparecer fenómenos de precipitación de diversos complejos o interacciones entre el ácido o la base y los compuestos presentes en la matriz, los cuales nos den problemas en la determinación electroquímica del metal.

3.4.2.2.-Factores que afectan a la etapa de detección

Medio de electrólisis y medida

Debido al proceso de oxidación que genera la redisolución del metal, el medio de medida más adecuado es un medio ácido, ya que los protones del medio debilitan la interacción del complejo metal-ligando, aunque medios fuertemente ácidos pueden provocar la formación de burbujas de hidrógeno en la superficie del electrodo durante la etapa de electrólisis debido a la reducción de los protones. Por otro lado, un electrolito adecuado es aquel que una vez que los iones metálicos se redisuelven, este evita que se forme de nuevo el complejo metal-ligando, por lo que interesan medios que formen compuestos poco solubles con el metal de forma que el metal pasa a no estar disponible para adsorberse nuevamente en la superficie del electrodo.

Con el fin de estudiar el medio más adecuado para llevar a cabo las etapas de electrolisis y medida se realizaron ensayos utilizando distintas disoluciones de electrolito para tal fin.

Tiempo de electrólisis

El tiempo de electrolisis es el tiempo en el que se aplica un potencial determinado sobre el electrodo de forma que se produzca la reducción del metal adsorbido sobre la superficie del electrodo en forma catiónica a metal en estado elemental. En general, un periodo de electrolisis grande conduce a una mayor cantidad de analito depositado en su forma elemental. Sin embargo, tiempos demasiado largos pueden ocasionar fenómenos perjudiciales como por ejemplo, la formación de burbujas de hidrógeno en la superficie del electrodo. Con el fin de permitir un adecuado proceso de electrodeposición, se probaron varios tiempos de electrólisis.

3.4.3.-Evaluación del electrodo

Una vez optimizados los parámetros operativos se lleva a cabo la evaluación del electrodo preparado, para lo cual se preparan curvas de calibrado con el fin de comprobar el rango lineal de concentraciones en el que nos permite medir, con los datos de linealidad se obtienen los valores de límite de detección y límite de cuantificación, utilizando el criterio de 3 y 10 veces el valor del ruido de fondo respectivamente. Además, se lleva a cabo la medida de reproducibilidad para un único electrodo tanto como para un solo día como para varios y la reproducibilidad interelectrodo.

3.4.4.-Aplicación del método

Optimizados los parámetros operativos y comprobados los límites de cuantificación y de detección y la reproducibilidad del método desarrollado, se llevó a cabo la aplicación de este a diversas muestras de agua (agua del grifo, agua de río y agua subterránea) para el análisis de Pb(II) o Hg(II). Dicho análisis se realizó mediante el método de adición estándar con el fin de compensar los efectos de matriz.

4.-CONCLUSIONES

Como principal conclusión de la realización de esta tesis cabe destacar la posibilidad de preparar materiales mesoporosos híbridos con base silícea y su incorporación a electrodos, haciendo posible la determinación de metales pesados en aguas mediante la técnica de voltamperometría de redisolución. Los métodos desarrollados presentan unos límites de detección lo suficientemente bajos como para realizar medidas en aguas de manera rápida, sencilla, con un bajo coste y con la posibilidad de realizar medidas de campo.

A continuación, se recogen las conclusiones más relevantes por capítulos:

Capítulo II

Se han preparado y caracterizado con éxito sílices mesoporosas con estructura tipo MSU-2. Los materiales preparados presentan distintas propiedades texturales en función de las condiciones de síntesis utilizadas, principalmente la relación fuente de silicio/surfactante.

Se ha conseguido funcionalizar con éxito las sílices preparadas con el ligando orgánico 5-mercapto-1-metiltetrazol (MTTZ) para obtener un material capaz de adsorber Pb(II) y Zn(II) de medios acuosos, con una buena capacidad de adsorción y cierta selectividad frente a otros metales como Cd(II), Cu(II) y Cr(VI).

Las propiedades texturales de la sílice mesoporosa híbrida preparada suponen una mejora en cuanto a la eficiencia de adsorción de Pb(II) y Zn(II) frente a otras sílices empleadas anteriormente, tipo HMS y MCM-41, lo que indica que estas características del soporte influyen drásticamente en el proceso de adsorción de los metales pesados.

Capítulo III

Las sílices mesoporosas funcionalizadas se muestran como una importante alternativa a la preparación de electrodos de pasta de carbono modificados para la determinación de metales pesados mediante la técnica voltamperométrica de redisolución anódica adsortiva. En este sentido, ha sido posible determinar la concentración de Pb(II) en disolución acuosa utilizando un electrodo de trabajo de pasta de carbono modificada con sílice mesoporosa híbrida (HMS-MTTZ).

Las condiciones óptimas de trabajo obtenidas han sido 5 minutos de tiempo de preconcentración, pH de la muestra igual a 6,5, y 120 s de tiempo de electrólisis en HCl 0,2 M. En estas condiciones el rango lineal obtenido ha sido para concentraciones

de Pb(II) de 1 a 100 ppb. El método desarrollado permite realizar análisis multicomponente, al menos para Pb(II), Cd(II) y Cu(II), sin que se observen interferencias entre estos metales.

Varias ventajas, como un límite de detección bajo (0,3 ppb con 5 minutos de tiempo de preconcentración) y una buena reproducibilidad hacen que este electrodo sea una buena opción para la determinación de Pb(II) en muestras de aguas, pudiéndose trabajar con concentraciones inferiores a las más restrictivas que propone la ley española (10 ppb a partir de enero de 2014 para aguas de consumo). El método desarrollado presenta además mejores límites de detección y reproducibilidad frente a otros desarrollados previamente. También es posible aplicar el método desarrollado a muestras de agua de grifo y agua subterránea con unos buenos resultado.

Capítulo IV

Se ha demostrado que la modificación de un electrodo serigrafiado de pasta de carbono con la sílice mesoporosa funcionalizada MTTZ-MSU-2 permite la determinación de Pb(II) en medios acuosos, utilizando la técnica voltamperometrica de redisolución anódica adsortiva.

Las condiciones óptimas de trabajo obtenidas para este electrodo han sido 5 minutos de tiempo de preconcentración y 120 s de tiempo de electrólisis en HCl 0,2 M. En estas condiciones el rango lineal obtenido ha sido para concentraciones de Pb(II) de 1 a 30 ppb. Además, queda probada la posibilidad de realizar un análisis multielemental con el Pb(II), Cu (II) y Cd (II).

La sensibilidad del método de redisolución anódica adsortiva (límite de detección de 0,1 ppb) es una de las principales ventajas que presenta, así como la posibilidad de realizar medidas de campo. Además, se ha aplicado el método desarrollado a la determinación de Pb(II) en muestras de agua de grifo, agua de río y agua subterránea con buenos resultados en todos los casos.

Capítulo V

El electrodo de pasta de carbono modificado con la sílice mesoporosa funcionalizada MTTZ-MSU-2 nos ha permitido también llevar a cabo la determinación de Hg(II) en medios acuosos, utilizando la técnica voltamperometrica de redisolución anódica adsortiva.

Se han establecido las condiciones óptimas del método analítico para la determinación de Hg(II), correspondientes a tiempo de preconcentración (5 minutos), medio de electrólisis y medida (HCl 0,1 M) y tiempo de deposición o de electrólisis (60 s) obteniendo una buena sensibilidad en las medidas. También se ha comprobado la

posibilidad de realizar la detección simultánea multielemental de iones metálicos como Cd(II), Pb(II) y Hg(II) sin interferencia alguna

El electrodo desarrollado presenta condiciones favorables en cuanto a estabilidad, precisión y reproducibilidad en las medidas, tanto para superficies únicas como para superficies múltiples con mínima degradación del mismo. El rango lineal obtenido (20 – 200 ppb) y el bajo limite de detección de 4,2 ppb indican que la técnica se considera viable al análisis de Hg(II) a nivel de trazas. Igualmente se ha comprobado la aplicación del método en muestras de agua de grifo y de río con resultados satisfactorios.

Capítulo VI

Ha sido posible preparar una película de sílice mesoporosa con el ligando MTTZ inmovilizado en la superficie mediante un proceso sol-gel en una sola etapa. Esto representa la innovación de introducir un ligando más complejo que el grupo aminopropilo o mercaptopropilo, empleados habitualmente como grupos funcionales en las películas de sílice mesoporosas.

Se ha logrado depositar la película mesoporosa híbrida formando un recubrimiento homogéneo sobre un electrodo de carbono vitrificado, presentando una buena porosidad y accesibilidad dicha película después de extraer el surfactante, como se ha demostrado con los ensayos realizados.

Se ha comprobado la posibilidad de utilizar el electrodo preparado en la determinación de Hg(II) en medios acuosos y se han establecido las condiciones óptimas del método analítico y la preparación del electrodo, correspondientes a la cantidad de ligando presente en el sol inicial (10%), tiempo de preconcentración (5 minutos), medio de electrólisis y medida (HCl 0,1 M) y tiempo de deposición o de electrólisis (60 s) obteniendo una buena sensibilidad en las medidas.

En las condiciones óptimas el rango lineal obtenido ha sido de 10 a 200 ppb, obteniéndose un límite de detección de 4,8 ppb para un tiempo de preconcentración de 5 minutos y de 0,4 ppb para un tiempo de preconcentración de 20 minutos.

5.-BIBLIOGRAFÍA

[1] Orozco Barrenetxea C., Pérez Serrano A., González Delgado M. N., Rodríguez Delgado F. J., Alfayate Blanco J. M.; **Contaminación ambiental: una visión desde la química.** *Ed. Thomson, Madrid, 2003.*

[2] Pérez-Quintanilla D., Sierra I., Fajardo M.; **Preparación de materiales adsorbentes de Hg(II) con fines analíticos y aplicaciones medioambientales.** *Ed. Dykinson, Madrid, 2006.*

[3] Ramos Castellanos P., en: Ramos Castellanos P., y col.; **Uso eficiente y sostenible de los recursos naturales.** *Ed. Universidad Salamanca, Salamanca, 2007.*

[4] Domènech X., Peral J.; **Química ambiental de sistemas terrestres.** *Ed. Reverté, Barcelona, 2006.*

[5] Moreno Grau M. D.; **Toxicología ambiental. Evaluación de riesgo para la salud humana.** *McGraw Hill, Madrid, 2003.*

[6] Baird C.; **Química ambiental.** *Editorial Reverté, Barcelona, 2001.*

[7] Manahan S. E.; **Introducción a la química ambiental.** *Ed. Reverté, Mexico D. F., 2007.*

[8] Mendoza Roca J. A., Montañes Sanjuan M. T., Palomares Gimeno A. E.; **Ciencica y tecnología del medio ambiente.** *Servicio de publicaciones de la Universidad Politécnica de Valencia, Valencia, 1998.*

[9] Storey M. V., van der Gaag B., BurnsB. P.; **Advances in on-line drinking water quality monitoring and early warning systems.** *Water Res. 45 (2011) 741–747.*

[10] **REAL DECRETO 140/2003, de 7 de febrero por el que se establecen los criterios sanitarios de la calidad del agua de consumo humano.** *BOE nº 45 (2003) 7228-7245.*

[11] Bradl H. B. (ed.); **Heavy metals in the environment: origin, interaction and remedation.** *Interface Science and Technology Vol. 6. Elsevier academic press, Reino Unido, 2005.*

[12] Guitart R., Thomas V. G.; **¿Es el plomo empleado en deportes (caza, tiro y pesca deportiva) un problema de salud pública infravalorado?** *Rev. Esp. Salud Pública 79 (2005) 621-632.*

[13] Naseri M. T., Hosseini M. R. M., Assadi Y., Kiani A.; **Rapid determination of lead in water samples by dispersive liquid-liquid microextraction coupled with electrothermal atomic absortion spectrometry.** *Talanta 75 (2007) 56-62.*

[14] Plan de Acción Regional de América del Norte (P.A.R.A.N.); **Expediente de nominación sobre el plomo.** *Presentado por Estados Unidos al Grupo de Trabajo sobre el Manejo Adecuado de las Sustancias Químicas, 6 de abril de 1998.*

[15] Anderson R. V.; **The effects of lead on oxygen uptake in the crayfish, Orconectes virilis (Hagen).** *Bull. Environ. Contam. Toxicol. 20 (1978) 394-400.*

[16] Yang C.-C., Kumar A. S., Zen J.-M.; **Precise blood lead analysis using a combined internal standard and standard addition approach with disposable screen-printed electrodes.** *Anal. Biochem. 338 (2005) 278–283.*

[17] Senkal B. F., Ince M., Yavuz R., Yaman M.; **The synthesis of new polymeric sorbent and its application in preconcentration of cadmium and lead in water samples.** *Talanta 72 (2007) 962-967.*

[18] Gil M., Torres A., Harvey M., Esteves J. L.; **Metales pesados en organismos marinos de la zona costera de la Patagonia argentina continental.** *Rev. Biol. Mar. Oceanogr. 41 (2006) 167-176.*

[19] Díaz O., Encina F., Recabarren E., del Valle S., Pastene S., Montes S., Figueroa A.; **Estudio de la concentración de arsénico, mercurio, plomo y fenantreno en la macha (*Mesodesma donacium*). Implicaciones alimentarias y toxicológicas.** *Rev. Chil. Nutr. 35 (2008) 53-60.*

[20] **WHO: Guidelines for drinking water quality.** *2nd Edn., Addendum to vol. 2 (1998) Health Criteria and Other Supporting Information, WHO/EOS/98.1, Geneva, p. 283.*

[21] **WHO: Guidelines for drinking water-quality.** *3rd Edn (2003).*

[22] Cotton F. A., Wilkinson G., Murillo C., Bochman M.; **Advanced Inorganic Chemistry.** *6ª Edición. Ed. John Wiley & Sons., New York, 1999.*

[23] Programa de las Naciones Unidas para el Medio Ambiente (U.N.E.P). **Evaluación mundial sobre el mercurio.** *Programa interorganismos para la gestión racional de las sustancias químicas. Suiza, 2002.*

[24] Departamento de Salud y Servicios Humanos (A.T.S.D.R). Agencia para Sustancias Tóxicas y el Registro de Enfermedades. **Servicio de salud pública, reseña toxicológica del mercurio.** *Atlanta G.A, 1999.*

[25] González de Buitrago J. M.; **Técnicas y métodos de laboratorio clínico.** *2ª Edición. Ed. Masson, Barcelona, 2005.*

[26] Ghaedi M., Ahmadi F., Shokrollahi A.; **Simultaneous preconcentration and determination of copper, nickel, cobalt and lead ions content by flame atomic absortion spectrometry.** *J. Hazard. Mater. 142 (2006) 272-278.*

[27] Skoog D. A., West D. M., Holler F. J., Crouch S. R.; **Fundamentos de química analítica.** *8ª Edición. Ed. Thomson, Madrid, 2005.*

[28] Yantasee W., Lin Y., Fryxell G. E., Busche B. J.; **Simultaneous detection of cadmium, copper and lead using a carbon paste electrode modified with carbamoylphosphonic acid self-assembled monolayer on mesoporous silica (SAMMS).** *Anal. Chim. Acta 502 (2004) 207-212.*

[29] Yantasee W., Deibler L. A., Fryxell G. E., Timchalk C., Lin Y.; **Screen-printed electrodes modified with functionalized mesoporous silica for voltammetric analysis of toxic metal ions.** *Electrochem. Commun. 7 (2005) 1170-1176.*

[30] Lu J., He X., Zeng X., Wan Q., Zhang Z.; **Voltammetric determination of mercury (II) in aqueous media using glassy carbon electrodes modified with novel calyx(4)arene.** *Talanta 59 (2003) 553-560.*

[31] Berchmans S., Arivukkodi S., Yegnaraman V.; **Self-assembled monolayers of 2-mercaptobenzimidazole on gold: Stripping voltammetric determination of Hg(II).** *Electrochem. Commun. 2 (2000) 226-229.*

[32] Walcarius A.; **Impact of mesoporous silica-based materials on electrochemistry and feedback from electrochemical science to the characterization of these ordered materials.** *C. R. Chim. 8 (2005) 693-712.*

[33] Kounaves S.; **Voltammetric techniques.** en Settle F. A.; **Handbook of instrumental techniques for analytical chemistry.** *Ed. Prentice Hall PTR, Upper Saddle River, New Jersey, 1997.*

[34] Kresge C. T., Leonowicz M. E., Roth W. J., Vartuli J. C., Beck J. S.; **Ordered mesoporous molecular sieves synthesized by a liquid-crystal template mechanism.** *Nature 359 (1992) 710-712.*

[35] Beck J. S., Vartulli J. C., Roth W. J., Leonowicz M. E., Kresge C. T., Schmitt K. D., Chu C. T. W., Olson D. H., Sheppard E. W., McCullen S. B., Higgins J. B., Schlenker J. L.; **A new family of mesoporous molecular sieves prepared with liquid crystal templates.** *J. Am. Chem. Soc. 114 (1992) 10834-10843.*

[36] Sogorb Sánchez M. A., Vilanova Gisbert E.; **Técnicas analíticas de contaminantes químicos. Aplicaciones toxicológicas, medioambientales y alimentarias.** *Ed. Díaz de Santos, Madrid, 2004.*

[37] Brown J., Mercier L., Pinnavaia T. J.; **Selective adsorption of Hg^{2+} by thiol-functionalized nanoporous silica.** *Chem. Commun. (1999) 69-70.*

[38] Brown J., Richer R., Mercier L.; **One-step synthesis of high capacity mesoporous Hg^{2+} adsorbents by non-ionic surfactant assembly.** *Microporous Mesoporous Mat. 37 (2000) 41-48.*

[39] Fryxell G. E., Lui J., Hauser T. A., Nie Z., Ferris K. F., Mattigod S., Gong M., Hallen R. T.; **Design and sythesis of selective mesoporous anions traps.** *Chem. Mater. 11 (1999) 2148-2154.*

[40] Hossain K. Z., Mercier L.; **Intraframework metal ion adsorption in ligand-functionalized mesoporous silica.** *Adv. Mater. 14 (2002) 1053–1056.*

[41] Yoshitake H., Yokoi T., Tatsumi T.; **Adsorption behavoir of arsenate at transition metal cations captured by amino-functionalized mesoporous silicas.** *Chem. Mater. 15 (2003) 1713-1721.*

[42] Bois L., Bonhommé A., Ribes A., Pais B., Raffin G., Tessier F.; **Functionalized silica for heavy metal ions adsorption.** *Colloid Surf. A- Physicochem. Eng. Asp. 221 (2003) 221-230.*

[43] Sayari A., Hamoudi S., Yang Y.; **Applications of pore-expanded mesoporous silica. 1. Removal of heavy metals cations and organic pollutants from wastewater.** *Chem. Mater. 17 (2005) 212-216.*

[44] Yang H., Xu R., Xue X., Li F., Li G.; **Hybrid surfactant-templated mesoporous silica formed in ethanol an its application for heavy metal removal.** *J. Hazard. Mater. 152 (2007) 690-698.*

[45] Jiang Y., Gao Q., Yu H., Chen Y., Deng F.; **Intensively competitive adsorption for heavy metal ions by PAMAM-SBA-15 and EDTA-PAMAM-SBA-15 inorganic-organic hybrid materials.** *Microporous Mesoporous Mat. 103 (2007) 316-324.*

[46] Bruzzoniti M. C., Prelle A., Sarzanini C., Onida B., Fiorilli S., Garrone E.; **Retention of heavy metal ions on SBA-15 mesoporous silica functionalized with carboxylic groups.** *J. Sep. Sci. 30 (2007) 2412-2420.*

[47] Jamali M. R., Assadi Y., Shemirani F., Salavati-Niasari M.; **Application of thiophene-2-carbaldehyde-modified mesoporous silica as a new sorbent for separation and preconcentration of palladium prior to inductively coupled**

plasma atomic emission spectrometric determination. *Talanta 71 (2007) 1524–1529.*

[48] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Preparation of 2-mercaptobenzothiazole-devivatized mesoporous silica and removal of Hg(II) from aqueous solution.** *J. Environ. Monit. 8 (2006) 214-222.*

[49] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **2-Mercaptothiazoline modified mesoporous silica for mercury removal from aqueous media.** *J. Hazard. Mater. B 134 (2006) 245-256.*

[50] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Mesoporous silica functionalized with 2-mercaptopyridine: Synthesis, characterization and employment for Hg(II) adsorption.** *Microporous Mesoporous Mat. 89 (2006) 58-68.*

[51] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Adsorption of cadmium(II) from aqueous media onto a mesoporous silica chemically modified with 2-mercaptopyridine.** *J. Mater. Chem. 16 (2006) 1757-1764.*

[52] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Cr(VI) adsorption of functionalized amorphous and mesoporous silica from aqueous and non-aqueous media.** *Mater. Res. Bull. 42 (2007) 1518-1530.*

[53] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **Functionalized HMS mesoporous silica as solid phase extractant for Pb(II) prior to its determination by flame atomic absorption spectrometry.** *J. Sep. Sci. 30 (2007) 1556-1567.*

[54] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **Preparation, characterization, and Zn^{2+} adsorption behavior of chemically modified MCM-41 with 5-mercapto-1-methyltetrazole.** *J. Colloid Interface Sci. 313 (2007) 551-562.*

[55] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **Preconcentration of Zn(II) in water samples using a new hybrid SBA-15-based material.** *J. Hazard. Mater. 166 (2009) 1449-1458.*

[56] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **Solid phase extraction of Pb(II) in water samples using a new hybrid inorganic-organic mesoporous silica prior to its determination by FAAS.** *Microchim. Acta 165 (2009) 291-298.*

[57] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **Synthesis and characterization of novel mesoporous silicas of the MSU-*X* family for environmental applications.** *J. Nanosci. Nanotechnol. 9 (2009) 4901-4909.*

[58] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **New hybrid materials as Zn(II) sorbents in water samples.** *Mater. Res. Bull. 45 (2010) 1177-1181.*

[59] Walcarius A., Despas C., Trens P., Hudson M. J., Bessière J.; **Voltammetric in situ investigation of an MCM-41-modified carbon paste electrode – a new sensor.** *J. Electroanal. Chem. 453 (1998) 249-252.*

[60] Walcarius A., Bessière J.; **Electrochemistry with mesoporous silica: Selective mercury(II) binding.** *Chem. Mater. 11 (1999) 3009-3011.*

[61] Walcarius A., Lüthi N., Blin J.-L., Su B.-L., Lamberts L.; **Electrochemical evaluation of polysiloxane-immobilized amine ligands for the accumulation of copper(II) species.** *Electrochim. Acta 44 (1999) 4601-4610.*

[62] Walcarius A., Devoy J., Bessière J.; **Silica-modified electrode for the selective detection of mercury.** *J. Solid State Electrochem. 4 (2000) 330-336.*

[63] Walcarius A., Etienne M., Sayen S., Lebeau B.; **Grafted silicas in electroanalysis: Amorphous versus ordered mesoporous materials.** *Electroanalysis 15 (2003) 414-421.*

[64] Walcarius A., Sayen S., Gérardin C., Hamdoune F., Rodehüser L.; **Dipeptide-functionalized mesoporous silica spheres.** *Colloid Surf. A-Physicochem. Eng. Asp. 234 (2004) 145-151.*

[65] Ganesan V., Walcarius A.; **Surfactant templated sulfonic acid functionalized silica microspheres as new efficient ion exchangers and electrode modifiers.** *Langmuir 20 (2004) 3632-3640.*

[66] Ganesan V., Walcarius A.; **Ion exchange and ion exchange voltammetry with functionalized mesoporous silica materials.** *Mater. Sci. Eng. B-Adv. Funct. Solid-State Mater. 149 (2008) 123-132.*

[67] Goubert-Renaudin S., Etienne M., Rousselin Y., Denat F., Lebeau B., Walcarius A.; **Cyclam-functionalized silica-modified electrodes for selective determination of Cu(II).** *Electroanalysis 21 (2009) 280-289.*

[68] Goubert-Renaudin S., Moreau M., Despas C., Meyer M., Denat F., Lebeau B., Walcarius A.; **Voltammetric detection of lead(II) using amide-cyclam-functionalized silica-modified carbon paste electrodes.** *Electroanalysis 21 (2009) 1731-1742.*

[69] Yantasee W., Lin Y., Zemanian T. S., Fryxell G. E.; **Voltammetric detection of lead(II) and mercury(II) with thiol self-assembled monolayer on mesoporous silica (SAMMS).** *Analyst 128 (2003) 468-472.*

[70] Yantasee W., Lin Y., Fryxell G. E., Wang Z.; **Carbon paste electrode modified with carbamoylphosphosnic acid functionalized silica: A new mercury-free sensor for uranium detection.** *Electroanalysis 16 (2004) 870-873.*

[71] Yantasee W., Timchalk C., Fryxell G. E., Dockendorff P., Lin Y.; **Automated portable analyzer for lead(II) based on sequential flow injection and nanostructured electrochemical sensors.** *Talanta 68 (2005) 256-261.*

[72] Yantasee W., Fryxell G. E., Conner M. M., Lin Y.; **Nanostructured electrochemical sensors based on functionalized nanoporous silica for**

voltammetric analysis of lead, mercury, and copper. *J. Nanosci. Nanotechnol. 5 (2005) 1537-1540.*

[73] Yantasee W., Fryxell G. E., Lin Y.; **Voltammetric analysis of europium at screen-printed electrodes modified with salicylamide self-assembled on mesoporous silica.** *Analyst 131 (2006) 1342-1346.*

[74] Yantasee W., Charnhattakorn B., Fryxell G. E., Lin Y., Timchalk C., Addleman R. S.; **Detection of Cd, Pb, and Cu in non-pretreated natural waters and urine with thiol functionalized mesoporous silica and Nafion composite electrodes.** *Anal. Chim. Acta 620 (2008) 55-63.*

[75] Cesarino I., Marino G., Matos J. R., Cavalheiro E. T. G.; **Evaluation of a carbon paste electrode modified with organofunctionalised SBA-15 silica in the determination of copper.** *Eclét. Quím. 32 (2007) 29-34.*

[76] Cesarino I., Marino G., Matos J. R., Cavalheiro E. T. G.; **Using the organofunctionalised SBA-15 nanostructured silica as a carbon paste electrode modifier: Determination of cadmium ions by differential anodic pulse stripping voltammetry.** *J. Braz. Chem. Soc. 18 (2007) 810-817.*

[77] Cesarino I., Marino G., Matos J. R., Cavalheiro E. T. G.; **Evaluation of a carbon paste electrode modified with organofunctionalised SBA-15 nanostructured silica in the simultaneous determination of divalent lead, copper and mercury ions.** *Talanta 75 (2008) 15-21.*

[78] Cesarino I., Cavalheiro E. T. G., Brett C. M. A.; **Characterization of graphite-polyurethane composite electrodes modified with organofunctionalised SBA-15 nanostructured silica in the presence of heavy metal ions. Application to anodic stripping voltammetry.** *Microchim. Acta 171 (2010) 1-9.*

[79] Cesarino I., Cavalheiro E. T. G., Brett C. M. A.; **Simultaneous determination of cadmium, lead, copper and mercury ions using organofunctionalised SBA-**

15 nanostructured silica modified graphite-polyurethane composite electrode. *Electroanalysis 22 (2010) 61-68.*

[80] Javanbakht M., Divsar F., Badiei A., Fatollahi F., Khaniani Y., Ganjali M. R., Norouzi P., Chaloosi M., Ziarani G. M.; **Determination of picomolar silver concentrations by differential pulse anodic stripping voltammetry at a carbon paste electrode modified with phenylthiourea-functionalized high ordered nanoporous silica gel.** *Electrochim. Acta 54 (2009) 5381-5386.*

[81] Javanbakht M., Divsar F., Badiei A., Ganjali M. R., Norouzi P., Ziarani G. M., Chaloosi M., Jahangir A. A.; **Potenciometric detection of mercury(II) ions using a carbon paste electrode modified with substituted thiourea-functionalized high ordered nanoporous silica.** *Anal. Sci. 25 (2009) 789-794.*

[82] Xia F., Zhang X., Zhou C., Sun D., Dong Y., Liu Z.; **Simultaneous determination of copper, lead, and cadmium at hexagonal mesoporous silica immobilized quercetin modified carbon paste electrode.** *J. Autom. Methods Manag. Chem. 2010 (2010) 1-6.*

[83] Popa D. E., Buleandra M., Mureseanu M., Ionica M., Tanase I. G.; **Carbon paste electrode modified with organofunctionalized mesoporous silica for electrochemical detection and quantitative determination of cadmium (II) using square wave anodic stripping voltammetry.** *Rev. Chim. 61 (2010) 162-167.*

[84] Popa D. E., Buleandra M., Mureseanu M., Ionica M., Tanase I. G.; **Carbon paste electrode modified with organofunctionalized mesoporous silica for electrochemical detection and determination of copper(II) using cyclic voltammetry and anodic stripping voltammetry.** *Rev. Roum. Chim. 55 (2010) 123-130.*

SYNTHESIS AND CHARACTERIZATION OF NOVEL MESOPOROUS SILICAS OF THE MSU-*X* FAMILY FOR ENVIRONMENTAL APPLICATIONS.

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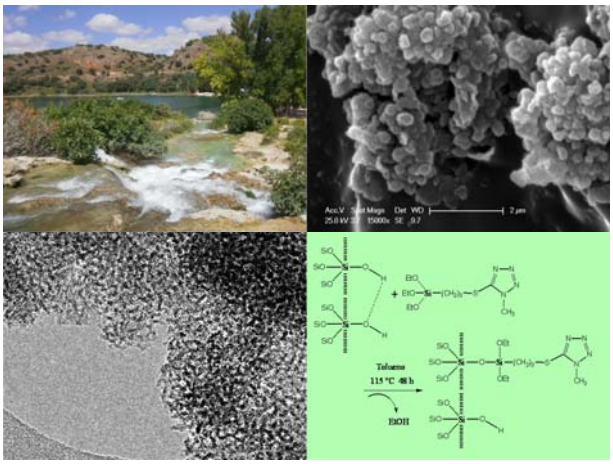


SYNTHESIS AND CHARACTERIZATION OF NOVEL MESOPOROUS SILICAS OF THE MSU-*X* FAMILY FOR ENVIRONMENTAL APPLICATIONS.

ABSTRACT

Submicron particles of mesoporous MSU-2 silica have been obtained by using Tergitol® NP-9 as nonionic surfactant and TEOS as silica precursor. The material has been chemically modified with 5-mercapto-1-methyltetrazole and characterized by powder X-ray diffraction, TEM, SEM, N_2 adsorption, FT-IR spectroscopy, ^{29}Si-NMR spectroscopy, thermogravimetric analysis and elemental analysis. The environmental application of such material (denoted MTTZ-MSU-2b) in the treatment of heavy metal-contaminated waters has been demonstrated. The maximum adsorption capacity of this material was found to be 0.73 ± 0.01 mmol/g for Pb(II) and 0.94 ± 0.01 mmol/g for Zn(II), and is higher than the capacity of most of the other functionalized mesoporous materials previously reported. The very high access of these metal ions to the binding sites in the adsorbent has also been demonstrated (adsorption efficiency equal to 0.88 and 1.13 for Pb(II) and Zn(II), respectively).

1.-INTRODUCTION

In the last few years, the development in the design of materials with particle size, shape and/or pores of nanometric scale (nanoparticles and nanoestructured materials) have made possible important advances in Nanoscience and Nanotechnology, and thus this period can be considered as a revolution of the nanometric scale. A clear example was the discovery of the M41S mesoporous silica family by Mobil researchers [1, 2] with well ordered pores of uniform size, high volume and high surface area (MCM-41, MCM-48, etc). From these studies, the field of so-called micelle templated structures (MTS) has been widely explored because depending on the preparation conditions (pH, temperature, type and concentration of the reagents, etc) it is possible to obtain materials with different structures, morphologies, surface properties, etc. Almost all kinds of surfactants (cationic, anionic, neutral and triblock copolymers) have been tested to obtain MTS. For example, the possibility of using neutral amine surfactants (S^0) and neutral inorganic precursors (I^0) to obtain hexagonal mesoporous metal oxides was demonstrated by Pinnavaia's group [3]. This S^0I^0 pathway tends to afford structures with thicker framework walls with improved stability and smaller elementary particle sizes (HMS mesoporous family). Despite the attractive benefits of mesostructure templating by neutral alkyl amines, they are not ideally suited for industrial applications (large-scale synthesis) because of their highly cost and toxicity.

Other surfactants proposed as promising neutral templating agents are polyethylene oxide (PEO) nonionic surfactants or block copolymers. These compounds are relatively inexpensive, biodegradable and the critical micelle concentration is approximately one-hundredth of those for ionic surfactants. Depending on the synthetic conditions, different mesoporous families can be prepared by using these surfactants (SBA and MSU-*X* mesoporous silicas). For example, the MSU-*X* family of mesoporous silica can be prepared by using alkyl-PEO ($X = 1$), alkylary-PEO ($X = 2$), polypropylene-oxyde-PEO block copolymers ($X = 3$) and Tween-type nonionic ethoxylated sorbitan esteres ($X = 4$) [4-10]. MSU-*X* materials lack regular channel packing order but exhibit 3D worm-hole porous framework with uniform channel diameters. Compared to MCM and SBA mesoporous families that exhibit either hexagonal or cubic symmetries, the structure of these materials is expected to enhance the diffusion rate of species into the pores [8].

