

APPLICATIONS

Extraction and Analysis of Endothall in Water by EPA Method 548.1 using Strata™-XL-A SPE and Zebron™ ZB-SemiVolatiles GC/MS Methods

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Endothall is an herbicide that is commonly used on crops and in water to control plant and algae growth. Due to its use in water and hydrophilicity, endothall is often present in drinking water and is regulated under EPA Method 548.1. In water samples that contain even low concentrations of other ions like calcium or sulfate, the recovery of endothall can drop to below 10% recovery due to interferences by endogenous ions in the water sample. EPA Method 548.1 consists of dilution and treatment steps with EDTA (ethylenediamine tetraacetic acid) to minimize the interference, but the extraction is still problematic and complicated by these extra steps.

This work details the use of an alternative extraction sorbent that achieves high recoveries, even under considerable calcium and sulfate concentrations, without the need for extra dilution or treatment steps. All other aspects of the method are unchanged so the alternative sorbent is not considered a deviation from the method. The improved procedure achieves greater consistency without the need to prescreen samples for extra possible interferences.

Introduction

Endothall is a di-carboxylic acid (**Figure 1**) that is used as an herbicide to regulate the growth of terrestrial and aquatic plants. Because of its acidic nature, it is very soluble in water and is commonly found in runoff as well as drinking water sources. However, endothall provokes adverse health effects in humans, and is therefore regulated under the Safe Drinking Water Act. The analysis of endothall is regulated in drinking water by EPA Method 548.1^[1]. By this method, Endothall is extracted from water using ion-exchange solid phase extraction (SPE), derivatized into the methyl ester, and then analyzed using GC/MS. Testing laboratories often encounter poor endothall recoveries due to interferences that are present in the sample. Section 4.2 of EPA Method 548.1 states that, "interferences in the ion-exchange procedure are other naturally occurring ions in the water sources, namely, dissolved calcium, magnesium, and sulfate." Unfortunately, these ions can be common in some drinking water sources and can cause recoveries to dip into the single digits. This is illustrated by the recovery data that is presented in EPA Method 548.1 and shared in **Figure 2**. To counteract the concentrations of these ions, the method recommends either diluting the sample, which decreases the sensitivity of the method, or treating the samples with EDTA to chelate with interfering ions.

The main cause of this poor recovery in EPA Method 548.1 may be due to the limited capacity of the recommended ion-exchange media, for endothall. The method describes this media as an intermediate strength, primarily tertiary amine ion-exchanger, which is consistent with the manufacturer's description^[2]. This weak anion-exchange resin has higher affinity for naturally occurring, stronger anions such as sulfates and therefore the capacity of the cartridge can be occupied by interfering anions rather than endothall. Additionally, endothall has higher affinity for interactions with dissolved calcium and magnesium cations rather than the sorbents

primary, secondary amine chemistry. As a result, endothall can fail to partition onto the SPE sorbent and can simply come through the extraction cartridge unretained with the load fraction. Typically, weak acids such as endothall, exhibit much stronger and more consistent retention in strong anion-exchange sorbents that contain quaternary amine chemistry. This is because quaternary amine chemistry is strongly basic and consists of permanent positive charges, which are highly efficient for the extraction of weakly acidic compounds such as endothall.

This work will explore the interactions between endothall and entirely tertiary or quaternary amine ion-exchange resins. The effectiveness of these resins will be evaluated individually for the extraction of endothall. The method will then be tested using both standard solutions as well as real world samples containing calcium, magnesium, and sulfate.

