Video Article

Sulfate Separation by Selective Crystallization with a Bis-iminoguanidinium Ligand

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Abstract

A simple and effective method for selective sulfate separation from aqueous solutions by crystallization with a bis-guanidinium ligand, 1,4-benzene-bis(iminoguanidinium) (BBIG), is demonstrated. The ligand is synthesized as the chloride salt (BBIG-Cl) by *in situ* imine condensation of terephthalaldehyde with aminoguanidinium chloride in water, followed by crystallization as the sulfate salt (BBIG-SO_4). Alternatively, BBIG-Cl is synthesized *ex situ* in larger scale from ethanol. The sulfate separation ability of the BBIG ligand is demonstrated by selective and quantitative crystallization of sulfate from seawater. The ligand can be recycled by neutralization of BBIG-SO_4 with aqueous NaOH and crystallization of the neutral bis-iminoguanidine, which can be converted back into BBIG-Cl with aqueous HCl and reused in another separation cycle. Finally, ^{35}S-labeled sulfate and β liquid scintillation counting are employed for monitoring the sulfate concentration in solution. Overall, this protocol will instruct the user in the necessary skills to synthesize a ligand, employ it in the selective crystallization of sulfate from aqueous solutions, and quantify the separation efficiency.

Video Link

The video component of this article can be found at http://www.jove.com/video/54411/

Introduction

Selective separation of hydrophilic oxoanions (*e.g.*, sulfate, chromate, phosphate) from competitive aqueous solutions represents a fundamental challenge with relevance to environmental remediation, energy production, and human health.[1,2] Sulfate in particular is difficult to extract from water due to its intrinsic reluctance to shed its hydration sphere and migrate into less polar environments.[3] Making aqueous sulfate extraction more efficient typically requires complex receptors that are difficult and tedious to synthesize and purify, often involving toxic reagents and solvents.[4,5]

Selective crystallization offers a simple yet effective alternative to sulfate separation from water.[6-9] Though some metal cations such as Ba^{2+}, Pb^{2+}, or Ra^{2+} form very insoluble sulfate salts, their use in sulfate separation is not always practical due to their high toxicity and sometimes-low selectivity. Employing organic ligands as sulfate precipitants takes advantage of the structural diversity and amenability to design characteristic to organic molecules. An ideal organic ligand for aqueous sulfate crystallization should be soluble in water, yet form an insoluble sulfate salt or complex in a relatively short time and in the presence of high concentrations of competing ions. Additionally, it should be easy to synthesize and recycle. One such a ligand, 1,4-benzene-bis(iminoguanidinium) (BBIG), self-assembled *in situ* from two commercially available precursors, terephthalaldehyde and aminoguanidinium chloride, was recently found to be extremely effective in aqueous sulfate separation.[10] The ligand is water-soluble in the chloride form, and selectively crystallizes with sulfate into an extremely insoluble salt that can be easily removed from solution by simple filtration. The BBIG ligand can then be recovered by deprotonation with aqueous NaOH and crystallization of the neutral bis-iminoguanidine, which can be converted back into the chloride form with aqueous HCl, and reused in another separation cycle. The efficacy of this ligand in removing sulfate from water is so great that monitoring the remaining sulfate concentration in solution is no longer a trivial task, requiring a more advanced technique that allows accurate measurement of trace amounts of the anion. For this purpose, radiolabeled ^{35}S sulfate tracer in conjunction with β liquid scintillation counting was employed, a technique commonly utilized in liquid-liquid extractive separations, and recently demonstrated to be effective in monitoring sulfate crystallization.[8]

This protocol demonstrates the one-pot *in situ* synthesis of the BBIG ligand and its crystallization as the sulfate salt from aqueous solutions. The *ex situ* synthesis of the ligand[11] is also presented as a convenient method for the production of larger amounts of BBIG-Cl, which can be stored in the crystalline form until ready to use. Sulfate removal from seawater using the previously prepared BBIG-Cl ligand is then demonstrated. Finally, the use of ^{35}S-labeled sulfate and β liquid scintillation counting for measuring the sulfate concentration in seawater is demonstrated. This protocol is intended to provide a tutorial for those broadly interested in exploring the use of selective crystallization for aqueous anion separation.