The remarkable progress in the synthesis of ordered mesoporous materials has been accompanied by the development of a wide variety of potential applications of these materials: adsorption, catalysis, separations, gas storage, drug delivery, environmental remediation, etc [11-13]. It is well known that the characteristics of these materials makes them potentially useful in those processes were the adsorption

of different kind of compounds is required. However, this adsorption is not selective and it can also be reversible [14]. To increase the selectivity and loading capacity, the chemical modification of these materials with organic functional groups to obtain new hybrid silica-based materials is necessary. For selective adsorption and extraction of heavy metals from waters, the design of these hybrid silicas is based on the emulation of the toxic effect of heavy metals on living cells [15]. Therefore a good understanding of the selective binding between such metals and some biomolecules is a key point to determine the best functional groups for silica modification, for example those containing oxygen, nitrogen and sulphur atoms. These new materials have important economical benefits for industry and society because the retained metals can be recovered and the materials reused.

In this work we describe the preparation and characterization of new MSU-type mesoporous silicas and their application in heavy metal adsorption in aqueous media.

2.-EXPERIMENTAL DETAILS

2.1.-Reagents and materials

Tetraethylorthosilicate (TEOS) 98% (*MW* = 208.33, *d* = 0.934 g/mL) and Tergitol® NP-9 (*MW* = 616.82) were purchased from Sigma-Aldrich (Germany), sodium fluoride 99% (*MW* = 41.99) was purchased from Fluka (Switzerland), 3-chloropropyltriethoxysilane 95% (CPTS) (*MW* = 240.81, *d* = 1.007 g/mL), chlorotrimethylsilane 99% (*MW* = 108.64), triethylamine (*MW* = 101.19, d = 0.726 g/mL) and 5-mercapto-1-methyltetrazole 98% (MTTZ) (*MW* = 116.15), all from Sigma-Aldrich, were used as supplied. Organic solvents (toluene, diethyl ether, ethanol and hexane) were purchased from SDS (France). These solvents were distilled and dried before use according to conventional literature methods. Buffer solutions were prepared in the laboratory using reagents purchased for Panreac (Spain). Standard stock solutions of Zn(II), Cr(VI), Cu(II), Cd(II) and Pb(II) were prepared by dissolving zinc chloride 98% (Sigma-Aldrich), extra pure potassium dichromate (Scharlau, Spain), extra pure copper(II) nitrate trihydrate (Scharlau), cadmium nitrate tetrahydrate 98% (Sigma-Aldrich) and lead(II) nitrate 98% (Panreac), in dilute HNO_3. Water (resistance 18.2 MΩ · cm) used in the preparation of materials and standard solutions was obtained from a Millipore Milli-Q-System (Waters, USA). Glassware was soaked in (1 + 1) HNO_3 overnight and cleaned with Milli-Q water before use. All other reagents were of at least analytical grade.

2.2.-Synthesis and functionalization of the mesoporous silicas

Novel mesoporous silicas of the MSU-*X* family (MSU-2-type) were prepared using Tergitol® NP-9 (Figure 1) as nonionic surfactant. The materials were synthesized by a two-step process: Firstly TEOS was added to a stirred 0.02 M (pH 4.5) or 0.08 M (pH 4.8) solution of Tergitol® NP-9 in Milli-Q water, at room temperature, to obtain a milky suspension (TEOS/surfactant solution molar ratio of 8/1 or 2/1, respectively). The resulting suspension was then aged without agitation for 20 h to give a clear solution. In the second step, a 0.24 M sodium fluoride solution was added dropwise with stirring to the TEOS/surfactant solution to obtain a NaF/TEOS molar ratio of 0.025/1. The solutions were placed in a bath with agitation at 55 ºC for 48 h. The final products (MSU-2a and MSU-2b), were filtered off, washed with Milli-Q water and air-dried at 100 ºC for 4 h. Finally, the surfactant was removed by calcination in air at 600 ºC for 12 h.

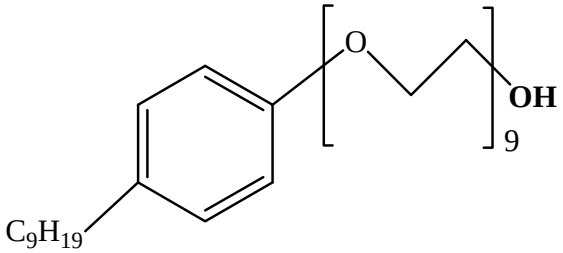


Figure 1. Tergitol® NP-9 structure.

20.00 mL of MTTZ derivative, synthesised and characterized as described elsewhere [16, 17], was reacted with 20.00 g of activated MSU-2b (5 h at 160 ºC under high vacuum) in dry toluene (200 mL) with mechanical stirring (48 h under reflux conditions and nitrogen atmosphere). The resulting modified mesoporous silica (MTTZ-MSU-2b) was filtered off and washed with toluene (2 × 100 mL), ethanol (2 × 100 mL) and diethyl ether (2 × 100 mL). Finally, the product was heated for 4 h at 110 ºC under vacuum.

2.3.-Characterization

X-ray diffraction (XRD) patterns of the silicas were obtained on a Philips X'Pert diffractometer at 45 kV and 40 mA, using the Cu-Kα line (λ = 1.5418 Å). N_2 gas adsorption-desorption isotherms were recorded using a Micromeritics TriStar 3000 manometric porosimeter. The surface specific areas were calculated by the Brunauer- Emmett-Teller (BET) method. Pore size distributions were obtained using the Barret-Joyner-Halenda (BJH) model on the adsorption branch. Scanning electron micrographs (SEM) and morphological analysis were obtained on a XL30 ESEM Philips microscope with an energy dispersive spectrometry system. The samples was treated with a sputtering method with the following parameters: sputter time 100 s, sputter current 30 mA, film thickness 20 nm, using a sputter coater BAL-TEC SCD 005. Conventional transmission electron microscopy (TEM) was carried out with a TECNAI 20 Philips electron microscope operating at 200 kV. Infrared spectra were recorded on a Thermo-Nicolet Avatar 380 FT-IR spectrophotometer in the region 4000 to 400 cm^{-1} using spectroscopic quality KBr powder. Solid state ^{29}Si MAS-NMR spectra were recorded on a Varian-Infinity Plus 400 MHz Spectrometer operating at 79.44 MHz under the following conditions: 4 μs 90º pulse, 1024 transients, spinning speed of 5 kHz. Elemental analysis (%C, %N and %S) was performed at Alcalá de Henares University (Madrid, Spain). The thermal stability of the modified mesoporous silicas was studied with a Setsys 18 A (Setaram) thermogravimetric analyzer and a platinum crucible of 100 μL (synthetic air atmosphere from 25 ºC to 800 ºC, 10 ºC/ min).

2.4.-Heavy metal adsorption

A sample of MTTZ-MSU-2b (0.1 g) was suspended in 35 mL of 0.014 mM Zn(II), Cr(VI), Cu(II), Cd(II) or Pb(II) solution at pH 8. Each solution was stirred mechanically (300 rpm) during 4 h at 25 ºC to attain equilibrium. Afterwards, the suspension was filtered through Whatman Nº 50 filter paper and the solid was washed with copious amounts of Milli-Q water. Finally, the total volume of the filtrate was made up to 100 mL with Milli-Q water using a volumetric flask. The unbound Zn(II), Cr(VI), Cu(II), Cd(II) and Pb(II) ions present in each solution was determined by flame atomic absorption spectrometry (FAAS, Varian Spectrometer A50) according to the standard guidelines of the manufacturers. The uptake of each heavy metal by the MTTZ-MSU-2b was calculated from the difference between the initial metal amount and that remaining in the supernatant.

2.4.1.-Maximum adsorption of Pb(II) and Zn(II)

A glass column (10 cm × 1 cm i.d.) was filled with 0.05 g MTTZ-MSU-2b. 5000 mL of 0.0125 mM of Pb(II) solution or 1000 mL of 0.06 mM of Zn(II) solution, adjusted to pH 8, was passed through the column at a flow rate of approximately 0.4 mL/min (20 rpm) with a peristaltic pump (Ismatec MV-Pumpsystem). Then, the column was washed with 20 mL of Milli-Q water. Finally, to desorb the bound-Pb(II) or Zn(II) ions, 25 mL of 1 M hydrochloric acid solution was passed at a flow rate of approximately 0.4 mL/min. The maximum amount of Pb(II) or Zn(II) taken up by the MTTZ-MSU-2b ($N_{f\,max}$) was calculated as follows:

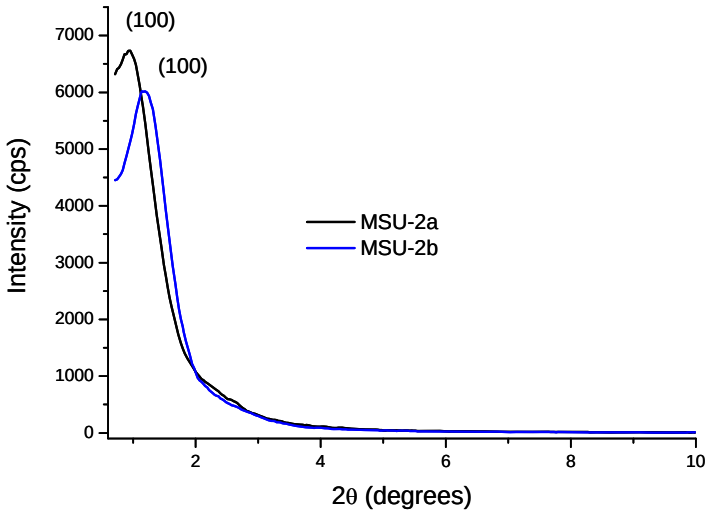
$$N_{f\ \max} = \frac{n_i - n_e}{m}$$

where n_i is the initial number of moles of Pb(II) or Zn(II) added to the system, n_e the amount remaining after the equilibrium and m is the mass in grams of the adsorbent. For comparison purposes, the same adsorption experiment was carried out with nonmodified MSU-2b silica.

3.-RESULTS AND DISCUSSION

3.1.-Characterization of the mesoporous silicas

The XRD patterns for unmodified MSU-2-type mesoporous silicas prepared in the presence of Tergitol® NP-9 as nonionic surfactant, with a TEOS/surfactant solution molar ratio of 8/1 (MSU-2a) and 2/1 (MSU-2b), are shown in Figure 2. Both materials exhibit a well-resolved pattern at low 2θ values with a single narrow (100) diffraction peak at 1.02 and 1.20º for MSU-2a and MSU-2b, respectively. The peak broadening and the intensity are very close in both patterns. Since these materials do not exhibit a long-range periodicity, the single peak observed in the XRD pattern can not be related with any actual *d* spacing of a crystalline cell. In these structures the d_{100} spacing, assigned to the pore-to-pore centre correlation distance, was 87.18 and 73.43 Å for MSU-2a and MSU-2b, respectively. Analogous XRD patterns have been observed for other MSU-type silicas prepared by neutral templating pathways, which have been described as typical of materials with uniform pores in the mesoporous range and non-symmetrical 3D wormhole-like structure of the porous framework [4-10].



Material	TEOS/Tergitol	(hkl)	2θ (degrees)	*d*-spacing(Å)
MSU-2a	8/1	100	1.01	87.18
MSU-2b	2/1	100	1.20	73.43

Figure 2. XRD patterns of (a) MSU-2a and (b) MSU-2b.

Evidence for disordered wormhole-like structure of the MSU-2b silica is provided by the transmission electron micrographs presented in Figure 3. These micrographs show that the silica particle contains a large number of channels that are regular in diameter, although they lack long-range packing order. TEM also reveals that the whole material is made of such a porous framework and that there is no amorphous component. SEM images of the MSU-2a and MSU-2b silicas (Figure 4) confirmed that in both cases the particle shape was similar (pseudo-spherical) but not the mean diameter (915 and 244 nm for MSU-2a and MSU-2b, respectively). A strong influence of the surfactant solution pH on the microostructure of the material obtained has been indicated previously by Boissière et al [8]. Results obtained in the present studies are in agreement with those obtained by these authors, who indicated that syntheses at pH up to 3 produce particles with non defined shape and nanometric size.

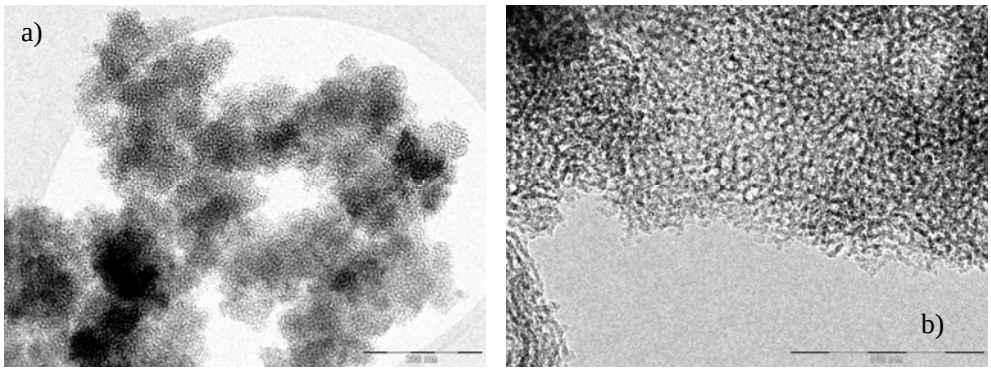


Figure 3. TEM images of the synthesized MSU-2b.

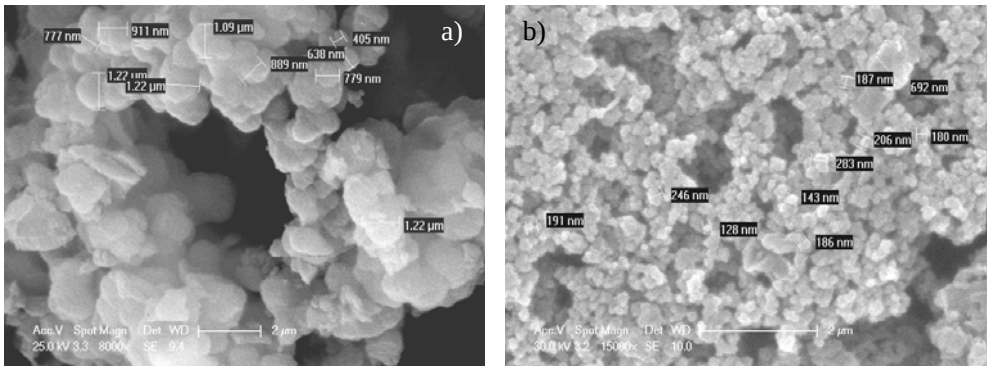


Figure 4. SEM images of (a) MSU-2a, (b) MSU-2b.

Figure 5 illustrates the nitrogen adsorption-desorption isotherms for the prepared materials. As can be seen, isotherms contain a well-defined step (type IV according to the I.U.P.A.C. classification) and have an H1 hysteresis loop that is indicative of framework-confined mesoporosity. In unmodified MSU-2 silicas, the

volume of nitrogen adsorbed increased at a relative pressure (P/P_0) of, approximately, 0.65 in MSU-2a and 0.5 in MSU-2b, which represents capillary condensation within the uniform mesopore structure. Table I summarizes structural parameters for the prepared materials that are in agreement with those reported previously for MSU-2-type mesoporous silicas prepared under neutral pH conditions and by using aqueous solution of the surfactant. It can be observed that the addition of a high amount of TEOS to the surfactant solution (TEOS: surfactant molar ratio of 8/1) a material with lower S_{BET} (558 m^2/g), higher pore volume (1.05 cm^3/g) and higher BJH pore diameter (74 Å) is obtain. This fact can be attributed to the higher mean hydrodynamic diameter of the micelles formed in this preparation (MSU-2a), in comparison when a lower amount of TEOS is added (TEOS: surfactant molar ratio of 2/1 in MSU-2b), as indicated previously by other authors [18]. Assuming that the d_{100} spacing is a direct measure of the average distance between channel centres, the average wall thickness in these preparations (13 and 25 Å for MSU-2a and MSU-2b, respectively) was deduced by subtracting the BJH pore diameter from the d_{100} spacing. The pore size distribution revealed a quite narrow pore diameter distribution for MSU-2b that attests their more uniform framework mesoporosity in comparison with the MSU-2a material (Figure 6).

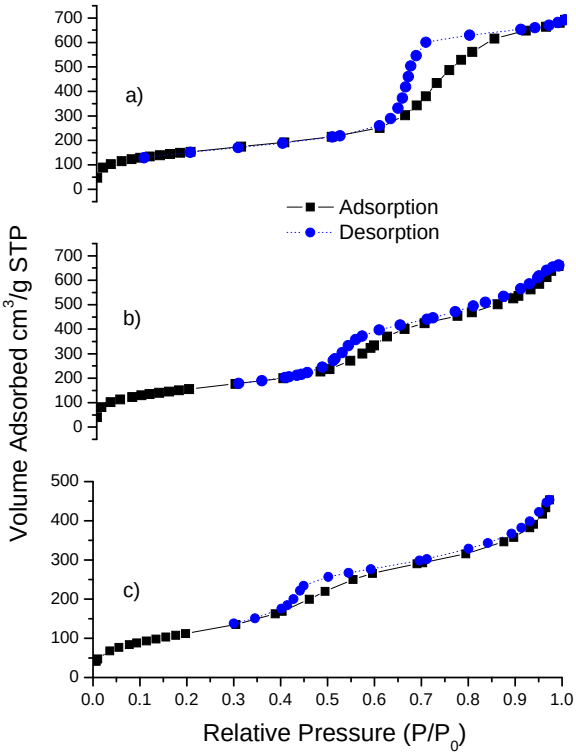


Figure 5. Nitrogen adsorption-desorption isotherms of (a) MSU-2a, (b) MSU-2b and (c) MTTZ-MSU-2b.

Table I
Mesoporous silica characterization data.

		MSU-2a	MSU-2b	MTTZ-MSU-2b
XRD	2θ (º)	1.01	1.20	1.20
patterns	*d*-spacing (Å)	87.18	73.43	73.43
	BET surface (m^2/g)	558	570	423
N_2	Pore volume (cm^3/g)	1.05	0.95	0.64
Isotherms	BJH pore diameter (Å)	74	48	37
	Wall thickness (Å)	13	25	36

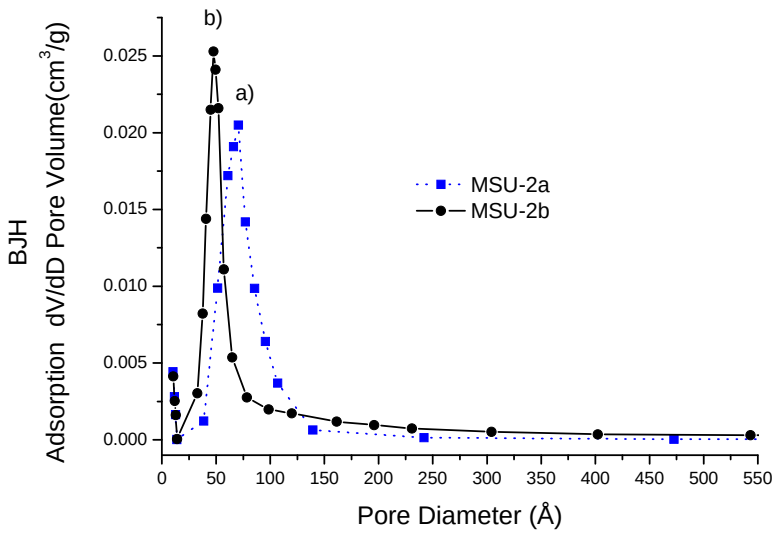


Figure 6. Pore size distribution for (a) MSU-2a and (b) MSU-2b.

To study the environmental application of the prepared mesoporous materials for the treatment of heavy metal contaminated waters, MSU-2b was functionalized with a ligand that contains nitrogen and sulphur atoms (MTTZ derivative). In the functionalized silica (MTTZ-MSU-2b), a decrease in the intensity of the (100) XRD diffraction peak was observed. This fact provides further evidence that grafting mainly occurs inside the mesopore channels, since the attachment of organic functional

groups in the mesopore channels tends to reduce the scattering power of the mesoporous silicate wall. The XRD pattern of the functionalized silica also suggests that the structural order of the synthesized material is maintained after functionalization. As can be seen in Table I, a decrease in the surface area (from 570 to 423 m^2/g), pore volume (from 0.95 to 0.64 cm^3/g) and pore diameter (from 48 to 37 Å) together with an increase in the wall thickness (from 25 to 36 Å) were produced after functionalization of the MSU-2b silica. Figure 5 shows that the inflection position of the nitrogen adsorption-desorption isotherms shifted slightly toward lower relative pressures (0.4) after functionalization, the volume of nitrogen adsorbed decreased and the hysteresis loop decreased. These changes can be attributed to the presence of pendant groups on the MTTZ-MSU-2b surface that partially blocks the adsorption of nitrogen molecules. A narrow pore size distribution was found for this material (centred at 37 Å) that provides evidence for a uniform framework mesoporosity after functionalization.

The IR patterns of prepared mesoporous silicas between 4000-400 cm^{-1} are shown in Figure 7. The main features of the MTTZ-MSU-2b spectra include a large broad band between 3600 and 3250 cm^{-1}, which is attributed to O-H stretching of the surface silanol groups and the remaining adsorbed water molecules. The siloxane (-Si-O-Si-) band appears as a broad strong peak centred at 1100 cm^{-1}. The band due to Si-O bond stretching of the silanol groups was observed at 800 cm^{-1}. The adsorption band at 1630 cm^{-1} is due to deformation vibrations of adsorbed water molecules. Characteristic bands for aliphatic C-H stretching vibrations due to pendant alkyl chains at around 3000-2800 cm^{-1} and bands at ca. 1460, 1390 and 700 cm^{-1} due to C=N, C-N and C-O stretching vibrations of the attached MTTZ group [19, 20] were observed.

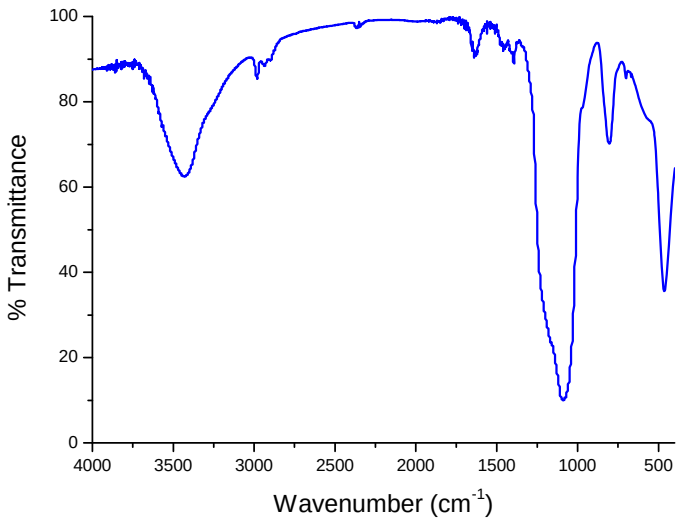


Figure 7. FT-IR spectra of MTTZ-MSU-2b.

The ^{29}Si MAS-NMR spectra in the solid state for MTTZ-MSU-2b confirmed the covalent bond formed between the ligand and the silanol groups dispersed on the silica surface. The unmodified MSU-2b silica spectrum (not shown) have two main peaks at -111 and -103(*sh*) ppm, assigned to Q^4 and Q^3 silanol sites, respectively. The spectrum of the functionalized silica (Figure 8) shows a marked decrease in the intensity of the Q^3 signal, which verifies the anchoring of the functional groups to Si-OH. In addition, two new peaks appear at -54 and -62 ppm, which are assigned to T^2 ($(SiO)_2$***Si*** OH-R) and T^3 ($(SiO)_3$***Si***-R) sites, respectively.

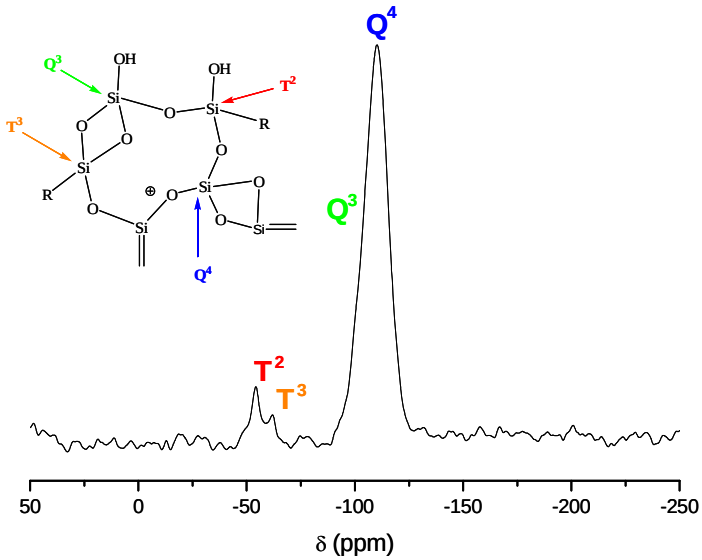


Figure 8. ^{29}Si MAS-NMR spectra of MTTZ-MSU-2b.

A very high Si-OH group content (α_{OH} = 6.85 mmol/g) on the pore channel walls was calculated for MSU-2b silica, after a silylation procedure with chlorotrimethylsilane according to Zhao et al. [21] Figure 9 shows the ^{29}Si MAS-NMR spectrum for the silated MSU-2b. At 14 ppm appears the M site (3.85% relative area) corresponding to the silicon atoms of the trimethylsilane ($(CH_3)_3$-***Si***-). The low intensity peak at -93 ppm (2.91%) corresponds to surface silicon atoms with two siloxane bonds and two silanol groups, Q^2 silanol sites ($(SiO)_2$***Si*** $(OH)_2$), the resonance at -101 ppm (12.69%) is attributed to surface silicon atoms with three siloxane bonds an one silanol group, Q^3 silanol sites ($(SiO)_3$***Si*** OH), while the resonance at -110 ppm (80.55) is ascribed to surface silicon atoms with four siloxane bonds, Q^4 sites ($(SiO)_4$***Si***). From these relative peak areas, we can calculate α_{OH} using the following expressions:

$$\alpha_{OH} = \frac{S \cdot (1 + f + f_g^r)}{f}$$

$$S = \frac{\Delta W \cdot N_A}{72 \cdot S_{BET}} \cdot 10^{-20}$$

$$f = \frac{\% M\ peak}{\% Q^2\ peak + \% Q^3\ peak}$$

$$f_g^r = \frac{\% Q^2\ peak}{\% Q^2\ peak + \% Q^3\ peak}$$

where ΔW is the weight gain after silylation (8.5 %), N_A is Avogadro's number and 72 is the effective molar weight of the attached trimethylsilane groups.

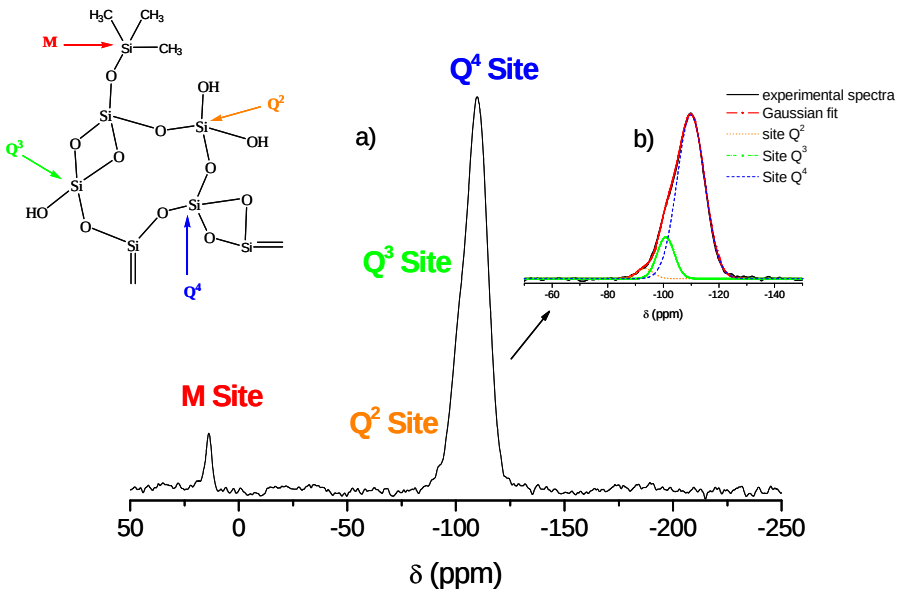


Figure 9. (a) ^{29}Si MAS-NMR spectra of ((CH$_3$)$_3$-Si-MSU-2b, (b) Gaussian deconvolution spectrum

The quantity of molecules attached to the mesoporous silica surface (L_0 = 0.83 mmol/g) was calculated from the percentage of sulphur in the functionalized mesoporous silica, as estimated by elemental analysis, using the following expression:

$$L_0 = \frac{\%\,\mathrm{S} \times 10}{\text{Sulphur atomic weight}}$$

The C/S molar ratio calculated from the elemental analysis of MTTZ-MSU-2b indicates a 1:1 stoichiometry between the silanol groups on the silica surface and the ligand. Taking into account L_o and S_{BET} of the mesoporous silicas, the average surface density, *d*, of the attached molecules and the average intermolecular distance, *l*, can be calculated by applying the following equations [22]:

$$d = N_A \frac{Lo}{S_{BET}} x10^{-21}$$

$$l = \left(\frac{1}{d}\right)^{1/2}$$

Results obtained (d = 0.88 molecules/nm^2 and l = 1.07 nm) confirm a high efficiency in the functionalization of the MSU-2b that can be explained as a consequence of its high α_{OH}.

The thermogravimetric curves (TGA) for the modified mesoporous silica allow information concerning the thermal stability to be obtained and also confirm the amount of immobilized ligand. The TGA curves of the MTTZ-MSU-2b (Figure 10) shows that an exothermic degradation process occurs between 225-600 ºC with a total weight loss of about 14 %. The weight loss that takes place between 225-275 ºC may be due to the break down of the ethoxy groups (-O-CH_2CH_3), between 275-360 ºC to the heterocyclic ring and between 360-430 ºC to the alquilic chain (exothermic processes). The thermal stability of these samples is in agreement with previous results given in the literature for other functionalized mesoporous silicas [16, 17].

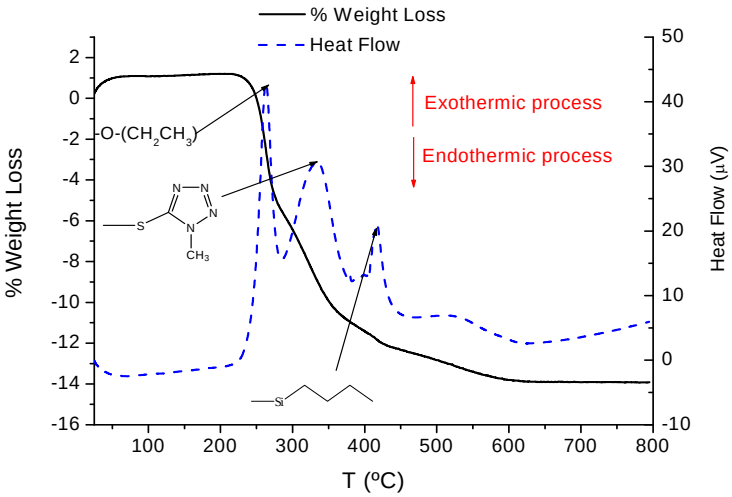


Figure 10. Thermogravimetric curves and heat flow of MTTZ-MSU-2b.

3.2.-Heavy metal adsorption on the mesoporous silicas

The adsorption capacity of the material is an important factor because it determines how much adsorbent is required to quantitatively remove a specific amount of heavy metals from contaminated media. The different adsorption capacity of a material for various metal ions is due to their differences in size, degree of hydratation and binding constant with the ligand immobilized onto the silica. To study the capacity of the MTTZ-MSU-2b material to extract Zn(II), Cr(VI), Cu(II), Cd(II) and Pb(II) from aqueous solutions, adsorption experiments were carried out in triplicate (n = 3) using a batch wise process. Each metal solution (pH 8) was stirred mechanically in presence of the adsorbent (0.1 g) during 4 h at 25 ºC to attain equilibrium. The amount of each heavy metal adsorbed by the synthesized material was determined by means of the equation:

$$\%\ adsortion = (n_i - n_e / n_i)x100$$

where n_i is the initial number of moles of each ion added to the system and n_e the amount remaining in the supernatant after the equilibrium. In each experiment standard and blank solutions were prepared and treated in the same way as that sample.

As can be seen in Figure 11, MTTZ-MSU-2b has a very high affinity towards Zn(II) and Pb(II) (adsorptions near to 100%), whereas for Cd(II) and Cr(VI) MTTZ-MSU-2b exhibits very low affinity (adsorptions lower than 10%). The % adsorption in the prepared material follows the order Zn(II) ~ Pb(II) > Cu(II) >> Cd(II) > Cr(VI).

