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I, Jennifer Z Menzies , hereby submit this original work as part of the requirements for the degree of Master of Science in Environmental Science.

It is entitled:

An Evaluation of Biodegradation Rates and Pathways of High Volume Surfactants in the Sewer System

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An Evaluation of Biodegradation Rates and Pathways of High Volume Surfactants in the Sewer System

A thesis submitted to the

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Submitted by

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ABSTRACT

Limited data has been published using the OECD 314A test method, a simulation test that assesses the biodegradability of chemicals in sewer wastewaters. This research used the OECD 314A method to obtain biodegradation rate and pathway information on a suite of high volume surfactants including homologues of alkyl ethoxylate (AE), alkyl ethoxysulfate (AES), alkyl sulfate (AS) and linear alkyl benzene sulfonate (LAS). In this test, trace levels of ^{14}C labeled test materials were incubated in raw wastewater at 15 ^{0}C while the dissolved oxygen in the system was maintained at 0.5 mg/L. Test results provided first-order kinetic rates for primary degradation and pathway information based on RAD-TLC analysis of metabolites. Comparisons of different alkyl chain lengths for the same surfactant were used to evaluate the impact of chain length on biodegradation rates and metabolite formation. Kinetic data for AE showed that parent half-lives in sewer wastewater ranged from 6 minutes for $C_{12}E_3$ to 1.5 hours for the longer alkyl chain length $C_{16}E_3$. Conversely, the number of ethoxylates does not have a measureable effect on rate. Homologs with ^{14}C labels in different positions were used to elucidate biodegradation pathway information. For example, a comparison of AES homologs with ^{14}C labels in different locations of the molecule showed that biodegradation begins with fission of the central ether followed by rapid mineralization of the resulting alcohol through both ω and β-oxidation and slower mineralization of the resulting sulfated polyethylene glycol. Similarly, both AS and LAS degraded in sewer wastewater. While the calculated half-life of AS was 1 min, LAS showed at least a 12 hour lag before degradation started and a half-life of 27 hours. This test method and the data it generates can be used to improve exposure assessments for down-the-drain chemicals including these surfactants as well as other labile materials.

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1. Introduction

1.1 Overview

The objective of this study was to determine the biodegradation rates and pathways for several high production volume surfactants in a wastewater conveyance system. Biodegradation of these surfactants in the sewer prior to reaching the municipal wastewater treatment plant (WWTP) decreases the total chemical load to the WWTP. As in-sewer removal decreases the WWTP influent concentration, the amount of chemical that exits the treatment plant in effluent and on sludge solids also decreases, thus lowering the exposure of the chemical to aqueous and terrestrial environments. Furthermore, in-sewer removal decreases chemical load to the plant and thus decreases the energy required to treat the wastewater. In direct discharge or inadequate wastewater treatment situations, e.g., combined sewer overflow or poorly maintained treatment plants, in-sewer removal is the only mechanism for decreasing the environmental exposure concentration prior to discharge into surface waters.

To date there has been limited inquiry into in-sewer removal rates. Monitoring of WWTP influents shows that the concentrations of these high volume surfactants are in general less than concentrations predicted based on surfactant use volumes and per capita water consumption[1-4]. This observation indicates a high level of in-sewer removal, however, there is little to no data describing biodegradation rates or pathways in the sewer.

The OECD 314A, *Simulation Tests to Assess the Biodegradability of Chemicals Discharged in Wastewater: Biodegradation in the Sewer System,* uses ^{14}C-labelled representative surfactants to quantify primary biodegradation and mineralization rates, and to identify metabolite formation.

Primary degradation (or primary biotransformation) is defined as loss of parent identity through a microbially mediated process. Mineralization is defined as complete degradation of metabolites to CO_2 and other inorganic substances. The surfactants evaluated in this study include homologues of the nonionic surfactant alkyl ethoxylate (AE) and homologs of the anionic surfactants linear alkyl benzene sulfonate (LAS), alkyl sulfate (AS), and alkyl ethoxysulfate (AES) (see Figure 1.1.1 for structures). Multiple AE homologs were used to evaluate the effect of chain-length and degree of ethoxylation on the biodegradation rate and pathway. Two homologs of AES with ^{14}C labels in different parts of the molecule were used to evaluate the biodegradation pathway. Relevant comparisons to surfactant biodegradation in activated sludge and river water provide information on how biodegradation changes in each compartment.

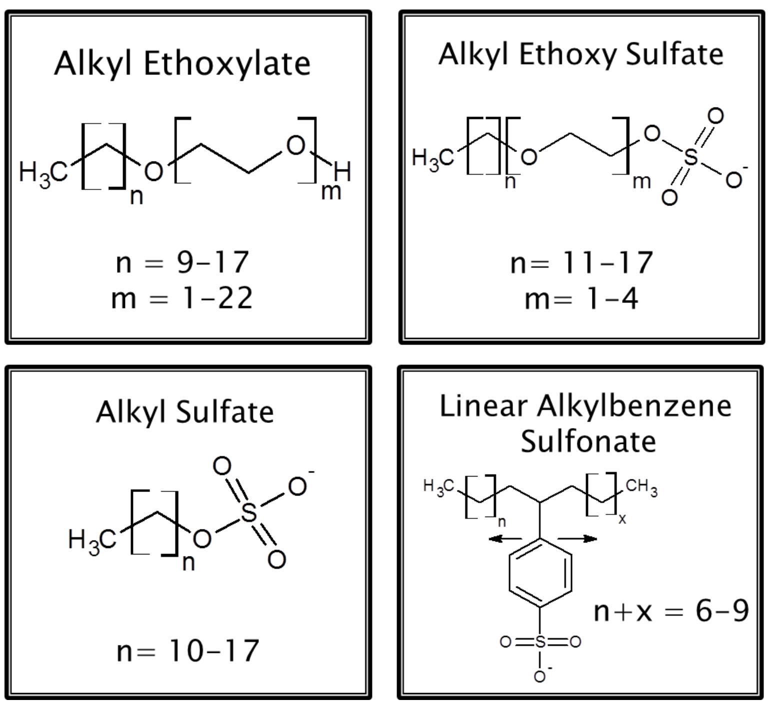


Figure 1.1.1. Generic structures of the surfactants evaluated in this research.