Protocol

1. Synthesis of 1,4-Benzene-bis(iminoguanidinium) Chloride (BBIG-Cl)

1. *In Situ* Synthesis of the 1,4-Benzene-bis(iminoguanidinium) Chloride Ligand (BBIG-Cl) and Its Crystallization with Sulfate
    1. Add 0.067 g of terephthalaldehyde and 2.2 ml of a 0.5 M aqueous solution of aminoguanidinium chloride to 10 ml of deionized water in a 25 ml round bottom flask equipped with a magnetic stir bar.
    2. Stir the solution magnetically for four hours at 20 °C. This will yield a slightly yellow solution of BBIG-Cl.
    3. Add 0.5 ml of a 1 M aqueous solution of sodium sulfate. This will result in the instant precipitation of BBIG-SO_4 as a crystalline white solid.
    4. Filter the solid using vacuum filtration to recover BBIG-SO_4. Wash the solid on the filter paper five times with 5 ml aliquots of water in order to obtain the pure sulfate salt.
    5. Check the phase purity of the crystalline BBIG-SO_4 obtained by powder X-ray diffraction[12]. Compare with the pattern shown in **Figure 1**.

2. *Ex Situ* Synthesis of 1,4-Benzene-bis(iminoguanidinium) Chloride[11]
    1. Add 4 g of terephthalaldehyde and 7.26 g of aminoguanidinium chloride to 20 ml of ethanol in a 50 ml round bottom flask equipped with a magnetic stir bar.
    2. Heat the solution to 60 °C using a hotplate, and stir with a magnetic stir bar for 2 hr. Cool the solution to 20 °C and let it sit for 3 hr, then collect the solid by vacuum filtration through a filter-paper equipped Büchner funnel.
    3. Suspend the obtained solid in 20 ml of ethanol and heat on a hotplate until boiling. If the solid does not go completely into solution at this point, add small aliquots (1 ml) of ethanol, allowing each time the solution to reach boiling temperature, until all solid is dissolved.
    4. Allow the flask to cool to room temperature, then place in a 0 °C freezer overnight. Collect the solid by filtering through a filter-paper equipped Büchner funnel using vacuum filtration.
    5. Confirm the identity and purity of BBIG-Cl by ^{1}H NMR spectroscopy[13]. Compare with the spectrum shown in **Figure 2**.

2. Sulfate Separation from Seawater

1. Sulfate crystallization as BBIG-SO_4
   NOTE: The amount of BBIG-Cl necessary to remove the sulfate depends on the exact amount of sulfate in the seawater. It was found that using 1.5 equivalents of BBIG-Cl relative to sulfate results in 99% removal of sulfate. The seawater used in this protocol has a concentration of 30 mM sulfate, as determined by titration with $BaCl_2$.
    1. Filter the seawater with a 0.22 µm syringe filter or filtration membrane with small pore size to remove suspended particulates and bio organisms.
    2. Make a 30 mM solution of BBIG-Cl using deionized water and solid BBIG-Cl prepared as described in the previous section.
    3. Add the BBIG-Cl solution to the seawater in a 1.5:1 (v/v) proportion.
    4. Stir the mixture for a few hours to ensure quantitative (> 99%) removal of sulfate.
    5. Collect the solid by filtering through a filter-paper equipped Büchner funnel using vacuum filtration. Wash the solid on the filter paper five times with 5 ml aliquots of water.
    6. Dry the isolated solid under vacuum and weigh it to determine the yield.

2. Ligand Recovery
    1. Add 53.1 mg of BBIG-SO_4 to a 2 ml solution of NaOH (10%) in a 20 ml scintillation vial equipped with a magnetic stir bar.
    2. Stir the mixture for two hours at 20 °C. A slightly yellow precipitate will form.
    3. Filter the solid through a filter-paper equipped Büchner funnel using vacuum filtration. Wash the solid on the filter paper with 0.2 ml of water, and dry under vacuum.
    4. Characterize the recovered solid by NMR[13] to confirm its identity as the bis(guanidine) free base. Compare with the NMR spectrum shown in **Figure 3**.