To find out the maximum adsorption capacity of the MTTZ-MSU-2b ($N_{f\ max}$) for Pb(II) and Zn(II), column experiments were carried out. In these experiments the functionalized material (0.05 g) was placed in a glass column and Pb(II) or Zn(II) solutions were passed through the column controlling the flow rate with a peristaltic pump. For comparison purposes, the same experiments were carried out with nonmodified MSU-2b silica. Based on the results shown in Table II, the functionalization of the MSU-2b with the MTTZ derivative leads to a significant increase in the binding capacity for both metals (MTTZ-MSU-2b $N_{f\ max}$ = 0.73 ± 0.01 and 0.94 ± 0.01 mmol/g for Pb(II) and Zn(II), respectively). As an indicator of the adsorption efficiency (φ), the fraction of surface attached ligand bonded to metal ions was calculated ($\varphi = N_{f\ max} / L_o$). The φ values obtained (0.88 and 1.13 for Pb(II) and Zn(II), respectively) indicates a high level of access for these ions to the complexing sites in the mesopore channels of the MTTZ-MSU-2b.

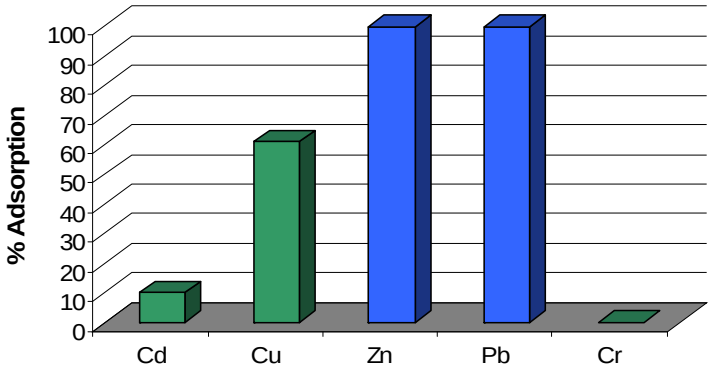


Figure 11. % Adsorption of heavy metals on the MTTZ-MSU-2b.

Table II
Maximum adsorption capacity ($N_{f\ max}$) and adsorption efficiency (φ) of the mesoporous silicas.

Material	$N_{f\,max}$ (mmol/g) [a]		φ [b]	
	Pb(II)	Zn(II)	Pb(II)	Zn(II)
MSU-2b	0.004 ± 0.001	0.008 ± 0.001	-	-
MTTZ-MSU-2b	0.73 ± 0.01	0.94 ± 0.01	0.88	1.13

[a] mmol of Pb(II) or Zn(II) adsorbed per g of material
[b] $\varphi = N_{f\ max} / L_o$
- Not calculated

Reported Pb(II) and Zn(II) adsorption data in aqueous media using functionalized mesoporous silicas are very scarce in the literature. Comparing the adsorption capacity of the MTTZ-MSU-2b material with those of other functionalized mesoporous silicas (Table III), it can be concluded that the material prepared in this work has better $N_{f\,max}$ value than most of them [16, 17, 23-28]. In previous published works [16, 17], we prepared HMS and MCM-41 mesoporuos materials functionalized with the MTTZ derivative. These hybrid materials showed very good adsorption capacities for Pb(II) (0.56 mmol/g of MTTZ-HMS) and Zn(II) (1.59 mmol/g of MTTZ-MCM-41) that indicated a high affinity of the MTTZ derivative for these metals. Interestingly, the MTTZ-MSU-2b material prepared in the present work shows better φ than both previously prepared materials. These results can be attributed to the 3D worm-hole porous framework with uniform large-diameter pore channels of the MSU silica, which is expected to enhance the access of the heavy metals to the binding sites.

In conclusion, it is clear the influence of the physical characteristics of the mesoporous silica support on the metal uptake capacity of these materials. The potential advantages of MSU-*X* mesoporous family over other materials are lower reagent and energy costs (process at ambient pressure and at moderate temperature), low surfactant toxicity and biodegradability. All these points decrease the overall cost and environment impact of this synthesis process.

Table 3
Bibliographic dates about functionalized mesoporous silicas for Pb(II) and Zn(II) adsorption in aqueous media

Support	Immobilized ligand	L_o (mmol/g)[a]	$N_{f\,max}$ (mmol/g)[b]		φ[c]		Reference
			Pb(II)	Zn(II)	Pb(II)	Zn(II)	
HMS	5-mercapto-1-methyl-1-H-tetrazol	1.09	0.56	-	0.51	-	[16]
HMS	1,4-bis-[3-(trimethoxysilyl)-propyl]ethylenediamine	0.31	-	0.0009	-	0.003	[23]
HMS	1,4-bis-[3-(trimethoxysilyl)-propyl]ethylenediamine	0.72	-	0.00011	-	0.00015	[23]
HMS	3-mercaptopropyltrimethoxysilane	1.3	≅0	-	≅0	-	[24]
MCM-41	5-mercapto-1-methyl-1-H-tetrazol	1.46	-	1.59	-	1.09	[17]
MCM-41	3-(2-aminoethylamino)-propyltrimethoxysilane	-	-	0.57	-	-	[25]
Mesoporous silica	3-aminopropyltriethoxysilane	-	0.41	0.36	-	-	[26]
MSU-2	3-mercaptopropyltrimethoxysilane	2.3	≅0	-	≅0	-	[27]
SBA-15	Polyamidoamine	-	0.025	0.21	-	-	[28]
SBA-15	EDTA-Polyamidoamine	-	0.048	0.15	-	-	[28]

[a] mmol of ligand per g of support
[b] mmol of Pb(II) or Zn(II)/ g material
[c] $\varphi = N_f / L_o$
- Not calculated or not indicated

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4.-REFERENCES AND NOTES

[1] Kresge C. T., Leonowicz M. E., Roth W. J., Vartuli J. C., Beck J. S.; **Ordered mesoporous molecular sieves synthesized by a liquid-crystal template mechanism.** *Nature 359 (1992) 710-712.*

[2] Beck J. S., Vartulli J. C., Roth W. J., Leonowicz M. E., Kresge C. T., Schmitt K. D., Chu C. T. W., Olson D. H., Sheppard E. W., McCullen S. B., Higgins J. B., Schlenker J. L.; **A new family of mesoporous molecular sieves prepared with liquid crystal templates.** *J. Am. Chem. Soc. 114 (1992) 10834-10843.*

[3] Tanev P. T., Pinnavaia T. J.; **A neutral templating route to mesoporous molecular sieves.** *Science 267 (1995) 865-867.*

[4] Bagshaw S. A., Prouzet E., Pinnavaia T. J.; **Templating of mesoporous molecular sieves by non-ionic polyethylene oxide surfactants.** *Science 269 (1995) 1242-1244.*

[5] Prouzet E., Pinnavaia T. J.; **Assembly of mesoporous molecular sieves containing wormhole motifs by a non-ionic surfactant pathway: Control of pore size by synthesis temperature.** *Angew. Chem.-Int. Edit. 36 (1997) 516-518.*

[6] Boissiere C., van der Lee A., el Mansouri A., Larbot A., Prouzet E.; **A double step synthesis of mesoporous micrometric spherical MSU-X silica particles.** *Chem. Commun. 20 (1999) 2047-2048.*

[7] Boissiere C., Larbot A., Prouzet E.; **Synthesis of mesoporous MSU-X materials using inexpensive silica sources.** *Chem. Mat. (2000) 12, 1937-1940.*

[8] Boissiere C., Larbot A., van der Lee A., Kooyman P- J., Prouzet E.; **A new synthesis of mesoporous MSU-X silica controlled by a two-step pathway.** *Chem. Mat. 12 (2000) 2902-2913.*

[9] Herrier G., Blin J.-L., Su B.-L.; **MSU-type mesoporous silicas with well-tailored pore sizes synthesized via an assembly of deca(ethylene oxide) oleyl ether surfactant and tetramethoxysilane silica precursor.** *Langmuir 17 (2001) 4422-4430.*

[10] Boissiere C., Martines M. A. U., Tokumoto M., Larbot A., Prouzet E.; **Mechanisms of pore size control in MSU-X mesoporous silica.** *Chem. Mat. 15 (2003) 509-515.*

[11] Nguyen T.-Q., Wu J., Doan V., Schwartz B. J., Tolbert S. H.; **Control of energy transfer in oriented conjugated polymer-mesoporous silica composites.** *Science 288 (2000) 652-656.*

[12] Wigth A., Davis M.; **Desing and preparation of organic-inorganic hybrid catalysts.** *Chem. Rev. 102 (2002) 3589-3614.*

[13] Muñoz B., Ramila A., Pérez-Pariente J., Díaz I., Vallet-Regí M.; **MCM-41 organic modification as drug delivery rate regulator.** *Chem. Mat. 15 (2003) 500-503.*

[14] Stein A., Melde B., Schroden R.; **Hybrid inorganic-organic mesoporous silicates-nanoscopic reactors coming of age.** *Adv. Mater. (2000) 12, 1403-1419.*

[15] Olkovyk O., Jaroniec M.; in: **Environment applications of nanomaterials,** ed. Fryxell G. E., Cao G.; *Imperial College Press, London, 2007, chap. 8 p. 179.*

[16] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Functionalized HMS mesoporous silica as solid phase extractant for Pb(II) prior to its determination by flame atomic absorption spectrometry.** *J. Sep. Sci. 30 (2007) 1556-1567.*

[17] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Preparation, characterization, and Zn^{2+} adsorption behavior of chemically modified MCM-41 with 5-mercapto-1-methyltetrazole.** *J. Colloid Interface Sci. 313 (2007) 551-562.*

[18] Prouzet E., Boissiere C.; **A review on the synthesis, structure and applications in separation processes of mesoporous MSU-X silica obtained with the two-step process.** *C. R. Chim. 8 (2005) 579-596.*

[19] Pretsch E., Clero T., Seibl J., Simon W.; **Tablas para la elucidación structural de compuestos orgánicos por métodos espectroscópicos.** *Alhambra-Longman, Madrid, 1991.*

[20] Pavia D., Lampman G., Kriz G.; **Introduction to spectroscopy.** *Harcourt College Publishers, Philadelphia, 2001.*

[21] Zhao X. S., Lu G. Q., Whittaker A. K., Millar G. J., Zhu H. Y.; **Comprehensive study of surface chemistry of MCM-41 using ^{29}Si CP/MAS NMR, FTIR, Pyridine-TPD, and TGA.** *J. Phys. Chem. B 101 (1997) 6525-6531.*

[22] Dias Filho N. L., Gushikem Y., Franco D. W., Schultz M. S., Vasconcellos L. C. G.; **Study of copper complexes adsorbed on a silica gel surface chemically modified with 2-amino-1,3,4-thiadiazole.** *Colloid Surf. A-Physicochem. Eng. Asp. 141 (1998) 181-187.*

[23] Hossain K. Z., Mercier L.; **Intraframework metal ion adsorption in ligand-functionalized mesoporous silica.** *Adv. Mater. 14 (2002) 1053-1056.*

[24] Brown J., Mercier L., Pinnavaia T. J.; **Selective adsorption of Hg^{2+} by thiol-functionalized nanoporous silica.** *Chem. Commun. (1999) 69-70.*

[25] Lu Y.-K., Yan X.-P.; **An imprinted organic-inorganic hybrid sorbent for selective separation of cadmium from aqueous solution.** *Anal. Chem. 76 (2004) 453-457.*

[26] Yang H., Xu R., Xue X., Li F., Li G.; **Hybrid surfactant-templated mesoporous silica formed in ethanol and its application for heavy metal removal.** *J. Hazard. Mater. 152 (2008) 690-698.*

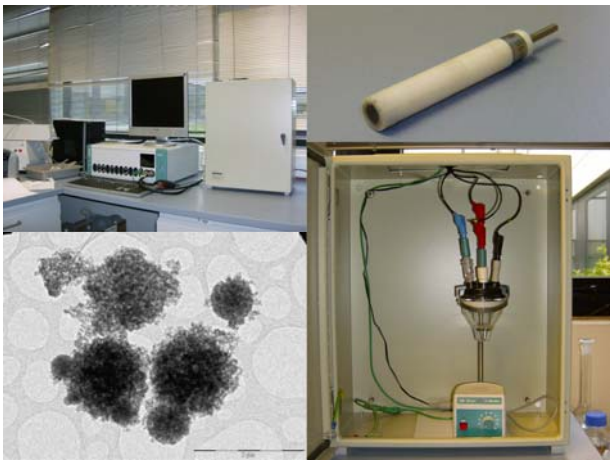
[27] Brown J., Richer R., Mercier L.; **One-step synthesis of high capacity mesoporous Hg^{2+} adsorbents by non-ionic surfactent assembly.** *Microporous Mesoporous Mat. 37 (2000) 41-48.*

[28] Jiang Y., Gao Q., Yu H., Chen Y., Deng F.; **Intensively competitive adsorption for heavy metal ions by PAMAM-SBA.15 and EDTA-PAMAM-SBA-15 inorganic-organic hybrid materials.** *Microporous Mesoporous Mat. 103 (2007) 316-324.*

VOLTAMMETRIC ANALYSIS OF Pb(II) IN NATURAL WATERS USING A CARBON PASTE ELECTRODE MODIFIED WITH 5-MERCAPTO-1-METHYLTETRAZOL GRAFTED ON HEXAGONAL MESOPOROUS SILICA.

Sonia Morante-Zarcero, Alfredo Sánchez, Mariano Fajardo, Isabel del Hierro and Isabel Sierra.

Microchimica Acta 169 (**2010**) 57-64.



VOLTAMMETRIC ANALYSIS OF Pb(II) IN NATURAL WATERS USING A CARBON PASTE ELECTRODE MODIFIED WITH 5-MERCAPTO-1-METHYLTETRAZOL GRAFTED ON HEXAGONAL MESOPOROUS SILICA.

ABSTRACT

A hexagonal mesoporous silica (HMS) functionalized with a 5-mercapto-1-methyltetrazole derivative was employed to prepare a chemically modified carbon paste electrode for Pb(II) detection in aqueous solution by square wave adsorptive stripping voltammetry. The optimal operating conditions were 5 min preconcentration time at pH 6.5, and 120 s electrolysis time in 0.2 mol L^{-1} HCl. Under these conditions, the voltammetric signal increased linearly with the preconcentration time in the range 1 to 10 min and with the Pb(II) concentration in the range 1 to 100 μg L^{-1}. The electrode was reproducible and sensitive. Simultaneous determination of Pb, Cd and Cu was also carried out with the electrode. The accuracy of the method was validated by analysing Pb(II) in tap water and groundwater samples.

1.-INTRODUCTION

The determination of heavy metals has a special importance in the environmental field due to its accumulative toxicity. Lead is one of the most abundant heavy metals and the greater levels found throughout the environment come from human activities. Lead exposure occurs when lead dust or fumes are inhaled, or when lead is ingested via contaminated hands, foods, waters, etc. More than 90% of the total body burden of lead is accumulated in the bones, and it may be released into the blood re-exposing organ systems long after the original exposure. Lead may cause brain and kidney irreversible damages. In addition, acetates and phosphates of lead are carcinogens [1, 2]. According to the UE, the limit for lead in water for human consumption will be 10 $\mu g\ L^{-1}$ in 2014, until then this limit is 25 $\mu g\ L^{-1}$.

There are several analytical techniques currently employed for trace determination of heavy metals in environmental samples, such as atomic absorption spectrometry, inductively coupled plasma-mass spectrometry, X-ray fluorescence, etc. However, these are expensive techniques that often require laborious sample pretreatment. Electrochemical techniques have also been used extensively for heavy metal ion analysis. In addition to their lower cost, these techniques have other advantages such as the possibility to perform analysis *in-situ*. This provides a significant saving in time and operating costs [3].

Anodic stripping voltammetry (ASV) is one of the most sensitive electroanalytical techniques to determine heavy metals [3, 4]. Carbon paste electrodes chemically modified with functionalized silica (SM-CPEs) have reached considerable attention for ASV during the last years, because they display significant improvements with respect to conventional electrodes [5, 6]. Normally, the functionalized silica is directly embedded in the paste of the electrode, which consists of a homogeneous mixture of graphite and mineral oil. These types of electrodes have the advantage of combining the mechanical stability of an inorganic framework with the chemical reactivity of the ligand covalently grafted to the silica support. Thus, it is possible to accumulate the metal ions onto the electrode (via ion exchange or chelation) without applying any potential. In addition, using the appropriate ligand, the selectivity of the analysis may increase [5, 6]. Other additional advantages of the SM-CPEs are non-toxicity, surface renewal, very low background currents, mainly in positive potential range, and easy fabrication [4].

In the last years, the development in the design of materials with particle size, shape and/or pores of nanometric scale (nanoparticles and nanostructured materials) have made possible relevant advances in Analytical Chemistry [7]. A clear example was the discovery of a mesoporous silica family by Mobil researchers [8, 9] with large BET surface area, high porosity, controllable and narrowly distributed pore size, and

ordered pore arrangement. From these initial works, functionalized mesoporous silicas has been widely used for environmental applications as stationary phases in chromatographic separations, as solid-phase extractants for preconcentration of several compounds and to prepare selective and sensitive electrodes [10]. The use of functionalized mesoporous silicas for modified CPEs development (MSM-CPEs) is a current research area in electroanalysis. In a recent review [11], Walcarius has indicated the higher sensitivity of this type of electrodes, in comparison to the corresponding electrodes prepared with functionalized amorphous silica that is attributed to a better preconcentration efficiency, a better accessibility to the binding sites and faster diffusion processes relative to disordered silica. Despite of these advantages, a limited number of functionalized mesoporous silicas have been used to prepare MSM-CPEs for voltammetric determination of heavy metals. Walcarius, Lin & Fryxell and, more recently, Cavalheiro groups have been working with mesoporous MCM-41 and SBA-15 silicas functionalized, mainly with amino and thiol groups [12–23]. It appears therefore interesting to investigate other functionalized mesoporous silicas displaying either higher ligand loadings or regular pore channels allowing unrestricted access of the analyte to all functional sites.

Previously our group has developed two mesoporous silicas with uniform pore channels using as surfactant a neutral alkylamine and a non-ionic alkylaryl-polyethylene oxide (HMS and MSU-2 silicas, respectively). Both silicas have been chemically modified by reaction with a 5-mercapto-1-methyltetrazole derivative and the resulting materials (MTTZ-HMS and MTTZ-MSU-2) have been employed successfully as adsorbent for the preconcentration of Pb(II) from aqueous media. The presence of stronger (sulphur) and weaker (nitrogen) donor atoms allows multiple Pb-ligand interaction, so the multifunctional ligand used in these studies assures Pb-ligand complexation [24–26]. In contrast with MCM-41-type materials, prepared by electrostatic assembly pathway with ionic surfactants, HMS and MSU-2 silicas obtained via non-electrostatic assembly pathways tends to afford structures with thicker framework walls (with improved stability) and with pore channel structure, particle size and morphology which improve their ability to interact with the adsorbate species [27].

In the current work, the MTTZ-HMS material has been used to prepare a MSM-CPE for Pb(II) detection in aqueous solution by square wave adsorptive stripping voltammetry technique (AdSV). Factors affecting the preconcentration and detection steps were investigated and, in the optimal operating parameters, the electrode performance was evaluated (linearity, detection and quantification limits, precision and selectivity). The MSM-CPE has been used for simultaneous detection of lead, cadmium and copper. To our knowledge, this is the first report on using functionalized HMS mesoporous silica for Pb(II) analysis by AdSV.

2.-EXPERIMENTAL

2.1.-Reagents

All chemicals (HCl, HNO_3, NaOH, $HClO_4$, KCl, CH_3COOH, $Ni(NO_3)_2$ $Ca(NO_3)_2$, $Mg(NO_3)_2$, NaCl, Na_2SO_4, Na_3PO_4, Na_2CO_3 and CH_3COONa) were of analytical grade and used without further purification. A certified standard stock solution of Pb(II) at 1 g L^{-1} was purchased from Metrohm (http://www.gomensoro.net/metrohm). ICP stock solutions of Cu(II), Zn(II), Cd(II) at 1 g L^{-1} were purchased from Fluka (http://www.sigmaaldrich.com/spain.html). Single-component lead solution and multicomponent metal solution were prepared daily by diluting the stock solution with Milli-Q water (resistance 18.2 MΩ cm obtained from a Millipore Milli-Q-System, Waters, USA http://www.millipore.com/index.do).

2.2.-Apparatus

Square wave voltammetry (SWV) experiments were performed in an Autolab ECO-CHEMIE Potentiostat-Galvanostat model PGSTAT302 using a conventional voltammetry cell equipped with three electrodes: a modified carbon paste/MTTZ-HMS electrode, as working electrode, a saturated Ag/AgCl electrode (Metrohm, No 6.0724.140), as reference electrode, and a platinum wire (Metrohm, No 6.0331.000), as auxiliary electrode. All measurements were made at room temperature. SWV was operated at a frequency of 100 MHz with pulse amplitude of 50 mV and a potential step height of 5 mV. pH measurements were done on a XS Instruments 510 pH-meter.

2.3.-Working electrode

The MSM-CPE was prepared by mixing 0.100 g (20%w:w) of the MTTZ-HMS with 0.300 g (60% w:w) of carbon paste (Metrohm, No 6.2801.000). Then, 0.100 g (20% w:w) of mineral oil (Aldrich, http://www.sigmaaldrich.com/spain.html) were added and mixed until obtain a uniform paste. The so obtained material was packed into a 5.0 cm long polyethylene tube (d.i. 3 mm), with a metallic wire as inner electrical contact, and conveniently polished with filter paper (this procedure also helps to eliminate the excess of mineral oil). When necessary, a new electrode surface was obtained by replacing the wetted paste by freshly prepared product. For comparison purpose, a CPE was prepared according to the same procedure but adding non-functionalized HMS to the mixture. All freshly-made electrodes were activated by performing 2–3 cycles of electrolysis/stripping. Figure 1 shows the schematic of the carbon paste electrode.

The preparation and characterization of the MTTZ-HMS was described elsewhere [24]. The MTTZ-HMS material possessed a S_{BET} of 557 m^2 g^{-1}, a pore volume of 0.57 cm^3 g^{-1}, a BJH pore diameter of 16 Å and a functionalization degree of 1.09 mmol MTTZ g^{-1}.

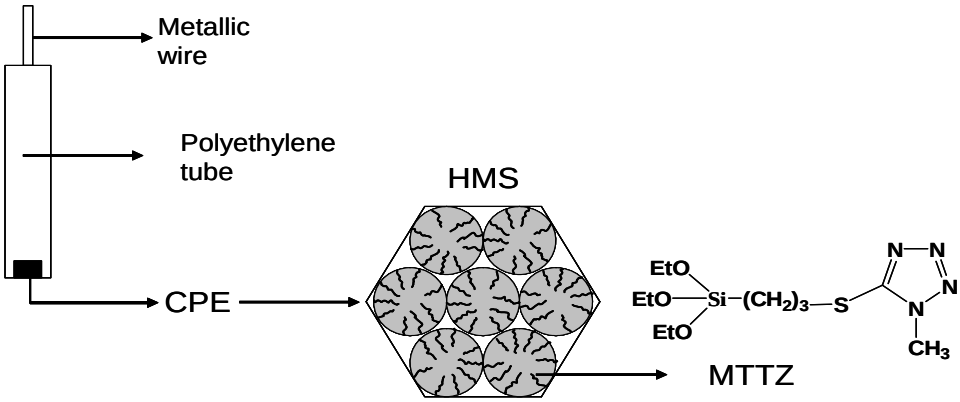


Figure 1. Schematic of the CPE modified with MTTZ-HMS

2.4.-Voltammetric detection procedure

Table 1 summarizes the operating conditions optimized for the voltammetric experiments. During preconcentration, the electrode was immersed in 15 mL of Pb(II) solution (pH 3–6.5) efficiently stirred at an open circuit for a desired period of time (1 – 10 min). The electrode was then rinsed with Milli-Q water and transferred to the voltammetric cell containing 15 mL of electrolyte solution (HCl, HNO_3, $HClO_4$, KCl, CH_3COOH). For cathodic electrolysis, a negative potential (−1.2 V) was applied to the electrode immediately after immersing into the electrolyte solution for a 0 – 150 s period, to reduce the desorbed metal into elemental form. Anodic stripping voltammetry was performed by sweeping a square wave potential toward positive values (from −1.2 V to +0.4 V). No stirring conditions were used in both cathodic electrolysis and stripping steps. All measurements were made at room temperature and under an atmospheric environment. After each measurement, to ensure that no metal was still left on the electrode surface, a regeneration of the electrode was performed by electrolysis and stripping steps in 0.2 mol L^{-1} HCl. If the electrode surface is clean, no peak will be detected in the second step. Each measurement was performed in triplicate and the average value was reported.

Table 1
Standard operating conditions for the voltammetric experiments during each step.

	Preconcentration	**Electrolysis**	**Stripping**	**Regeneration**
Solution/ electrolyte	1-100 μg L^{-1}	HCl 0.2 mol L^{-1}	HCl 0.2 mol L^{-1}	HCl 0.2 mol L^{-1}
Immersion time	5 min	120 s	-	-
Solution volume	15 mL (stirred)	15 mL	15 mL	15 mL
Applied potencial	Open circuit	-1.2 V	-1.2 to +0.4 V	Electrolysis/ stripping

3.-RESULTS AND DISCUSSION

Functionalized mesoporous silicas are good candidates to prepare chemically modified carbon paste electrodes. The selectivity of these electrodes is directly governed by the reactivity of the ligand grafted on the silica support towards the analyte, while the sensitivity of the detection is related to the efficiency of the preconcentration step. So, high sensitivity and low detection limit can be achieved using as electrode modifier functionalized mesoporous silica containing a large amount of accessible binding sites, which could be easily reached by the analyte in a short time. A well-defined three-dimensional structure allows an easier and faster access of the analyte to the binding sites, when they are grafted on the walls of regularly-assembled channels of monodisperse size [11].

Recently we have developed a highly efficient adsorbent by reaction of HMS mesoporous silica with a 5-mercapto-1-methyltetrazole derivative [24]. From batch adsorption experiments (pH 6 and 25 ºC), the maximum adsorption capacity ($N_{f\,max}$) of this material was equal to 0.56 mmol g^{-1} of Pb(II), that is better to most other chelating adsorbents used for Pb(II) adsorption [24]. This good adsorption capacity was due to the very high number of organofunctional groups attached to the surface (L_0) equal to 1.09 mmol g^{-1}. The adsorption efficiency, φ ($N_{f\,max}$ / L_0), was found to be 0.56 that indicates a high level of access for Pb(II) ions to the complexing sites in the mesopore channels of the MTTZ-HMS. In addition, a reasonably fast kinetics of the matrix-metal ion interaction was observed, that reflected the good accessibility of the chelating sites of the modified matrix and high binding constants of Pb(II) with the immobilized MTTZ on the HMS mesoporous silica surface.

These previous experiments suggested that MTTZ-HMS can be used as modifier to prepare a MSM-CPEs for voltammetric determination of Pb(II). To achieve the most accurate and sensitive Pb(II) measurements, optimization of the operating parameters in preconcentration and detection steps was performed.

3.1.-Electrode composition

The electrode composition must be uniform to maintain a good level of reproducibility after mechanical replacement of the surface electrode between measurements. This fact and their insulator character prompt us to develop the strategy of used a carbon paste matrix to provide conductive properties. The modified carbon paste electrode was prepared by mixing MTTZ-HMS or HMS silica with carbon paste and mineral oil until obtain a uniform paste. Table S1 (see ESM) shows the effect of the MTTZ-HMS or HMS content in the modified CPE on the Pb(II) peak current after 5 min preconcentration time and 120 s electrolysis time in 0.2 mol

L^{-1} HCl. From this Table, we can see that the optimum composition was 20% MTTZ-HMS, 20% mineral oil and 60% carbon paste (electrode E). For comparative purposes similar carbon paste electrodes were prepared according to the same procedure, but adding non-functionalized HMS to the mixture (Electrodes G to H, see Table S1 in ESM). Comparing electrodes G and C (10% of MTTZ-HMS and HMS, respectively, 20% mineral oil, 70% carbon paste), as it is shown in Table S1 the voltammetric response was lower in electrode G, so the use of MTTZ-HMS silica as modifier in the CPE clearly enhanced its sensitivity for Pb(II) detection.

3.2.-Preconcentration time

The effect of preconcentration time on the voltammetric response for Pb(II) detection using electrode E (composed by 20% MTTZ-HMS, 20% mineral oil and 60% carbon paste) is shown in Figure S1 (see ESM). The experiments were performed using 100 and 10 μg L^{-1} Pb(II) solutions (120 s electrolysis time and 0.2 mol L^{-1} HCl). As it can be seen in Figure S1, peak currents and areas increased proportionally with time from 1 to 10 min ($y = 0.7388 \cdot x + 1.0255$, $R^2 = 0.996$, for 100 ppb solution, and $y = 0.4385 \cdot x + 0.4704$, $R^2 = 0.975$, for 10 ppb solution, where y = peak area and x = time). These results suggest that the CPE modified with MTTZ-HMS have high content of functional groups, and it is not saturated easily by Pb(II) ions. For a rapid analysis of lead, a 2 min preconcentration time was found to be sufficient for the studied concentration range, but longer time (5 min) may be required in samples with lower Pb(II) concentration.

3.3.-pH of the precontration solution

The pH effect on the voltammetric response for Pb(II) detection electrode E was studied in the range within 3–6.5 (100 μg L^{-1} Pb(II) solution, 5 min of preconcentration, 120 s of electrolysis time in 0.2 mol L^{-1} HCl). The pH of the solutions was adjusted by addition of either HNO_3 and/or NaOH; no buffer was employed to avoid any eventual complexation of lead by one of the buffer components. As can be seen in Figure 2, the peak currents and areas increased rapidly with increasing the pH (from 3 to 6.5). Experiments above pH 6.5 were avoided due to the formation of metal hydroxides complexes that may precipitate either on the wall of the electrolytic cell or on the electrode surface, thereby causing a significant decrease in the quantity of solution phase metal that may reach the electrode surface [17]. The low peak currents and areas observed at pH < 6.5 can be attributed to the protonation of the nitrogen atom of the MTTZ ligand, which is responsible for the coordination of the metal ion. So at low pH, only the sulphur atom of the MTTZ ligand is available for Pb(II) coordination.

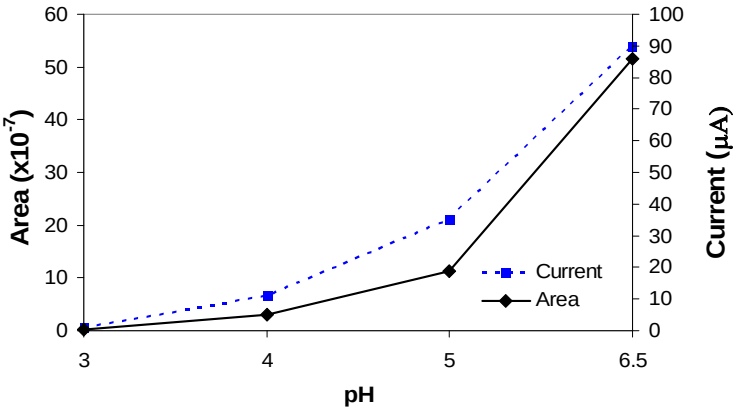


Figure 2. Effect of the pH on the voltammetric responses (area and current) of 100 µg L^{-1} Pb(II) solution. Other conditions as in Table 1.

3.4.-Electrolysis and stripping media and HCl concentration

Desorption of previously accumulated Pb(II) ions from the electrode surface to the electrode/solution interface must take place to enable the voltammetric detection. Concurrent with the desorption process, an electrolysis was performed by applying a negative potential of −1.2 V to reduce the desorbed Pb(II) ions to Pb(0), followed by the quantification of lead via SWV, which oxidizes the previously reduced Pb(0) to Pb(II). Thus, different electrolysis and stripping media were investigated (0.2 mol L^{-1} HCl, HNO_3, $HClO_4$, CH_3COOH and KCl). The experiments were performed with electrode E in a 100 µg L^{-1} Pb(II) solution (pH = 6.5) for 5 min of preconcentration time and 120 s of electrolysis time. Results shown in Figure 3a indicated that 0.2 mol L^{-1} CH_3COOH was ineffective as electrolysis and stripping media due to the incomplete leaching of the adsorbed Pb(II) from the chelating sites in the MTTZ-HMS to the electrode/solution interface. In consequence, in this medium the sensitivity of the electrode to Pb(II) ions was negligible. On the other hand, the most effective Pb(II) detection was accomplished by performing electrolysis and stripping steps in 0.2 mol L^{-1} HCl. As it can be seen in Figure 3b, a high peak current with very good shape was obtained in this medium.

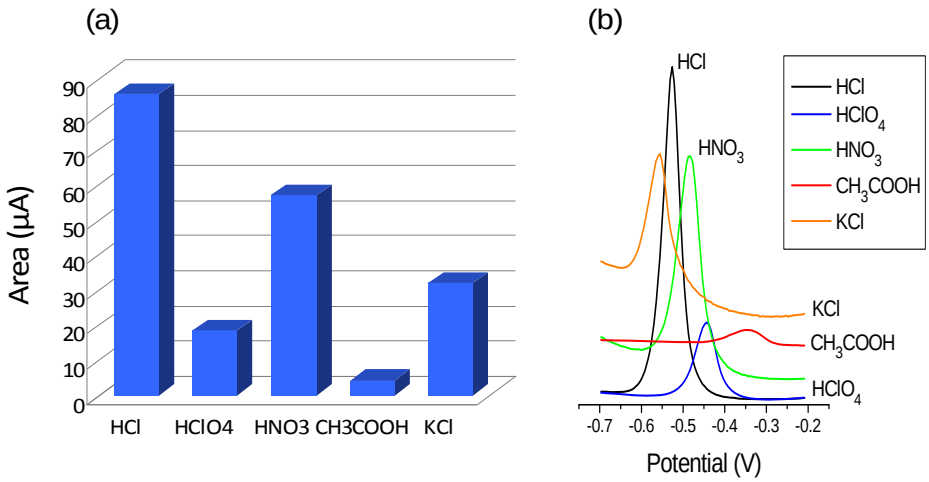


Figure 3. Effect of the electrolysis and stripping media on (a) the peak area and (b) the peak shape of 100 µg L^{-1} Pb(II) solution. Other conditions as in Table 1.