1.2 Surfactant Overview

Surfactants are amphipathic chemicals, i.e., highly soluble in both water and organic solvents, due to the presence of both a hydrophobic "tail" and a hydrophilic "head group". This solubility makes them excellent cleaning agents. The surfactants evaluated in this research are primarily used in household cleaning applications disposed of down-the-drain. This project includes representative homologs of widely used nonionic and anionic surfactants, namely: the nonionic alkyl ethoxylate (AE) and anionic linear alkyl benzene sulfonte (LAS), alkyl sulfate (AS), and alkyl ethoxysulfate (AES). All of these surfactants are classified as Ready Biodegradable per the environmental regulation definition and have been shown to be highly removed during wastewater treatment[5-9].

1.3 Use Volumes and Homolog Distributions

The North American volumes of the surfactants used in this study are reported in the Chemical Economics Handbook[10] and are compiled in Table 1.3.1.

Table 1.3.1. Annual AE, AES, and LAS surfactant volumes of use in North America[10].

Surfactant	2012 North America Use (Thousand Metric Tons)
AE	216
AES	666.0
AS	68
LAS	194

Each surfactant is a mixture of homologs of varying chain length. The chain length homolog distribution used in Western Europe is listed in Table 1.3.2.[5-7,9] The distribution in Table 1.3.2 is an approximation of the homolog distribution used in consumer products and as such was used to identify relevant test materials.

Table 1.3.2. Chain length homolog distribution of the major surfactants[5-7,9].

	AS	AES	LAS	AE
C10			13%	7%
C11	1.4%		30%	5%
C12	29.30%	60.9%	33%	31%
C13	10.60%	8.9%	24%	22%
C14	25.60%	24.8%		15%
C15	6.00%	2.4%		11%
C16	11.70%	2.2%		2.50%
C18	13.20%	0.8%		3%
C20+	1.90%			

In addition to alkyl chain length, AE and AES homologs vary by ethoxylate number. In an AE surfactant mixture, the number of ethoxylate groups attached to the alkyl chain ranges from 1- 22 and is evenly distributed across this range.[6] The number of ethoxylate groups range from 1-4 in the AES surfactant mixture [5]. In the LAS surfactant mixture, the phenyl substitution is distributed along the alkyl backbone. The most nonpolar homologs of LAS are the isomers containing a 2- phenyl substitution, which comprise 18-29% of the commercial mixture[7].

1.4 The Sewer as a Biological Removal System

To estimate in-sewer removal of surfactants, wastewater treatment plant influent monitoring data can be compared to predicted influent concentrations derived from chemical and water per capita use data. This type of analysis has shown that there is variable but measurable surfactant loss in the sewer. Monitoring data from 2003 in North America showed a 98% loss of AES, 4% AE, and 50% loss of LAS in the sewer.[8] Earlier published data from the Netherlands estimated an in-sewer removal of 0-48% for AES, 28-58% for AE, and 18-85% for AS.[1] Additionally, monitoring in Spain and Switzerland in 1990 showed that 30-70% of LAS did not reach the municipal sewage treatment plant.[2] Given the labile nature of these materials, biodegradation was believed to be a dominant removal mechanism. Published laboratory data showed that AE, AES and LAS can be degraded in wastewater[11]; however, the test methods did not include controlled oxygen or temperature conditions. Also, the sample time points of these studies were not sufficient to calculate kinetic rates.

In addition to surfactant removal, there are other areas of research that consider the biological processes and potential for in-sewer removal. This research includes engineering the sewer collection system as a purification process for removing the organic load prior to centralized treatment.[12,13]

1.5 Sewer Conditions: Temperature, Dissolved Oxygen and Biofilms

An actual sewer system varies in temperature, dissolved oxygen, organic load, and inherent biological activity. Given this range of characteristics, it is impossible to create a laboratory system that represents all of these conditions. Thus, the OECD 314A attempts to create a conservative, but realistic sewer, conditions. Realistic sewer conditions are defined as 15 °C, 0.5 mg/L DO and a biofilm free test vessel at test initiation. The OECD defined conditions were evaluated and determined to be conservative by a literature search into sewer conditions.

Temperature in the sewer is an important and highly variable condition[14]. It depends on the geographical climate and season. Wastewater temperature in the United States fluctuates between 3 to 27 °C with 15.6 °C suggested as a representative temperature[15]. The Arrhenius equation can be used to predict rates at other temperatures, but it cannot account for extreme changes due to the fact that certain microbial communities are active within different temperature ranges.

Dissolved oxygen levels are inversely related to wastewater temperature. As the temperature decreases, DO increases due to increased oxygen solubility and decreased microbial metabolic activity. Monitoring in Dutch sewers demonstrated this inverse relationship.[14] This study also showed that DO concentrations in the sewer fluctuate diurnally and seasonally with flow and temperature. The DO fluctuation over the course of the study were wide. During the two month campaign, the measured DO values range from occasional anaerobic periods at 14 °C during low flow to 6 mg/L at peak flow at 9 °C. The majority of the DO measurements over the course of the day ranged from 0.5- 2 mg/L. Thus, a DO of 0.5 mg/L is realistic and conservative.