3. Determination of the Amount of Sulfate Removed from Seawater by β Liquid Scintillation Counting
   CAUTION: This technique involves the use of radioisotopes, which pose a different class of hazards than what is normally encountered in most labs. Special radiation protection equipment is usually required when handling the radionuclides. Thus, it is essential that the procedure is followed carefully and that a safety officer is consulted for advice and guidance.
    1. Calculate the volume of the stock solution of the sulfur-35 radioisotope (5 mCi/ml) used to ensure there is more than 5 million counts per minute (cpm) per milliliter of seawater solution, using the following equations (cpm and curies (Ci) are both units of measure for radioactivity):
    $$2.22 \times 10^{12}\ cpm = 1\ Ci$$
    $$\frac{5.0 \times 10^{6}\ cpm/mL}{2.22 \times 10^{12} cpm} \times 1\ Ci = 2.25 \times 10^{-6}\ Ci/mL = 2.25 \times 10^{-3}\ mCi/mL$$
    $$\frac{2.25 \times 10^{-3}\ mCi/mL \times 25mL}{5.0\ mCi/mL} = 1.12 \times 10^{-2}\ mL$$
    2. Spike 25 ml of the seawater with 0.0112 ml of 5.0 mCi/ml solution of ^{35}S radiolabeled sodium sulfate solution.
    3. Prepare 0, 15, 30, 33, 45, and 60 mM solutions of BBIG-Cl in deionized water and combine 0.750 ml of these solutions with an equal volume of ^{35}S-radiolabeled sulfate spiked seawater in a 2 ml centrifuge tube.
    4. Stir the mixture via a rotating wheel or vortex in an incubator/air-box maintained at a constant temperature of 25 ± 0.2 °C for 24 hr.
    5. Centrifuge the solutions at 1,500 x g for 10 min at 25 °C.

6. After centrifugation, remove 1.2 ml of each solution using a syringe, then filter it through a 0.22 µm syringe filter to remove the suspended precipitate. Pipette 1.0 ml from each of these solutions into 20 ml of scintillation cocktail in polypropylene scintillation vials. The solution containing no BBIG-Cl (the control solution) should be diluted ten-fold with deionized water prior to addition to the scintillation cocktail.
7. Place the scintillation vials containing the samples and the scintillation cocktail on a liquid scintillation counter and let it sit for 1 hr prior to counting to allow the samples to dark-adapt.
   NOTE: Prior to counting the samples, calibrate the instrument and allow each sample to count for 30 min. Count additional vials containing only scintillation cocktail in order to allow for a background correction that is used when determining the concentrations of sulfate in solution.
8. Determine the amount of sulfate removed, using the following equations:

$$SO_4^{2-}{}_{sample-cpm} - SO_4^{2-}{}_{background-cpm} = SO_4^{2-}{}_{corrected-cpm}$$

$$\frac{SO_4^{2-}{}_{initial-cpm} - SO_4^{2-}{}_{sample-cpm}}{SO_4^{2-}{}_{initial}} \times 100 = \%\, SO_4^{2-} removed$$

$$[SO_4^{2-}]_{initial} \times \left(1 - \frac{\%\ SO_4^{2-}\ removed}{100}\right) = [SO_4^{2-}]_{remaining}$$

Representative Results

The powder X-ray diffraction pattern of BBIG-SO_4 (**Figure 1**) allows for unambiguous confirmation of the identity of the crystallized solid. In comparing the obtained pattern versus the reference one, peak intensity matters less than peak positioning. All strong peaks shown in the reference should be present in the obtained sample. The appearance of strong peaks in the sample that are absent in the reference pattern indicates the presence of impurities.

^{1}H-NMR of BBIG-Cl and the recovered ligand (**Figures 2** and **3**) enable an assessment of both the identity of the compounds as well as their purity to about 5%. Comparison with these spectra help to ensure that the ligand was fully formed and that any impurities were adequately removed during the filtrations and/or recrystallizations. In comparing the obtained spectrum versus the reference, it is important to make sure all peaks are present in the exact positions shown. Use the same solvents used in the reference spectra so that the relative shift of the peaks do not change.

The results of sulfate separation from seawater are shown in **Table 1**, with over 99% of the sulfate being removed using only 1.5 molar equivalents of BBIG-Cl. This represents near-quantitative removal of sulfate from seawater despite the high ionic strength of the medium, demonstrating the efficacy of the technique described.

BBIG-Cl was obtained in a 70% yield via the *ex situ* method, while BBIG-SO_4 was obtained in 86% yield via the *in situ* synthesis of BBIG-Cl. The ligand recovery yield was 93%. All organic reactions carried out in this procedure are high yielding and operationally simple, making the compounds easily accessible even to a novice chemist.