Figure 1.
Structure of Endothall

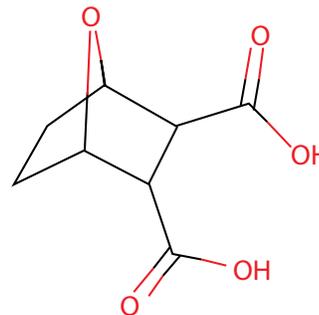
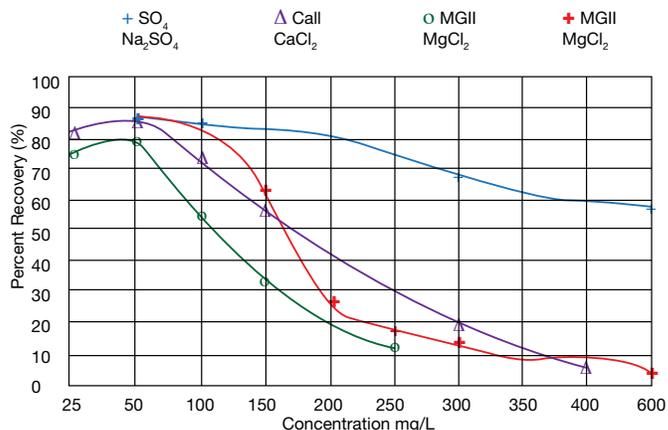


Figure 2.
Endothall Recovery from Reagent Water with SO₄²⁻, Mg²⁺, and Ca²⁺ Ions Present as Provided from EPA Method 548.1



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Materials and Methods

All extraction protocols were followed as stated by EPA Method 548.1. An outline of the method is shown below.

Solid Phase Extraction (SPE) Method

Cartridge: Strata™-XL-A, 2g/12 mL
Part No.: 8B-S053-KDG
Condition: 10 mL Methanol
 10 mL DI Water
 10 mL 10% Sulfuric acid in methanol
 10 mL DI Water
 20 mL 1 N Sodium hydroxide
 20 mL DI Water
 (Do not dry cartridge between steps)
Load: 100 mL of sample without allowing to dry
Wash: 10 mL Methanol
Dry: With air for 5 minutes
Elute: 8 mL of 10% Sulfuric acid in methanol
 6 mL of Methylene chloride

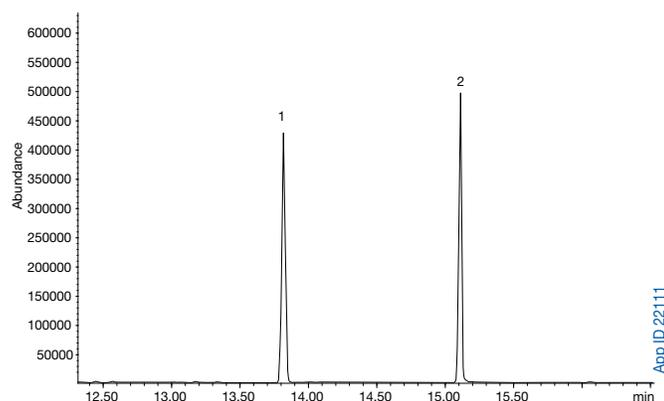
Sample Derivatization and Partition

- Cap and react sample at 50 °C for 1 hour
- Place contents in 125 mL separatory funnel
- Rinse tube with 2x 0.5 mL aliquots of methylene chloride and add to separatory funnel
- Add 20 mL of sodium sulfate in water • Shake
- Drain off lower organic layer into centrifuge tube
- Repeat extraction with 2x 2 mL aliquots of methylene chloride and add to centrifuge tube
- Fortify with 250 µL IS (acenaphthene-d10 @ 10 ppm in methanol) and concentrate to 1.0 mL under dry nitrogen
- Analyze by GC/MS

GC/MS Method

Column: Zebtron™ ZB-SemiVolatiles
Dimensions: 30 meter x 0.25 mm x 0.25 µm
Part No.: 7HG-G027-11
Injection: Pulsed 2 µL @ 200 °C
Carrier Gas: Helium @ 1 mL/min (constant flow)
Oven Program: 80 °C for 5 min to 260 °C @ 10 °C/min for 10 min
Detector: MSD @ 320 °C, 45-450 amu
Note: Pulsed splitless injection @ 30 psi for 0.55 min
Sample: 1. Acenaphthene-d10
 2. Endothall (derivatized)

Figure 3.



