Instructions for Supelco Multi-Layer Silica Gel Column and Dual-Layer Carbon Reversible Column

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The Supelco Dioxin Prep System

The Supelco Dioxin Prep system provides a highly efficient means of extracting and isolating dioxins, furans, and coplanar PCBs from stack gases, wastewater, soil, food, blood, and milk. The prep system design reduces solvent usage, decreases prep time by 1-2 days, and results in extraction recoveries greater than 85%.

The convenient multi-layer silica gel column is key to the extraction process. Seven layers of treated silica oxidize, reduce, and separate polar interferents. For very dirty samples, bulk treated silica gels and empty glass tubes are available to customize packings to meet individual sample needs.

A unique dual-layer carbon reversible tube isolates the PCBs, dioxins, and furan groups from other non-polar interferences. Isolation and separation is based on the two layers of carbon having different affinities for such compounds.

An integrated glassware and hardware design makes it convenient for analysts to select a few pieces or the entire prep system for their extraction needs. A vacuum manifold and a vacuum adapter provide the option of running a single sample or multiple samples at one time, using vacuum or gravity feed.

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SUPELCO
Bellefonte, PA

How to Use the Multi-Layer Silica Gel Column*

*The multi-layer silica gel column was developed with the assistance of Mr. Masaaki Maeoka at JQA.

The Supelco multi-layer silica gel column is designed to meet the requirements of Japanese Industrial Standard Methods K-0311 and K-0312. The column has a 15 mm internal diameter and is 35 cm in length. It contains 7 layers of treated silica gels as described in the JIS methods under dry packing conditions. The design of the column allows for easy connection to various components including stopcock valves and separatory flasks through the use of commercially available connectors.

Conditioning the Column

Prior to sample addition, the column is rinsed with 200mL of n-hexane. This rinse is designed to:

1. remove air trapped between and within the particles of silica, allowing the sample solution to contact the surfaces of the various coated silica gels and thus remove any interferences more efficiently,
2. establish a steady and consistent flow of n-hexane by removing air bubbles in the column, and
3. ensure the cleanliness of the column packing and remove background contamination.

After conditioning with n-hexane, the column should allow a flow of about 2.0-2.5mL/min using gravity feed. Two optional devices, the vacuum manifold (Cat. No. 28403-U) (Figure1) or the vacuum adapter (Cat. No. 28408-U) (Figure 2) are available to perform this conditioning quickly and more effectively. The flow after vacuum assisted conditioning will be about 3mL/min.

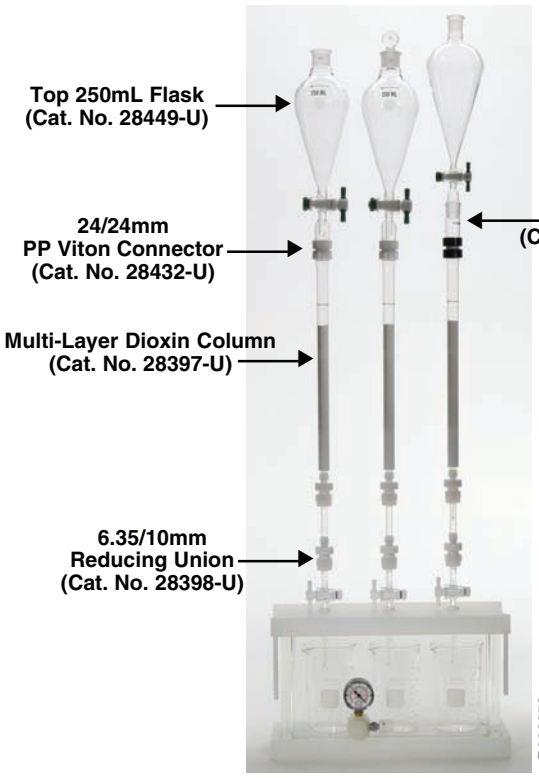
The column is then ready to accept an n-hexane extract of the sample. The analytes (coplanar PCB/PCDD/PCDFs) in the extract will pass through the column with minimal retention while interferences and carry-over contamination from the extraction will be trapped and retained on the column. The analytes can then either be collected in the n-hexane eluate for additional processing by a rotary evaporator or Kuderna-Danish concentrator, or trapped and desorbed with minimum solvent using the dual-layer Carbon column, or another suitable concentration method.

