Journal of Environmental Chemical Engineering 4 (2016) 1862–1868

Contents lists available at ScienceDirect

Journal of Environmental Chemical Engineering

journal homepage: www.elsevier.com/locate/jece

# Discrimination for inorganic and organic mercury species by cloud point extraction of polyethylene glycol

Pallabi Samaddar, Kamalika Sen*

*Department of Chemistry, University of Calcutta, 92 APC Road, Kolkata 700009, India*

ARTICLE INFO

*Article history:*
Received 24 December 2015
Received in revised form 2 March 2016
Accepted 7 March 2016
Available online 8 March 2016

*Keywords:*
Cloud point extraction
Polyethylene glycol
Confocal microscopy
Zeta potential
Speciation

ABSTRACT

Speciation and extraction of mercury has been studied using cloud point extraction of polyethylene glycol (PEG). A solution of PEG itself is reported to form cloud at a considerably high temperature which is unfavorable for the extraction and separation of volatile analytes. Different inorganic salt solutions ($Na_2SO_4$, $NaH_2PO_4$, NaCl, NaOAc) when used as additives were found to lower the cloud point temperature of PEG (#6000) effectively. The lowest temperature for cloud formation was observed at 35 °C with 0.8 M $Na_2SO_4$. The cloud so formed was found suitable to discriminate between inorganic and organic mercury species. 1-(2-pyridylazo)-2-napthol (PAN) was used for spectrophotometric detection of inorganic and organic mercury. Confocal microscopic images and zeta potential values reveal the actual interactions of the inorganic Hg species with the organized PEG micelles in aqueous medium. Environmental samples were also analyzed using the present method.



## 1. Introduction

Speciation of mercury has become significant as mercury bio accumulation in different tissues mainly depends on its chemical form and has important implications on tissue-specific toxicity. The most prominent species of mercury are inorganic mercury ($Hg^+$, $Hg^{2+}$) and organic mercury (monomethyl mercury, dimethyl mercury) which enters the water bodies of the nature through several routes. Mercury is exposed to the atmosphere from coal fired power plants as it is highly volatile in nature. It is also released in the air due to volcanic eruptions. Several organisms including a class of seaweeds dwelling near industrial sites accumulate mercury. Water currents of the sea as well as other aquatic organisms broaden the distribution of mercury in natural water bodies. Consequently, drinking water becomes one of the routes of mercury invasion into other living organisms. Due to high bioaccumulation in fish, mercury directly enters the food chain e.g., predatory fish can have up to $10^6$ times higher mercury concentrations than ambient water [1]. This was the cause of the well known event in Minamata Bay in Japan. The World Health Organization (WHO) recommends a maximum intake of methyl-mercury of 1.6 $\mu g\,kg^{-1}$ body weight per week [2]. Therefore, precise monitoring of each mercury species and understanding of species transformations are essential for reliable risk deliberation.

Direct analysis of mercury from aqueous media is possible using several sophisticated instruments such as inductively coupled plasma optical emission spectrometry (ICP-OES) [3,4], inductively coupled plasma mass spectrometry (ICP-MS) [5], cold vapor atomic absorption spectrometry (CV-AAS) [6] and electrothermal atomic absorption spectrometry (ETAAS) [7–9] have been developed to detect Hg species after extraction by cloud point technique. However, these instruments are very expensive to purchase and operate which is further complicated by the exhaustive sample preparation methods. Additionally, these instruments have already inbuilt interferences like solvent dependent nebulization rate, background interferences, etc.

Cloud point extraction is based on the fact that polymer/surfactants in aqueous solutions form micelles and become opaque when it is allowed to heat at a specific temperature (cloud point temperature) or in the presence of an electrolyte. Most of the previous works on cloud point extraction (CPE) method have been done by using Triton X-100 and Triton X-114. Many metal species had been successfully extracted in the Triton rich phase with different chelating agents at different conditions [10–13]. Triton X-100 is inherently toxic as revealed in several experiments [14,15]. Conversely, reports on cloud point extraction using polyethylene glycol (PEG) are very limited [16–18]. PEG is mostly used as an additive to maintain the viscosity of paint, as an additive in the production of paper, to wrap the surfaces of different materials [19], as ingredient of various commercial products like sanitizer, shampoo etc., and even in pharmaceuticals and laxatives. These polymers are commercially available over a wide range of

* Corresponding author.
*E-mail address:* kamalchem.roy@gmail.com (K. Sen).

http://dx.doi.org/10.1016/j.jece.2016.03.010
2213-3437/

molecular weights from 300 g/mol to 10,000,000 g/mol and form various types of vesicular arrangement depending on the number of repetitive polymeric chains present in it [20]. Hence there remains a considerable possibility of extraction related study of different species in a variety of vesicular arrangements of PEG cloud. Moreover different mercury species were already found to get extracted in PEG/salt aqueous biphasic systems [21] with no possibility of speciation. However, to the best of our knowledge extraction of mercury species in PEG cloud is still unexplored.

The article presents the first report on the speciation and extraction behavior of organic and inorganic mercury using cloud point extraction method in PEG. In the present work only PEG solution (4%) is used for the formation of cloud as well as to form complex with the Hg(II) ions in HCl medium. The results of cloud point extraction of mercury species are compared with previously reported aqueous biphasic extraction results. Certain salts were used as additives to lower the cloud point temperature of PEG. For spectral detection of Hg species we have chosen 1-(2-pyridylazo)-2-napthol (PAN) indicator which efficiently quantify the extraction percent at very low mercury concentration in the micellar medium. Zeta potential data of the cloud containing solution has been taken for better understanding of electrokinetic stability of the system. Finally the effect on the structural modifications of the cloud after CPE of Hg has been visualized using confocal microscopy.