Results and Discussion

An instrument performance check solution containing 5 µg of endothall derivatized into the dimethyl ester form was analyzed by GC/MS. The resulting chromatogram and conditions used are shown in **Figure 3**. Internal standard, acenaphthene-d10 eluted at ~13.75 minutes and the derivatized endothall eluted at ~15.2 minutes. Ample separation and good peak shape were obtained for both compounds. After demonstrating instrument capability, the next step was to determine the extent of interaction between endothall and two types of anion-exchange SPE media: weak anion-exchange and strong anion-exchange sorbents. For this, the EPA procedure was followed exactly to extract 50 µg endothall from 1L samples of water using a weak anion-exchange resin containing primary, secondary amine chemistry, and a strong anion-exchange resin containing quaternary amine chemistry. The results are shown in **Table 1**. No recovery from the weak anion-exchange resin suggested that the sorbent failed to retain endothall via a weak anion-exchange mechanism. The strong anion-exchange sorbent exhibited more reliable and stronger interactions to retain endothall via a strong anion-exchange mechanism provided by the quaternary amine chemistry.

Experiments were then conducted to compare the performance of the recommended Bio-Rex® 5 sorbent against Phenomenex Strata™-XL-A, a polymeric strong anion-exchange sorbent. Each sorbent was tested using two different matrices, de-ionized (DI) water and tap water. The dissolved calcium, magnesium, and sulfate content of the local tap water was approximated using the local Annual Water Quality Report [3], and the values are listed in **Table 2**. Though these levels are not extremely high, they are still significant enough to cause dramatically reduced recoveries according to the EPA recovery data in **Figure 2**. All samples were then spiked with 5 µg of endothall resulting in a 50 ppb solution and then extracted using the EPA prescribed protocol.

Five control samples were spiked with 5 µg of endothall, but were not exposed to SPE for the purposes of obtaining a baseline for recovery information. The results of the recoveries are shown in **Table 3**. These results show good recovery for both ion-exchange resins from the de-ionized water matrix when there are no competing ions present. The recoveries are both over 100%, and this is likely due to the emulsion that occurred in the post-derivatization liquid extraction step with the SPE extracted samples, but is not present with the control samples. It is believed that this emulsion assisted in the liquid-liquid extraction of the derivatized endothall. Since the control did not form an emulsion, the liquid-liquid extraction efficiency is not as high. However, both samples have % RSD values below 10%.

The significant difference in the data was observed when analyzing tap water samples. In this instance, the percent recoveries for the weak anion-exchange resin (Bio-Rex 5) are half of the recoveries for the strong anion-exchange resin (Strata-XL-A). This is likely due to competition from dissolved ion interferences for the capacity of the anion-exchange resin.

Because the recoveries in the de-ionized water were above 100 %, this portion of the experiment was repeated by a different analyst on a different day using a different instrument. Unfortunately, the tap water was also taken on a different day so the exact concentrations of salts may have varied. The data for this repeated experiment is presented in **Table 4**. This data also shows that the recoveries for de-ionized water were above 100 % for both sorbents and they shared similar results. The recovery data for tap water shows a significant deviation from the previous data because the percent recoveries for the Strata™-XL-A strong anion-exchange resin were five times (5x) the recovery of Bio-Rex® 5, a weak anion-exchange resin. Under these circumstances, it would have been appropriate to undertake the EPA recommended steps of further dilution of the sample or even treatment with EDTA with only Bio-Rex 5 material. The samples were consistent with all data sets having less than 10 % RSD.

Table 1.
Percent Recovery of Endothall from De-ionized Water

Matrix	Weak Anion-Exchange	Strong Anion-Exchange
De-ionized Water	0	68.1

Table 2.
Concentrations of Calcium, Magnesium, and Sulfate in Tap Water

Ion	Concentration (ppm)
Calcium	150
Magnesium	8
Sulfate	133

Table 3.
Percent Recoveries and RSD Values (n=5) for 50 ppb Samples

Matrix	Bio-Rex 5 % Recovery	% RSD	Strata-XL-A % Recovery	% RSD
De-ionized Water	123.6	3.5	112.8	7
Tap Water	42.4	12.2	84.8	14.4

Table 4.
Percent Recoveries and RSD Values (n=5) for 50 ppb Samples Using a Different Analyst, Different Day, and Different Instrument