Column Conditioning Using Vacuum Manifold

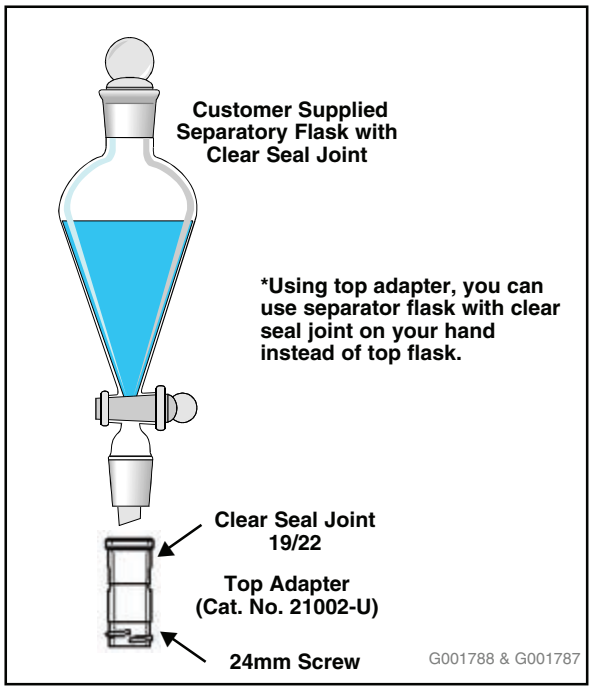
Preferred method (see Figure 1)

Position the manifold beakers to retain the eluted n-hexane inside the vacuum manifold. Connect the multi-layer columns to the manifold stopcocks with 6.35/10mm Reducing Union (Cat. No. 28398-U) and to a support with clamps. Turn the stopcock valves to the open position. Place the specified amount of clean anhydrous sodium sulfate into the top of each column and tap the columns to settle the particles. Attach a top flask (Cat. No. 28449-U) or a top adapter (Cat. No. 21002-U) with a customer-supplied clear seal ground joint flask to the top of each multi-layer column with a 24/24mm connector (Cat. No. 28432-U). Attach a vacuum source to the manifold and adjust the amount of vacuum to 100-400mm Hg (0.013-0.053 MPa). Add the specified amount of n-hexane to each top flask or separatory flask, and allow the n-hexane to flow through the columns under vacuum. When the n-hexane level has dropped but is still above the layer of sodium sulfate, turn the stopcocks to the closed position to stop the flow of n-hexane and to keep air out of the column layers. The column is now ready for use.

Figure 1. Vacuum Manifold



Vacuum Manifold for Dioxin
(Cat. No. 28403-U)



Column Conditioning Using the Vacuum Adapter

Secondary method (see Figure 2)

Attach a round bottom flask with clear seal joint 24/40 (Cat. No. 21269-U) or other similar container and a stopcock to the vacuum adapter. Connect a multi-layer column to the stopcock with a 6.35/10mm Reducing Union (Cat. No. 28398-U) and to a support with clamps. Turn the stopcock valve to the open position. Place the specified amount of clean anhydrous sodium sulfate into the top of the column and tap the column to settle the particles. Attach a top flask (Cat. No. 21001-U) or a top adapter (Cat. No. 21002-U) with a customer-supplied clear seal ground joint flask to the top of the multi-layer column with a 24/24 mm connector (Cat. No. 28432-U). Attach a vacuum source to the adapter and adjust the amount of vacuum to 100-400mm Hg (0.013-0.053 MPa). Add the specified amount of n-hexane to each top flask or separatory flask, and allow the n-hexane to flow through the columns under vacuum. When the n-hexane level has dropped but is still above the layer of sodium sulfate, turn the stopcock to the closed position to stop the flow of hexane and to keep air out of the column layers. The column is now ready for use.