It is commonly accepted that biofilms are present and active in the sewer system. Two studies found the bacterial population counts per gram dry weight of biofilm is comparable to that of activated sludge.[16,17] These two studies quantified two different biofilms, one was 100-200 µm thick and located on a cement gravity sewer pipe, while the second was on the surface of sewer sediment and determined to be 10-20 µm thick. Biofilms are expected to be pervasive throughout the sewer system. However, the OECD 314A requires a homogenous batch system and thus the growth of a biofilm is not considered in this test. The presence of a biofilm would likely increase the rates of primary biodegradation and mineralization due to increased microbial activity. Thus the use of clean systems is considered conservative.

2. Materials and Methods

2.1 Test Materials

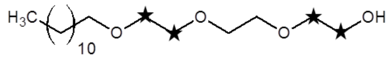
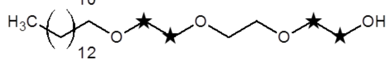
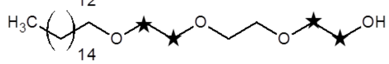
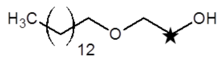
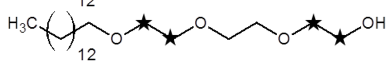
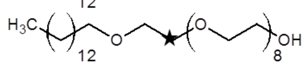
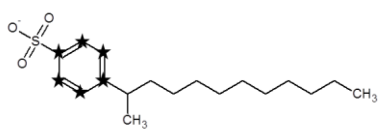
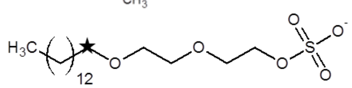
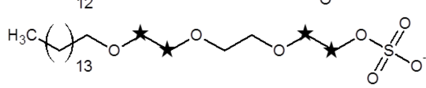
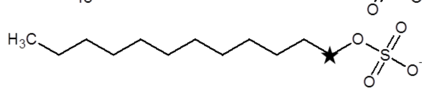
The research was conducted in three phases. The first and second phases focused on the nonionic surfactant linear alkyl ethoxylate (LAE), also known simply as alkyl ethoxylate (AE). All of the AE homologs were labeled in the most recalcitrant portion of the molecule, namely the ethoxylate. The first phase included a suite of AE homologs that varied in alkyl chainlength while remaining constant in extent of ethoxylation. These homologs ($C_{12}E_3$, $C_{14}E_3$, and $C_{16}E_3$) were labeled in the first and third ethoxylate groups away from the alkyl chain.

The second phase of the study included a suite of AE homologs that varied in ethoxylate number while remaining constant in alkyl chainlength. In this phase $C_{14}E_1$, $C_{14}E_3$, and $C_{14}E_9$ were tested. The same $C_{14}E_3$ was used in both phases. The $C_{14}E_1$ and $C_{14}E_9$ were labeled in the first ethoxylate adjacent to the alkyl chain.

The final phase of research included four anionic surfactants, namely: linear alkyl benzene sulfonate (LAS), two alkyl ethoxy sulfate (AES) homologs, and alkyl sulfate (AS). The LAS homolog tested was 2-phenyl, dodecyl LAS and contained ^{14}C labels throughout the benzene ring (Wizard Laboratories Inc., Lot 960820). The homologs of AES were each labeled in different halves of the molecule. $C_{14}E_2S$ was labeled on the alkyl chain carbon adjacent to the ethoxylate chain while $C_{15}E_3S$ was labeled in the first and third ethoxylate. Both the AES molecules were synthesized in the Procter and Gamble Organic Synthesis Laboratory. 1-^{14}C Dodecyl sulfate AS was labeled in the carbon adjacent to the sulfate and was purchased from American Radiolabeled Chemicals (ARC 0833, Lot 141111).

The structure of each test material, the location of the ^{14}C label, its specific activity, chemical purity at test initiation, and concentration at which it was tested are provided in Table 2.1.1. The chemical purity was measured by radio-thin layer chromatography (rad-TLC) using normal phase silica plates (Analtech Inc. Newark, DE) and analyzed by a Bioscan AR 2000 Imaging System (Eckert and Ziegler, Washington D.C.). The plates were developed in the following mobile phases: chloroform / methanol / formic acid (90/10/1) for all nonionics and chloroform / methanol / water/ formic acid (75/25/3/2) for all anionics.

Table 2.1.1. Test Materials: Structure, Label, Purity and Test Concentration

Test Material	Structure with ^{14}C Label (★)	Molecular Weight (µg/µmol)	Specific Activity (µCi/mg)	Purity	Test Concentration (µg/L)
PHASE 1					
$C_{12}E_3$		319	17.0	96.8%	103
$C_{14}E_3$		347	13.7	91.5%	121
$C_{16}E_3$		375	14.0	92.4%	165
PHASE 2					
$C_{14}E_1$		259	23.2	97.7%	61.8
$C_{14}E_3$		347	13.7	> 99%	144
$C_{14}E_9$		611	8.0	98.4%	102
PHASE 3					
LAS		326	146	> 99%	13.1
$C_{14}E_2S$		358	29.0	> 99%	56.6
$C_{15}E_3S$		440	9.3	> 99%	185
AS		365	191	93.2%	8.15

2.2 Biodegradation Testing

Testing was conducted in accordance with the OECD 314A guideline[18]. Briefly, trace levels of radiolabeled test material were incubated in raw wastewater with constant mixing in simulated conservative sewer conditions. Periodically samples were collected, flash frozen, and freeze dried. The freeze dried solids were subsequently extracted with appropriate solvents and the liquid extracts were concentrated and analyzed for metabolites by rad-TLC. The extracted solids were combusted using a Model 307 Sample Oxidizer (Perkin Elmer, Waltham, MA) to account for nonextractable radioactivity. The radioactivity in each fraction was quantified by liquid scintillation counting (LSC) on a Tri-Carb 2900TR (Perkin Elmer, Waltham, MA).

