EPA Method 1668A: Chlorinated Biphenyl Congeners by HRGC/HRMS

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Scope

This application note will outline the recommended methods for the extraction of chlorinated biphenyl congeners as outlined in EPA Method 1668A using Biotage automated or manual SPE solutions, Atlantic® C18 SPE disks, and the DryVap® Concentrator System. The first section will highlight the use of the Biotage® Horizon 5000 fully automated extraction system and the method used for this application. Additionally, there will be an Application Modification section that will highlight the use of the Biotage® VacMaster™ Disk for this application.

Introduction

Method 1668A is used with the purpose of determining the level of chlorinated biphenyl congeners by high resolution gas chromatography with high resolution mass spectrometry (HRGC/HRMS) in aqueous samples (samples containing less than 1% solids). Stable isotopically labeled analogs of the toxic PCBs are spiked into a 1 L sample, and the sample is extracted using the Biotage® Horizon 5000 Automated Extraction System along with the Atlantic® C18 solid-phase extraction (SPE) disk. Samples are then dried and concentrated using the DryVap® Concentrator System with DryDisk® technology.

Instrumentation

Biotage Instruments:

- Biotage® Horizon 5000 Automated Extraction System
- DryVap® Concentrator System
- DryDisk® Separation Membranes
- Atlantic® C18 SPE Disks (47 mm)

Method Summary

1. Prepare the sample according to EPA Method 1668A.
2. Purge the Biotage® Horizon 5000 extraction system using the method listed in table 1.
3. Place the sample bottle on the Biotage® Horizon 5000 Extraction System and place the Atlantic® C18 disk in the standard 47mm disk holder.
4. Extract the sample using the method in table 2 and collect the final sample extract.
5. Using the DryVap® Concentrator System with a DryDisk®, dry the extract and concentrate it down to the appropriate final volume.
6. Analyze the extract using a HRGC/HRMS system.

Table 1. Biotage® Horizon 5000 Purge method.

Step	Select Solvent	Volume (mL)	Purge (s)	Vacuum	Saturate (s)	Soak (s)	Drain/ Elute (s)	Sample Delay (s)
Wash Sample Container	Dichloromethane	8	15	2	1	15	60	
Wash Sample Container	Acetone	8	15	2	1	15	60	

Table 2. Biotage® Horizon 5000 extraction method.

Step	Select Solvent	Volume (mL)	Purge (s)	Vacuum	Saturate (s)	Soak (s)	Drain/ Elute (s)	Sample Delay (s)
Condition SPE Disk	Dichloromethane	15	60	2	1	90	180	
Condition SPE Disk	Acetone	11	60	2	1	90	120	
Condition SPE Disk	Methanol	11	60	2	1	90	5	
Condition SPE Disk	Reagent water	15	60	2	1	90	5	
Load Sample				2				45
Air Dry Disk				6			600	
Elute Sample Container	Acetone	8	15	2	1	90	120	
Elute Sample Container	Dichloromethane	8	15	2	1	90	180	
Elute Sample Container	Dichloromethane	8	15	6	1	90	240	

Application Modifications

Biotage® VacMaster™ Disk Method Summary

1. Repeat the following steps for each active Biotage® VacMaster™ Disk station.
2. Setup the VacMaster Disk manifolds ensuring all waste lines and vacuum lines are attached. Set the vacuum pump to -24”Hg.
3. Prepare the disk holder assembly (47mm): ensure the support screen is flat in the center of the disk holder. Place the Atlantic® C18 Disk on top of the support screen with the ripples of the disk on top and add any prefilters on top of the disk. Place the disk holder assembly on the VacMaster Disk manifold ensuring there is a tight seal with the luer fitting.
4. If using the multifunnel, place onto the disk holder assembly. If not using the multifunnel, omit those directions throughout the method.
5. Condition the SPE Disk:
   a. Guide for each conditioning step in table 3 below:
      I. Measure the appropriate VOLUME of SOLVENT into a graduated cylinder and pour into the disk holder assembly.
      II. Using a Nalgene Wash Bottle (phthalate free), rinse the multifunnel and disk holder in a circle for about 3 seconds using the same SOLVENT (approximately 5 additional mL).
      III. SATURATE the disk for the indicated time (in SECONDS). (Saturate means: quickly turn the knob to the appropriate waste destination and back to the “OFF” position. This brings the solvent into the disk media bed).
      IV. SOAK the disk for the indicated time (in SECONDS).
      V. DRAIN to the appropriate waste destination for the indicated time (in SECONDS). Switch to the “OFF” position.
6. Load the Sample:
   a. For multifunnel: quickly and efficiently angle the bottle to rest on the multifunnel upside-down.
   b. For no multifunnel: pour a portion of the sample into the disk holder.
   c. Adjust the vacuum between -10”Hg and -15”Hg for sample load (please note, if the sample is flowing too slowly, the vacuum can be increased). Drain the sample to “AQUEOUS” waste. Continue to pour the sample into the disk holder ensuring the disk does not go dry or overflow for the duration of sample load.