Column Conditioning Using Gravity Feed

Least preferred method

Connect the multi-layer column to a support with a clamp. Attach a stopcock valve to the column with a 6.35/10mm Reducing Union (Cat. No. 28398-U). Turn the valve to the open position. Place a container under the stopcock to retain the eluted hexane. Place the specified amount of clean anhydrous sodium sulfate into the top of the column and tap the column to settle the particles. Attach a top flask (Cat. No. 21001-U) or top adapter (Cat. No. 21002-U) with a customer-supplied clear seal ground joint flask to the top of multi-layer column with a 24/24 mm connector (Cat. No. 28432-U). Add the specified amount of n-hexane to the flask and allow the n-hexane to flow through the column and stopcock into the container below. When the n-hexane level has dropped but is still above the layer of sodium sulfate, turn the stopcock to the closed position to stop the flow of n-hexane and to keep air out of the column layers. The column is now ready for use.

Figure 2. Vacuum Adapter

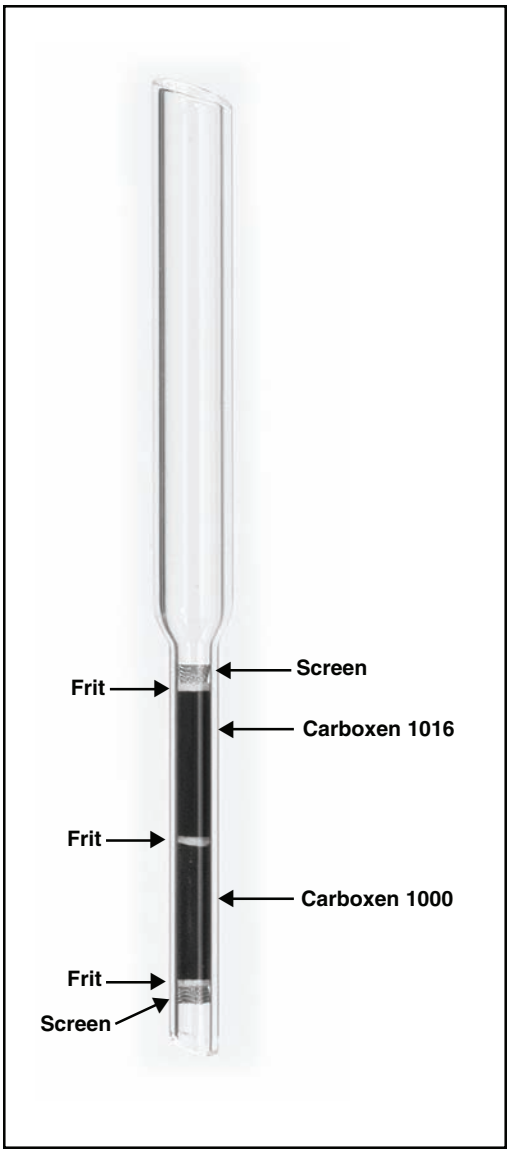
The Dual-Layer Carbon Column*

The Dual-Layer carbon column is composed of two 100mg carbon layers, Carboxen 1016 (Surface area 75m^2/g) and Carboxen 1000 (Surface area 1200m^2/g). The carbon layers are held in place with wire screens and frits between each layer and at the ends of the column.

The direction of the initial sample flow has the small 6.35mm end of the column pointing down. The top bed of the dual-layer column contains Carboxen 1016 and the lower bed Carboxen 1000.

The sample should flow from the 10mm end to the 6.35mm end of the column. The retained analytex are eluted from the column by a flow in the reverse direction, into the 6.35mm end and out the 10mm end of the column.

*The Dual-Layer carbon column was developed with Kawajyu Techno Service, with the assistance of Mr. Yukihiro Nishimura and Mr. Kouji Takayama.



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How to Use the Dual-Layer Carbon Column

Caution. Please note the following warnings:

Exercise extreme care when removing the dual-layer carbon column from its package. As you unscrew the green storage caps on the dual-column ends, do not drop or bump the column as the glass frit may be expelled or the packing beds may be disturbed.

Before connecting dual-layer carbon column to other parts, i.e. stopcock, Luer adapter, etc., inspect each end of the column. Remove any silica gel particles, if present, with a piece of clean lab paper, Q-tip, or with a rinse of n-hexane from a squeeze bottle.

Conditioning

The purpose of conditioning is to remove pockets of air between and within the particles of carbon and allow a consistent solvent flow. This will also wet the surface of the carbons, allowing the analytes to achieve maximum contact with the surface of the packing material, and will remove background contamination that may be present in the packing and glass column.

For this conditioning, three techniques are available: the vacuum manifold, the vacuum adapter, and the syringe Luer adapter with syringe (syringe not supplied). (see below).