## 2. Experimental

### 2.1. Materials

All the inorganic salts $HgCl_2$, $Na_2SO_4$, NaCl, $NaH_2PO_4$, $Bi(NO_3)_3$, $Pb(NO_3)_2$, $CdCO_3$, $Na_3AsO_4$, $Na_2SeO_4$, NaI, NaOAC, PEG#6000 were purchased from Merck. $CH_3HgCl$ was obtained from Sigma-Aldrich. All the solutions were made in triple-distilled water. The complexing agent 1-(2-pyridylazo)-2-napthol (PAN) and all other chemicals were of analytical grade. Fluorescein was procured from Sigma Aldrich.

### 2.2. Apparatus

The absorption spectra were obtained using an Agilent 8453 diode array spectrophotometer. Mettler Toledo seven compact pH/Ion meter S220 was used to measure and adjust the pH of different solutions. Hermle microprocessor controlled universal refrigerated high speed Table top centrifuge (model Z 36 K) with an adjustable speed range of 200–30000 was used for centrifugation. Zeta potentials were measured using Malvern Instrument zetasizer nano (Zn).

### 2.3. The cloud point extraction (CPE):

Cloud point extraction of two different mercury species, $HgCl_2$ and $CH_3HgCl$ were studied. We have taken PEG#6000 for cloud formation. 0.5 mL of 0.5 mM $HgCl_2$/1 mL of 1 mM $CH_3HgCl$, 3 mL sodium sulfate (varying pH and concentration), 1 mL of 20%(w/v) PEG(#6000) were taken and the volumes were made upto 5 mL. This results in a 4%(w/v) final concentration with respect to PEG (#6000). pH (2.5–10.8) was adjusted using dilute HCl solution in the acidic range and dilute NaOH solution in the basic range. The solutions were heated in a water bath for 15 min at 35–40 °C. The solution appeared cloudy, which was then centrifuged at 3000 rpm for 5 min. The centrifuge tubes were then cooled in ice water for 10 min. The surfactant rich phase appeared at the upper surface of the solution and was separated out carefully. Then the separated portion was dissolved in distilled water and taken for absorption, zeta potential and confocal micrcoscopic studies. For absorption studies, the cloud dissolved in water was treated with pH 9.1 borate buffer followed by addition of 0.5 mM PAN. At this condition a new complex of Hg-PAN was formed which was spectrophotometrically estimated at its $\lambda_{max}$ 560 nm wavelength. 0.5 mM, 1 mM and 1.5 mM solutions of NaCl, $NaH_2PO_4$, $Bi(NO_3)_3$, $Pb(NO_3)_2$, $CdCO_3$, $Na_3AsO_4$, $Na_2SeO_4$ and NaI were prepared in triple distilled water for interference studies. These ions were mixed with equimolar concentrations of Hg solution and analyzed for their interference. For confocal microscope imaging, the cloud dissolved solutions were spiked with a small volume of diluted fluorescein solution. A drop of this solution was placed on a glass slide, covered with a cover-slip, observed under the confocal microscope and the images were recorded.

### 2.4. Analysis of environmental samples

Water samples and river sediment were collected from Hooghly river from Babughat, Kolkata, India and a pond water sample was collected from Baruipur, West Bengal, India. The freshly collected samples were concentrated by evaporation and filtered before analysis. Concentrated HCl extract of the sediment was obtained after keeping the sediment immersed in the acid for overnight and then slow heating of the mixture for 30 min. The extract was cooled and filtered through Whatman 40. The filtrate was collected, evaporated to dryness and taken in small volume of distilled water to be analyzed after CPE. Our proposed CPE method was also applied to both the water samples. Standard addition method has been used to find mercury concentration in these samples.

## 3. Results and discussion

PEG is chiefly a thermo separating polymer but cloud point temperature of PEG in an aqueous binary system is very high (above 373.15 K) [22]. The cloud point temperature of micellar solutions can be controlled by addition of salts, alcohols, organic compounds, etc. We have taken four different salts as additives to lower the cloud point temperature of PEG. The variations of cloud point temperatures versus concentration of four different salts e.g. $Na_2SO_4$, NaCl, $NaH_2PO_4$, NaOAc are shown in Table 1. Among the four salts, sodium sulfate efficiently reduces cloud point of PEG to 35 °C. This observation can be explained on the basis of ionic interactions. Generally, chaotropes ($Cl^-$, $I^-$) are large singly charged ions, with low charge density and they exhibit weaker interactions with water molecules. Conversely, small or multiply-

**Table 1**
Variations of cloud point temperatures of PEG with concentration of four different salts (NaCl, $NaH_2PO_4$, NaOAc, $Na_2SO_4$).

| Concentration of PEG | Concentration of salts | Cloud point temperature |
|---|---|---|
| 4% PEG | 1.6 M NaCl | 100° C |
| | 1.8 M NaCl | 85° C |
| | 2.4 M NaCl | 80° C |
| | 2.8 M NaCl | 70° C |
| 4% PEG | 0.2 M $NaH_2PO_4$ | 100° C |
| | 0.4 M $NaH_2PO_4$ | 100° C |
| | 0.6 M $NaH_2PO_4$ | 80° C |
| | 0.8 M $NaH_2PO_4$ | 70° C |
| 4% PEG | 0.8 M NaOAc | 100° C |
| | 1.2 M NaOAc | 90° C |
| | 1.6 M NaOAc | 85° C |
| | 2.0 M NaOAc | 80° C |
| 4% PEG | 0.2 M $Na_2SO_4$ | 100° C |
| | 0.4 M $Na_2SO_4$ | 70° C |
| | 0.6 M $Na_2SO_4$ | 40° C |
| | 0.8 M $Na_2SO_4$ | 35° C |