The effect of HCl concentration on the voltammetric response for Pb(II) detection was also investigated in the range from 0.1 to 0.5 mol L^{-1}. The results obtained indicated that increasing the HCl concentration did not significantly improve the sensitivity of the electrode to lead ions. Therefore, 0.2 mol L^{-1} HCl was chosen as electrolysis and stripping media because with it we achieved good stability and reproducibility in the voltammetric responses (less interference from hydrogen generation during electrolysis with respect to high HCl concentration), and on the other hand the need for regeneration of the electrode surface was lower with respect to 0.1 mol L^{-1} HCl.

3.5.-Electrolysis time

Figure S2 (see ESM) shows the effect of the electrolysis time on the voltammetric response for Pb(II) detection. The experiments were performed using electrode E in a 100 µg L^{-1} Pb(II) solution (pH = 6.5, 5 min of preconcentration time and 0.2 mol L^{-1} HCl as electrolysis and stripping media). The peak area increased as the electrolysis time increased from 0 to 30 s, and remained practically constant up to 30 s. However, the best data reproducibility was observed with 120 s, thus this electrolysis period was chosen as the optimal for lead deposition. The maximum voltammetric response obtained at 30 s of electrolysis time suggests a rapid desorption and diffusion rate of lead ions from inside the mesoporous of MTTZ-HMS to the electrode/solution interface. The poor reproducibility obtained at 150 s of electrolysis time is probably due to the generation of the hydrogen bubbles in the electrode surface that affected the stability of the voltammetric response.

3.6.-Calibration, limit of detection, limit of quantification and reproducibility

Figure S3 (see ESM) shows the calibration curve obtained for the voltammetric response measured, y = peak current, versus the metal ion concentration under the optimized conditions, x ($y = 0.8626 \cdot x + 0.0311$). As it can be seen in Figure S3a, the voltammetric response was linear to the Pb(II) concentration with a R^2 of 0.998 in the range of concentration studied (from 1 to 100 μg L^{-1}).

The limit of detection (LOD = 0.3 μg L^{-1}) and limit of quantification (LOQ = 1 μg L^{-1}) were determinated as the concentration of Pb(II) that would give a signal 3 or 10 times the background noise, respectively. The background noise was measured under the optimized conditions. It is well known that in CPE modified with silica materials the LOD improves significantly when the preconcentration time increases. It is interesting to note that very low LOD and LOQ were obtained in this work with a relatively low preconcentration time (5 min).

Finally, the developed electrode showed very good reproducibility for Pb(II) detection. For a single electrode surface, the RSD for the detection of 100 μg L^{-1} Pb(II) solution after 5 min preconcentration time was less to 5% for seven consecutive determinations. Using the same electrode surface for three consecutively days, the RSD was less to 10% (three determinations for each day). For a six different electrode surfaces, used in different days, the RSD was less to 9% (three determinations for each electrode surface).

Some other studies using mesoporous silicas as CPE modifiers have been published for Pb(II) determination as shown in Table S2 (see ESM). Over the existing MSM-CPEs, the electrode prepared in this work has to main advantages: a lower LOD (0.3 μg L^{-1} after 5 min preconcentration time) and a better reproducibility (RSD < 9 for six electrode surfaces used in different days). Long term stability was also obtained since the same electrode surface can be reused for 25 measurements with minimal degradation.

3.7.-Interferences and detection of other metals ions

Cations may be considered as interferences to the voltammetric detection of Pb(II) if they compete with Pb(II) for the binding sites on the MTTZ-HMS material during the preconcentration step or if its peak response overlaps the Pb(II) peak in the detection step. On the other hand, some anions present in real samples have the capability to complex with many metals ions. Consequently, they may compete with the immobilized MTTZ ligand for the Pb(II) ions reducing the voltammetric detection of lead. Consequently, the selectivity of the developed electrode was evaluated by

intentionally introducing some cations and anions into the Pb(II) solution during the preconcentration step. These ions were chosen because they exhibit redox activity in roughly the same potential range as Pb(II) and/or existing in real waters samples. Table 2 summarizes the interference effect of these ions on the Pb(II) detection. The experiments were performed under the optimised conditions with a solution containing 100 μg L^{-1} of Pb(II) and 100 μg L^{-1} of the interference. Based on the results showed in Table 2, Cu(II), Ca(II) and Mg(II) did not interfere with lead detection, so these metals do not compete with lead for the binding sites on the MTTZ-HMS at the assayed concentration. On the other hand, Zn(II), Ni(II) and Cd(II) did interfere producing an increase in the lead signal.

Table 2
Interference study for the detection of 100 μg L^{-1} Pb(II) in the presence of 100 μg L^{-1} of other ions.

Interference	Pb peak area (%)
none	100 ± 5
Cu^{2+}	109 ± 7
Cd^{2+}	124 ± 7
Ni^{2+}	178 ± 13
Zn^{2+}	190 ± 13
Ca^{2+}	87 ± 11
Mg^{2+}	93 ± 14
SO_4^{2-}	49 ± 10
PO_4^{3-}	28 ± 11
CO_3^{2-}	67 ± 5
$HCOO^-$	73 ± 6
Cl^-	121 ± 6

In order to simulated waters of medium to high hardness, additional experiments were carried out increasing the level of Ca(II) and Mg(II) (1, 10 and 100 mg L^{-1}) and decreasing the level of Pb(II) to 10 μg L^{-1} (according the future European regulations). Result obtained shown that Ca(II) did not interfere with lead detection at 1 mg L^{-1} (Pb peak area = 97 ± 5%) while Mg(II) at the same concentration reduces the signal considerably (63 ± 5%). On the other hand, when Ca(II) and Mg(II) concentration was 10 mg L^{-1} the Pb(II) signal was reduced to 48 ± 6% and 32 ± 13%, respectively. In these conditions the calculated LOD and LOQ increased to 1.4 and 8.6 μg L^{-1}, respectively ($y = 4.8 \cdot 10^{-10} \cdot x + 8.4 \cdot 10^{-10}$, $R^2 = 0.998$, where y is the peak area and x the Pb(II) concentration in μg L^{-1}). Increasing the Ca(II) and Mg(II) concentration to 100 mg L^{-1} the Pb(II) signal disappeared. Although the mechanism of the interference is not definitely known, the most likely explanation is that, when present in sufficient excess, other cations possessing a

limited capacity for complexation with Pb(II) for the available organofunctional groups attached to the surface of the MTTZ-HMS.

In relation with anions, with the exception of Cl^-, the presence of these ionic species in the medium produced an important reduction in the Pb(II) peak area (between 39 and 77% of reduction). The means that these anions have the capability to complex the Pb(II) ions under the assayed conditions and the bound fraction of Pb(II) is not accessible to determination.

As the redox potential of copper and cadmium are different from lead, the CPE modified with MTTZ−HMS was tested for simultaneous detection of different metal ions. Figure 4 shows a representative voltammogram of 100 ppb of Cd(II), Pb(II) and Cu(II) (5 min of preconcentration time, 0.2 mol L^{-1} HCl as electrolysis and stripping media, 120 s of electrolysis time). The high functionalization degree of the MTTZ-HMS minimizes the competition among these metal ions, allowing them to be detected simultaneously without sacrificing the response intensity.

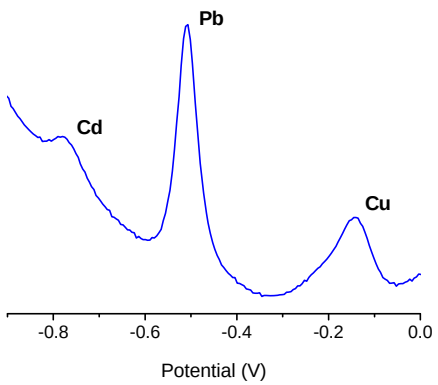


Figure 4. Representative voltammogram of the simultaneous detection of 100 µg L^{-1} of Pb(II), 100 µg L^{-1} of Cu(II) and 100 µg L^{-1} of Cd(II). Other conditions as in Table 1.

3.8.-Determination of Pb(II) in the spiked real water samples

In order to evaluate the performance of the developed MSM-CPE by practical analytical applications, the determination of Pb(II) was carried out in tap water (Canal Isabel II, Madrid) and underground water (Toledo) samples, without any pre-treatment. The Pb(II) concentration was determined by the standard addition method to compensate the matrix effect from real water that contains high concentration of nitrates and other ions. Accuracy was assessed by recovery test and

Pb(II) was added to the water samples to achieve a final concentration of 10 μg L^{-1} (according the future European Regulation). As can be seen in Table 3, good correlations between amounts determined and added were obtained (recovery values of 100 ± 4% for tap water and 98 ± 4% for underground water), suggesting that the MSM-CPE developed can be used for practical analysis of Pb(II) in real water samples.

Table 3
Determination of Pb(II) on a carbon paste electrode modified with functionalized mesoporous silica in tap and underground water samples [a].

Sample	Spiked (μg L^{-1})	Found (μg L^{-1})	Recovery (%)	RSD (n = 3)
Tap water	0.0	0.0		
	10.0	10.0	100	4
Underground water	0.0	0.0		
	10.0	9.8	98	4

[a] Square-wave voltammetric stripping scans a frequency of 100 MHz with a pulse amplitude of 50 mV and a potential step height of 5 mV. Experiments were performed using electrode E (20% MTTZ-HMS, 20% mineral oil and 60% carbon paste), 5 min of preconcentration time, 0.2 mol L^{-1} HCl as electrolysis and stripping media and 120 s as the electrolysis time

4.-CONCLUSIONS

Functionalized mesoporous silicas are an intelligent solution to prepare chemically modified electrodes because its high surface area, high porosity, controllable and narrowly distributed pore size, ordered pore arrangement and high functionalization degree. In this study, we have demonstrated that a carbon paste electrode modified with MTTZ-HMS is a good option to determine Pb(II) by anodic stripping voltammetry in natural waters. The electrode prepared has two main advantages: a low LOD (0.3 μg L^{-1} after 5 min preconcentration time) and a good reproducibility, thus has a great potential to be used in the development of portable analyzers for monitoring heavy metal ions in water samples.

Acknowledgements

We gratefully acknowledge financial support from the Comunidad de Madrid and URJC (project URJC-CM-2007-CET-1491) and Ministerio de Ciencia e Innovación (project CTQ2008-05821/PPQ).

5.-REFERENCES

[1] Baird C., in **Environmental chemistry,** ed. W. H. Freeman and Company, *University of Western Ontario, New York,1999.*

[2] USDHHS; **Toxicological profile for lead.** *Agency for Toxic Substances and Disease Registry, Atlanta, 2005.*

[3] Wang J.; **Analytical electrochemistry,** *Willey, New York, 2000.*

[4] Wang J.; **Stripping analysis.** *VCH Publishers Inc., Florida,1985.*

[5] Walcarius A.; **Analytical applications of silica-modified electrodes. A comprehensive review.** *Electroanalysis 10 (1998) 1217-1235.*

[6] Walcarius A.; **Electroanalysis with pure, chemically modified and sol-gel-derived silica-based materials.** *Electroanalysis 13 (2001) 701-718.*

[7] He L., Toh C.-S.; **Recent advances in analytical chemistry – A material approach.** *Anal. Chim. Acta 556 (2006) 1-15.*

[8] Kresge C. T., Leonowicz M. E., Roth W. J., Vartulli J. C., Beck J. S.; **Ordered mesoporous molecular sieves synthesized by a liquid-crystal template mechanism.** *Nature 359 (1992) 710-712.*

[9] Beck J. S., Vartulli J. C., Roth W. J., Leonowicz M. E., Kresge C. T., Schmitt K. D., Chu C. T. W., Olson D. H., Sheppard E. W., McCullen S. B., Higgins J. B., Schlenker J. L.; **A new family of mesoporous molecular sieves prepared with liquid crystal templates.** *J. Am. Chem. Soc. 114 (1992) 10834-10843.*

[10] Fryxell G. E., Cao G.; **Environmental applications of nanomaterials.** *Imperial College Press, London, 2007.*

[11] Walcarius A.; **Impact of mesoporous silica-based materials on electrochemistry and feedback from electrochemical science to the characterization of these ordered materials.** *C. R. Chim. 8 (2005) 693-712.*

[12] Walcarius A., Lüthi N., Blin J. L., Su B. L., Lamberts L.; **Electrochemical evaluation of polysiloxane-inmobilized amine ligands for the accumulation of copper(II) species.** *Electrochim. Acta 44 (1999) 4601-4610.*

[13] Yantasee W., Lin Y., Zemanian T. S., Fryxell G. E.; **Voltammetric detection of lead(II) and mercury(II) using a carbon paste electrode modified with thiol self-assembled monolayer on mesoporous silica (SAMMS).** *Analyst 128 (2003) 467-472.*

[14] Yantasee W., Lin Y., Li X., Fryxell G. E., Zemanian T. S. Viswanathan V. V.; **Nanoengineered electrochemical sensor based on mesoporous silica thin film functionalized with thiol-terminated monolayer.** *Analyst 128 (2003) 899-904.*

[15] Walcarius A., Etienne M., Sayen S., Lebeau B.; **Grafted silicas in electroanalysis: amorphous versus ordered mesoporous materials.** *Electroanalysis 15 (2003) 414-421.*

[16] Walcarius A., Sayen S., Gérardin C., Hamdoune F., Rodehüser L.; **Dipeptide-functionalized mesoporous silica spheres.** *Colloid Surf. A-Physicochem. Eng. Asp. 234 (2004) 145-151.*

[17] Yantasee W., Lin Y., Fryxell G. E., Busche B. J.; **Simultaneous detection of cadmium, copper, and lead using a carbon paste electrode modified with carbamoylphosphonic acid self-assembled monolayer on mesoporous silica (SAMMS).** *Anal. Chim. Acta 502 (2004) 207-212.*

[18] Yantasee W., Fryxell G. E., Conner M. M., Lin Y.; **Nanostructured electrochemical sensors based on functionalized nanoporous sílica for voltammetric analysis of lead, mercury and copper.** *J. Nanosci. Nanotechnol. 5 (2005) 1537-1540.*

[19] Yantasee W.,Timchalk C., Fryxell G. E., Dockendorff B. P., Lin Y.; **Automated portable analyzer for lead(II) based on sequential flow injection and nanostructures electrochemical sensors.** *Talanta 68 (2005) 256-261.*

[20] Yantasee W., Deibler L. A., Fryxell G. E., Timchalk C., Lin Y.; **Screen-printed electrodes modified with functionalized mesoporous silica for voltammetric analysis of toxic metal ions.** *Electrochem. Commun. 7 (2005) 1170-1176.*

[21] Cesarino I., Marino G., Matos J. R., Cavalheiro E. T. G.; **Evaluation of a carbon paste electrode modified with organofunctionalised SBA-15 sílica in the determination of copper.** *Eclet. Quim. 32 (2007) 23-28.*

[22] Cesarino I., Marino G., Matos J. R., Cavalheiro E. T. G.; **Evaluation of a carbon paste electrode modified with organofunctionalised SBA-15 nanostructured sílica in the simultaneous determination of divalent lead, copper and mercury ions.** *Talanta 75 (2008) 15-22.*

[23] Yantasee W., Charnhattakom B., Fryxell G. E., Lin Y., Timchalk C., Addleman R. S.; **Detection of Cd, Pb, and Cu in non-pretreated natural waters and urine with thiol functionalized mesoporous silica and Nafion composite electrodes.** *Anal. Chim. Acta 620 (2008) 55-63.*

[24] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Functionalized HMS mesoporous silica as solid phase extractant for Pb(II) prior to its determination by flame atomic absorption spectrometry.** *J. Sep. Sci. 30 (2007) 1556-1567.*

[25] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Synthesis and characterization of novel mesoporous silicas of the MSU-*X* family for environmental applications.** *J. Nanosci. Nanotechnol. 9 (2009) 4901-4909.*

[26] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Solid phase extraction of Pb(II) in water samples using a new hybrid inorganic-organic**

mesoporous silica prior to its determination by FAAS. *Microchim. Acta 165 (2009) 291-298.*

[27] Mercier L., in **Studies in surface science and catalysis,** ed. Sayari A.; *Elsevier Science vol. 129, Amsterdam, 2000.*

6.-ELECTRONIC SUPPLEMENTARY MATERIAL (ESM)

Table S1
Effect of MTTZ-HMS or HMS content of the modified carbon paste electrodes on the peak current of Pb(II), 5 min preconcentration time, 0.2 mol L^{-1} HCl and 120 s electrolysis time.

Electrode	Silica % (w/w)	Mineral oil % (w/w)	Carbon paste % (w/w)	Pb concentration (µg L^{-1})	Peak current (µA)
A	5[a]	20	75	500	0
B	10[a]	0	90	500	0
C	10[a]	20	70	500	0.085
D	10[a]	30	60	500	0
E	20[a]	20	60	100	0.5
F	30[a]	20	50	100	0.045
G	10[b]	20	70	666	0.02
H	10[b]	30	60	666	0
I	20[b]	20	60	666	0

[a] MTTZ –HMS.
[b] HMS.

When the weight percent of MTTZ-HMS was kept constant at 10%, the best voltammetric response was obtained with a 20% of oil (electrode C). With a 0% of oil (electrode B) no voltammetric signal could be observed, because the electrode material was not compact enough. On the other hand, the voltammetric response decreased with more than 20% of oil, which can be attributed to a very high hydrophobicity of the electrode surface that reduces the interaction between the metal ions and the ligand [26]. Then, keeping constant the weight percent of oil at 20%, the voltammetric response increased by about six-fold when the MTTZ-HMS content increased from 10% (electrode C) to 20% (electrode E) due to the increase in the number of the binding sites for Pb(II). With higher percentage of MTTZ-HMS (30% in electrode F), the reliability of the electrode was poor and the electrode material was not compact enough.

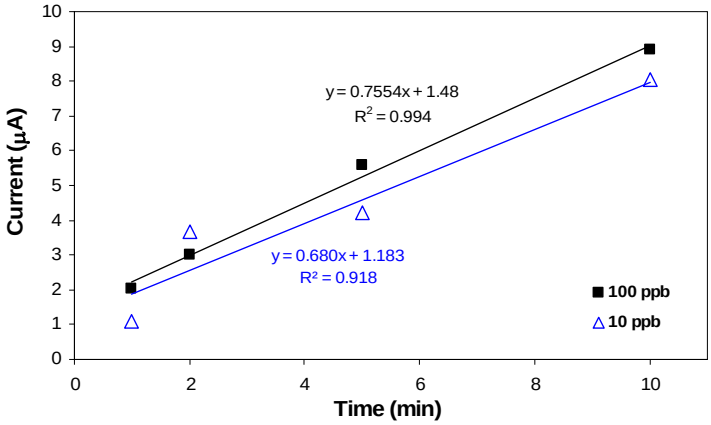


Figure S1. Effect of preconcentration time on the voltammetric response, current, of 10 µg L^{-1} and 100 µg L^{-1} Pb(II) solutions. Other conditions as in Table 1.

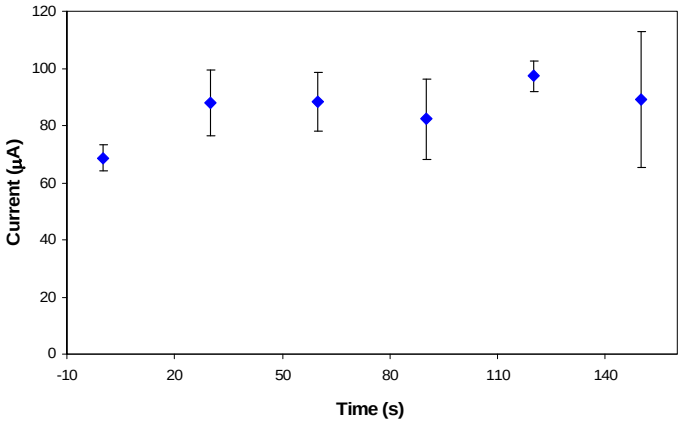


Figure S2. Effect of electrolysis time on the voltammetric response (current) of 100 µg L^{-1} Pb(II) solution. Other conditions as in Table 1.

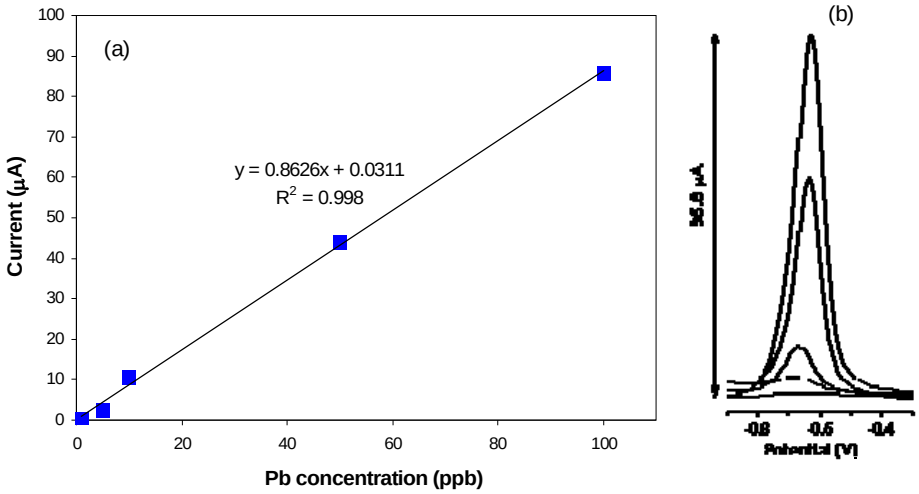


Figure S3. (a) Calibration curve for Pb(II) measured after 5 min preconcentration time. (b) Representative voltammograms of different Pb(II) solutions in ascending order of concentration. Other conditions as in Table 1.

Comparison of the CPE modified with MTTZ-HMS with others MSM-CPEs of Pb (II) detection

Some other studies using mesoporous silicas as CPE modifiers have been published for Pb(II) determination as shown in Table S2. Yantasee et al. [13] used SH-SAMMS (thiol terminated self-assembled monolayer on mesoporous silica) to prepare a MSM-CPE for simultaneous detection of Pb(II) and Hg(II). The linear response obtained for Pb(II) was between 10 - 1500 µg L^{-1} and the LOD was 0.5 µg L^{-1}, for 20 min of preconcentration time. In other study [17], these authors used the same mesoporous silica modified with carbamoylphosphonic acid (Ac-Phos) to develop a MSM-CPE for simultaneous detection of Cd(II), Cu(II) and Pb(II). The voltammetric response of this electrode increased linearly with Pb(II) concentrations ranging from 10 to 200 µg L^{-1} with a LOD of 0.5 µg L^{-1} after 20 min preconcentration time. This chemically modified CPE had good reproducibility for both single and various electrode surfaces (5 and 12%, respectively). The same material has been used to develop an automated portable analyzer for Pb(II) based on sequential flow injection [19] and to develop a screen-printed CPE for voltammetric analysis of this metal ion [20]. In other study, Yantasee et al. [18] have used glycinylurea-SAMMS to prepare a MSM-CPE electrode for the determination of Pb(II), obtaining a linear response from 2.5 to 50 µg L^{-1} after 2 min preconcentration time with a LOD of 1 µg L^{-1} in the same conditions. Recently, in 2008, Cesarino et al. [22] have used a CPE modified with 2-benzothiazolethiol-SBA-15 silica for simultaneous detection of Pb(II), Cu(II), and Hg(II). Calibration curve for Pb(II) was found linear in the range 60 – 1.400 µg L^{-1} with a LOD equal to 8 µg L^{-1} after 3 min of preconcentration time.

Table S2
Comparison of CPE modified with MTTZ-HMS with others CPE modified with functionalized mesoporous silica.

Analytes	Electrode	Pb (II) linear range	LOD – Preconcentration time	Reproducibility	Reference
Pb(II), Hg(II)	SH-SAMMS	10-1500 μg L^{-1}	0.5 μg L^{-1} - 20 min	-	[13]
Cd(II), Cu(II), Pb(II)	Ac-Phos SAMMS	10-200 μg L^{-1}	0.5 μg L^{-1} - 20 min	1 electrode surface (n=7): 5% RSD 5 electrode surfaces: 12% RSD	[17]
Pb(II)[a]	Ac-Phos SAMMS	1-25 μg L^{-1}	-	1electrode surface (n=7): 2.5% RSD	[19]
Pb(II)[b]	Ac-Phos SAMMS	0-100 μg L^{-1}	0.91 μg L^{-1} -5 min	1 electrode surface (n=6): 5% RSD 5 electrode surfaces: 10% RSD	[20]
Pb(II), Cu(II), Hg(II)	Gly-UR SAMMS	2.5-50 μg L^{-1}	1 μg L^{-1} - 2 min	-	[18]
Pb(II), Cu(II), Hg(II)	2-BTT-SBA-15	60 – 1.400 μg L^{-1}	8 μg L^{-1} - 3 min	-	[22]
Pb(II), Cu(II), Cd(II)	MTTZ-HMS	1-100 μg L^{-1}	0.3 μg L^{-1} - 5 min	1 electrode surface (n=7): 5% RSD 1 electrode surface 3 days: 10% RSD 6 electrode surfaces: 9% RSD	Present work

Abreviations: SH-SAMMS: thiol terminated self-assembled monolayer on mesoporous silica (MCM-41); Ac-Phos SAMMS: acetamide phosphonic acid self-assembled monolayer on mesoporous silica (MCM-41); Gly-UR SAMMS: glycinylurea self-assembled monolayer on mesoporous silica (MCM-41); 2-BTT-SBA-15: 2-benzothiazolethiol-SBA-15;
-: Not data.
[a] Flow injection
[b] Sreen-printed

DEVELOPMENT OF SCREEN-PRINTED CARBON ELECTRODES MODIFIED WITH FUNCTIONALIZED MESOPOROUS SILICA NANOPARTICLES: APPLICATION TO VOLTAMMETRIC STRIPPING DETERMINATION OF Pb(II) IN NON-PRETREATED NATURAL WATERS.

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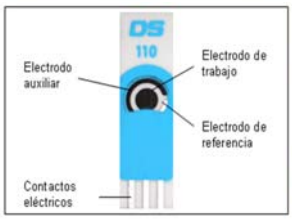

DEVELOPMENT OF SCREEN-PRINTED CARBON ELECTRODES MODIFIED WITH FUNCTIONALIZED MESOPOROUS SILICA NANOPARTICLES: APPLICATION TO VOLTAMMETRIC STRIPPING DETERMINATION OF Pb(II) IN NON-PRETREATED NATURAL WATERS.

ABSTRACT

A screen-printed carbon electrode modified with functionalized mesoporous silica nanoparticles (MTTZ-MSU-2) was developed and evaluated for reliable quantification of trace Pb(II) ions by anodic stripping square wave voltammetry in non-pretreated natural waters. The optimal operating conditions were 5 min preconcentration time and 120 s electrolysis time in HCl 0.2 M. The electrode displayed excellent linear behavior in the concentration range examined (1 – 30 μg L^{-1}) with a limit of detection of 0.1 μg L^{-1}. The screen-printed carbon modified electrode has long service time and good single and inter-electrode reproducibility. Applicability to spiked drinking water, river water and groundwater was demonstrated without any sample pre-treatment (recoveries between 97% and 106%, RSD 4 – 7%). On the basis of the present data, mercury-free screen-printed electrodes modified with functionalized mesoporous silicas have the potential to become the next-generation analyzers for decentralized heavy metal monitoring in environmental samples.

1.-INTRODUCTION

The determination of heavy metals in natural waters is of environmental concern due to its toxicity and their accumulation in the environment and living organisms [1]. Because of the risk to human health and a variety of environmental problems, the extent of heavy metal pollution needs to be assessed as it occurs, so that it can be remedied before it becomes hazardous. To carry out this, there is a need to develop simple and cheap sensing devices, which are field-portable and facilitate quicker decision-making for remediation of polluted sites [2]. On the other hand, to perform on-site analyses at the point of interest is of special importance the development of "environmentally friendly" analytical methodologies that reduce or eliminate the use of toxic reagents [3].

Analysis of heavy metals in environmental samples remains a challenging task since these metal ions are present at very low levels in the samples being very complex in nature. The search is therefore ongoing to find new sensing materials with suitable recognition elements that can respond selectively and reversibly to specific metal ions [4]. This avoids or reduce sample pre-treatment that is time-consuming, inappropriate for on-site environmental monitoring and may risk adding metals to the samples from reagents and equipments.

Conventional analytical techniques for heavy metals determination in environmental samples include atomic absorption spectroscopy (AAS), inductively coupled plasma-atomic emission spectroscopy (ICP-AES) and inductively coupled plasma-mass spectrometry (ICP-MS). Although these techniques are very powerful for heavy metal analysis, they are known to be expensive and not suitable for on-site monitoring owing to their size, and require highly trained personnel. Electroanalytical techniques, especially anodic stripping voltammetry (ASV), satisfy many of the requirements for on-site monitoring of heavy metals due to the low cost, low-power requirements, speed of analysis and sensitivity. The remarkable sensitivity obtainable with stripping analysis (in low ppb levels) is attributed to the preconcentration step, in which trace metals are accumulated onto the working electrode surface [5]. This is then followed by the measurement step, in which the metals are stripped away from the electrode during an appropriate potential scan.

With the advent of battery-powered computers and potentiostats/ galvanostats, as well as the increasing application of micro-fabrication technologies like screen-printing to produce electrodes, portable and compact stripping analyzers are gaining popularity for heavy metals analysis because of their easy-of use and simplicity. Replacement of conventional electrochemical cells by screen-printed carbon electrodes (SPCEs) connected to miniaturized potentiostats is a main trend in the shift of lab electrochemical equipments to hand-held field analyzers [6].

SPCEs produced by printing different inks on plastic or ceramic supports are gaining widespread applicability for electrochemical analysis, especially in environmental, biomedical and industrial samples. The applicability of screen-printed electrochemical sensors for heavy metals determination was extensively reviewed by Palchetti et al. [7] and by Honeychurch and Hart [6]. More recently, Palchetti et al. [8] developed a new miniaturized electrochemical carbon modified sensor for stripping analysis of Pb(II), Cd(II) and Cu(II). The graphite working electrode was modified with cellulose-derivative mercury coating and on-site environmental applications were given. As Hg is toxic, its incorporation into sensors poses problems, particularly from the viewpoint of disposability and use; consequently, other mercury-free SPCEs have been developed by employing alternative metals films (e.g. gold, bismuth) which are less toxic. Palchetti group [9] proposed a gold-based SPCEs able to detect Pb(II) with good sensitivity and reproducibility in river water samples. Kadara and Tothill [10] prepared a Bi_2O_3-modified electrode by directly modifying the screen-printing graphite-carbon ink with the solid oxide for the voltammetric stripping analysis of Pb(II) in soil extracts and wastewater samples. More recently, Granado et al. [11] modified commercial SPCEs by passive adsorption of metallic Bi nanoparticles onto the carbon working electrode surface for the voltammetric stripping detection of Zn(II), Cd(II) and Pb(II) in water samples.

In the last few years, research work has progressed on to the development of electrochemical sensors based on modification of the screen-printed working electrode with other compounds. This is carried out by incorporating these modifiers into the graphite ink prior to the printing step or drop coating the modifying reagents onto the transducer surface. These electrode surfaces may exploit a chemical reaction in open circuit for the accumulation of a selected species prior to its voltammetric quantification. Several modifiers have been used in the construction of screen-printed carbon modified electrodes (SPCMEs) and the selection is determined by its application. Modifiers commonly used are organic polymers [12, 13], ligands, [14, 15] and, more recently, functionalized silicas [16, 17].

Functionalized silicas have gained considerable interest in the field of electroanalytical chemistry in recent years [18, 19] because they combine in a single solid both the mechanical stability of a rigid inorganic framework with the chemical reactivity of organic groups covalently bonded to the porous structure. They have found applications as electrode modifiers for the accumulation/voltammetric analysis of electroactive analytes [18, 19]. In this case, the resort to order mesoporous functionalized silica has been reported as one possibility to increase the sensitivity of the detection by improving diffusion processes that constitute the rate-determining step in preconcentration electroanalysis. These materials are characterized by a regular three-dimensional arrangement of mesoporous channels of monodisperse dimension (2 – 10 nm). The use of functionalized mesoporous silicas in electrochemical sensors is relatively new and yet attractive, since they enable the development of solid-state and metal-free electrodes. At present, there are few reports on SPCEs modified with

functionalized mesoporous silicas. Yantasee et al. [16] reported the preparation of a SPCME with Ac-Phos SAMMS (acetamide phosphonic acid self-assembled monolayer on mesoporous MCM-41 silica) for Pb(II) analysis. Later, the same authors [17] reported the preparation of a SPCME with salicylamide on MCM-41 mesoporous silica (Sal-SAMMS) for Eu(III) determination. Although these works demonstrate the advantages of SPCMEs with functionalized mesoporous silica (e.g. mercury-free, reusable, low cost and as sensitive as Hg-based SPCEs), both were carried out with lab-simulated samples (metal spiked buffered solutions) instead real complex samples.