Using the Vacuum Manifold

(See Figure 3)

Place an empty beaker inside the manifold to retain the eluted toluene in this procedure. Attach a 10/10mm union (Cat. No. 28412-U) to the 10mm stopcock (Cat. No. 28425-U). Insert the 10mm end of the dual-layer carbon column into the other side of the union and tighten snugly. Attach the 6.35mm end of the dual-layer carbon column to one side of a 6.35mm union (Cat. No. 28411-U) and tighten snugly. To the other side attach an empty dioxin tube (Cat. No. 28404-U or Cat. No. 28409-U) or a customer-supplied solvent reservoir and tighten snugly. It is advisable to support the empty dioxin tube or customer-supplied reservoir with a clamp and stand.

Add a small amount of toluene to the empty tube or reservoir, turn on and adjust the vacuum to about 100-400mm Hg (0.013-0.053 MPa). Check for leaks as the toluene flows through the column. Tighten the union connections if necessary. **Do not overtighten.** Add 40mL of toluene and elute the solvent through the dual-layer carbon column.

Discard this toluene flush. Next, add 50mL of n-hexane and elute through the column to remove any residual toluene. Discard the n-hexane rinse. Repeat this step a second time and discard the second rinse. Remove the 10mm union and connect the stopcock and the dual-layer column with a reducing union. Connect the column inlet to a solvent reservoir with a reducing union. Cap the solvent reservoir after adding a small amount of n-hexane above the bed of the reversible column.

Capping the reservoir and closing the stopcock will minimize evaporation and keep the column bed wetted.

Note: The beds of the dual-layer column should remain wetted with the non-polar solvent after conditioning.

With the Vacuum Adapter

(See Figure 4)

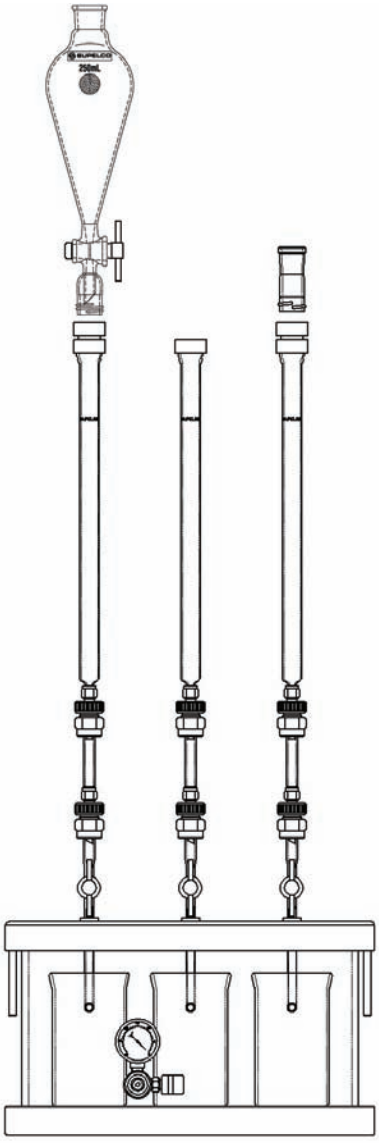
Attach a 250mL round bottom flask (Cat. No. 21296-U) or other suitable vacuum vessel to the vacuum adapter (see Figure 4) to retain the eluted toluene in this procedure. Attach a 10/10mm union (Cat. No. 28412-U) to the adapter and to the 10mm end of dual-layer column and tighten snugly. Attach the 6.35mm end of the dual-layer column to an empty dioxin tube (Cat. No. 28404-U or Cat. No. 28409-U) with a 6.35mm union (Cat. No. 28411-U) and tighten snugly. Add a small amount of toluene to the empty tube, turn on and adjust the vacuum to about 100-400mm Hg (0.013-0.053 MPa). Check for leaks as you draw the toluene through the reversible dual-layer column. It is advisable to support the empty column with a clamp and stand. Add 40mL of toluene and elute the solvent through the dual-layer carbon column.

Discard this toluene flush. Next, add 50mL of n-hexane and elute through the column to remove any residual toluene. Discard the n-hexane rinse. Repeat this step a second time and discard the second n-hexane rinse.