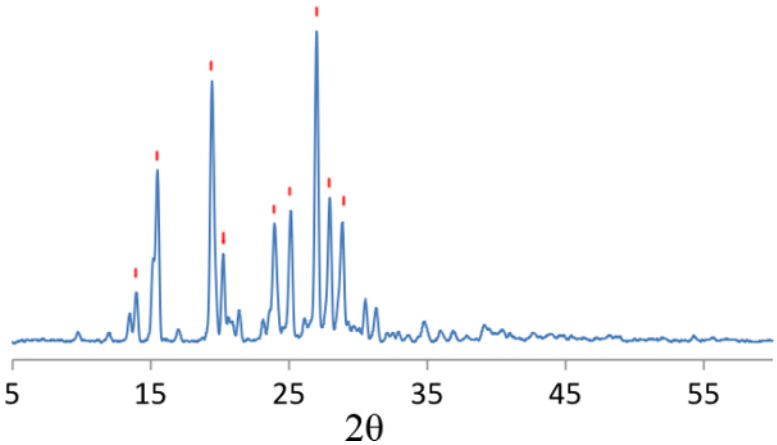

Figure 1: Powder X-ray diffraction pattern of BBIG-SO_4. The pattern was obtained with a powder X-ray diffractometer using a flat sample stage in reflection mode. The strongest peaks are marked in red. Please click here to view a larger version of this figure.

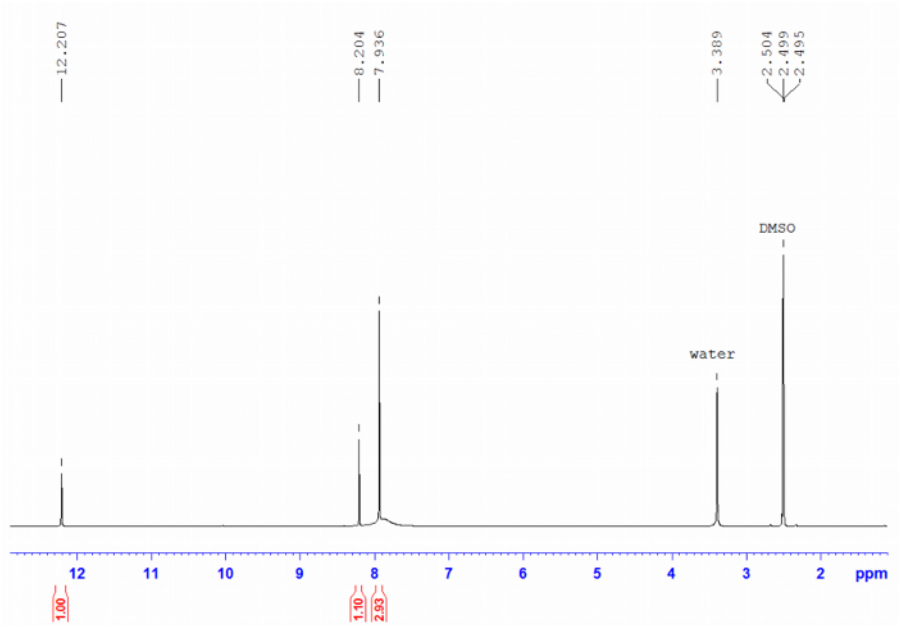

Figure 2: ^{1}H-NMR spectrum of BBIG-Cl. The spectrum was taken in DMSO-d_6 with a 400 MHz NMR instrument. Please click here to view a larger version of this figure.