2.3 Test System

Raw wastewater was collected from the influent of a wastewater treatment plant receiving greater than 90% domestic waste (Fairfield Wastewater Treatment Plant, Fairfield, OH). The collection point was located after the grinders and main pump but before the mechanical screen and grit removal. Upon collection, the wastewater was characterized by temperature, pH, chemical oxygen demand (Hach TNT 822), ammonia (Hach TNT 832), and suspended solids levels (Standard Methods[19]).

For each test material 1.5 L of wastewater was dispensed into a 2 L wide mouth Erlenmeyer flask. The test flasks where placed in a circulating water bath that was chilled to 15 °C. The flasks were capped with rubber stoppers while a sample port, two gas lines, and a dissolved oxygen probe were plumbed through the stoppers. Nitrogen gas continuously sparged the system through one gas line and air was intermittently allowed to flow into the system through the second gas line. The DO in the system was continuously monitored and controlled at 0.4-0.6 mg/L by a Model 1630 Oxygen Controller (Engineered Systems and Designs Inc., Newark, DE). The electronic probes were programed such that when the DO dropped below 0.5 mg/L, a set point valve opened to allow air to flow through the gas line into the system. See Figures 2.3.1 and 2.3.2 for pictures and a diagram of the set up. Four test flasks as described above could be set up simultaneously, which allowed for simultaneous test material comparisons in identical wastewater.

Figure 2.3.1. Laboratory test apparatus.

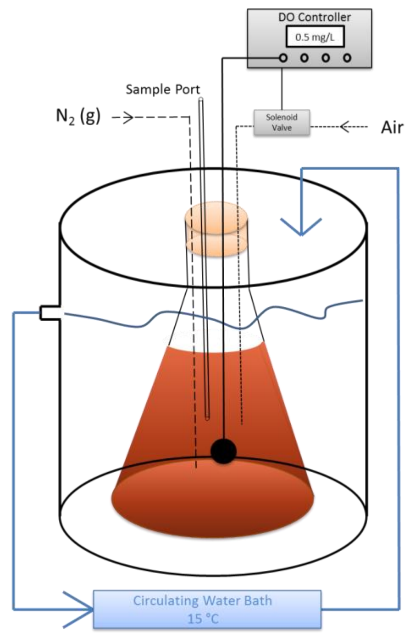


Figure 2.3.2. Diagram of test apparatus.

Abiotic controls were achieved by autoclaving wastewater for 60 min and then amending the media with 0.1 mg/L $HgCl_2$ to inhibit microbial growth. Abiotic controls (0.5 L) of each test material were incubated in an open test system at controlled room temperature (25 °C) in parallel with the biotic treatment. The wastewater was collected from the treatment plant the day prior to test initiation to allow the biotic systems to equilibrate to test conditions and permit time to sterilize the abiotic controls. The test materials were dosed in a 10% methanol solution in dose volumes of at least 1 mL. While the majority of the sampling occurred during the first 12 hours, each test was run for four days.

2.4 Mineralization

Periodically the amount of test material mineralized, or loss of ^{14}C from the system, was quantified by acidifying 1 mL of test media with 0.5% HCl and incubating it overnight to allow any $^{14}CO_2$ to degas. Then, 15 mL of Ultima Gold scintillation cocktail (Perkin Elmer, Shelton CT) was added and the remaining radioactivity quantified by LSC. The biotic and abiotic systems were compared to account for chemical hydrolysis, quenching or sorption onto the test vessel.

2.5 Metabolite Analysis

To measure the degradation of parent and appearance of metabolites, test media were periodically sampled in duplicate. 10 mL of media were collected through the sample port and placed into 15 mL disposable glass conical centrifuge tubes. Samples were immediately flash frozen in a bath of dry ice and acetone. The solids were stored at -80 °C until they were freeze dried. The dried solids were sequentially extracted twice with methanol (10 mL and then 5 mL) and then twice with 5 mL of water. In the case of the AE homologs, the water extractions were conducted at 40 °C to improve extraction efficiency. Like solvent extracts were combined, dried, and resuspended in 1 mL solvent for rad-TLC analysis. With the exception of AS, all TLC analyses were conducted on 60 Å hard silica normal phase plates (Analtech Inc. Newark, DE). AS analysis was conducted on 60 Å soft silica normal phase plates from the same supplier. While most of the AE extracts were developed in chloroform:methanol:formic acid (90:10:1). Due to the position of parent migration, the plates containing $C_{14}E_1$ extracts were developed in ethyl acetate:hexanes (1:1) to better separate parent from metabolites. The plates containing extracts of the sulfated test materials were developed in chloroform:methanol:water:formic acid (75:25:3:2). Radioactivity disposition was reported as a percentage of radioactivity dosed. Duplicate samples were averaged.