Mesoporous silicas like MCM-41, SBA-15 and HMS have been widely used for the adsorption of different heavy metals in waters [20–26], but adsorption studies using functionalized MSU-X silicas are comparatively scarce. Compared to MCM-41 and SBA-15 materials, that exhibit either hexagonal or cubic symmetries, the structure and particle morphology of MSU-X silicas is expected to enhance the diffusion rate of the heavy metals into the pores [27–30]. Other potential advantages of MSU-X mesoporous family over other materials are lower reagent and energy costs (synthesis at ambient pressure and moderate temperature), low surfactant toxicity and biodegradability. All these points decrease the overall cost and environment impact of this synthesis process.

The goal of the present study is to prepare a new solid-state and metal-free electrode by modifying the surface of a commercial screen-printed carbon electrode with nanoparticles of functionalized mesoporous silica (MTTZ-MSU-2). This work also aims to broaden the area of analysis of real samples by applying the developed SPCME to real natural water samples.

2.-EXPERIMENTAL

2.1.-Chemicals and reagents

HCl Hiperpur (35%) and HNO_3 Hiperpur (69%), $NaH_2PO_4 \cdot 2H_2O$ (99%) and NaCl (99%) were obtained from Panreac. Na_2CO_3 (99%), $Ca(NO_3)_2 \cdot 4H_2O$ (98%), $Mg(NO_3)_2 \cdot 6H_2O$ (98%) and CH_3–COONa (99%) were obtained from Scharlab, $Ni(NO_3)_2 \cdot 6H_2O$ (99%, Merck), $Zn(NO_3)_2 \cdot 6H_2O$ (98%, Accros) and Na_2SO_4 (99%, Probus) were of analytical grade and used without further purification. A certified ICP standard stock solution of Pb(II) at 1.000 g L^{-1} ±0.5% was purchased from Metrohm. ICP stock solutions of Cu(II) and Cd(II) at 1.000 g L^{-1} were purchased from Fluka. Single-component lead solution and multi-component metal solution (lead, copper and cadmium) were prepared daily by diluting the stock solutions with ultrapure water (resistivity 18.2 MΩ cm) obtained from a Millipore Milli-Q-System (Waters, USA). For hybrid mesoporous silica preparation, tetraethylorthosilicate (TEOS, 98%) and Tergitol® NP-9 were purchased from Sigma–Aldrich, sodium fluoride (99%, Fluka), 3-chloropropyltriethoxysilane (CPTS, 95%), triethylamine and 5-mercapto-1-methyltetrazole (MTTZ, 98%), all from Sigma–Aldrich, were used as supplied. Organic solvents (toluene, diethyl ether, ethanol and hexane) were purchased from SDS (France). These solvents were distilled and dried before use according to conventional literature methods [29]. Glassware was soaked in HNO_3 (50% diluted with Milli-Q water) overnight and cleaned with Milli-Q water before use.

2.2.-Samples

A drinking water sample was collected from a running water tap in Madrid (Canal de Isabel II) and used immediately. River water (Alberche river) and groundwater (collected in Escalona, Toledo) were filtered through a 0.45 µm membrane filter (Millipore Membrane filters 0.45 µm HA) to remove suspended particles and stored at −26 ºC prior to use in polyethylene bottles. The three waters were analysed without pre-treatment and pH was not adjusted to show the versatility of the developed SPCME. This will minimize the potential sample contamination leading to time and reagents savings.

2.3.-Apparatus

Anodic stripping square wave voltammetry (SW-ASV) measurements were performed on a hand-held, battery-powered Bipotentiostat model µSTAT200 (Dropsens, Spain) equipped with a sensor connector (model DSC Dropsens, Spain) for connecting the screen-printed electrode strip electronically to the bipotentiostat. A PC with DropView software for Windows was used to control the instrument, plot

the measurements and perform the analysis of results. Figure 1 shows a screen-printed electrode (SPCE) strip (model 110 Dropsens) consisting of three built-in electrodes on a ceramic substrate (length 34 mm x width 10 mm x height 0.5 mm). The strips present a 4 mm diameter disk screen-printed carbon working electrode, a screen-printed carbon counter electrode and a silver reference electrode. An insulating layer serves to delimit the working area and the silver electric contacts.

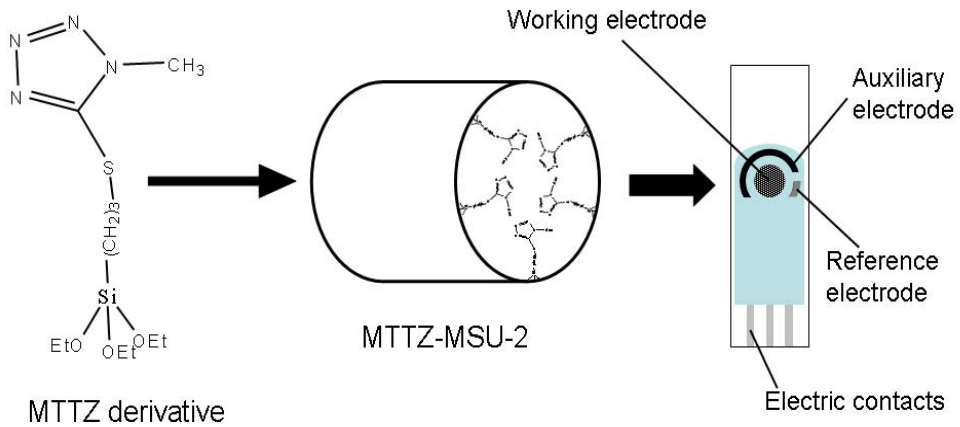

Figure 1. MTTZ-MSU-2 modified screen-printed carbon electrode strip.

For hybrid mesoporous silica characterization, X-ray diffraction (XRD) patterns were obtained on a Philips X'Pert diffractometer. N_2 gas adsorption–desorption isotherms were recorded using a Micromeritics TriStar 3000 manometric porosimeter. The surface specific areas were calculated by the Brunauer–Emmett–Teller (BET) method. Pore size distributions were obtained using the Barret–Joyner–Halenda (BJH) model on the adsorption branch. Scanning electron micrographs (SEM) and morphological analysis were obtained on a XL30 ESEM Philips microscope with an energy dispersive spectrometry system. Conventional transmission electron microscopy (TEM) was carried out with a TECNAI 20 Philips electron microscope operating at 200 kV. Elemental analysis (%C, %N and %S) was performed at Alcalá de Henares University (Madrid, Spain). The thermal stability of the modified mesoporous silicas was studied with a Setsys 18A (Setaram) thermogravimetric analyzer 100 µL (synthetic air atmosphere from 25 ºC to 800 ºC, 10 ºC min^{-1}).

2.4.-Synthesis of hybrid mesoporous silica nanoparticles

Hybrid mesoporous silica nanoparticles of the MSU-X family (MSU-2-type) were prepared according to Perez-Quintanilla et al. [29]. Briefly, TEOS was added to a stirred 0.08 M solution of Tergitol® NP-9 in Milli-Q water to obtain a milky

suspension that was aged without agitation for 20 h. In a second step, a 0.24 M sodium fluoride solution was added dropwise with stirring to obtain a NaF/TEOS molar ratio of 0.025/1. The solution was placed in a bath with agitation at 55 ºC for 48 h. The obtained product was filtered off, washed with Milli-Q water, dried and calcined in air at 600 ºC for 12 h. 20.00 g of activated MSU-2 silica (5 h at 160 ºC under high vacuum) were reacted with 20.00mL of a 5-mercapto-1-methyltetrazole derivative in dry toluene for 48 h under reflux conditions and nitrogen atmosphere. The resulting hybrid mesoporous silica nanoparticles (MTTZ-MSU-2) were filtered off and washed with toluene, ethanol and diethyl ether. Finally, the product was heated for 4 h at 110 ºC under vacuum.

2.5.-Preparation of working electrode

To modify a commercial SPCE strip with MTTZ-MSU-2 (Figure 1) a cellophane mask was used to cover all the elements of the strip leaving the working electrode uncovered. Then a known quantity of MTTZ-MSU-2 was mixed with a known quantity of carbon paste (Metrohm, No 6.2801.000) and mineral oil (Aldrich) to achieve a 20:60:20% (w/w/w), respectively. The paste was then manually printed for some seconds onto the working electrode surface as a thin layer and polished with filter paper. Finally, the mask was removed very carefully. For comparative purpose, a modified SPCE strip was prepared according to the same procedure but adding non-functionalized MSU-2 silica to the paste. Prior to use, freshly made electrodes were activated by an electrolysis step, applying a potential of −1.2 V for 120 s in 50 μL of 0.2 M HCl, and a stripping step registered by sweeping a square wave potential toward positive values (from −1.2 V to +0.4 V) until the background was stable (2 – 3 cycles).

2.6.-Voltammetric measurements

SW-ASV was operated at a frequency of 100 Hz with pulse amplitude of 50 mV and a potential step height of 5 mV. Table 1 summarizes the operating conditions optimized for the voltammetric experiments. During preconcentration, the SPCME strip was immersed in 15 mL of Pb(II) solution efficiently stirred for 5 min (300 rpm) at open circuit, leaving the electrical connection pads out of the solution. The SPCME strip was then rinsed with Milli-Q water and connected to the potentiostat via the sensor connector. A negative potential (−1.2 V) was applied to the electrode for 120 s as soon as 50 μL of 0.2 M HCl was dropped onto the SPCME strip, covering the three electrodes. Anodic stripping voltammetric curves were registered by sweeping a square wave potential toward positive values (from −1.2 V to +0.4 V). All measurements were made at room temperature and under an atmospheric environment. After each measurement, to ensure that no metal was still left on the electrode surface, a regeneration of the electrode was performed by an electrolysis step, applying a potential of −1.2 V for 120 s in 50 μL of 0.2 M HNO_3, and a stripping step registered by sweeping a square wave potential toward positive values

(from −1.2 V to +0.4 V). Each measurement was performed in triplicate and the average value was reported.

Table 1
Operating conditions optimized for the square wave voltammetry experiments.

	Preconcentration	Electrolysis	Stripping	Regeneration
Solution/ electrolyte	1 - 50 μg L^{-1}	HCl 0.2 mol L^{-1}	HCl 0.2 mol L^{-1}	HNO_3 0.2 mol L^{-1}
Immersion time	5 min	120 s	-	-
Solution volume	15 mL (stirred)	50 μL	50 μL	50 μL
Applied potential	Open circuit	-1.2 V	-1.2 to +0.4 V	Electrolysis/ stripping

3.-RESULTS AND DISCUSSION

3.1.-Characterization of hybrid mesoporous silica nanoparticles

Table 2 shows the main characterization data for the prepared MTTZ-MSU-2 material. The materials exhibit a well-resolved XRD pattern at low 2θ values with a single narrow (100) diffraction peak at 1.23º. In this material the d_{100} spacing, assigned to the pore-to-pore centre correlation distance, was 71.67 Å. Evidence for disordered wormhole-like structure of the material is provided by the transmission electron micrographs presented in Figure 2A. These micrographs show that the silica particles contain a large number of channels that are regular in diameter, although they lack long-range packing order. TEM also reveals that the whole material is made of such a porous framework and that there is no amorphous component. SEM images (Figure 2B) confirmed that in both cases the particle shape was pseudo-spherical and the mean diameter was 190 nm

Table 2
Characterization data for MSU-2 and MTTZ-MSU-2 silicas.

		MSU-2	**MTTZ-MSU-2**
XRD Patterns	2θ (º)	1.23	1.23
	d-spacing (Å)	71.67	71.67
N_2 Isotherms	BET surface (m^2 g^{-1})	628	508
	Pore volume (cm^3 g^{-1})	0.93	0.64
	BJH pore diameter (Å)	43.5	36.4
	Wall thickness (Å)	39.2	46.4

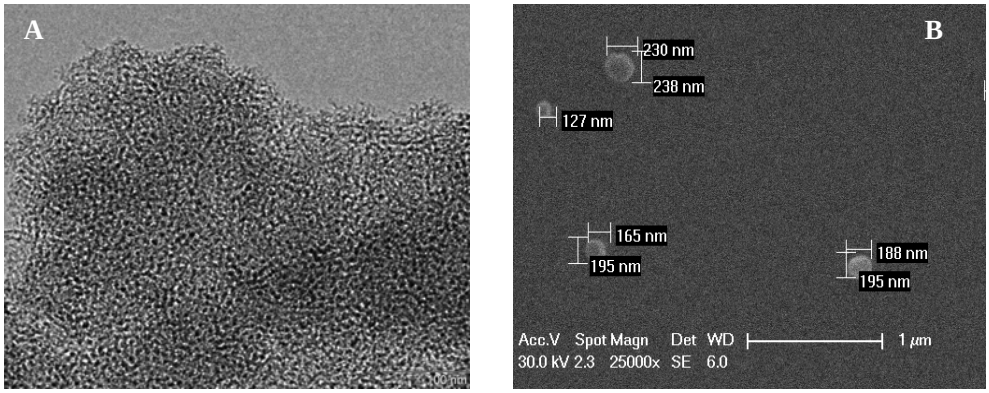

Figure 2. (A) TEM and (B) SEM images of MTTZ-MSU-2.

The nitrogen adsorption–desorption isotherms (Figure 3A) show a well-defined step (type IV according to the I.U.P.A.C. classification) and have an H1 hysteresis loop that is indicative of framework-confined mesoporosity. A narrow pore size distribution was found for this material that provides evidence for a uniform framework mesoporosity (Figure 3B). The physicochemical properties of the MTTZ-MSU-2 such the Brunauer–Emmett–Teller surface area (S_{BET}), total pore volume, Barrett, Joyner and Halenda (BJH) average pore diameter and wall thickness were 508 $m^2\ g^{-1}$, 0.64 $cm^3\ g^{-1}$, 36.4 Å and 46.4 Å, respectively (Table 2). The quantity of molecules attached to the mesoporous silica surface (L_o), calculated from the percentage of sulphur in the functionalized mesoporous silica (estimated by elemental analysis) was equal to 0.73 mmol g^{-1}. Taking into account L_o and S_{BET} of the mesoporous silica, the average surface density, d of the attached molecules and the average intermolecular distance, l was 0.86 molecules per nm^2 and 1.07 nm, respectively that confirm a high efficiency in the functionalization of the MSU-2. Thermal stability of the MTTZ-MSU-2 was studied by thermogravimetric analysis (TGA). TGA curves of the MTTZ-MSU-2 shows that an exothermic degradation process occurs between 225 ºC and 600 ºC due to the breakdown of the ethoxy groups, the heterocyclic ring and the alquilic chain (Figure 4). The chemical stability of the MTTZ-MSU-2 was examined in 1, 3 and 6 M HCl, solutions at room temperature during 24 h. Results obtained indicate that after acid treatment, the functionalization degree in the functionalized mesoporous silica was not modified.

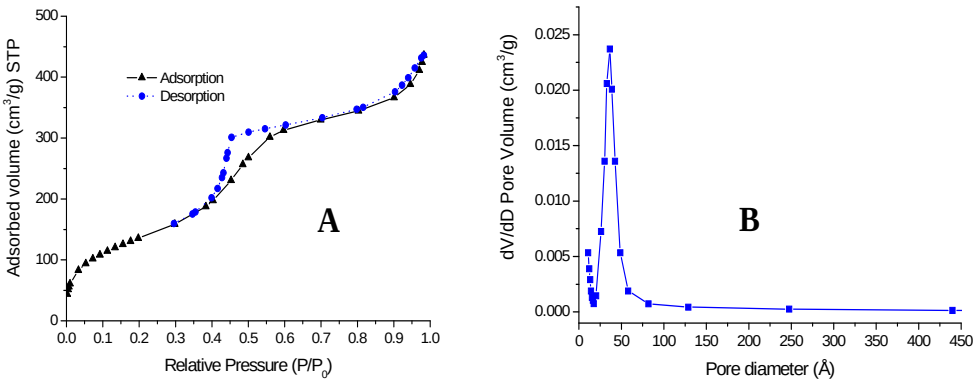

Figure 3. (A) Nitrogen adsorption-desorption isotherms and (B) pore size distribution of MTTZ-MSU-2.

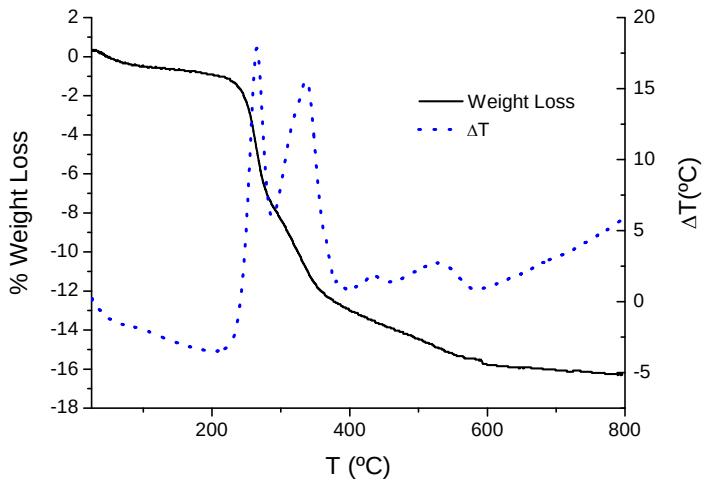

Figure 4. Thermogravimetric curves of MTTZ-MSU-2.

3.2.-Voltammetric measurements optimization

To achieve the most accurate and sensitive Pb(II) measurements, optimization of the operating parameters in preconcentration and detection steps was performed. Factors affecting the preconcentration of Pb(II) onto the electrode surface include electrode composition and preconcentration time. Factors affecting the detection of Pb(II) onto the electrode surface include electrolysis/stripping solution composition and electrolysis time.

3.2.1.-Electrode composition

Different pastes were prepared by mixing MTTZ-MSU-2 with carbon paste and mineral oil in order to modify the working electrode of the commercial SPCE strip. Voltammetric curves were obtained after 5 min preconcentration time and 120 s electrolysis time in 0.2 M HCl. Preliminary studies indicated that pastes prepared with a 0% of mineral oil were not compact enough to modify the SPCE strip. More than 20% of mineral oil in the paste produced a reduction on the voltammetric response, due to the high hydrophobicity of the electrode surface that reduced the interaction between Pb(II) and binding sites. On the other hand, the voltammetric response increased significantly when the MTTZ-MSU-2 content in the paste increased from 10% to 20% due to an increase in the binding sites for Pb(II). Higher % of MTTZ-MSU-2 resulted in a mixture too thick and dry. Therefore a 20% MTTZ-MSU-2, 60% carbon paste and 20% mineral oil was selected as the most appropriate mixture to give reproducible results. For comparative purpose, modification of the working electrode was carried out by mixing MSU-2 with carbon paste and mineral oil until (20:60:20%, w/w/w) but in this conditions a very small peak of Pb(II) was obtained, so the use of MTTZ-MSU-2 silica as modifier in the SPCE clearly enhanced its sensitivity for Pb(II) detection.

3.2.2.-Preconcentration time

The effect of preconcentration time (1, 2, 5 and 10 min) on the voltammetric response for Pb(II) detection was studied. The experiments were performed using 20 $\mu g\ L^{-1}$ Pb(II) solutions for the preconcentration step and 120 s of electrolysis time in 50 µL of 0.2 M HCl. Results obtained indicate that peak currents (and peak areas) increased proportionally with time from 1 min to 10 min. These results suggest the SPCE modified with 20% MTTZ-MSU-2 have high content of functional groups, and it is not saturated easily by Pb(II) ions. It is worth noting that with one drop of acidic solution (50 µL of 0.2 M HCl) as electrolysis and stripping media and only 2 min of preconcentration time, this system can detect Pb(II) near the EPA and Spanish action level for public drinking water supplies (15 and 25 ppb, respectively) [31, 32]. This short preconcentration time can be attributed to a fast adsorption rate, due to the structure, particle morphology and size of MSU-2 silica, that enhance the diffusion rate of the Pb(II) ions into the pores [27–30]. For a rapid analysis of lead, a 2 min preconcentration time was found to be enough for the studied concentration (20 $\mu g\ L^{-1}$), but longer time (5 min) may be required in samples with lower Pb(II) concentration (< 10 $\mu g\ L^{-1}$).

3.2.3.-Electrolysis/stripping solution

Desorption of previously accumulated Pb(II) ions from the electrode surface to the electrode/solution interface must take place to enable the voltammetric detection. Concurrent with the desorption process, an electrolysis was performed by applying a negative potential of −1.2 V to reduce the Pb(II) ions to Pb(0), followed by the quantification of lead via SWV, which oxidizes the previously reduced Pb(0) to Pb(II). Thus, as electrolysis and stripping media HCl solutions of different

concentration were investigated (0.1, 0.2, 0.3 and 0.5 M HCl). The experiments were performed with a 20 µg L^{-1} Pb(II) solution for 5 min of preconcentration time and 120 s of electrolysis time. Results shown in Figure 5 indicated that increasing the HCl concentration (from 0.1 M to 0.3 M) did not significantly improve the sensitivity of the electrode to Pb(II) ions. Therefore, 0.2 M HCl was chosen as electrolysis and stripping media because good reproducibility was achieved in the voltammetric responses. At higher HCl concentration (0.5 M) interferences from hydrogen generation reduced the voltammetric response.

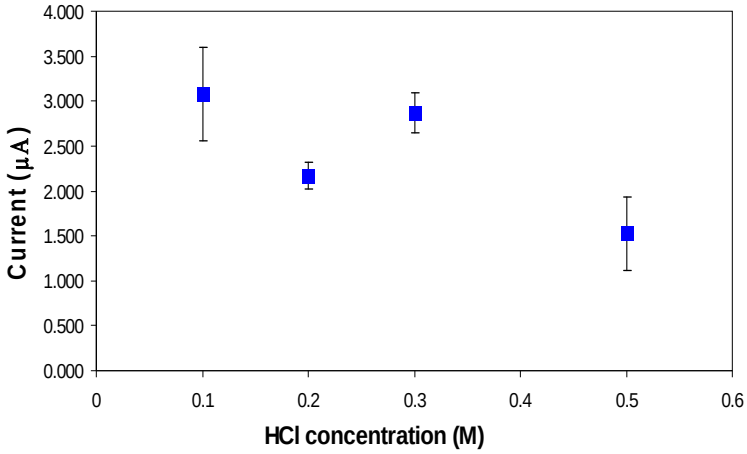

Figure 5. Effect of the electrolysis and stripping media on the voltammetric response (peak currents) measured by anodic stripping square wave voltammetry after 5 min preconcentration from 20 µg L^{-1} Pb(II) solution and 120 s of electrolysis time at -1.2 V.

3.2.4.-Electrolysis time

Figure 6 shows the effect of the electrolysis time on the voltammetric response for Pb(II) detection. The experiments were performed with a 20 µg L^{-1} Pb(II) solution, 5 min of preconcentration time and 0.2 M HCl as electrolysis and stripping media. As it can be seen in Figure 6, the peak current increased as the electrolysis time increased from 0 s to 120 s, and levelled off after 120 s. The best reproducibility was observed with 120 s of electrolysis, thus this electrolysis period was chosen as the optimal for lead deposition. This electrolysis time was found to be optimum in other studies carried out with carbon electrodes modified with functionalized mesoporous silicas [16, 33].

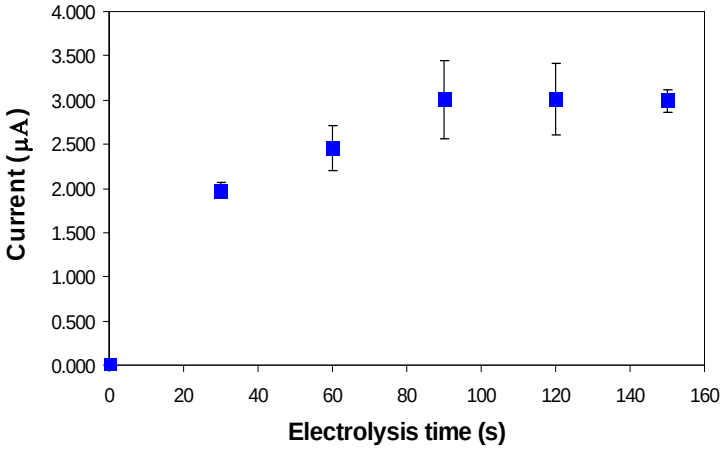

Figure 6. Effect of the electrolysis time on the voltammetric response (peak currents) measured by anodic stripping square wave voltammetry after 5 min preconcentration from 20 µg L^{-1} Pb(II) solution obtained in 0.2 M HCl as electrolysis and stripping media.

3.3.-Multi-component metal ions detection and interferences study

Cations may be considered as interferences to the voltammetric detection of Pb(II) if they compete with these metal ions for the binding sites on the MTTZ-MSU-2 material during the preconcentration step or if its peak response overlaps the Pb peak in the detection step. On the other hand, some anions present in real samples have the capability to complex with many metals ions. Consequently, they may compete with the immobilized MTTZ ligand for the Pb(II) ions, reducing the voltammetric detection of lead. Consequently, the selectivity of the developed electrode was evaluated by intentionally introducing some cations and anions into the Pb(II) solution during the preconcentration step. These ions were chosen because they exhibit redox activity in roughly the same potential as Pb(II) and/or existing in real waters samples. The experiments were performed under the optimized conditions with a solution containing 20 µg L^{-1} of Pb(II) and 100 µg L^{-1}, 1 mg L^{-1}, 10 mg L^{-1} and 100 mg L^{-1} of the interference. Based on the results obtained, the presence of 100 µg L^{-1} of Ca(II), Mg(II), Cd(II), Zn(II), Ni(II) or Cu(II) did not interfere significantly or produce a small reduction in the Pb peak current (recoveries in the range 80 – 110% with RSD < 10%), so these metals do not compete with lead for the binding sites on the MTTZ-MSU-2 at this level (5-fold higher). On the other hand, in presence of higher amounts of interferences (1, 10 and 100 mg L^{-1}) the Pb peak current was reduced significantly (recoveries in the range 10 – 20% for 5000-fold higher). In relation with anions, the presence of these ionic species in the medium

produced an important reduction in the Pb peak current (recoveries in the range 60 – 80% in presence of 100 µg L^{-1} of the anionic interference) that means that Cl^-, SO_4^{2-}, PO_4^{3-}, CO_3^{2-} and CH_3COO^- have the capability to complex the Pb(II) ions under the assayed conditions.

As the redox potential of copper and cadmium are different from lead, the SPCE modified with MTTZ-MSU-2 was tested for simultaneous detection of multi-components metal ions. Figure 7 shows a representative voltammogram of 20 µg L^{-1} of Pb(II) and 100 µg L^{-1} of Cd(II) and Cu(II) (5 min of preconcentration time, 0.2 M HCl as electrolysis and stripping media, 120 s of electrolysis time). As we can see in this figure, the high functionalization degree of the MTTZ-MSU-2 minimizes the competition among these metal ions, allowing them to be detected simultaneously without sacrificing the response intensity.

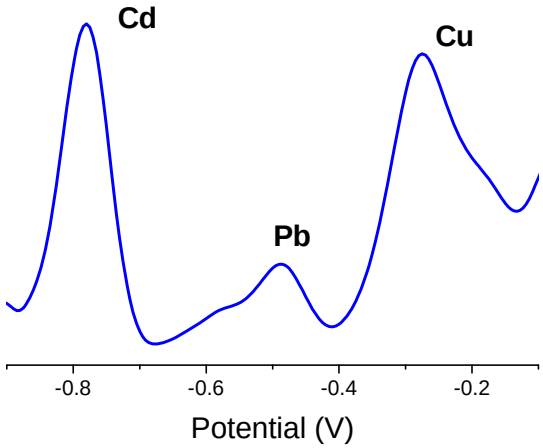

Figure 7. Representative voltammograms of the simultaneous detection of 20 µg L^{-1} of Pb(II), 100 µg L^{-1} of Cu(II) and 100 µg L^{-1} of Cd(II) measured by anodic stripping square wave voltammetry after 5 min preconcentration and 120 s electrolysis at -1.2 V in 0.2 M HCl.

3.4.-Analytical performance

Signal stability was first studied as a key parameter to test the practical usability of the developed electrode. Results obtained indicated that analytical signals for a single electrode surface were stable for 20 measuring cycles (RSD 4 – 7%), so a

single modified electrode can be used for a complete set of measurements (calibration curve, blanks and sample analysis). For a single electrode surface, the RSD of three replicate measurements of a solution containing 20 μg L^{-1} of Pb(II) was less to 5%. Using the same electrode surface for three consecutive days, the RSD was less to 10% (three determinations for each day). To test the reproducibility of the working electrode modification procedure, six different electrodes were prepared and assayed in different days. The RSD obtained was 10% (three determinations for each electrode). These results indicated very good reproducibility for Pb detection with the developed electrode, similar to others SPCMEs previously prepared by other authors [16]. Higher inter-electrode reproducibility can be easily achieved through precise manufacturing of the electrode surface instead of manual coating.

Figure 8 shows a calibration curve obtained for the voltammetric response measured (peak current) versus the metal ion concentration under the optimized conditions. As it can be seen in Figure 8, the voltammetric response was linear to the Pb(II) concentration with a R^2 of 0.999 in the range of 1 – 30 μg L^{-1}. To increase the linearity range a shorter preconcentration time (2 min) can be used. The limit of detection (LOD = 0.1 μg L^{-1}) and limit of quantification (LOQ = 0.4 μg L^{-1}) were calculated as the concentration of Pb(II) that would give a signal 3 or 10 times the background noise, respectively. The background noise was measured under the optimized conditions. It is well known that in stripping analysis the LOD improves significantly when the preconcentration time increases. It is worth noting that very low LOD (0.1 μg L^{-1}) was obtained with the SPCME developed in this work with a relatively low preconcentration time (5 min).

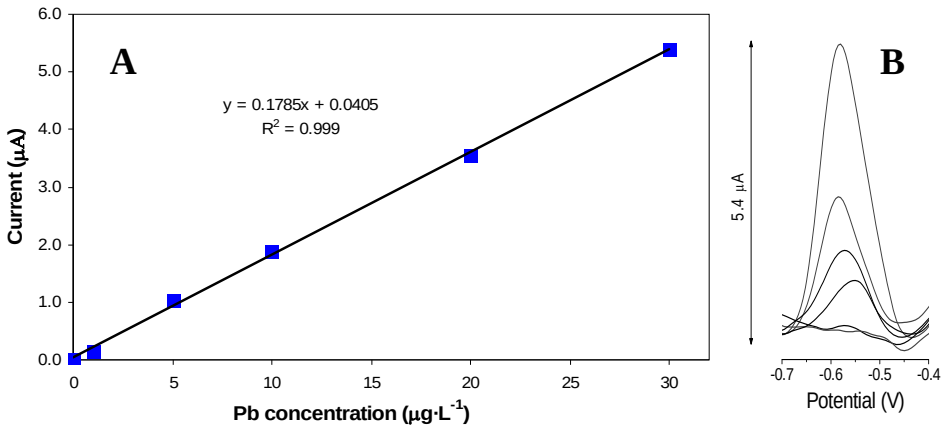

Figure 8. (A) Calibration curve for Pb(II) measured by anodic stripping square wave voltammetry after 5 min preconcentration time and 120 s of electrolysis time in 0.2 M HCl. (B) Representative voltammograms of different Pb(II) solutions in ascending order of concentration.

SPCMEs have been investigated as disposable sensors for the analysis of trace levels of Pb(II) by some authors. The SPCME with MTTZ-MUS-2 developed in the present work show better detection limits than other previously reported in the literature. Wang and Tian [34] analyzed Pb(II) in water samples with a SPCE electroplated with a thin film of gold. A detection limit of 0.6 μg $L^{-1}$was estimated following 4 min deposition. Palchetti et al. [35] used SPCMEs for Pb(II) detection. The detection limits were 12 μg L^{-1} and 15 μg L^{-1} for dithizone-SPE and Nafion®-SPE, respectively (10 min accumulation in open circuit). Honeychurch et al. analyzed Pb(II) in water using 1-(2-pyridylazo)-2-naphthol modified SPCE [14] and calixarene modified SPCE [15]. The detection limits were 15 μg L^{-1} (5 min preconcentration in open circuit) and 5 μg L^{-1} (10 min preconcentration in open circuit), respectively. Masawat et al. [36] analyzed Pb(II) in waters using gold-sputtered SPCEs. The detection limit was 0.8 μg L^{-1} at 120 s deposition. Yantasee et al. [16] calculated a limit of detection of 0.91 μg L^{-1} Pb(II) using SPCEs modified with acetamide phosphonic acid self-assembled monolayer on mesoporous MCM-41 silica (5 min preconcentration in open circuit). Laschi et al. [9] used a gold-based SPCE for Pb(II) detection in waters. The detection limit was 0.5 μg L^{-1} at 120 s deposition. More recently, Granado et al. [11] reported the use of a SPCME with Bi nanoparticles for the detection of Pb(II) in waters. The detection limit was 0.5 μg L^{-1} at 120 s deposition.