Note: The beds of the dual-layer column should remain wetted with the non-polar solvent after conditioning.

Closing the stopcock and capping the empty tube or column will prevent the evaporation of the solvent from the column. The column may be reversed and a small amount of hexane placed at the head of the column to keep the column bed wetted. When reversing the direction of the column, disconnect the unions attached to the reversible column. Attach two reducing unions at both ends of the column and a solvent reservoir to the top union. Capping the reservoir and the column outlet will minimize evaporation.

Figure 3.



Dioxin Sample Vacuum Manifold

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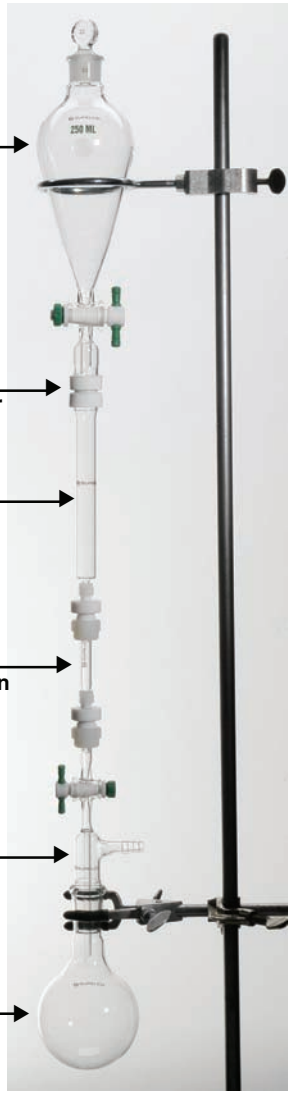
Figure 4.



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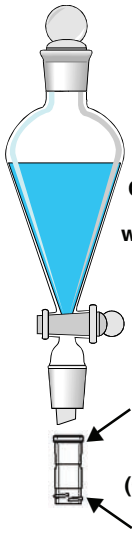

*Using top adapter, you can use separator flask with clear seal joint on the ring stand instead of top flask.

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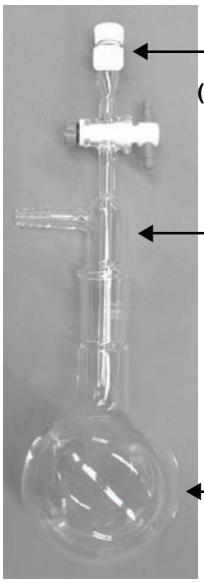

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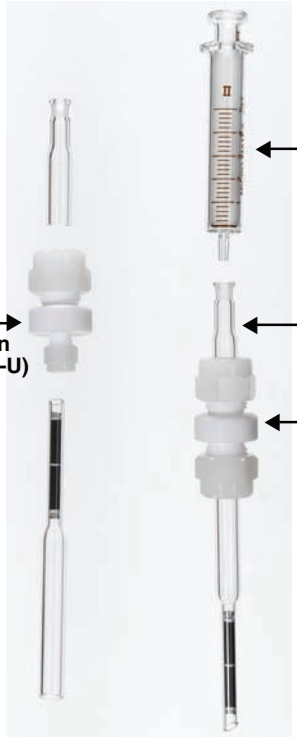
With the Syringe Luer Adapter

(See Figure 5)

Connect a syringe Luer Adapter to the bottom end of a dual layer column with a 6.35/10mm Reducing Union (Cat. No. 28398-U) and tighten snugly. Connect the 10mm end of the column to a stopcock (Cat. No. 28402-U) with a 10mm union (Cat. No. 28412-U) and tighten snugly. Using a clean glass syringe, elute 40mL of toluene through the column, followed by 100mL of n-hexane.

Check for leaks during this procedure. Discard the eluate. **Note:** The beds of the dual-layer column should remain wetted with the non-polar solvent after conditioning. Closing the stopcock and capping the column will prevent the evaporation of the solvent from the dual-layer carbon column. Be certain you load the sample onto the reversible dual-layer carbon column from the 10mm end.

Figure 5.



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Sample Processing with the Multi-Layer Silica Gel and Dual-Layer Carbon Reversible Columns

The extraction of dioxins from the sample is performed according to the method parameters. The extract is routinely concentrated and/or reconstituted into a non-polar solvent for cleanup, elution, and isolation of dioxins using the Multi-Layer Silica Gel and the Dual-Layer Carbon Columns. This may be done in two steps or in a single step procedure.