2.6 Calculation of Kinetics

The kinetic rates of biodegradation were calculated using Table Curve 2D software version 5.01 (SYSTAT Software Inc. San Jose, CA). A variety of model fits were evaluated but ultimately, the primary biodegradation was best fit to a first-order model (Equation 1) where the amount of parent remaining (y) is a function of initial concentration (A_o), time (t), and the rate constant (k_1). Mineralization data was evaluated against a suite of production models, but they could not be fit to any of the models due to incomplete mineralization over the four day study. The first-order rates were used to estimate the half-life of surfactants in the sewer (Equation 2).

$$y = A_o e^{-k_1 t} \qquad (1)$$

$$t_{\frac{1}{2}} = \ln(2)/k_1 \qquad (2)$$

3. Results

3.1 Wastewater Parameters

A comparison of wastewater characteristics for each test reveals that the total suspended solids of the wastewaters used remained fairly consistent between tests (Table 3.1.1). Additionally, wastewaters collected for the August and November tests had similar COD, temperature, and pH values. The Phase 2 wastewater, collected in February was distinct in that it was colder at the time of collection than the other wastewaters and contained less COD and NH_3. While it could not be measured at the collection point, the dissolved oxygen in the February wastewater was likely higher than in the other wastewaters due to the temperature difference. All wastewaters were collected in the late morning from the same treatment plant, however more work is necessary to understand the inherent variability of wastewater and how it could affect biodegradation rates.

Table 3.1.1. Parameters of the wastewater used in each test phase measured at the time of collection or in the laboratory within 1 hour of collection.

Parameter	Phase 1 August 2014	Phase 2* February 2015	Phase 3 November 2014
Temperature	20.8 °C	12.0 °C	18.8 °C
pH	7.38	7.6	7.42
TSS	160 mg/L	158 mg/L	227 mg/L
COD	551 mg/L	356 mg/L	516 mg/L
$N\text{-}NH_3$	31.7 mg/L	21.4 mg/L	41 mg/L

*Test material alky sulfate was tested in February 2015 but is discussed with the other anionic surfactants.

3.2 Phase 1 Results—Effect of Chainlength on Degradation Rates of AE

The distribution of radioactivity for $C_{12}E_3$ in the biotic treatment and the average for the abiotic treatment over the course of the test are provided in Table 3.2.1. The distribution of radioactivity for all time points is shown graphically in Figure 3.2.1. Less than 60% of the RAM dosed was present as parent after 5 minutes and only 9% remained as parent at 20 minutes. Correspondingly, metabolite formation was also rapid with 70% of the RAM dosed present as metabolites at 20 min. In addition, 10% of the RAM dosed was not extractable from the solids during the first 2 days of the test and 18% was not extractable on day 3 and 4. The final, day 4, abiotic sample contained 98% parent. No mineralization was observed in the abiotic sample. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 94.4% ± 4.6 and 101% ± 3.4, respectively.

Table 3.2.1. Percentage distribution of $C_{12}E_3$ RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0		99.05			
0.08	10.81	58.37	27.12	1.45	86.13
0.33	1.54	9.88	69.98	7.63	86.53
1	14.12	9.19	72.42	9.35	83.37
2	16.64	2.41	74.22	9.17	80.74
3	19.33	2.98	68.12	9.90	76.97
4	16.04	1.49	73.94	10.50	79.29
6	18.06	1.67	70.01	11.36	78.21
8	15.24	1.52	68.50	7.60	72.48
10	20.67	1.42	67.16	10.47	75.04
13	19.41	1.53	67.70	10.53	74.97
15	17.39	1.91	65.45	10.38	76.15
22.6	23.24	1.09	63.24	10.44	73.04
29	24.25	1.51	59.08	10.00	70.57
32.6	27.35	2.10	57.46	10.23	67.59
48	27.87	1.49	52.23	9.60	63.14
72	40.74	1.15	32.24	18.92	51.67
96	54.12	1.41	24.32	18.11	42.27
Abiotic Avg. (± StDev)	0.00 (± 2.1)	98.59 (± 3.09)	1.15 (± 1.5)	0.62 (± 1.44)	101 (± 3.4)

The distribution of radioactivity for $C_{14}E_3$ in the biotic treatment and the average for the abiotic treatment over the course of the test are provided in Table 3.2.2. The distribution of radioactivity for all time points is shown graphically in Figure 3.2.2. For this homolog, 40% of the RAM dosed was present as parent after 1 hour and less than 10% remained as parent after 3 hours. Up to 12% of the RAM dosed was not extractable from the solids during the course of the test. At test initiation, TLC of the test material showed that it was 92% pure. Similarly, throughout the test 86 ± 3% of the dosed RAM was extracted as parent and 9% was extracted as metabolites from the abiotic solids. The quantity of each metabolite remained constant in the abiotic extracts throughout the test. No mineralization was observed in the abiotic sample. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 92.6% ± 4.5 and 96.0% ± 3.8, respectively.

Table 3.2.2. Percentage distribution of $C_{14}E_3$ RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0		90.71			
0.08	-1.09	91.01	7.67	1.03	96.03
0.33	6.86	75.46	13.05	0.63	89.12
1	4.96	40.05	36.38	3.66	79.02
2	9.97	15.40	61.49	5.66	82.16
3	6.28	6.02	70.56	8.35	85.23
4	13.58	5.32	66.83	8.01	82.73
6	10.64	2.74	74.42	9.05	84.27
8	7.83	1.68	76.42	11.85	85.13
10	13.63	1.31	76.26	9.72	84.23
13	11.86	1.50	68.92	12.10	82.05
15	8.13	0.99	69.11	10.74	80.54
22.6	11.71	1.07	68.39	8.87	77.38
29	13.61	0.52	59.42	4.81	72.90
32.6	19.59	0.71	63.15	12.98	76.56
48	23.85	0.27	60.24	6.44	65.24
72	38.10	0.00	37.38	4.89	56.63
96	53.09	0.81	35.79	8.07	42.69
Abiotic Avg. (± StDev)	0.0 (± 2.2)	86.16 (± 3.0)	9.34 (± 2.6)	0.65 (± 0.68)	96.04 (± 3.8)