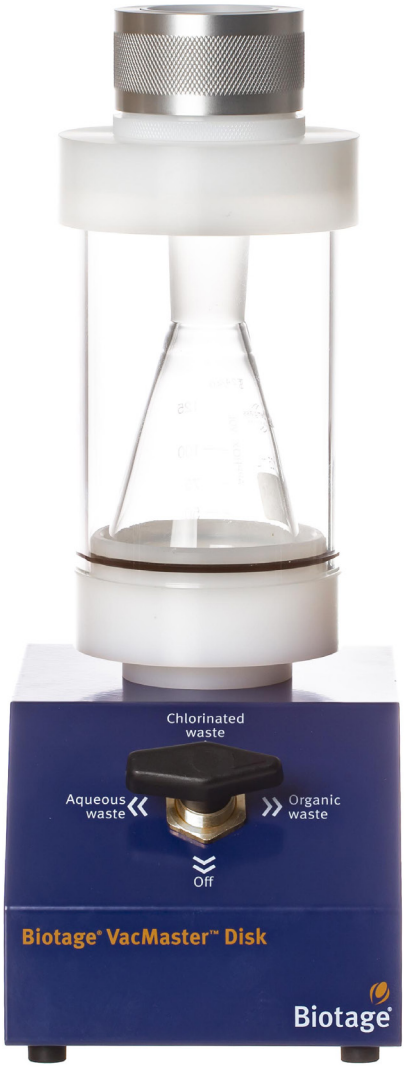
Table 3. Disk Conditioning.

Solvent	Vol. (mL)	Saturate (sec.)	Soak (sec.)	Waste Destination	Drain (sec.)
DCM	15	1	90	Chlorinated	180
Acetone	11	1	90	Organic	120
MeOH	11	1	90	Organic	5
Reagent Water	15	1	90	Organic	5

Table 4. Disk Elution.

Solvent	Vol. (mL)	Saturate (sec.)	Soak (sec.)	Waste Destination	Elute (sec.)
Acetone	8	1	90	Organic	120
DCM	8	1	90	Organic	180
DCM	8	1	90	Organic	240

1. Air Dry the SPE Disk:
   a. Return the vacuum to -24”Hg and continue to air dry the SPE disk to “AQUEOUS” waste for an additional 600 SECONDS. Switch to the “OFF” position.
   b. Remove the sample bottle from the multifunnel if it was used.
2. Elute the SPE Disk: (Please note: the elution solvent will go into the collection flask inside the chamber, not to waste containers).
   a. Place a clean 125mL 24/40 tapered Erlenmeyer flask or 40 mL VOA vial using the VOA vial insert into the Biotage® VacMaster™ Disk collection chamber. Place the cover on the chamber. Remove the disk holder assembly and place the disk holder assembly into the luer fitting on top of the collection chamber. Attach the luer fitting of the collection chamber assembly onto the manifold.
   b. Guide for each elution step in table 4 below:
      I. Measure the appropriate VOLUME of SOLVENT into a graduated cylinder, pour into the sample bottle, and swirl around. Pour the solvent in the sample bottle into the disk holder assembly.
      II. Using a Nalgene Wash Bottle (phthalate free), rinse the multifunnel and disk holder in a circle for about 3 seconds using the same SOLVENT (approximately 5 additional mL).
      III. SATURATE the disk for the indicated time (in SECONDS) to “ORGANIC”.
      IV. SOAK the disk for the indicated time (in SECONDS).
      V. DRAIN to “ORGANIC” for the indicated time (in SECONDS). Switch to the “OFF” position.
      VI. Remove the chamber lid to release the vacuum from inside the chamber.



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