The Two Step Procedure

This procedure consists of two steps. In the first step, the non-polar solution is passed through the multi-layer column into a suitable collection vessel. This eluate may be concentrated. In the second step, the concentrated eluate is passed through the dual-layer carbon column to trap the analytes of interest. The analytes are then recovered from the dual-layer column with a minimum of solvent.

First Step: Dioxin analytes elute and contaminants are trapped on multi-layer column.

Uncap the conditioned multi-layer column. Place an appropriate collection vessel at the bottom of the multi-layer column. Add the sample solution to the column. Attach solvent reservoir to top of column. Add appropriate amount of the method elution solvent. Open the stopcock and allow the solvent to flow completely through the column into the collection vessel.

After the sample extraction solution is collected from the multi-layer column, check the dual-layer carbon column direction and verify that the 6.35mm end is pointing down.

Second Step: Pass the solvent solution through a pre-conditioned dual-layer carbon column to trap the dioxins.

Attach the sample reservoir to the dual-layer column. Take the collected eluate from the multi-layer column and add this to the dual-layer column reservoir. Turn the stopcock so the collected solvent solution flows through the dual-layer column.

Note: If the sample extraction solution was concentrated before application to the dual-layer column it may be advisable to rinse the dual-layer column with another solvent mixture in the same direction as when loading with sample to remove possible interferences from the column. A solution of n-hexane containing 3.3% methylene chloride has been found to be useful for this purpose. Pass about 30mL of this solvent through the dual layer column.

Now reverse the dual-layer column and pass 40 to 100 mL of toluene through the column to recover the dioxins from the column. Collect the eluate containing the dioxins in a suitable container. This eluate solution may be concentrated and/or reconstituted before GC analysis.

Alternately, the dual-layer carbon column may be attached directly to the multi-layer column and the trapping may be performed in a single step.

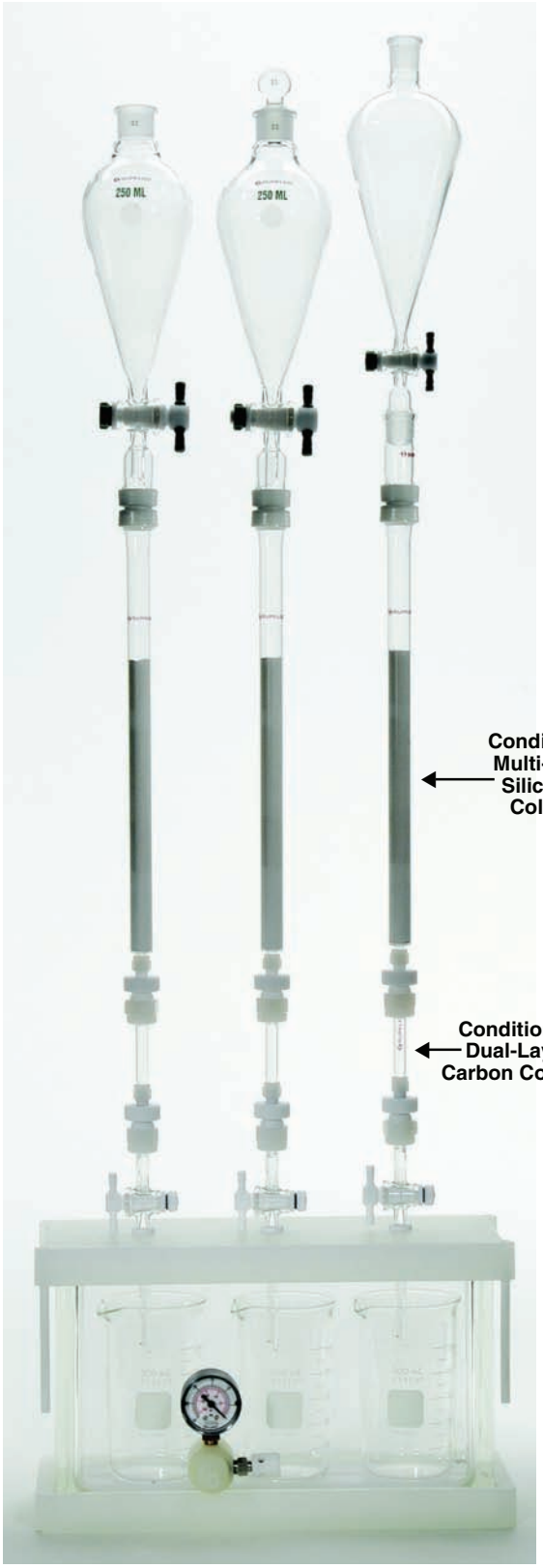
The Single Step Procedure*

(See Figure 6)