Matrix	Bio-Rex 5 % Recovery	% RSD	Strata-XL-A % Recovery	% RSD
De-ionized Water	156.1	8.0	164.3	1.4
Tap Water	21.9	5.4	95.6	2.9

Conclusion

Endothall is a polar herbicide with two carboxylic acid groups that is extracted and analyzed using EPA Method 548.1. The EPA method prescribes the use of a weak anion-exchange SPE sorbent for extraction. However, this work shows that the weak anion-exchange interaction is unable to extract endothall. The retention is solely due to the minor content of strong anion-exchange that is present in the prescribed material. In the presence of naturally occurring dissolved ions in real world samples, the capacity of the weak anion-exchange sorbent is overloaded up by the dissolved ions such as calcium, magnesium, or sulfate and the recovery of endothall is dramatically compromised.

This work explored the differences in recoveries between the prescribed material and a strong anion-exchange material, Strata-XL-A from Phenomenex, under real world conditions. Using tap water from surface water sources, we determined that the Strata-XL-A material furnished up to 5x more recovery than Bio-Rex 5. The Strata-XL-A material gave near 100 % recovery when the Bio-Rex 5 resin resulted in only 21 % recovery. Under these circumstances, a laboratory using Bio-Rex 5 resin may need to re-run the samples using a dilution step or a treatment step with EDTA to remove the competing salts. This was not necessary when using Strata-XL-A, saving the laboratory time and costs associated with re-extraction, treatment, and re-analysis.

References

- [1] EPA Method 548.1; http://water.epa.gov/scitech/methods/cwa/bioindicators/upload/2007_11_06_methods_method_548_1.pdf
- [2] Bio-Rad Website; <http://www.bio-rad.com/en-us/product/bio-rex-5-anion-exchange-resins>
- [3] Torrance Water Quality Report for 2012; www.torranceca.gov/PDF/Water-QualityReport2012-Revised.pdf



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Ordering Information

Strata™-XL-A

Format	Sorbent Mass	Part Number	Unit
Tube			
	30 mg	8B-S123-TAK**	1 mL (100/box)
	30 mg	8B-S123-TBJ	3 mL (50/box)
	60 mg	8B-S123-UBJ	3 mL (50/box)
	100 mg	8B-S123-EBJ	3 mL (50/box)
	100 mg	8B-S123-ECH	6 mL (30/box)
	200 mg	8B-S123-FBJ	3 mL (50/box)
	200 mg	8B-S123-FCH	6 mL (30/box)
	500 mg	8B-S123-HBJ	3 mL (50/box)
	500 mg	8B-S123-HCH	6 mL (30/box)
Giga™ Tube			
	500 mg	8B-S123-HDG	12 mL (20/box)
	1 g	8B-S123-JDG	12 mL (20/box)
	1 g	8B-S123-JEG	20 mL (20/box)
	2 g	8B-S123-KEG	20 mL (20/box)
	5 g	8B-S123-LFF	60 mL (16/box)

**Tabless tube available. Contact Phenomenex for details
Additional sizes, formats and sorbent masses available.

Zebtron™ ZB-SemiVolatiles GC columns

ID (mm)	df (µm)	Temperature Limits (°C)	Part No.
20-Meter			
0.18	0.18	-60 to 325/350	7FD-G027-08
0.18	0.36	-60 to 325/350	7FD-G027-53
30-Meter			
0.25	0.25	-60 to 325/350	7HG-G027-11
0.25	0.50	-60 to 325/350	7HG-G027-17
30-Meter with 5-Meter Guardian™ Integrated Guard			
0.25	0.25	-60 to 325/350	7HG-G027-11-GGA
30-Meter with 10-Meter Guardian Integrated Guard			
0.25	0.25	-60 to 325/350	7HG-G027-11-GGC
60-Meter			
0.25	0.25	-60 to 325/350	7KG-G027-11



If Phenomenex products in this technical note do not provide at least an equivalent separation as compared to other products of the same phase and dimensions, return the product with comparative data within 45 days for a FULL REFUND.

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Strata-X is patented by Phenomenex. U.S. Patent No. 7,119,145

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