The distribution of radioactivity for $C_{16}E_3$ in the biotic treatment and the average for the abiotic treatment over the course of the test are provided in Table 3.2.3. The distribution of radioactivity for all time points is shown graphically in Figure 3.2.33. For this homolog, 63% of the RAM dosed was present as parent after 1 hour and less than 10% remained as parent after 4 hour. Approximately 5-6% of the RAM dosed was not extractable from the solids during the first 2 days of the test and 18% was not extractable on day 3 and 4. At test initiation, rad-TLC of the test material showed that it was 92% pure. Similarly, throughout the test 83 ± 5% of the dosed RAM was extracted as parent and 8.5 ± 5% was extracted as metabolites from the abiotic solids. The quantity of each metabolite remained constant in the abiotic extracts throughout the test. No mineralization was observed in the abiotic sample. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 92.2% ± 6.5 and 95.2% ± 5.,2 respectively.

Table 3.2.3. Percentage distribution of $C_{16}E_3$ RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0		89.09			
0.08	-0.71	81.26	8.35	0.15	87.07
0.33	-0.92	94.01	11.12	0.61	99.91
1	-4.35	67.32	29.88	1.71	95.61
2	-5.78	34.90	61.34	3.64	96.52
3	-4.68	18.04	72.51	4.78	93.17
4	-0.86	10.30	76.04	5.72	91.94
6	1.12	5.75	83.73	6.24	94.42
8	-2.94	2.90	81.36	5.89	91.81
10	0.59	2.24	84.90	5.69	91.70
13	-2.76	1.93	79.27	6.10	89.67
15	-2.20	1.81	79.40	3.83	87.82
22.6	0.17	1.25	81.08	4.95	87.41
29	3.00	0.72	71.31	5.23	87.50
32.6	5.92	1.07	76.87	6.13	86.37
48	11.68	0.54	75.32	6.97	81.88
72	27.63	0.56	48.57	19.64	79.50
96	42.94	1.21	32.59	17.15	58.17
Abiotic Avg. (± StDev)	0.0 (± 7.3)	83.79 (± 5.2)	8.54 (± 4.6)	0.55 (± 1.0)	95.23 (± 5.2)

Representative TLC graphs of all three test material are shown in Figures 3.2.4-3.2.6. Figure 3.2.7 shows a comparison of the TLC chromatographs of the methanol extracts for each test material at illustrative time points throughout the test. Figure 3.2.8 is a similar comparison of the water extracts. The biodegradation of each test material produced a similar metabolite pattern. As parent decreased, two major polar metabolites appeared. The 1 hour TLC graph of $C_{12}E_3$ contains a noticeably smaller parent peak, translating to a lower percentage of parent material, than the corresponding graphs of $C_{14}E_3$ and $C_{16}E_3$ (Figure 3.2.7). This visual difference in remaining parent confirms the difference in primary biodegradation rate calculated below. Comparisons of the TLC graphs at later time points show that the longer alkyl chainlengths $C_{14}E_3$ and $C_{16}E_3$ had a high abundance of minor metabolites than $C_{12}E_3$ (Figure 3.2.7). This is discussed in more detail in Section 4.1.

The percent of parent remaining was fit to a first-order decay model and the results of this kinetic analysis are provided in Table 3.2.4. Primary degradation for each material was well described by first order kinetics (Equation 1). The estimated starting concentration was greater than 85% of the dosed RAM (A_o in Table 3.2.4). The r-square values were greater than 0.99 indicating a strong fit of the model to the data. The modeled kinetic rate (k_1) for $C_{12}E_3$ was 6.8 hr^{-1}. This rate is an order of magnitude larger than the k_1 of $C_{14}E_3$ and $C_{16}E_3$, which was 0.84 and 0.49 respectively. These kinetic rates translate into estimated parent half lives in sewer conditions of 0.1-1.4 hours. Complete mineralization did not occur during the 96 hour test and mineralization kinetics could not be calculated.

Table 3.2.4. First-order kinetics of primary degradation of ^{14}C - $C_{12}E_3$ $C_{14}E_3$ and $C_{16}E_3$ in raw sewage.

Test Material	k_1 (hrs^{-1})	A_o	r^2	Half Life (hours)
$C_{12}E_3$	6.8 $\pm$ 0.3	99.3% $\pm$ 1.6	0.996	0.10
$C_{14}E_3$	0.84 $\pm$ 0.03	94.9% $\pm$ 1.4	0.997	0.83
$C_{16}E_3$	0.49 $\pm$ 0.02	87.1% $\pm$ 1.2	0.996	1.41