Precondition the multi-layer and the dual-layer columns separately. After conditioning, attach the dual-layer column to the bottom of the multi-layer column using a 6.35/10mm reducing union (Cat. No. 28398-U). Uncap the conditioned multi-layer column. Add a small amount of the method elution solvent. Open the stopcock and watch the solution level as it drops toward the bed of sodium sulfate. Check for leaks. Close the stopcock. Add the extracted sample solution. Attach a solvent reservoir to the top of the column. Open the stopcock and watch the solvent level drop close to the bed of sodium sulfate. Add the remainder of the method elution solvent. Close the stopcock to stop the flow of solvent through the column when the solvent level approaches the sodium sulfate bed. Disconnect solvent reservoir and the multi-layer column from the dual-layer column. Depending upon the sample matrix it may be advisable to rinse the dual-layer column with a solvent mixture to remove possible interferences. A solution of n-hexane containing 3.3% methylene chloride has been found to be useful for this purpose. If so, the use of the wash step and the strength of the solvent needed must be determined by experiment. If it is determined to be necessary attach a solvent reservoir to the dual-layer carbon column and pass about 30mL or so of this solvent through the dual-layer column. Then close the stopcock and reverse the dual-layer column and pass 40 to 100mL of toluene through the column to recover the PCBs and dioxins from the column. Collect the eluate in a suitable container.

*The One-Step Method using dual-layer carbon column and multi-layer silica gel column in series was developed with the assistance of Mr. Masaaki Maeoka at Japan Quality Assurance Organization.

Figure 6.



Ordering Information

Description	Cat. No.
Dioxin Sample Preparation Kit	28423-U
Kit includes all glassware and connectors (as listed below).	
NOTE: Requires, but does not include, multi-layer silica gel columns and dual-layer carbon reversible columns.	

Required Consumables

6.35mm Multi-Layer Silica Gel Column, pk. of 5	28397-U
6.6.35/10mm Dual-Layer Carbon Reversible Column, pk. of 10	28399-U

Replacement Kit Parts

Glassware

TTop 250mL Flask	28449-U
6.35mm Empty Dioxin Tube, pk. of 5	28404-U
6.35mm Syringe Luer Adapter, pk. of 3	28405-U
10mm Vacuum Adapter	28408-U
Beaker, 300mL, pk. of 3	21266-U
250mL Round Flask, pk. of 3	21269-U
Vacuum Manifold (includes Stopcocks)	28403-U

Connectors

6.35mm/10mm Reducing Union, pk. of 3	28398-U
10mm/10mm Union, pk. of 3	28412-U
24mm/24mm PP Viton Connector, pk. of 6	28432-U
6.35mm Union, pk. of 3	28411-U

Optional Components

Glassware

Top Adapter, pk. of 3	21002-U
10mm Short Stem Stopcock, pk. of 3	28402-U
10mm Longstem Stopcock, pk. of 3	28425-U
6.35mm Empty 20cm Glass Tube w/o frit, pk. of 5	28409-U

Bulk Treated Silica Gels and Sulfate

2% KOH Coated Silica Gel, 100g	21318-U
10% Silver Nitrate Coated Silica Gel, 100g	21319-U
44% H_2SO_4 Coated Silica Gel, 100g	21334-U
22% H_2SO_4 Coated Silica Gel, 100g	21341-U
Washed Silica Gel, 250g	21342-U
Sodium Sulfate Granular, 500g	239313-500G

Reference

11th Symposium on Environmental Chemistry Programs and Abstracts, 2002 June, page 298-299

Study on short time pre-treatment for analysis of dioxin, Masaaki Maeoka, Itaru Inoue, Hisano Shimono, Nobumasa Morita (Japan Quality Assurance Organization)

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