3.5.-Sample analysis

To test the proposed methodology, tap, river and underground water were assayed without any pre-treatment. Water samples were spiked with Pb(II) to obtain a measurable level of 10 μg L^{-1} according the future Spanish action level for public drinking water supplies (10 μg L^{-1} in 2014) [32]. To compensate matrix effects, Pb(II) was determined in the real water samples by standard addition method (Figure 9) and accuracy was assessed by recovery test. Figure 9 and Table 3 shows linear parameters of Pb analytical curves measured on SPCME with MTTZ-MSU-2 in the range of 0 – 30 μg L^{-1} in river, tap and underground water, which were obtained after 5 min of preconcentration time. For the same Pb(II) concentration, the signals were larger in river water than in underground water and smaller in tap water. As can be seen in Table 4, good correlations between amounts determined and added were obtained (recovery values 97 – 106, RSD 4 – 7%), suggesting that methodology developed can be used for practical analysis of Pb(II) in real water samples.

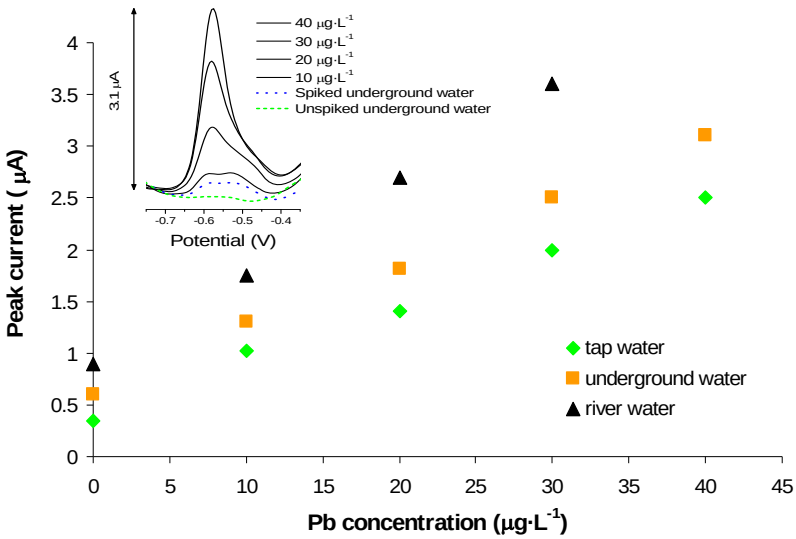

Figure 9. Pb(II) determination in tap, underground and river water samples by standard addition method measured by anodic stripping square wave voltammetry after 5 min preconcentration time and 120 s of electrolysis time in 0.2 M HCl. Inset shows representative voltammograms in underground water of different Pb(II) solutions in ascending order of concentration.

Table 3

Linear parameters of Pb(II) analytical curves measured at screen-printed electrode modified with hybrid mesoporous silica in various matrices [a].

Sample	Slope [b]	Intercept [b]	Data range (µg L^{-1})	R^2
Milli-Q water	0.1785	0.0405	0-30	0.999
Tap water	0.0427	0.4571	0-40	0.998
Underground water	0.0574	0.5888	0-40	0.997
River water	0.0905	0.8805	0-30	0.999

[a] 5 min preconcentration time and 120 s of electrolysis time in 0.2 M HCl.
[b] Peak current (µA) versus Pb concentration (µg L^{-1}).

Table 4
Determination of Pb(II) on a screen-printed electrode modified with hybrid mesoporous silica in real water samples [a].

Sample	Spiked (μg L^{-1})	Found (μg L^{-1})	Recovery (%)	RSD (n = 3)
Tap water	0.0	-[b]	-	-
	10.0	10.6	106	4
Underground	0.0	- [b]	-	-
water	10.0	10.2	102	6
River water	0.0	- [b]	-	-
	10.0	9.7	97	7

[a] 5 min preconcentration time and 120 s of electrolysis time in 0.2 M HCl.
[b] No detected

4.-CONCLUSIONS

Functionalized mesoporous silicas are a very good option to prepare chemically modified screen-printed carbon electrodes because of its high surface area, high porosity, controllable and narrowly distributed pore and particle size, ordered pore arrangement and high functionalization degree. In this study, we have demonstrated that a screen-printed carbon electrode modified with MTTZ-MSU-2 can be used to analyze Pb(II) by anodic stripping voltammetry in non-pretreated natural waters. The electrode developed has as main advantages a very low LOD (0.1 $\mu g\ L^{-1}$ after 5 min preconcentration time) and a good reproducibility, thus has a great potential to be used in the development of portable analyzers for monitoring heavy metal ions in water samples.

Acknowledgements

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5.-REFERENCES

[1] Baird C., in **Environmental Chemistry,** ed. W. H. Freeman and Company, *University of Western Ontario, New York,1999.*

[2] Allan I. J., Vrana B., Greenwood R., Mills G. A., Roig B., Gonzalez C.; **A "toolbox" for biological and chemical monitoring requirements for the European Union's Water Framework Directive.** *Talanta 69 (2006) 302-322.*

[3] Armenta S., Garrigues S., de la Guardia M.; **Green analytical chemistry.** *Trac-Trends Anal. Chem. 27 (2008) 497-511.*

[4] He L., Toh C.-S.; **Recent advances in analytical chemistry – A material approach.** *Anal. Chim. Acta 556 (2006) 1-15.*

[5] Wang J.; **Stripping analysis.** *VCH Publishers Inc., Florida,1985.*

[6] Honeychurch K. C., Hart J. P.; **Screen-printed electrochemical sensors for monitoring metal pollutants.** *Trac-Trends Anal. Chem. 22 (2003) 456-469.*

[7] Palchetti I., Marrazza G., Mascini M.; **New procedures to obtain electrochemical sensors for heavy metal detection.** *Anal. Lett. 34 (2001) 813-824.*

[8] Palchetti I., Laschi S., Mascini M.; **Miniaturised stripping-based carbon modified sensor for in field analysis of heavy metals.** *Anal. Chim. Acta 530 (2005) 61-67.*

[9] Laschi S., Palchetti I., Mascini M.; **Gold-based screen-printed sensor for detection of trace lead.** *Sens. Actuator B-Chem. 114 (2006) 460-465.*

[10] Kadara R. O., Tothill I. E.; **Development of disoponsable bulk-modified screen-printed electrode based on bismuth oxide for stripping chronopotentiometric analysis of lead (II) and cadmium (II) in soil and water samples.** *Anal. Chim. Acta 623 (2008) 76-81.*

[11] Granado Rico M. A., Olivares-Marín M., Pinilla Gil E.; **Modification of carbon screen-printed electrodes by adsorption of chemically synthesized Bi nanoparticles for the voltammetric stripping detection of Zn(II), Cd(II) and Pb(II).** *Talanta 80 (2009) 631-635.*

[12] Somerset V., Iwuoha E., Hernandez L.; **Stripping voltammetric measurement of trace metal ions at screen-printed carbon and carbon paste electrodes.** *Proc. Chem. 1 (2009) 1279-1282.*

[13] Somerset V., Leaner J., Mason R., Iwuoha E., Morrin A.; **Development and application of a poly(2,2'-dithiodianiline) (PDTDA)-coated screen-printed carbon electrode in inorganic mercury determination.** *Electrochim. Acta 55 (2010) 4240-4246.*

[14] Honeychurch K. C., Hart J. P., Cowell D. C.; **Voltammetric studies of lead at a 1-(2-pyridylazo)-2-naphthol modified screen-printed carbon electrode and its trace determination in water by stripping voltammetry.** *Anal. Chim. Acta 431 (2001) 89-99.*

[15] Honeychurch K. C., Hart J. P., Cowell D. C., Arrigan D. W. M.; **Voltammetric studies of lead at calixarene modified screen-printed carbon electrodes and its trace determination in water by stripping voltammetry.** *Sens. Actuator B-Chem. 77 (2001) 642-652.*

[16] Yantasee W., Deibler L. A., Fryxell G. E., Timchalk C., Lin Y.; **Screen-printed electrodes modified with functionalized mesoporous silica for voltammetric analysis of toxic metal ions.** *Electrochem. Commun. 7 (2005) 1170-1176.*

[17] Yantasee W., Fryxell G. E., Lin Y.; **Voltammetric analysis of europium at screen-printed electrodes modified with salicylamide self-assembled on mesoporous silica.** *Analyst 131 (2006) 1342-1346.*

[18] Lin Y., Yantasee W., Fryxell G. E.; **Electrochemical sensors based on functionalized nanoporous silica,** in: Dekker encyclopedia of nanoscience and nanotechnology, *Marcel Dekker, New York, 2004*.

[19] Yantasee W., Lin Y., Fryxell G. E., in **Environmental applications of nanomaterials: synthesis, sorbents and sensors**, ed. Fryxell G. E., Cao G., *Imperial College Press, London, 2007*.

[20] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Mesoporous silica functionalized with 2-mercaptopyridine: Synthesis, characterization and employment for Hg(II) adsorption.** *Microporous Mesoporous Mat. 89 (2006) 58-68*.

[21] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Preparation of 2-mercaptobenzothiazole-derivatized mesoporous silica and removal of Hg(II) from aqueous solution.** *J. Environ. Monit. 8 (2006) 214-222*.

[22] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Adsorption of cadmium(II) from aqueous media onto a mesoporous silica chemically modified with 2-mercaptopyrimidine.** *J. Mater. Chem. 16 (2006) 1757-1764*.

[23] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **2-Mercaptothiazoline modified mesoporous silica for mercury removal from aqueous media.** *J. Hazard. Mat. B 134 (2006) 245-256*.

[24] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Preparation, characterization, and Zn^{2+} adsorption behavior of chemically modified MCM-41 with 5-mercapto-1-methyltetrazole.** *J. Colloid Interface Sci. 313 (2007) 551-562*.

[25] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Functionalized HMS mesoporous silica as solid phase extractant for Pb(II) prior to its determination by flame atomic absorption spectrometry.** *J. Sep. Sci. 30 (2007) 1556-1567*.

[26] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Preconcentration of Zn(II) in water samples using a new hybrid SBA-15-based material.** *J. Hazard. Mat. 166 (2009) 1449-1458.*

[27] Brown J., Richer R., Mercier L.; **One-step synthesis of high capacity mesoporous Hg^{2+} adsorbents by non-ionic surfactant assembly.** *Microporous Mesoporous Mat. 37 (2000) 41-48.*

[28] Mercier L., in **Studies in surface science and catalysis,** ed. Sayari A., *Elsevier Science vol. 129, Amsterdam, 2000.*

[29] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Synthesis and characterization of novel mesoporous silicas of the MSU-*X* family for environmental applications.** *J. Nanosci. Nanotechnol. 9 (2009) 4901-4909.*

[30] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Solid phase extraction of Pb(II) in water samples using a new hybrid inorganic-organic mesoporous silica prior to its determination by FAAS.** *Microchim. Acta 165 (2009) 291-298.*

[31] EPA; **Consumer Factsheet on: Lead.**

http://www.epa.gov/OGWDW/dwh/t-ioc/lead.html.

[32] BOE nº 45; **Real Decreto 140/2003 de 7 de febrero.** *21 febrero de 2003.* **http://www.msc.es/ciudadanos/saludAmbLaboral/docs/rd_140_2003.pdf**

[33] Morante-Zarcero S., Sánchez A., Fajardo M., del Hierro I., Sierra I.; **Voltammetric analysis of Pb(II) in natural waters using a carbon paste electrode modified with 5-mercapto-1-methyltetrazol grafted on hexagonal mesoporous silica.** *Microchim. Acta 169 (2010) 57-64.*

[34] Wang J., Tian B.; **Mercury-free disponsable lead sensors based on potentiometric stripping analysis of gold-coated screen-printed electrodes.** *Anal. Chem. 65 (1993) 1529-1532.*

[35] Palchetti I., Upjohn C., Turner A. P. F., Mascini M.; **Disponsable screen-printed electrodes (Spe) mercury-free for lead detection.** *Anal. Lett. 33 (2000) 1231-1246.*

[36] Masawat P., Liawruangrath S., Slater J. M.; **Flow injection measurement of lead using mercury-free disponsable gold sputtered screen-printed carbon electrodes (SPCE).** *Sens. Actuator B-Chem. 91 (2003) 52-59.*

DETERMINATION OF Hg(II) IN NATURAL WATERS USING A CARBON PASTE ELECTRODE MODIFIED WITH HYBRID MESOSTRUCTURED SILICA NANOPARTICLES.

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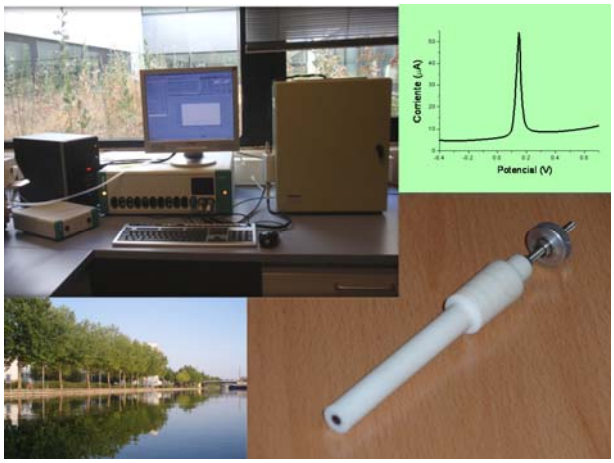
Sensors and Actuators B-Chemical (Submitted).



DETERMINATION OF Hg(II) IN NATURAL WATERS USING A CARBON PASTE ELECTRODE MODIFIED WITH HYBRID MESOSTRUCTURED SILICA NANOPARTICLES.

ABSTRACT

Mesostructured silica nanoparticles (MSU-2) functionalized with a 5-mercapto-1-methyltetrazole derivative was employed to prepare a chemically modified carbon paste electrode for Hg(II) detection in aqueous media by square wave adsorptive anodic stripping voltammetry. Under optimized operating conditions, 60 s of electrolysis time in HCl 0.1 M, the voltammetric signal increased with increasing preconcentration time from 1 to 10 min. The areas of the peak responses were linear with respect to Hg(II) concentrations in the range of 0.1 to 1 μM and a detection limit of 0.021 μM was obtained after 5 min preconcentration time. The carbon paste modified electrode had good single and inter-electrode reproducibility (RSD of 2 and 12%, respectively). A simultaneous multi-element analysis of Hg(II), Cd(II) and Pb(II) was also carried out with the electrode. Applicability to spiked drinking water and river water was demonstrated without any sample pretreatment (recoveries of 92 and 98%, RSD 4-7%, for river and tap water respectively).

1.-INTRODUCTION

In the last few years, the development in the design of materials with particle size, shape and/or pores of nanometric scale has made possible important advances in Nanoscience and Nanotechnology. A clear example was the discovery of the M41S mesostructured silica family by Mobil researchers [1, 2] with well ordered pores of uniform size, high volume and high surface area. It is well known that the characteristics of these materials makes them potentially useful in those processes were the adsorption of different kind of compounds is required. However, this adsorption is not selective and it can be reversible [3] so, to increase the selectivity and loading capacity, chemical modification of these materials with organic functional groups is necessary. Hybrid mesotructured silicas (MCM-41, SBA-15 and HMS) have been widely used for the adsorption of different heavy metals in waters [4-10], but adsorption studies using functionalized MSU-X silicas are comparatively scarce. Compared to MCM-41 and SBA-15 materials, that exhibit either hexagonal or cubic symmetries, the structure and particle morphology of MSU-X silicas is expected to enhance the diffusion rate of the heavy metals into the pores [11-14]. Other potential advantages of MSU-X mesoporous family over other materials are lower reagent and energy costs (synthesis at ambient pressure and moderate temperature), low surfactant toxicity and biodegradability. All these points decrease the overall cost and environment impact of this synthesis process.

Carbon paste electrodes chemically modified with hybrid silica have reached considerable attention for anodic stripping voltammetry during the last years, because they display significant improvements with respect to conventional electrodes [15, 16]. The use of hybrid mesostructured silicas to modifie carbon paste electrodes (MSM-CPEs) is a current research area in electroanalysis. In a recent review [17], Walcarius indicated the higher sensitivity of this type of electrodes, in comparison to the corresponding electrodes prepared with hybrid amorphous silica that is attributed to a better preconcentration efficiency, a better accessibility to the binding sites and faster diffusion processes relative to disordered silica. Despite of these advantages, a limited number of hybrid mesostructured silicas have been used to prepare MSM-CPEs for voltammetric determination of heavy metals. Recently we have developed some carbon paste electrodes modified with hybrid mesostructured silicas. These MSM-CPEs were evaluated for reliable quantification of trace Pb(II) ions by square-wave anodic stripping voltammetry (SW-ASV) in non-pretreated natural waters and results obtained indicated a great potential of these electrodes to be used in the development of portable analyzers for monitoring heavy metal ions in water samples [18, 19].

Mercury belongs to the class of poisons that presents the greatest hazard to human health and therefore its concentration must be kept under permanent control. Mercury has a particular toxicological interest because it occurs naturally in the

environment and exists in several chemical and physical forms that determine mercury's absorption and metabolism. There are several analytical techniques currently employed for trace determination of mercury in environmental samples, but electrochemical techniques have been used extensively because of to their lower cost and the possibility to perform analysis *in-situ*. This provides a significant saving in time and operating costs [20].

In the current work, the MTTZ-MSU-2 material has been used to prepare a MSM-CPE for Hg(II) detection in aqueous solution by SW-ASV technique. Factors affecting the preconcentration and detection steps were investigated and, in the optimal operating parameters, the electrode performance was evaluated (linearity, detection and quantification limits and precision). The MSM-CPE has been used for simultaneous detection of lead, cadmium and mercury. To our knowledge, this is the first report on using hybrid MSU-2 mesostructured silica nanoparticles for Hg(II) analysis by SW-ASV.

2.-EXPERIMENTAL

2.1.-Chemicals and reagents

A certified ICP standard stock solution of Hg(II) at 1.000 g L^{-1} ± 0.2% was purchased from Panreac. ICP stock solutions of Pb(II) and Cd(II) at 1.000 g L^{-1} were purchased from Metrohm and Fluka, respectively. Single-component mercury solution and multicomponent metal solution (mercury, lead and cadmium) were prepared daily by diluting the stock solutions with ultrapure water (resistivity 18.2 MΩ cm) obtained from a Millipore Milli-Q-System (Waters, USA). HCl Hiperpur (35%), NaCl (99%) obtained from Panreac and $HClO_4$ of analytical grade (70%) obtained from Scharlau were used for electrolysis/stripping solutions. The preparation and characterization of the MTTZ-MSU-2 was described elsewhere [13]. The MTTZ-MSU-2 material possessed a S_{BET} of 508 m^2 g^{-1}, a pore volume of 0.64 cm^3 g^{-1}, a BJH pore diameter of 37 Å and a functionalization degree of 0.73 mmol MTTZ g^{-1}.

2.2.-Samples

A drinking water sample was collected from a running water tap in Madrid (Canal de Isabel II) and used immediately. River water (Alberche river) was filtered through a 0.45 µm membrane filter (Millipore Membrane filters 0.45 µm HA) to remove suspended particles and stored at -26 ºC prior to use in polyethylene bottles. The two waters were analysed without pre-treatment and pH was not adjusted to show the versatility of the developed MSM-CPE. This will minimize the potential sample contamination driving also to time and reagents savings.

2.3.-Apparatus

SW-ASV measurements were performed in an Autolab ECO-CHEMIE Potenciostat-Galvanostat model PGSTAT302 using a conventional voltammetry cell equipped with three electrodes: a CPE modified with MTTZ-MSU-2 as working electrode, a saturated Ag/AgCl electrode (Metrohm, No 6.0724.140) as reference electrode, and a platinum wire (Metrohm, No 6.0331.000) as auxiliary electrode. All measurements were made at room temperature. A PC with GPES software for Windows was used to control the instrument, plot the measurements and perform the analysis of results.

2.4.-Working electrode

The MSM-CPE was prepared by mixing 0.100 g (20% w:w) of the MTTZ-MSU-2 with 0.300 g (60% w:w) of carbon paste (Metrohm, No 6.2801.000). Then,

0.100 g (20% w:w) of mineral oil (Aldrich) were added to the carbon paste/MTTZ-MSU-2 mixture and mixed until obtain a uniform paste. The so obtained material was packed into the end of a homemade Teflon cylindrical tube (d.i. 3 mm) equipped with a screwing stainless steel piston providing an inner electrical contact. Appropriate packing was achieved by pushing the electrode surface against a sheet of paper (this procedure also helps to eliminate the excess of mineral oil). Smoothing of the electrode surface was made by hand-polishing on a filter paper. When necessary, a new electrode surface was obtained by pushing an excess of paste out of the tube and polishing it again on a filter paper. All freshly-made electrodes were activated by performing 2-3 cycles of electrolysis/stripping. Figure 1 shows the schematic of the MSM-CPE developed.

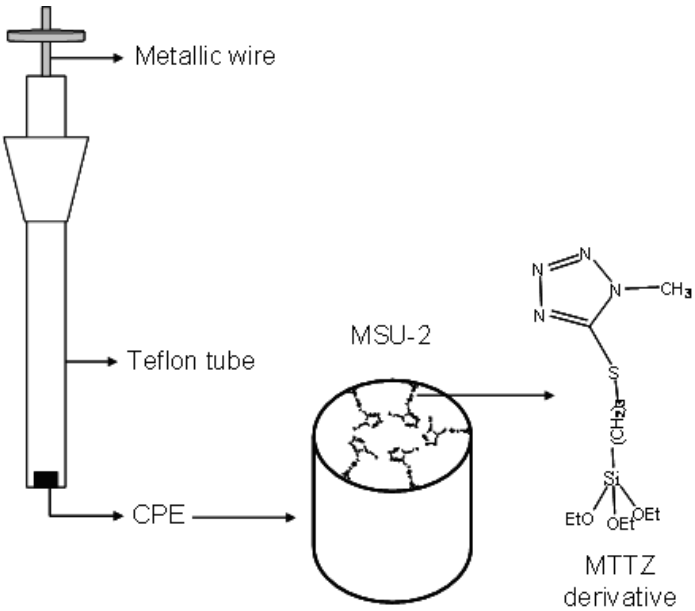


Figure 1. Schematic of the CPE modified with MTTZ-MSU-2.

2.5.-Voltammetric measurements

SW-ASV was operated at a frequency of 100 Hz with pulse amplitude of 50 mV and a potential step height of 5 mV. Table 1 summarizes the operating conditions optimized for the voltammetric experiments. During preconcentration, the MSM-CPE was immersed in 100 mL of Hg(II) solution efficiently stirred for a desired period of time (1-10 min) (300 rpm) at open-circuit. The MSM-CPE was then rinsed with Milli-Q water and transferred to the voltammetric cell containing 100 mL of electrolyte solution (HCl, $HClO_4$ or NaCl). For cathodic electrolysis, a negative potential (-0.8 V)

was applied to the electrode immediately after immersing into the electrolyte solution for a 0-120 s period, to reduce the adsorbed metal into elemental form. Anodic stripping voltammetry was performed by sweeping a square wave potential toward positive values (from -0.8 V to +0.8 V). No stirring conditions were used in both cathodic electrolysis and stripping steps. All measurements were made at room temperature and under an atmospheric environment. After each measurement, to ensure that no metal was still left on the electrode surface, a regeneration of the electrode was performed by electrolysis and stripping steps in 0.1 M HCl. If the electrode surface is clean, no peak will be detected in the second step. Each measurement was performed in triplicate and the average value was reported.

Table 1
Operating conditions optimized for the square wave voltammetry experiments.

	Preconcentration	**Electrolysis**	**Stripping**	**Regeneration**
Solution/ electrolyte	0.1 - 1 μM Hg(II)	HCl 0.1 M	HCl 0.1 M	HCl 0.1 M
Immersion time	5 min	60 s	-	60 s
Solution volume	100 mL (stirred)	100 mL	100 mL	100 mL
Applied potential	Open circuit	-0.8 V	-0.8 to +0.8 V	Electrolysis/ stripping

3.-RESULTS AND DISCUSSION

3.1.-Voltammetric measurements optimization

To achieve the most accurate and sensitive Hg(II) measurements, optimization of the operating parameters in preconcentration and detection steps was performed. One of the most important factor which affecting the preconcentration of Hg(II) onto the electrode surface is the preconcentration time. Factors affecting the detection of Hg(II) onto the electrode surface include electrolysis/stripping solution composition and electrolysis time.

3.1.1.-Preconcentration time

The effect of preconcentration time (1, 2, 5 and 10 min) on the voltammetric response for Hg(II) detection was studied. The experiments were performed using 0.5 μM Hg(II) solutions for preconcentration step and 60 s of electrolysis time in 100 mL of 0.1 M HCl. Results obtained (Figure 2) indicate that peak currents (and peak areas) increased with time from 1 to 10 min. These results suggest the MSM-CPE modified with MTTZ-MSU-2 (20%) have high content of functional groups, and it is not saturated easily by Hg(II) ions. These results are different from those obtained using a silica thin film that contains the same organic ligand (MTTZ) [21], in that case the signal was saturated at 5 min, even working at lower concentrations, indicating that in the CPE modified with MTTZ-MSU-2 the amount of accessible ligand is higher.

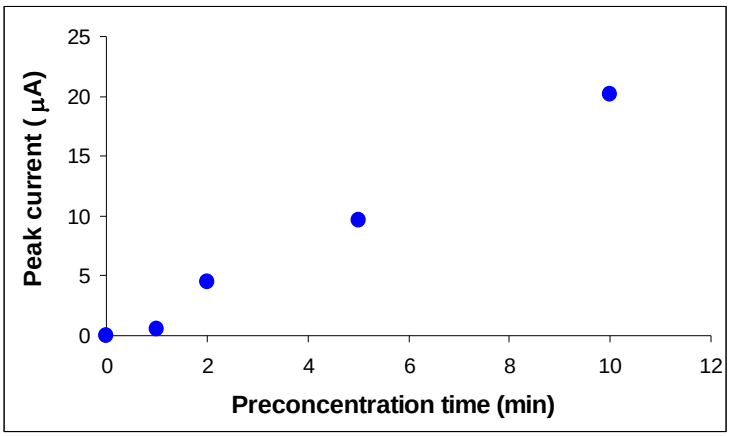


Figure 2. Effect of the preconcentration time on the voltammetric response (peak currents) measured by anodic stripping square wave voltammetry after preconcentration from 0.5 μM Hg(II) solution and 60 s of electrolysis time at -0.8 V in HCl 0.1 M.

For a rapid analysis of mercury, a 2 min preconcentration time was found to be enough for the studied concentration (0.5 μM), but longer time (5 min) may be required in samples with lower Hg(II) concentration. This short preconcentration time can be attributed to a fast adsorption rate, due to the structure, particle morphology and size of MSU-2 silica, that enhance the diffusion rate of the Hg(II) ions into the pores [11-14]. These results are in agreement with results obtained by other researchers groups [21-25].

3.1.2.-Electrolysis/stripping solution

Desorption of previously accumulated Hg(II) ions from the electrode surface to the electrode/solution interface must take place to enable the voltammetric detection. Concurrent with the desorption process, an electrolysis was performed by applying a negative potential of -0.8 V to reduce the Hg(II) ions to Hg(0), followed by the quantification of mercury via SW-ASV, which oxidizes the previously reduced Hg(0) to Hg(II). Thus, as electrolysis and stripping media $HClO_4$ (0.1 M), NaCl (0.1 M) and HCl solutions of different concentration (0.1, 0.3 and 1.0 M HCl) were investigated. The experiments were performed with a 0.5 μM Hg(II) solution for 5 min of preconcentration time and 60 s of electrolysis time. Results shown in Figure 3 indicated that increasing the HCl concentration (from 0.1 to 1.0 M) produced a decrease in the signal, which is in agreement with previous works [18, 19, 21]. On the other hand, 0.1 M $HClO_4$ and 0.1 M NaCl did not improved the sensitivity of the electrode to Hg(II) ions compared to the 0.1 M HCl media [26, 27]. Therefore, 0.1 M HCl was chosen as electrolysis and stripping media because good signal and reproducibility was achieved in the voltammetric responses.

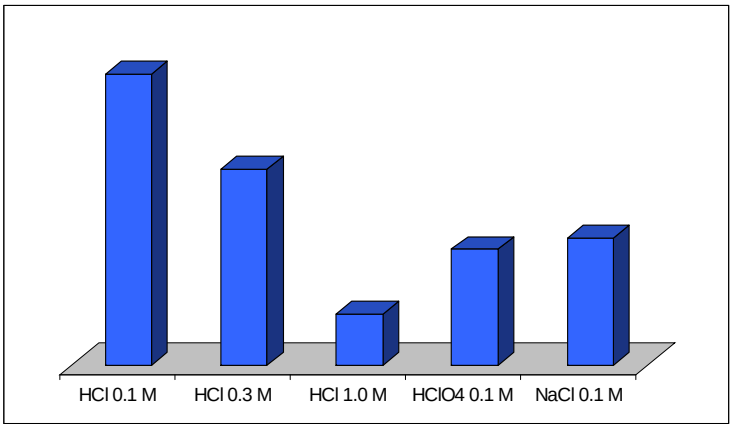


Figure 3. Effect of the electrolysis and stripping media on the voltammetric response (peak currents) measured by anodic stripping square wave voltammetry after 5 min preconcentration from 0.5 μM Hg(II) solution and 60 s of electrolysis time at -0.8 V.

3.1.3 Electrolysis time

Figure 4 shows the effect of the electrolysis time on the voltammetric response for Hg(II) detection. The experiments were performed with a 0.5 μM Hg(II) solution, 5 min of preconcentration time and 0.1 M HCl as electrolysis and stripping media. As it can be seen in Figure 4, the peak current increased as the electrolysis time increased from 0 to 60 s, and levelled off after 90 s. The best reproducibility was observed with 60 s of electrolysis, thus this electrolysis period was chosen as the optimal for mercury deposition. This electrolysis time was found to be optimum in other studies carried out with electrodes modified with functionalized mesoporous silicas [21, 28, 29].

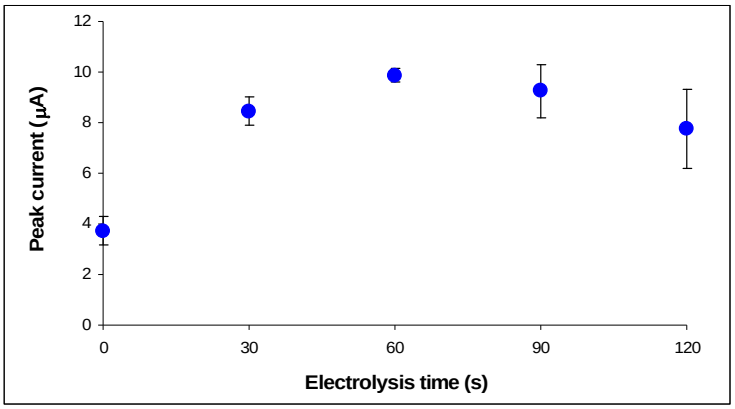


Figure 4. Effect of electrolysis time on the voltammetric response (peak currents) measured by anodic stripping square wave voltammetry after 5 min preconcentration from 0.5 μM Hg(II) solution obtained in 0.1 M HCl as electrolysis and stripping media.

3.2.-Multi-component metal ions detection

Hg(II) determination by ASV involves a first step were the Hg(II) cations are adsorbed on the electrode surface at open circuit potential. This preconcentration step reduces interferences during the stripping step because many species can be left in the sample solutions in function of the affinity between the organic ligand and the specie. On the other hand, it is possible to adsorb on the electrode surface some species which have high affinity against the organic ligand.

As the redox potential of cadmium and lead are different from mercury, the CPE modified with MTTZ-MSU-2 was tested for simultaneous detection of multi-components metal ions. Figure 5 shows a representative voltammogram of 0.5 μM of Hg(II), 0.9 μM of Cd(II) and 0.1 μM of Pb(II) (5 min of preconcentration time, 0.1 M

HCl as electrolysis and stripping media, 60 s of electrolysis time). As we can see in this figure, the high functionalization degree of the MTTZ-MSU-2 minimizes the competition among these metal ions, allowing them to be detected simultaneously without sacrificing the response intensity.

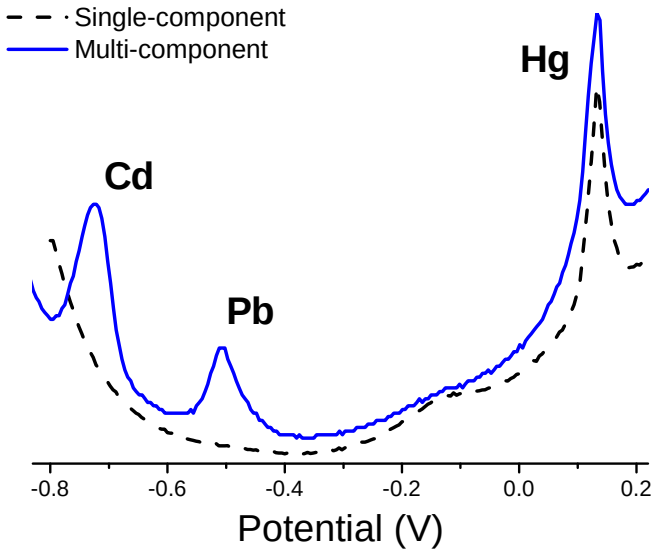


Figure 5. Representative voltammograms of the simultaneous detection of 0.5 μM of Hg(II), 0.9 μM of Cd(II) and 0.1 μM of Pb(II) and detection of 0.5 μM of Hg(II) alone, measured by anodic stripping square wave voltammetry after 5 min preconcentration and 60 s electrolysis at -1 V in 0.1 M HCl.