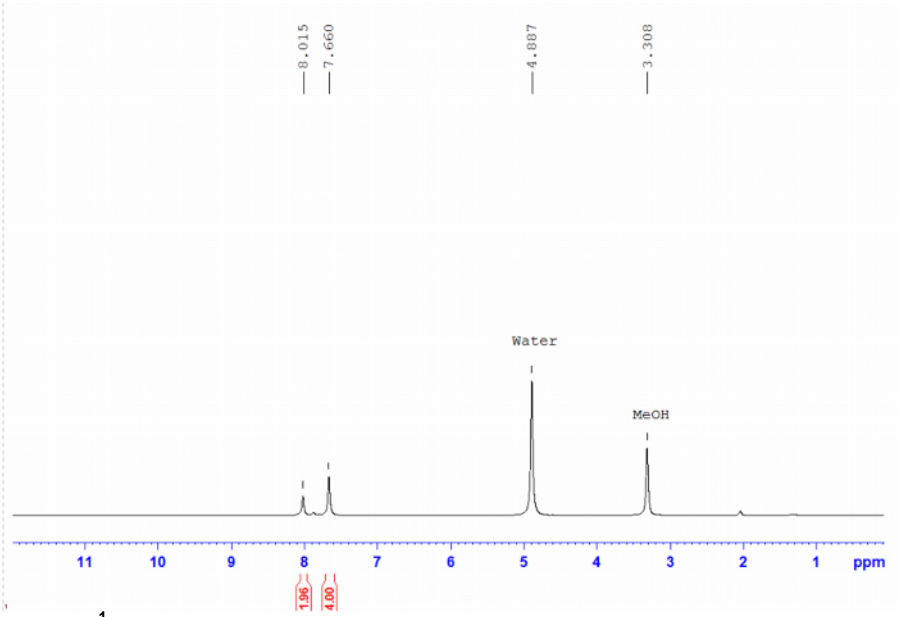

Figure 3: ^{1}H-NMR spectrum of the recovered BBIG ligand. The spectrum was taken in MeOD with a 400 MHz NMR instrument. Please click here to view a larger version of this figure.

BBIG equiv	[Sulfate] left (mM)	Sulfate removed (%)
1	3.5	88
1.1	1.6	95
1.5	0.3	99
2	0.3	99

Table 1: Representative results from sulfate separation from seawater. The data shows removal of up to 99% of sulfate from seawater using only 1.5 M equivalents of BBIG-Cl. The initial sulfate concentration in seawater was 30 mM.

Discussion

This technique is rather tolerant to many deviations from the written procedure, which makes it quite robust. There are however two critical steps that must be followed. First, the BBIG-Cl ligand needs to be as pure as possible. Impurities will not only affect the crystallization and the solubility of the resulting sulfate salt, but will also make it difficult to calculate the amount required for quantitative sulfate removal from solution. Second, all steps in the β liquid scintillation counting section need to be followed meticulously, as this technique may be very sensitive to subtle changes.

Due to the simplicity of the crystallization technique, troubleshooting will most likely not be needed. Some common issues are discussed as follows. In the case that the BBIG-Cl ligand does not appear to be removing the sulfate present in solution, one of two issues is the most likely culprit. If using the *ex situ* synthesized BBIG-Cl, confirm its identity and purity. Take an ^{1}H-NMR of the starting material and compare it with the reference spectrum in **Figure 2**. Another common culprit of this problem is the pH of the solution. If synthesizing the BBIG-Cl ligand *in situ*, make sure the pH of the solution is slightly acidic (pH = 5-6). Due to the fact that the active species is the protonated ligand, the method is sensitive to the solution pH. Basic solutions will deprotonate the guanidinium groups, yielding a neutral ligand that is incapable of crystallizing sulfate. If the pH is basic, a simple adjustment with HCl to a pH of about 5-6 will provide the optimal conditions for quantitative sulfate removal. This problem does highlight one of the main limitations of this technique, in that it is unable to remove sulfate from basic solutions. However, the BBIG ligand is quite acid-stable, so adjusting the pH of the solution provides a simple remedy to this problem. Another variable that may affect the sulfate crystallization efficiency is the ionic strength of the solution. While sulfate separation from seawater proved very efficient, it is possible the yield of sulfate separation to be lower when this method is applied to solutions with very high ionic strength.

The technique demonstrated in this protocol is extremely efficient, selective, green, and cost effective. By comparison, alternative sulfate removal methods involve expensive and high-maintenance membranes or ion exchange columns with low separation selectivity.[3] Furthermore, compared to existing methods, the technique presented here is very simple and requires little technical knowledge and experience in separation chemistry.

This crystallization technique offers a general approach to quantitative removal of sulfate from aqueous solutions. While seawater was used in this protocol to demonstrate the technique, this crystallization method is not limited to seawater, and could be used for sulfate removal from virtually any aqueous solutions. Since the class of bis(iminoguanidinium) ligands employed can be easily synthesized in one step from readily available dialdehyde and aminoguanidinium precursors, there are potentially many other simple combinations that may result in an effective precipitation agent for sulfate or other oxoanions. Thus, mastering the techniques presented in this protocol will enable one to develop his/her own crystallization ligand with potentially even better selectivity and efficacy than the BBIG ligand presented here.

Disclosures

The authors have nothing to disclose.

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