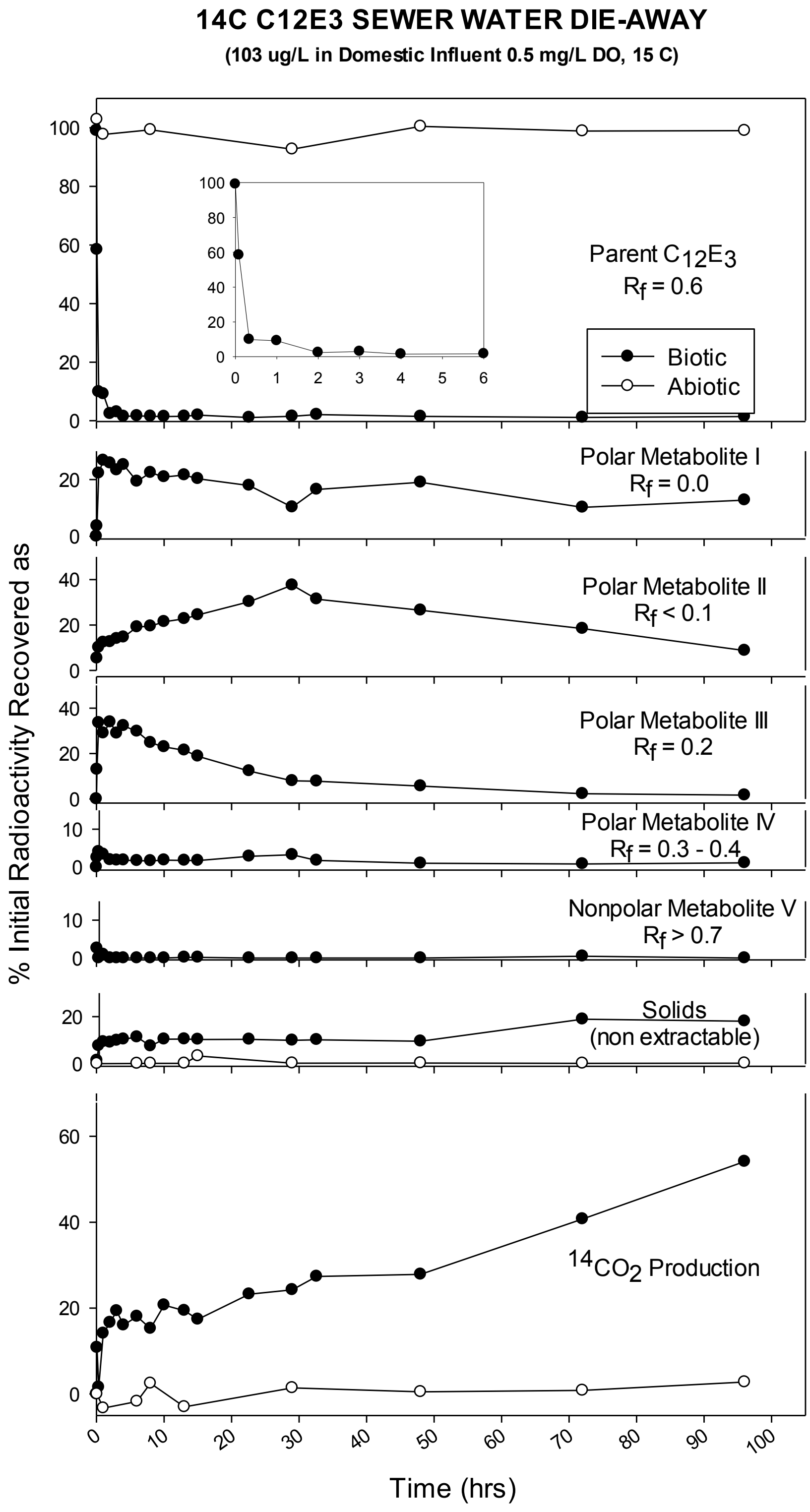


Figure 3.2.1. Radioactivity disposition of $C_{12}E_3$ sewer die away.