3.3.-Analytical performance

Signal stability was first studied as a key parameter to test the practical usability of the developed MSM-CPE. Results obtained indicated that analytical signals for a single electrode surface were stable for at least 23 measuring cycles. For a single electrode surface, the RSD of three replicate measurements of a solution containing 0.5 μM of Hg(II) was less to 2%. Using the same electrode surface for three consecutively days, the RSD was less to 10% (three determinations for each day). To test the reproducibility of the working electrode modification procedure, six different electrodes were prepared and assayed in different days. The RSD obtained was 12% (three determinations for each electrode). These results indicated very good reproducibility for Hg(II) detection with the developed electrode.

Figure 6 shows a calibration curve obtained for the voltammetric response measured (peak area) versus the metal ion concentration under the optimized conditions. As it can be seen in Figure 6, the voltammetric response was linear to the Hg(II) concentration with a R^2 of 0.999 in the range of 0.1 to 1 μM. To increase the linearity range a shorter preconcentration time (2 min) can be used. The limit of detection (LOD) and limit of quantification (LOQ) were calculated as the concentration of Hg(II) that would give a signal 3 or 10 times the background noise, respectively. The background noise was measured under the optimized conditions and values obtained after 5 min of preconcentration time were 0.021 μM and 0.070 μM for LOD and LOQ respectively. It is well known that in MSM-CPE the LOD improves significantly when the preconcentration time increases. It is worth noting that very low LOD was obtained in this work with a relatively low preconcentration time.

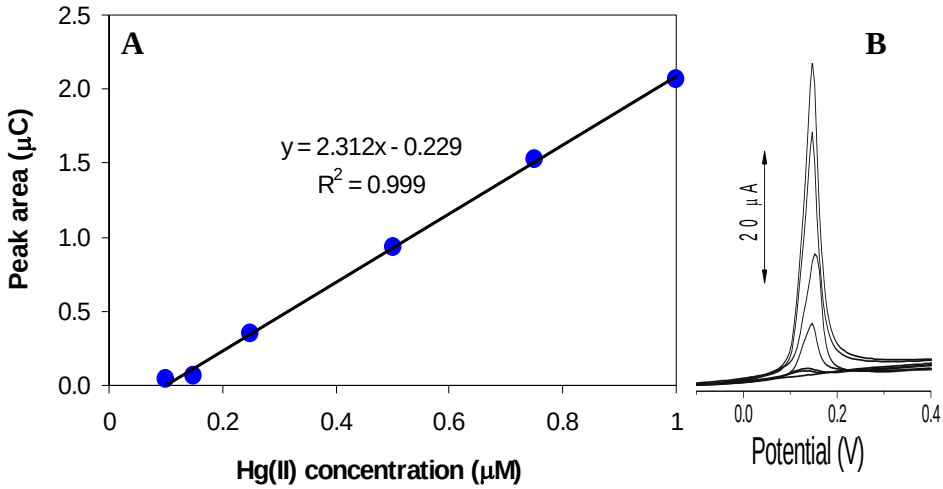


Figure 6. (A) Calibration curve for Hg(II) measured by anodic stripping square wave voltammetry after 5 min preconcentration time and 60 s of electrolysis time in 0.1 M HCl. (B) Representative voltammograms of different Hg(II) solutions in ascending order of concentration.

The MSM-CPE developed has been investigated as a disposable sensor for the analysis of trace levels of Hg(II) by some authors. Yantasee et al. [28] prepared a CPE modified with thiol terminated self-assembled monolayer on mesoporous silica for simultaneous determination of Pb(II) and Hg(II). The areas of the peak responses were linear with respect to Hg(II) concentration in the range of 0.1 to 8 μM and the LOD was 0.015 μM after a 20 min preconcentration period. Cesarino et al. [30] studied the performance of a CPE modified with SBA-15 nanostructrured silica organofunctionalized with 2-benzothiazolethiol in the simultaneous determination of Pb(II), Cu(II) and Hg(II). The voltammetric responses were linear with respect to Hg(II) concentration in the range of 2 to 10 μM and the LOD was 0.4 μM after 3 min

of preconcentration time. More recently, Sánchez and Walcarius [19] deposited a surfactant-templated mesoporuos silica film functionalized with MTTZ onto a glassy carbon electrode for Hg(II) analysis. The voltammetric responses were linear with respect to Hg(II) concentration in the range of 0.05 to 1 μM and the LOD was 0.024 μM after 5 min of preconcentration time. In this sense, the CPE modified with MTTZ-MUS-2 developed in the present work show better LOD than other electrodes reported previously in the literature, moreover with a very good reproducibility and long term stability.

3.4.-Sample analysis

To test the proposed methodology, tap and river water samples were assayed without any pre-treatment. To check the matrix effects, a calibration curve obtained for the voltammetric response measured (peak area) versus the metal ion concentration under the optimized condition in the different samples was measured. As can be seen in Figure 7, for the same Hg(II) concentration, the signals were larger in tap water than in river water what are expectable because of higher pollution and possible presence of surfactants in that kind of water. Table 2 shows linear parameters of Hg(II) of analytical curves measured on CPE modified with MTTZ-MSU-2 in the range of 0.1-1 μM in Milli-Q and tap waters and in the range 0.1-0.75 μM in river water, which were obtained after 5 min of preconcentration time. As can be seen in Table 2, good correlations between peak areas and Hg(II) concentration had been achieved.

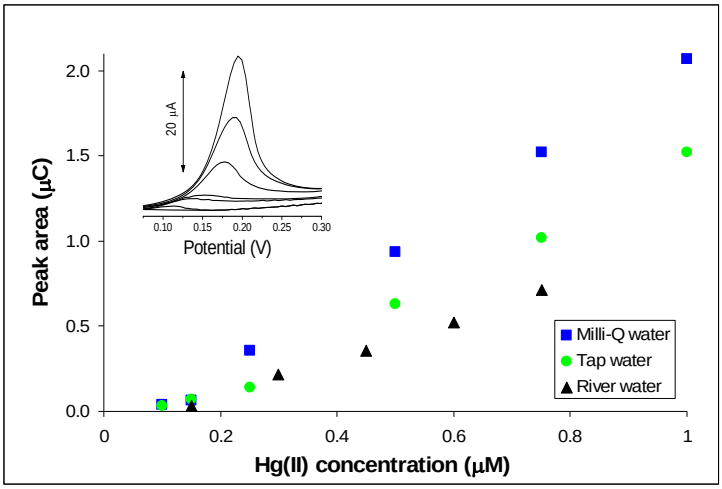


Figure 7. Matrix effect in Hg(II) determination in different samples (Milli-Q, tap and river water) measured by anodic stripping square wave voltammetry after 5 min preconcentration time and 60 s of electrolysis time in 0.1 M HCl. Inset shows representative voltammograms in tap water of different Hg(II) solutions in ascending order of concentration.

After prove that the matrix have an important effect on the signal, water samples were spiked with Hg(II) to obtain a measurable level of 0.15 μM. To compensate matrix effects, Hg(II) was determined in the real water samples by standard addition method and accuracy was assessed by recovery test. In this conditions, recovery values of 92 and 98, RSD 4-7% (n = 3) were obtained for river and tap water, respectively, suggesting that methodology developed can be used for practical analysis of Hg(II) in real water samples.

Table 2
Linear parameters of Hg(II) analytical curves measured at CPE modified with MTTZ-MSU-2 in various matrices [a].

Sample	Linear range (μM)	R^2
Milli-Q water	0.1 – 1	0.999
Tap Water	0.1 – 1	0.992
River water	0.1 – 0.75	0.998

[a] 5 min preconcentration time and 60 s of electrolysis time in 0.1 M HCl.

4.-CONCLUSIONS

Hybrid mesostructured silicas are useful to prepare chemically modified electrodes because its high surface area, high porosity, controllable and narrowly distributed pore size, ordered pore arrangement and high functionalization degree. In this study, we have demonstrated that a CPE modified with MTTZ-MSU-2 can be used to determine Hg(II) by anodic stripping voltammetry in natural waters. The electrode prepared has as main advantages a low LOD (0.021 μM after 5 min preconcentration time) and a good reproducibility, thus has a great potential to be used in the development of portable analyzers for monitoring heavy metal ions in water samples.

Acknowledgements

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5.-REFERENCES

[1] Kresge C., Leonowicz M., Roth W., Vartulli J., Beck J.; **Ordered mesoporous molecular sieves synthesized by a liquid-crystal template mechanism.** *Nature 359 (1992) 710-712.*

[2] Beck J. S., Vartulli J. C., Roth W. J., Leonowicz M. E., Kresge C. T., Schmitt K. D., Chu C. T. W., Olson D. H., Sheppard E. W., McCullen S. B., Higgins J. B., Schlenker J. L.; **A New Family of Mesoporous Molecular Sieves Prepared with Liquid Crystal Templates.** *J. Am. Chem. Soc. 114 (1992) 10834-10843.*

[3] Stein A., Melde B., Schroden R.; **Hybrid inorganic-organic mesoporous silicates-nanoscopic reactors coming of age.** *Adv. Mater. 12 (2000) 1403-1419.*

[4] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Mesoporous silica functionalized with 2-mercaptopyridine: Synthesis, characterization and employment for Hg(II) adsorption.** *Microporous Mesoporous Mat. 89 (2006) 58-68.*

[5] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Preparation of 2-mercaptobenzothiazole-derivatized mesoporous silica and removal of Hg(II) from aqueous solution.** *J. Environ. Monit. 8 (2006) 214-222.*

[6] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **Adsorption of cadmium(II) from aqueous media onto a mesoporous silica chemically modified with 2-mercaptopyrimidine.** *J. Mater. Chem. 16 (2006) 1757-1764.*

[7] Pérez-Quintanilla D., del Hierro I., Fajardo M., Sierra I.; **2-Mercaptothiazoline modified mesoporous silica for mercury removal from aqueous media.** *J. Hazard. Mater. B 134 (2006) 245-256.*

[8] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Preparation, characterization, and Zn^{2+} adsorption behavior of chemically modified MCM-41 with 5-mercapto-1-methyltetrazole.** *J. Colloid Interface Sci. 313 (2007) 551-562.*

[9] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Functionalized HMS mesoporous silica as solid phase extractant for Pb(II) prior to its determination by flame atomic absorption spectrometry.** *J. Sep. Sci. 30 (2007) 1556-1567.*

[10] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Preconcentration of Zn(II) in water samples using a new hybrid SBA-15-based material.** *J. Hazard. Mater. 166 (2009) 1449-1458.*

[11] Brown J., Richer R., Mercier L.; **One-step synthesis of high capacity mesoporous Hg^{2+} adsorbents by non-ionic surfactant assembly.** *Microporous Mesoporous Mat. 37 (2000) 41-48.*

[12] Mercier L.; in **Studies in surface science and catalysis,** ed. Sayari A., *Elsevier Science vol. 129, Amsterdam, 2000.*

[13] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Synthesis and characterization of novel mesoporous silicas of the MSU-*X* family for environmental applications.** *J. Nanosci. Nanotechnol. 9 (2009) 4901-4909.*

[14] Pérez-Quintanilla D., Sanchez A., del Hierro I., Fajardo M., Sierra I.; **Solid phase extraction of Pb(II) in water samples using a new hybrid inorganic-organic mesoporous silica prior to its determination by FAAS.** *Microchim. Acta 165 (2009) 291-298.*

[15] Walcarius A.; **Analytical applications of silica-modified electrodes – A comprehensive review.** *Electroanalysis 10 (1998) 1217-1235.*

[16] Walcarius A.; **Electroanalysis with pure, chemically modified, and sol-gel-derived silica-based materials.** *Electroanalysis 13 (2001) 701-718.*

[17] Walcarius A.; **Impact of mesoporous silica-based materials on electrochemistry and feedback from electrochemical science to the characterization of these ordered materials.** *C. R. Chim. 8 (2005) 693-712.*

[18] Morante-Zarcero S., Sanchez A., Fajardo M., del Hierro I., Sierra I.; **Voltammetric analysis of Pb(II) in natural waters using a carbon paste electrode modified with 5-mercapto-1-methyltetrazol grafted on hexagonal mesoporous silica.** *Microchim. Acta 169 (2010) 57-64.*

[19] Sanchez A., Morante-Zarcero S., Pérez-Quintanilla D., Sierra I., del Hierro I.; **Development of screen-printed carbon electrodes modified with functionalized mesoporous silica nanoparticles: Application to voltammetric stripping determination of Pb(II) in non-pretreated natural waters.** *Electrochim. Acta 55 (2010) 6983-6990.*

[20] Wang J.; **Analytical electrochemistry,** *Willey, New York, 2000.*

[21] Sanchez A., Walcarius A.; **Surfactant-templated sol-gel silica thin films bearing 5-mercapto-1-methyl-tetrazole on carbon electrode for Hg(II) detection.** *Electrochim. Acta 55 (2010) 4201-4207.*

[22] Aleixo L. M., Souza M. F. B., Godinho O. E. S., de Oliveira Neto G., Gushikem Y., Moreira J. C.; **Development of a chemically modified electrode based on carbon paste and functionalized silica gel for preconcentration and voltammetric determination of mercury(II).** *Anal. Chim. Acta 271 (1993) 143-148.*

[23] Souza M. F. B., Bertazzoli R.; **Preconcentration and voltammetric determination of mercury(II) at a chemically modified glassy carbon electrode.** *Anal. Chem. 68 (1996) 1258-1261.*

[24] Walcarius A., Despas C., Trens P., Hudson M. J., Bessière J.; **Voltammetric in situ investigation of an MCM-41-modified carbon paste electrode – a new sensor.** *J. Electroanal. Chem. 453 (1998) 249-252.*

[25] Dias Filho N. L., do Carmo D. R., Rosa A. H.; **An electroanalytical application of 2-aminothiazole-modified silica gel after adsorption and separation of**

Hg(II) from heavy metals in aqueous solution. *Electrochim. Acta 52 (2006) 965-972.*

[26] Tonle I. K., Ngameni E., Walcarius A.; **Preconcentration and voltammetric analysis of mercury(II) at a carbon paste electrode modified with natural smectite-type clays grafted with organic chelating groups.** *Sens. Actuator B-Chem. 110 (2005) 195-203.*

[27] Yantasee W., Deibler L. A., Fryxell G. E., Timchalk C., Lin Y.; **Screen-printed electrodes modified with functionalized mesoporous silica for voltammetric analysis of toxic metal ions.** *Electrochem. Commun. 7 (2005) 1170-1176.*

[28] Yantasee W., Lin Y., Zemanian T. S., Fryxell G. E.; **Voltammetric detection of lead(II) and mercury(II) using a carbon paste electrode modified with thiol self assembled monolayer on mesoporous silica (SAMMS).** *Analyst 128 (2003) 467-472.*

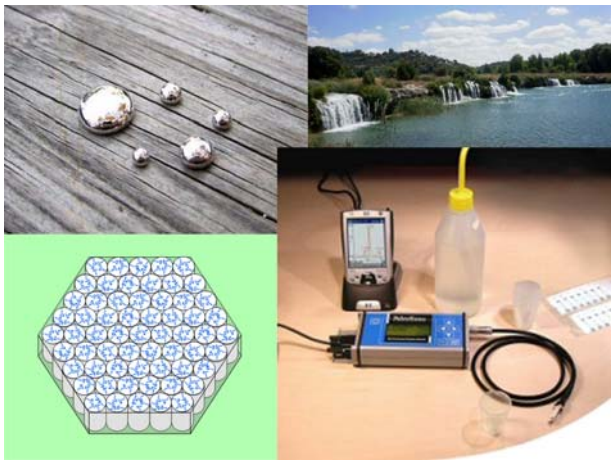
[29] Tchinda A. J., Ngameni E., Walcarius A.; **Thiol-functionalized porous clay heterostructures (PCHs) deposited as thin films on carbon electrode: Towards mercury(II) sensing.** *Sens. Actuator B-Chem. 121 (2005) 113-123.*

[30] Cesarino I., Marino G., do Rosario Matos J., Gomes Cavalherio E. T.; **Evaluation of a carbon paste electrode modified with organofunctionalised SBA-15 nanostructured silica in the simultaneous determination of divalent lead, copper and mercury ions.** *Talanta 75 (2008) 15-21.*

SURFACTANT-TEMPLATED SOL-GEL SILICA THIN FILMS BEARING 5-MERCAPTO-1-METHYL-TETRAZOLE ON CARBON ELECTRODE FOR Hg(II) DETECTION.

Alfredo Sánchez and Alain Walcarius.

Electrochimica Acta 55 (**2010**) 4201-4207.



SURFACTANT-TEMPLATED SOL-GEL SILICA THIN FILMS BEARING 5-MERCAPTO-1-METHYL-TETRAZOLE ON CARBON ELECTRODE FOR Hg(II) DETECTION.

ABSTRACT

Surfactant-templated mesoporous silica films functionalized with 5-mercapto-1-methyl-1-H-tetrazole (MTTZ) groups have been deposited in one step onto glassy carbon electrodes. This was made by evaporation-induced self-assembly co-condensation of triethoxysilyl-propyl-5-mercapto-1-methyltetrazole and tetraethoxysilane in the presence of cetyltrimethylammonium bromide. After template extraction, the film was highly porous and enabled fast mass transport of redox probes from the surrounding solution to the electrode surface. This attractive feature, associated to the easy access to MTTZ binding sites in such ordered mesoporous environment, can be exploited for effective analyte preconcentration and sensitive subsequent electrochemical detection. This has been illustrated using Hg(II) as a model analyte. Interestingly, effective Hg(II) accumulation was possible at low pH values (i.e., where other metal ions are not bonded to the hybrid material) and detection was made by anodic stripping square wave voltammetry after medium exchange to an HCl-containing solution where analyte desorption was going on. After optimization of both preconcentration and detection steps, the sensor exhibited good sensitivity, a linear response in the sub-micromolar concentration range, and a detection limit down to the nanomolar one.

1.-INTRODUCTION

Silica-based organic-inorganic hybrid materials have become increasingly used as electrode modifiers because of their attractive properties [1, 2]. These latter arise from the combination, in a single solid, of a rigid inorganic framework (ensuring mechanical stability) with the intrinsic reactivity of organic groups covalently attached to the inorganic matrix (tailor-made and durable functionality). Two main strategies have been applied to confine such hybrid materials at electrode/solution interfaces: (1) their dispersion as dry powders into carbon-based composites (e.g., carbon paste or ceramic-carbon composite electrodes) [3, 4], or (2) thin film deposition on solid electrode surfaces [5]. (Organo)silica-modified electrodes have found applications in various fields, such as sensing and biosensing, electrocatalysis, power sources, protection against corrosion [6-13].

Most of their applications to date in electrochemical sensing are based on modified carbon paste electrodes [8]. This is notably the case of preconcentration electroanalysis in which the hybrid material is exploited to accumulate the analyte prior to its voltammetric detection (see, e.g., [14-17]). For such application, the resort to ordered mesoporous materials obtained by the surfactant-template route [18-22] has proven to be of particular interest to increase the sensitivity of (organo)silica-modified electrodes [23, 24]. Actually, the regular mesostructure of the accumulation material induces very fast mass transport of the target analyte to the binding sites, enhancing thereby the preconcentration efficiency and thus the electrode response in comparison to non-ordered related porous solids [25]. This has been especially demonstrated for mercury binding to thiol-functionalized materials and extended afterwards to other analytes (heavy metal ions, uranium species) at mesoporous adsorbents bearing other complexing ligands (carbamoyl phosphonic acid, benzothiazolethiol, phenylthiourea, cyclam derivatives) [16, 26-31].

Electrochemical sensing devices designed from the integration of functionalized mesoporous materials under the form of continuous thin films on electrode surfaces are less widespread [32-35]. This is probably because generating homogeneous organic-inorganic hybrid silica films with controllable mesostructures and tuneable functionality is rather recent [36]. The only examples to date are based on very simple ligands. Thiol-functionalized mesoporous silica film electrodes prepared by evaporation-induced self-assembly have been applied to the detection of Pb(II) [32], Ag(I) [33], or Hg(II) [34]. Cu(II) sensing has also been reported using oriented mesoporous silica films bearing aminopropyl groups, which were prepared by electro-assisted self-assembly onto electrode surfaces [37].

Recently, a more sophisticated ligand, 5-mercapto-1-methyl-1-H-tetrazole (MTTZ), has been grafted on mesoporous silica particles via 3-

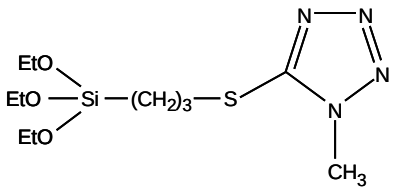
chloropropyltriethoxysilane acting as a linker between MTTZ and the silica surface, and the resulting hybrid material has been used to adsorb Pb(II) and Zn(II) species [38-41]. MTTZ was also attached to the surface of clay minerals by the same way and applied to thorium adsorption [42].

In this work, we have prepared surfactant-templated MTTZ-functionalized silica films on glassy carbon electrode, via a one-step synthesis method, by evaporation-induced self-assembly co-condensation of tetraethoxysilane and triethoxysilyl-propyl-5-mercapto-1-methyl-tetrazole in the presence of cetyltrimethylammonium bromide. After basic physico-chemical characterization and template extraction, the novel porous film electrode was applied to the preconcentration electroanalysis of Hg(II) species (chosen as model analyte) in acidic medium, according to a two-step procedure (voltammetric detection after open-circuit accumulation). The influence of various factors likely to affect both the accumulation and detection steps has been thoroughly investigated.

2.-EXPERIMENTAL

2.1.-Chemicals and reagents

Tetraethoxysilane (TEOS, 98%) and cetyltrimethylammonium bromide (CTAB, 98%) were purchased from Fluka. The MTTZ-siloxy derivative (triethoxysilyl-propyl-5-mercapto-1-methyl-tetrazole) was synthesized as previously described [38] and its structure (Scheme 1) confirmed by ^{13}C and ^{1}H NMR. Ethanol (95–96%, Merck) and HCl (37%, Aldrich) were used for precursor hydrolysis and film deposition, as well as for extraction of the surfactant template. $Ru(NH_3)_6Cl_3$ (Aldrich), $K_3Fe(CN)_6$ (Fluka) and $Fc(MeOH)_2$ (Alfa Aesar) have been used as redox probes, usually in solutions containing potassium hydrogen phthalate (KHP, Merck) as supporting electrolyte. Analytical grade $Hg(NO_3)_2$ (Prolabo) and HNO_3 (65%, Merck) were used for the preparation of diluted mercury solutions; their concentration was checked using a certified standard solution (Hg^{2+} 1.001±0.002 g L^{-1} in HNO_3 3 M, Merck). All solutions were prepared with high-purity water (18 MΩ cm) from a millipore milli-Q water purification system.



Scheme 1

2.2.-Preparation of the film electrodes

MTTZ-modified silica films were deposited by spin-coating on freshly polished surfaces of glassy carbon electrode (GCE, purchased from EG&G, Princeton Applied Research), according to a procedure adapted from Ref. [43]. The sol solutions were typically prepared by heating a mixture of 2.08 g TEOS (including selected equivalent fractions of MTTZ derivative, from 0 to 20% relative to TEOS), 5.5 g EtOH, 0.5 g water and 0.4 g of a 0.1 M HCl solution, at 70 ºC for 1 h. 0.78 g of CTAB dissolved in 10 g of EtOH was then added to this mixture, which was stirred again for 1 h at room temperature. GCE surfaces have been carefully polished before deposition of the film, using successively alumina suspensions made of Al_2O_3 powders (Buehler) of 1, 0.3 and 0.05 µm particle sizes; they were then washed with ethanol and water. Hundred microliters of sol (prepared as above) was deposited on the electrode surface, and left 30 s before the electrode was rotated at 7000 rpm for 30 s (optimized

values). This was performed using a disk electrode rotator (model 636, EG&G, Princeton Applied Research). The spin-coated films were then treated for 20 min at 60 °C. The extraction of the surfactant was performed in an ethanol solution containing 0.1 M HCl under stirring conditions for 15 min. The film electrodes were used in the electrochemical experiments without further treatment.

2.3.-Apparatus

Basic characterization of the film properties was carried out using various physico-chemical techniques. The film structure and morphology was examined by transmission electron microscopy (TEM) using a Philips CM20 microscope at an acceleration voltage of 200kV, and X-ray diffraction (XRD) by collecting data at room temperature using a classical diffractometer (X'PERT PRO, Philips), equipped with a Cu anode (quartz monochromator, $K\alpha_1$ radiation, λ = 0.154056 nm, scanning range: 0.5 - 10° (2θ), step size: 0.02° per 2 s). X-ray photoelectron spectroscopy (XPS) analyses were carried out on a KratosAxis Ultra spectrometer (Kratos Analytical, UK) with a hemispherical energy analyzer using a monochromatic Al $K\alpha$ source (1486.6 eV). All electrochemical experiments were carried out with a Palm-Sens potentiostat and PSLite electrochemical analysis system, in a conventional three-electrode cell. The working electrodes were the MTTZ film-modified glassy carbon disk electrodes, a Pt wire served as the counter electrode and an Ag/AgCl electrode (Metrohm) was used as reference. The Metrohm 691 pH-meter was used for pH monitoring.

2.4.-Electrochemical and analytical procedures

The film permeability properties, before and after template extraction, were characterized by cyclic voltammetry. The measurements have been carried out in buffered solution (50 mM KHP, pH 4.1) containing 1 mM $Ru(NH_3)_6^{3+}$, $Fe(CN)_6^{3-}$ or $Fc(MeOH)_2$. The curves have been recorded at a scan rate of 50 mV·s^{-1} if not specified otherwise.

Preconcentration electroanalysis of Hg(II) was performed following a two-step methodology. In a first step, the film electrode was immersed into a preconcentration cell containing Hg(II) at the desired concentration, most often at pH 1 (0.1 M HNO_3) if not specified otherwise, and the accumulation was carried out at open-circuit under constant stirring for selected periods of time. It was then removed from this solution, rinsed with milli-Q water and transferred to a detection medium (optimum composition: 0.1 M HCl) where a potential-controlled electrolysis was applied (-0.4 V for 60 s); this step was followed by a final anodic stripping voltammetry analysis performed in the square wave mode (in the -0.4 V to +0.8 V potential range). Square wave voltammetry was applied with optimized parameters of modulation amplitude (50 mV), frequency (100 Hz) and step height (5 mV). No stirring conditions were used in both cathodic electrolysis and stripping steps. All

measurements were made at room temperature and under an atmospheric environment. After each series of measurements, a second "checking" scan was applied to ensure that no metal was left on the electrode surface. Each measurement was performed in triplicate and the average value was reported.

3.-RESULTS AND DISCUSSION

3.1.-Characterization of MTTZ-functionalized thin films

Figure 1 depicts typical TEM picture and XRD pattern that have been obtained for a MTTZ-functionalized silica film generated by spin-coating in the presence of CTAB surfactant template. Even if imaging by TEM is not so easy as pieces of the film have to be removed by scratching and deposited on a small grid for analysis, part A of the figure shows the presence of mesopore channels, which seem to be spatially arranged parallel to each other (as noticeable on the thinnest part of the sample). The mesostructural order is confirmed by XRD data exhibiting a sharp diffraction line at 2θ equal 3 (see part B in Figure 1), indicating the existence of a hexagonal phase with lattice parameter of 2.9 nm (as calculated from the Bragg law). This value is lower than that reported for pure silica CTAB-based mesoporous silica films [44–46], indicating that introducing MTTZ groups in the material induced some lattice contraction. The additional less-defined signal of much lower intensity centered at 2θ values around 2.4 is due to the underlying GCE support. These results demonstrate that a mesoporous films has been indeed build around the CTAB template, which would result in a functionalized porous material upon surfactant removal and thus possible application to analyte preconcentration and subsequent electrochemical detection.

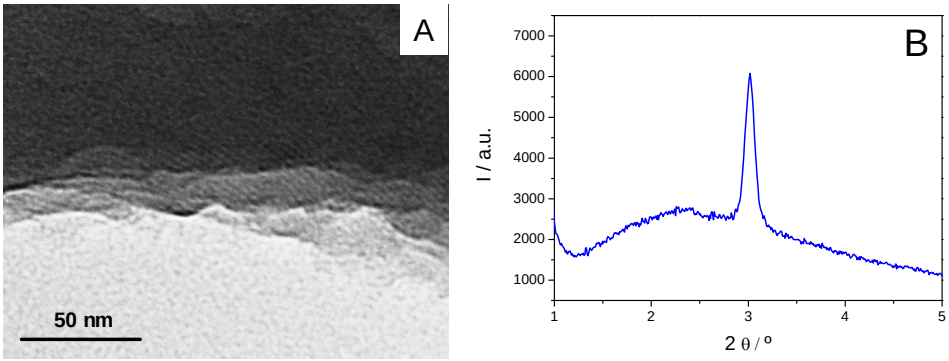


Figure 1. (A) Transmission electron micrograph and (B) X-ray diffraction pattern obtained for a 10%-MTTZ-functionalized silica thin film generated by spin-coating on glassy carbon electrode.

XPS was then used to point out the presence of MTTZ groups in the template-extracted mesoporous films. An illustrative spectrum is depicted in Figure 2, showing the presence of the silica framework (Si and O signals) along with N and S components arising from MTTZ groups (C element cannot be considered in the

analysis because of the well-known presence of C contamination in XPS experiments). Interestingly, the narrow scanned spectrum obtained in the S2p region (see top right inset in Figure 2) indicates a single component ($S2p_{3/2}$ at 164.0 eV and $S2p_{1/2}$ at 165.5 eV), which can be attributed to S^{-II} [47], and no contributions at higher binding energies (which would have occurred in case of oxidized samples). The N-to-S molar ratio was nearly 6, which is more than the theoretical value of 4 expected for MTTZ groups (1 S for 4 N). This N excess can be explained by the presence of some residual cetyltrimethylammonium groups that have not been washed out of the film during the template extraction step, which is sustained by the presence of two components in the N1s signal (see top left inset in Figure 2).

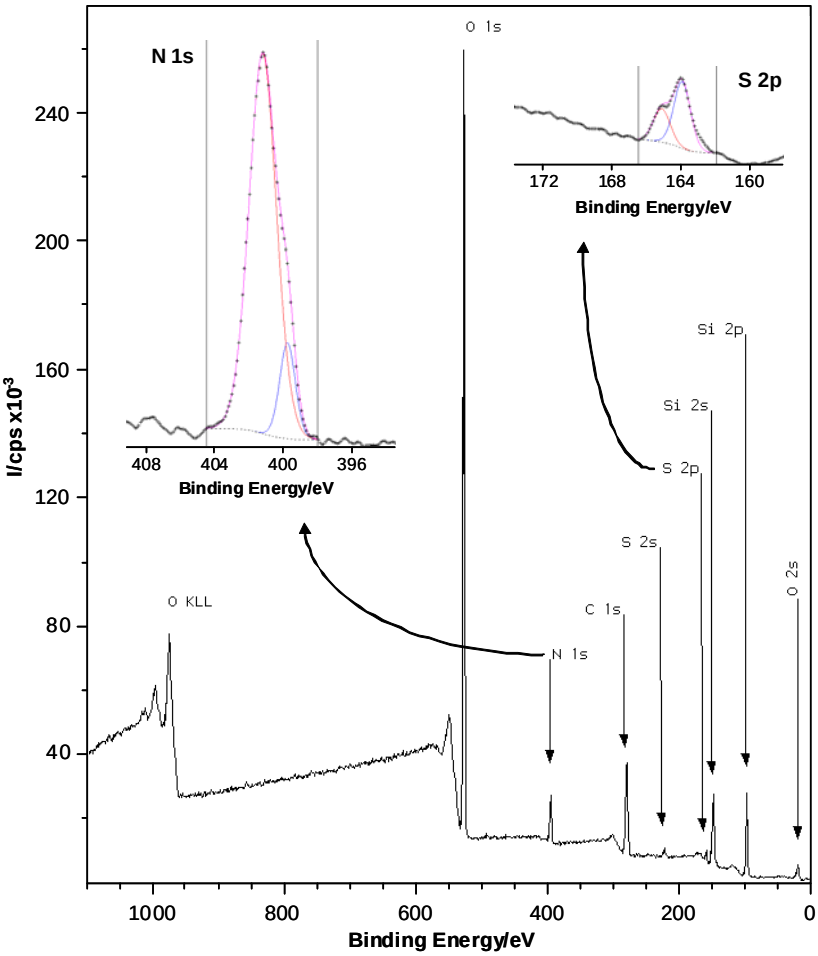


Figure 2. X-ray photoelectron spectra recorded for a 10%-MTTZ-functionalized silica thin film; insets represent narrow scanned spectra, respectively for nitrogen (N1s) and sulphur (S2p).

Figure 3 shows the effect of template removal on the permeability properties of the MTTZ-functionalized films to selected redox probes. Before surfactant extraction, both $Ru(NH_3)_6^{3+}$ and $Fe(CN)_6^{3-}$ gave rise to almost no noticeable voltammetric signals, confirming the presence of a rather homogeneous film with very few defects onto the electrode surface. After removal of the template, the porosity was released so that redox probes were now able to diffuse across the film from the solution to the electrode surface, generating thereby voltammetric signals of the same order of magnitude as on bare glassy carbon. Signal enhancement was lower with negatively charged $Fe(CN)_6^{3-}$ and slightly higher with positively charged $Ru(NH_3)_6^{3+}$ because the silica surface is negative in the conditions applied here (i.e., pH 4.1, in agreement with the point of zero charge of silica reported to be in the range 2–3 [48]). This can induce significant electrostatic attraction/repulsion effects in such confined environments [33, 45], leading to possible cation accumulation and anion rejection. The behaviour of $Fc(MeOH)_2$ was different as this neutral probe is likely to solubilise in surfactant micelles, so that it gave rise to a well-defined voltammetric curve prior to removal of CTAB from the mesoporous film (see curve (b) in Figure 3C). Peak potentials were shifted to the anodic direction with respect to solution-phase $Fc(MeOH)_2$ because this analyte was stabilized in the liquid crystalline phase and its oxidation product (positively charged ferricinium moieties) was subject to electrostatic repulsion by the ammonium groups [49], in agreement with previous observations made on other kinds of surfactant-templated silica films [33, 50]. Again, after template extraction, solution-phase $Fc(MeOH)_2$ species are likely to cross the film and to be detected onto the underlying electrode surface. Note that the high porosity of mesoporous films gave rise to systems which did not behave as electrodes with partially blocked surface as the voltammetric process at the MTTZ-modified electrodes exhibit the same reversibility as at the bare GCE, at least at the low scan rate used here (50mV s^{-1}). Much higher potential scan rates were necessary to evidence some larger peak potential separations at the modified electrode with respect to bare one. All the above results demonstrate that MTTZ-functionalized silica films prepared by the surfactant-template route are porous enough to enable fast mass transport and are thus good candidates for applications where diffusion constitutes the rate-determining step, such as preconcentration electroanalysis.

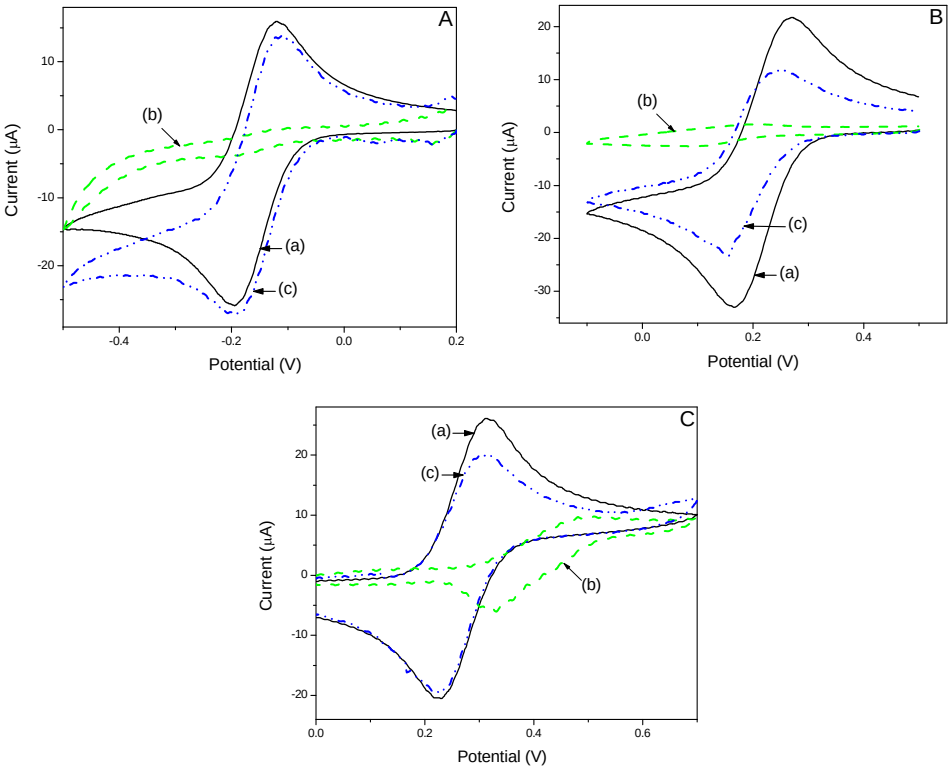


Figure 3. Cyclic voltammetric curves recorded in 0.05 M hydrogen phthalate solutions (pH 4.1) containing (A) 1 mM $Ru(NH_3)_6^{3+}$, (B) 1 mM $Fe(CN)_6^{3-}$ and (C) 1 mM $Fc(MeOH)_2$ at a glassy carbon electrode covered with a 10%-MTTZ-functionalized silica thin film, respectively before (b) and after (c) surfactant extraction; curves (a) represent the electrochemical responses sampled on bare glassy carbon. Potential scan rate was 50 mV s^{-1}.

3.2.-Factors affecting preconcentration electroanalysis of Hg(II)

Hg(II) was used as a model analyte to evaluate the analytical potentialities of the novel mesoporous film electrode. Figure 4 shows that the MTTZ-functionalized film enabled the sensitive detection of this analyte as well-defined anodic stripping square wave voltammetric signals have been observed after only 5 min accumulation from a 1×10^{-7} M Hg(II) solution. Comparison between curves obtained for mesoporous silica films prepared with and without MTTZ clearly demonstrates that the Hg(II) binding process is preferably due to complexation to the incorporated ligands and not noticeably to interaction with the silica surface (Hg(II) in the form of

$Hg(OH)_2$ is known to accumulate on mesoporous silica by reacting with surface silanol groups and then to be electrochemically detected, but this is only possible at concentrations high enough due to rather low equilibrium constant values [51]). Moreover, Hg(II) binding to MTTZ ligands is strong enough to ensure effective accumulation of this analyte in highly acidic media (e.g., pH 1), which are usually used to treat and/or to store samples containing heavy metal ions. For example, similar intensity was observed for preconcentration experiments performed at pH values 1, 4, and 6. This is promising for selective recognition as other metal ions likely to be complexed to MTTZ require much higher pH values (typically above 6 [39, 41]).

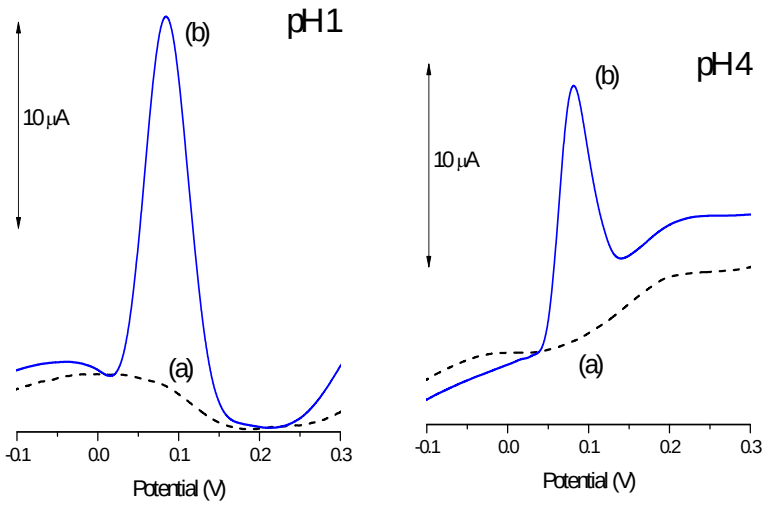


Figure 4. Square wave voltammograms obtained at glassy carbon electrodes covered with (a) a non-functionalized silica film or (b) 10%-MTTZ-functionalized silica thin film, after 5 min preconcentration from 0.1 µM Hg(II) solutions in 0.1 M HNO_3 (pH 1) or in 10^{-4} M HNO_3 + 0.1 M $NaNO_3$ (pH 4); detection was made after medium exchange to a 0.1 M HCl solution and 1 min electrolysis at -0.4 V prior to anodic stripping. SWV parameters were 50 mV modulation amplitude, 100 Hz frequency, and 5 mV step height.

Because of the insulating character of silica, the accumulated Hg(II) species have to be removed from the adsorbent prior to detection. This has been achieved by transferring the electrode to a separate medium whose composition has been adjusted to dissociate the MTTZ-Hg(II) complex, and this was made while applying a negative potential to the electrode (reducing Hg(II) into metallic mercury) in order to avoid any leaching of desorbed analyte out of the film in the surrounding solution, which would lead to non effective detection. HCl was found to operate quite well in that role but its concentration must be adjusted carefully to get sensitive detection signals (Figure 5A). The optimal HCl concentration was found to lie in the 0.1 – 0.2 M range. Lower values could be explained by either a too low complexation power to desorb Hg(II)

species from the MTTZ material or a change in the anodic stripping detection mechanism from a two-electron process to one-electron reoxidation with calomel formation [52, 53] (high [Cl^-] is known to favour $Hg^{(0)}$ oxidation into $HgCl_4^{2-}$ whereas $Hg^{(0)}$ oxidation in less concentrated chloride medium stops to Hg(I) in the form of calomel [53]). The presence of some calomel has been indeed identified by examining the forward and backward components of the SWV signals, which exhibited a reversible contribution (i.e., $Hg^{(0)}/Hg_2Cl_2$, as the $Hg^{(0)}/HgCl_4^{2-}$ system is not reversible [53]). The sensitivity decrease at higher HCl concentrations is not clearly understood but might be due to the fact that complexes formed in such conditions are negatively charged ($HgCl_3^-$ or $HgCl_4^{2-}$ instead of $HgCl_2$ [54]), suffering thus from more restricted mass transport rates through the negatively charged mesopores, as discussed above for $Fe(CN)_6^{3-}$ species (Figure 3B). Another explanation can also be found in the fact that Hg(II) species are more strongly complexed in highly concentrated chloride solutions, resulting in less quantitative reduction at the electrolysis potential applied here (−0.4 V), in agreement with the irreversible behaviour of the $Hg^{(0)}/HgCl_4^{2-}$ system [53]. The time spent for the electrolysis prior to anodic stripping is also expected to affect the detection step as it must be long enough to ensure complete desorption of the previously accumulated species. Figure 5B shows that it was very fast, thanks to the regular mesoporous structure of the film, as maximum current responses were already observed after 30 s electrolysis and maintained constant over 2 min. Finally, the optimal potential to be applied for electrolysis was found to be −0.4 V. A HCl concentration of 0.1 M in the detection medium where electrolysis at −0.4 V was applied for 1 min prior to anodic stripping square wave voltammetry constitute the conditions chosen for further experiments.

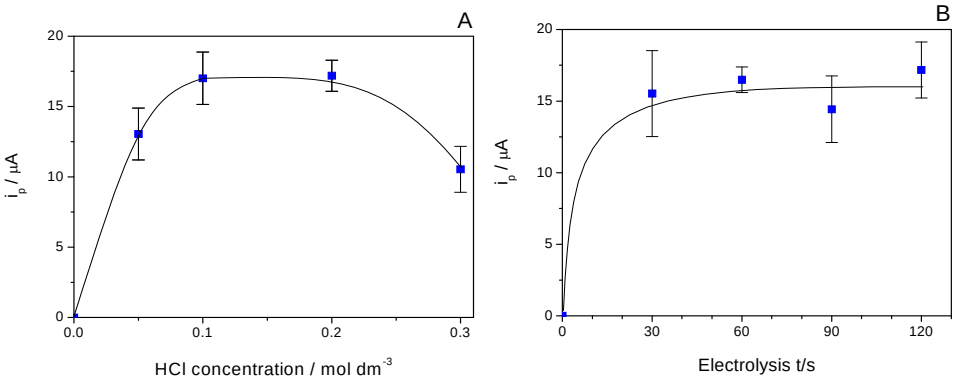


Figure 5. Influence of (A) HCl concentration in the detection medium and (B) electrolysis time on the stripping peak currents measured by square wave voltammetry after 5 min preconcentration from 0.1 μM Hg(II) solution in 0.1 M HNO_3. Data reported in part (A) have been obtained with 1 min electrolysis time and those in part (B) have been obtained in 0.1 M HCl. The glassy carbon electrode was covered with a 10%-MTTZ-functionalized silica thin film.

Figure 6 shows that the MTTZ content in the film has dramatic influence on the electrode sensitivity. No signal was observed for unmodified silica film, demonstrating the critical role played by MTTZ ligands in the preconcentration step and showing that the eventual presence of some residual surfactant groups has no influence on the accumulation/detection process. A minimum content of 5% was necessary to observe a significant response, which was then increasing to reach a maximum at 10% MTTZ and finally decreased continuously for higher functionalization levels. The first increase results from the increasing number of binding sites in the material while the subsequent loss in sensitivity at higher MTTZ contents is less easy to explain but it can be due to slower mass transport rates in a less porous and/or less organized environment (high functionalization levels are usually associated to porosity decrease and loss of mesostructural order [18–22]), as also reported for other electrodes modified with mesoporous organosilica materials [33, 37, 55]. The 10%-MTTZ film electrode was then exclusively used afterwards.

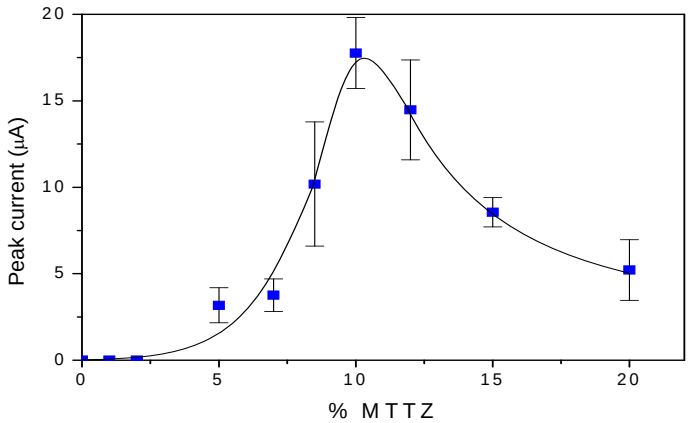


Figure 6. Influence of the content of MTTZ in the starting sol on the stripping peak current measured by square wave voltammetry after 1 min electrolysis at -0.4 V in 0.1 M HCl. Hg(II) species have been preconcentrated for 5 min from a solution containing 0.1 μM of the analyte in 0.1 M HNO_3. The glassy carbon electrodes were modified with MTTZ-functionalized silica thin film prepared from the same sol composition but by varying the MTTZ/(TEOS+MTTZ) ratio from 0.01 (1 %) to 0.20 (20 %).

Another important parameter likely to influence the preconcentration step is the accumulation time. This is illustrated in Figure 7 for three distinct Hg(II) concentrations. As expected, peak currents were of higher intensities at longer accumulation times as a result of larger amounts of accumulated species but this trend tended to level off as more rapidly as high was the analyte concentration. The limitation observed at "high" concentration (see curve (c) in Figure 7) can be explained by saturation of the binding sites in the thin film but this is apparently not

the only reason as some (yet less) restriction was also observed at lower Hg(II) concentrations (see curves (a) and (b) in Figure 7), suggesting the existence of another limiting process. On the basis of previous observations [25, 56], a possible explanation can be the formation of a positively charged MTTZ-Hg^{2+} complex in confined mesopores, which act as an electrostatic barrier slowing down the ingress of additional Hg^{2+} species, limiting thereby the effectiveness of the preconcentration step. Another feature that can be noticed in Figure 7 is the lack of linear increase between peak currents and accumulation times at short preconcentration time (lower response as expected), especially at low Hg(II) concentration. Such "delay" which was not observed with modified carbon paste electrodes [15, 30, 31], can be explained by the film configuration of the electrode: the accumulation starts on the upper part of the film contacting the solution, which results in very low amounts of accumulated species when operating in dilute solutions, some of them being lost after desorption in the detection medium and therefore not detectable on the electrode surface. This effect is much less pronounced at higher Hg(II) concentration and/or longer accumulation times because the entire volume of the film is concerned with the preconcentration event and the electrode response become linear with the accumulation time.

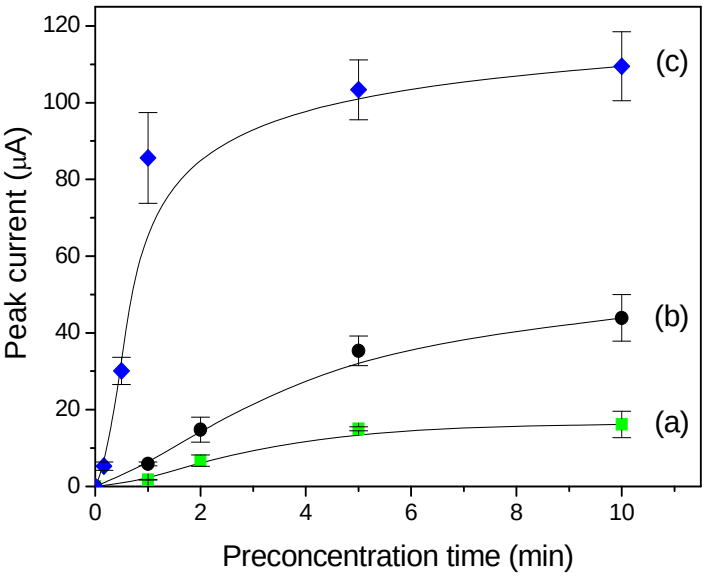


Figure 7. Influence of the accumulation time on the stripping peak currents measured at a glassy carbon electrode covered with a 10%-MTTZ-functionalized silica thin film, for several Hg(II) concentrations. The accumulation media consisted of 0.1 M HNO_3 containing $Hg(NO_3)_2$ at (a) 0.1 μM, (b) 0.3 μM, or (c) 1.0 μM. Detection was made in 0.1 M HCl after 1 min electrolysis at -0.4 V.

Figure 8 shows a typical calibration curve obtained in the sub-micromolar concentration range after 5 min preconcentration at pH 1. The electrode response was linear in the 5×10^{-8} M to 1×10^{-6} M Hg^{2+} concentration range, with detection limits (S/N = 3) of 2.4×10^{-8} M and 2×10^{-9} M, as calculated respectively for 5 min and 20 min accumulation times. Such analytical performance is comparable to that of related electrochemical sensors based on other organically modified mesoporous silica [25, 28, 34, 57].

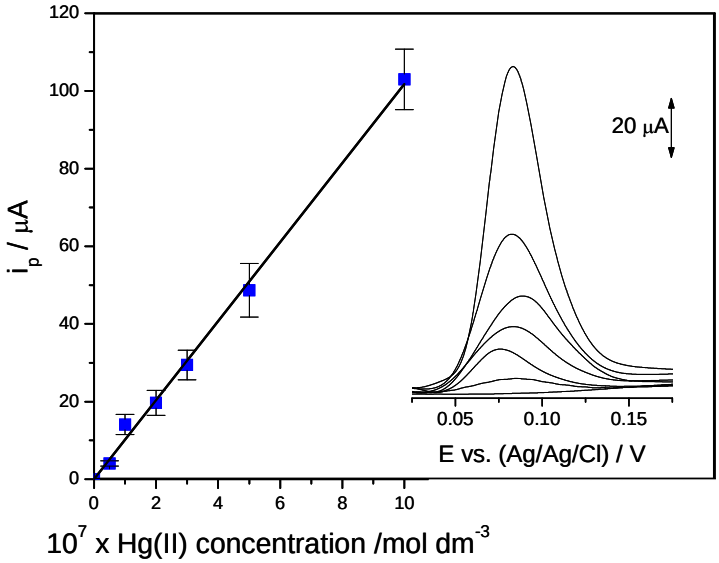


Figure 8. Typical calibration plot for Hg(II) at the 10%-MTTZ-functionalized silica thin film electrode, as obtained for 5 min preconcentration time, and corresponding voltammetric curves (inset); other conditions as in Figure 7.

4.-CONCLUSIONS

We have successfully prepared surfactant-templated silica films with immobilized 5-mercapto-1-methyl-tetrazole groups, in one step, and applied the resulting mesoporous organic–inorganic hybrid layer coated onto glassy carbon electrode in preconcentration electroanalysis. This constitutes the first example of thin film electrode made of a homogeneous mesoporous layer functionalized with a rather sophisticated ligand, which was generated in a single step (to the best of our knowledge, previous electrodes coated with silica films incorporating ligands other than aminopropyl- or mercaptopropyl-ones were either not mesostructured or prepared according to a two-step method). The film was highly porous after template removal, as demonstrated by cyclic voltammetry with the aid of several redox probes, and it was likely to accumulate Hg(II) species, very efficiently down to pH 1. Such preconcentration event was dramatically dependent on the functionalization level of the film and 10% MTTZ was found to be the optimal composition. Detection of this model analyte was possible in the sub-micromolar concentration range, down to a detection limit of 2×10^{-9} M (20 min accumulation).

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5.-REFERENCES

[1] Walcarius A.; **Electrochemical applications of silica-based organic-inorganic hybrid materials.** *Chem. Mater. 13 (2001) 3351-3372.*

[2] Walcarius A., Mandler D., Cox J. A., Collinson M. M., Lev O.; **Exciting new directions in the intersection of functionalized sol–gel materials with electrochemistry.** *J. Mater. Chem. 15 (2005) 3663-3689.*

[3] Rabinovich L., Lev O.; **Sol-gel derived composite ceramic carbon electrodes.** *Electroanalysis 13 (2001) 265-275.*

[4] Sayen S., Etienne M., Bessière J., Walcarius A.; **Tuning the sensitivity of electrodes modified with an organic-inorganic hybrid by tailoring the structure of the nanocomposite material.** *Electroanalysis 14 (2002) 1521-1525.*

[5] Thenmozhi K., Narayanan S. S.; **Electrocatalytic reduction of nitrite ion on a Toluidine Blue sol-gel thin film electrode derived from 3-Aminopropyl trimethoxy silane.** *Electroanalysis 19 (2007) 2362-2368.*

[6] Lev O., Wu Z., Bharathi S., Glezer V., Modestov A., Gun J., Rabinovich L., Sampath S.; **Sol-gel materials in electrochemistry.** *Chem. Mater. 9 (1997) 2354-2375.*

[7] Wang J.; **Sol-gel materials for electrochemical biosensors.** *Anal. Chim. Acta 399 (1999) 21-27.*

[8] Walcarius A.; **Electroanalysis with pure, chemically modified, and sol-gel-derived silica-based materials.** *Electroanalysis 13 (2001) 701-718.*

[9] Ruiz-Hitzky E., Casal B., Aranda P., Galvan J. C.; **Inorganic–organic nanocomposite materials based on macrocyclic compounds.** *Rev. Inorg. Chem. 21 (2001) 125-159.*

[10] Tan S. N., Hua L.; **Electrochemical detection base don sol-gel carbon composite electrodes for capillary electrophoresis.** *Curr. Top. Anal. Chem. 3 (2002) 83-93.*

[11] Hong S.-A., Nam S. W.; **Application of sol-gel techniques in fabrication of fuel cells.** *Stud. Surf. Sci. Catal. 159 (2006) 79-84.*

[12] Aurobind S. V., Amirthalingam K. P., Gomathi H.; **Sol-gel based surface modification of electrodes for electro analysis.** *Adv. Colloid Interface Sci. 121 (2006) 1-7.*

[13] Pagliaro M., Ciriminna R., Palmisano G.; **Silica.based hybrid coatings.** *J. Mater. Chem. 19 (2009) 3116-3126.*

[14] Aleixo L. M., Souza M. F. B., Godinho O. E. S., de Oliveira Neto G., Gushikem Y., Moreira J. C.; **Decelopment of a chemically modified electrode based on carbon paste and functionalized silica gel for preconcentration and voltammetric determination of mercury(II).** *Anal. Chim. Acta 271 (1993) 143-148.*

[15] Etienne M., Bessière J., Walcarius A.; **Voltammetric detection of copper(II) at a carbon paste electrode containing an organically modified silica.** *Sens. Actuator B-Chem. 76 (2001) 531-538.*

[16] Yantasee W., Fryxell G. E., Conner M. M., Lin Y.; **Nanostructured electrochemical sensors based on functionalized nanoporous silica for voltammetric analysis of lead, Mercury and copper.** *J. Nanosci. Nanotechnol. 5 (2005) 1537-1540.*

[17] Dias Filho N. L., do Carmo D. R., Rosa A. H.; **An electroanalytical application of 2-aminothiazole-modified silica gel after adsorption and separation of Hg(II) from heavy metals in aqueous solution.** *Electrochim. Acta 52 (2006) 965-972.*

[18] Stein A., Melde B. J., Schroden R. C.; **Hybrid inorganic-organic mesoporous silicates-nanoscopic reactors coming of age.** *Adv. Mater. 12 (2000) 1403-1419.*

[19] Lebeau B., Patarin J., Sanchez C.; **Desing and properties of hierarchically organized hybrid organic-inorganic nanocomposites.** *Adv. in Tech. of Mat. and Mat. Proc. J. 6 (2004) 298-307.*

[20] Vinu A., Hossain K. Z., Ariga K.; **Recent advances in functionalization of mesoporous silica.** *J. Nanosci. Nanotechnol. 5 (2005) 347-375.*

[21] Hoffmann F., Cornelius M., Morell J., Froeba M.; **Silica-based mesoporous organic-inorganic hybrid materials.** *Angew. Chem.-Int. Edit. 45 (2006) 3216-3251.*

[22] Wan Y., Zhang D., Hao N., Zhao D.; **Organic groups functionalised mesoporous silicates.** *Int. J. Nanotechnol. 4 (2007) 66-99.*

[23] Walcarius A.; **Impact of mesoporous silica-based materials on electrochemistry and feedback from electrochemical science to the characterization of these ordered materials.** *C. R. Chim. 8 (2005) 693-712.*

[24] Walcarius A.; **Electroanalytical applications of microporous zeolites and mesoporous (organo)silicas: recent trends.** *Electroanalysis 20 (2008) 711-738.*

[25] Walcarius A., Etienne M., Sayen S., Lebeau B.; **Grafted silicas in electroanalysis: Amorphous versus ordered mesoporous materials.** *Electroanalysis 15 (2003) 414-421.*

[26] Yantasee W., Lin Y., Fryxell G. E., Busche B. J.; **Simultaneous detection of cadmium, copper, and lead using a carbon paste electrode modified with carbamoylphosphonic acid self-assembled monolayer on mesoporous silica (SAMMS).** *Anal. Chim. Acta 520 (2004) 207-212.*

[27] Cesarino I., Marino G., Matos J. R., Cavalheiro E. T. G.; **Using the organofunctionalised SBA-15 nanostructured silica as a carbon paste**

electrode modifier: Determination of cadmium ions by differential anodic pulse stripping voltammetry. *J. Braz. Chem. Soc. 18 (2007) 810-817.*

[28] Cesarino I., Marino G., Matos J. R., Cavalheiro E. T. G.; **Evaluation of a carbon paste electrode modified with organofunctionalised SBA-15 nanostructured silica in the simultaneous determination of divalent lead, copper and mercury ions.** *Talanta 75 (2008) 15-22.*

[29] Javanbakht M., Divsar F., Badiei A., Fatollahi F., Khaniani Y., Ganjali M. R., Norouzi P., Chaloosi M., Ziarani G. M.; **Determination of picomolar silver concentrations by differential pulse anodic stripping voltammetry at a carbon paste electrode modified with phenylthiourea-functionalized high ordered nanoporous silica gel.** *Electrochim. Acta 54 (2009) 5381-5386.*

[30] Goubert-Renaudin S., Etienne M., Rousselin Y., Denat F., Lebeau B., Walcarius A.; **Cyclam-functionalized silica-modified electrodes for selective determination of Cu(II).** *Electroanalysis 21 (2009) 280-289.*

[31] Goubert-Renaudin S., Moreau M., Despas C., Meyer M., Denat F., Lebeau B., Walcarius A.; **Voltammetric detection of lead(II) using amide-cyclam-functionalized silica-modified carbon paste electrodes.** *Electroanalysis 21 (2009) 1731-1742.*

[32] Yantasee W., Lin Y., Li X., Fryxell G. E., Zemanian T. S., Viswanathan V. V.; **Nanoengineered electrochemical sensor based on mesoporous silica thin-film functionalized with thiol-terminated monolayer.** *Analyst 128 (2003) 899-904.*

[33] Etienne M., Cortot J., Walcarius A.; **Preconcentration electroanalysis at surfactant-templated thiol-functionalized silica thin films.** *Electroanalysis 19 (2007) 129-138.*

[34] Cesarino I., Cavalheiro E. T. G.; **Thiol-functionalized silica thin film modified electrode in determination of mercury ions in natural water.** *Electroanalysis 20 (2008) 2301-2309.*

[35] Walcarius A., Kuhn A.; **Ordered porous thin films in electrochemical analysis.** *Trac-Trends Anal. Chem. 27 (2008) 593-603.*

[36] Sanchez C., Boissière C., Grosso D., Laberty C., Nicole L.; **Desing, synthesis, and properties of inorganic and hybrid thin films having periodically organized nanoporosity.** *Chem. Mater. 20 (2008) 682-737.*

[37] Etienne M., Goux A., Sibottier E., Walcarius A.; **Oriented mesoporous organosilica films on electrode: A new class of nanomaterials for sensing.** *J. Nanosci. Nanotechnol. 9 (2009) 2398-2406.*

[38] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **Functionalized HMS mesoporous silica as solid phase extractant for Pb(II) prior to its determination by flame atomic absorption spectrometry.** *J. Sep. Sci. 30 (2007) 1556-1567.*

[39] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **Preparation, characterization, and Zn^{2+} adsorption behavior of chemically modified MCM-41 with 5-mercapto-1-methyltetrazole.** *J. Colloid Interface Sci. 313 (2007) 551-562.*

[40] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **Preconcentration of Zn(II) in water samples using a new hybrid SBA-15-based material.** *J. Hazard. Mater. 166 (2009) 1449-1458.*

[41] Pérez-Quintanilla D., Sánchez A., del Hierro I., Fajardo M., Sierra I.; **Solid phase extraction of Pb(II) in water samples using a new hybrid inorganic-organic mesoporous silica prior to its determination by FAAS.** *Microchim. Acta 165 (2009) 291-298.*

[42] Guerra D. L., Viana R. R., Airoldi C.; **Adsorption of thorium cation on modified clays MTTZ derivative.** *J. Hazard. Mater. 168 (2009) 1504-1511.*

[43] Etienne M., Walcarius A.; **Evaporation induced self-assembly of templated silica and organosilica thin films on various electrode surfaces.** *Electrochem. Commun. 7 (2005) 1449-1456.*

[44] Han C.-K., Jung S.-B., Park H.-H.; **Effects of tetraethoxysilane vapour treatment on the cetyltrimethylammonium bromide-templated silica mesoporous low-k thin film with 3D close-packed array of spherical pores.** *Appl. Surf. Sci. 237 (2004) 405-410.*

[45] Etienne M., Quach A., Grosso D., Nicole L., Sanchez C., Walcarius A.; **Molecular transport into mesostructured silica thin films: Electrochemical monitoring and comparison between p6m, $P6_3/mmc$, and Pm3n structures.** *Chem. Mater. 19 (2007) 844-856.*

[46] Zhang X., Wu W., Wang J., Liu C.; **Effects of sol aging on mesoporous silica thin films organization.** *Thin Solid Films 515 (2007) 8376-8380.*

[47] Wagner C. D., Naumkin A. V., Kraut-Vass A., Allison J. W., Powell C. J., Rumble Jr. J. R.; **NIST Standard Reference Database 20.** *NIST XPS Database, version 3.5 (web version, 2009).*

[48] Persello J., in **Adsorption on silica surfaces,** ed. Papirer E., *Surf. Sci. Ser. 90 (2000), Marcel Dekker, New York, pp.317-318.*

[49] Ryabov A. D., Amon A., Gorbatova R. K., Ryabova E. S., Gnedenko B. B.; **Mechanism of a "jumping off" ferricenium in glucose oxidase-D-glucose-ferrocene micellar electrochemical systems.** *J. Phys. Chem. 99 (1995) 14072-14077.*

[50] Walcarius A., Sibottier E., Etienne M., Ghanbaja J.; **Electrochemically assisted self-assembly of mesoporous silica thin films.** *Nat. Mater. 6 (2007) 602-608.*

[51] Walcarius A., Bessière J.; **Electrochemistry with mesoporous silica: Selective mercury(II) binding.** *Chem. Mater. 11 (1999) 3009-3011.*

[52] Agraz R., Sevilla M. T., Hernandez L.; **Voltammetric quantification and speciation of mercury compounds.** *J. Electroanal. Chem. 390 (1995) 47-57.*

[53] Moretto L. M., Mazzocchin G. A., Ugo P.; **Electroanalytical study on the ion-exchange voltammetric behaviour of Hg(II) at Tosflex®-coated glassy carbon electrodes.** *J. Electroanal. Chem. 427 (1997) 113-121.*

[54] Baes Jr. C. F., Mesmer R. E.; **The hydrolysis of cations.** *Wiley, New York, (1976), pp. 301-312.*

[55] Walcarius A., Delacôte C., Sayen S.; **Electrochemical probing of mass transfer rates in mesoporous silica-based organic-inorganic hybrids.** *Electrochim. Acta 49 (2004) 3775-3783.*

[56] Delacôte C., Gaslain F., Lebeau B., Walcarius A.; **Factors affecting the reactivity of thiol-functionalized mesoporous silica adsorbents toward mercury(II).** *Talanta 79 (2009) 877-886.*

[57] Yantasee W., Lin Y., Zemanian T. S., Fryxell G. E.; **Voltammetric detection of lead(II) and mercury(II) using a carbon paste electrode modified with thiol self-assembled monolayer on mesoporous silica (SAMMS).** *Analyst 128 (2003) 467-47.*

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