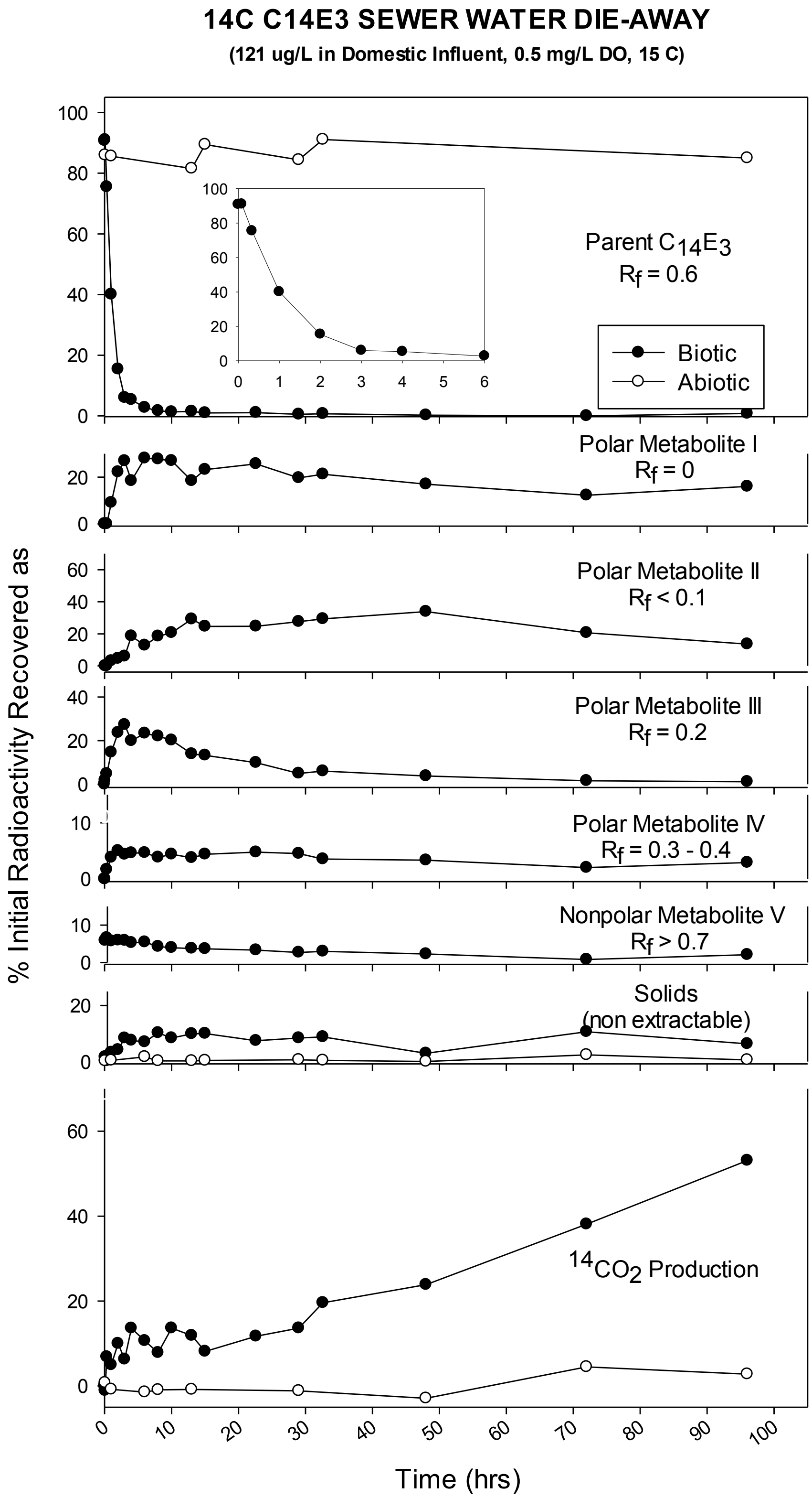


Figure 3.2.2. Radioactivity disposition of $C_{14}E_3$ sewer die away.

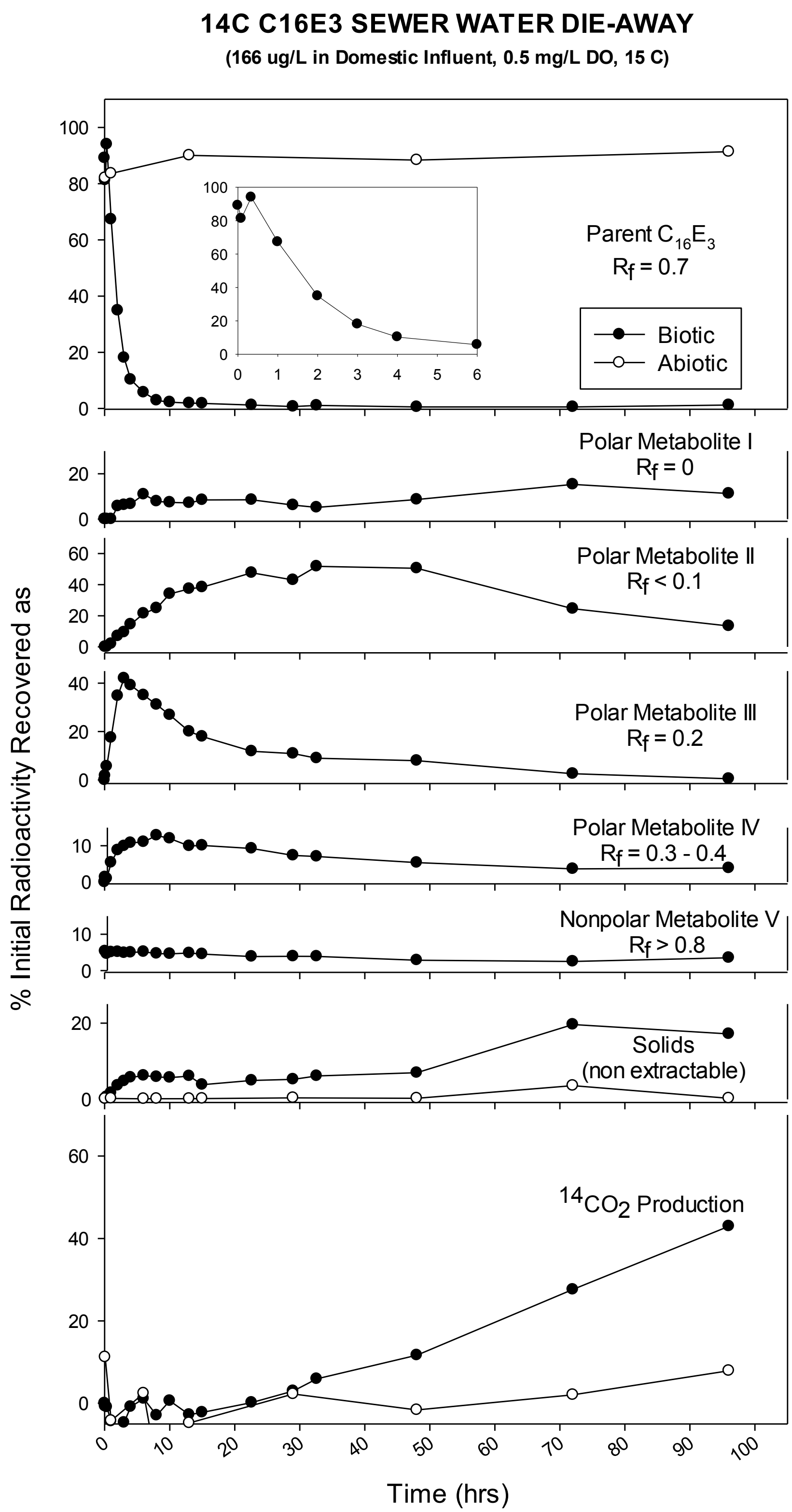


Figure 3.2.3. Radioactivity disposition of $C_{16}E_3$ sewer die away.

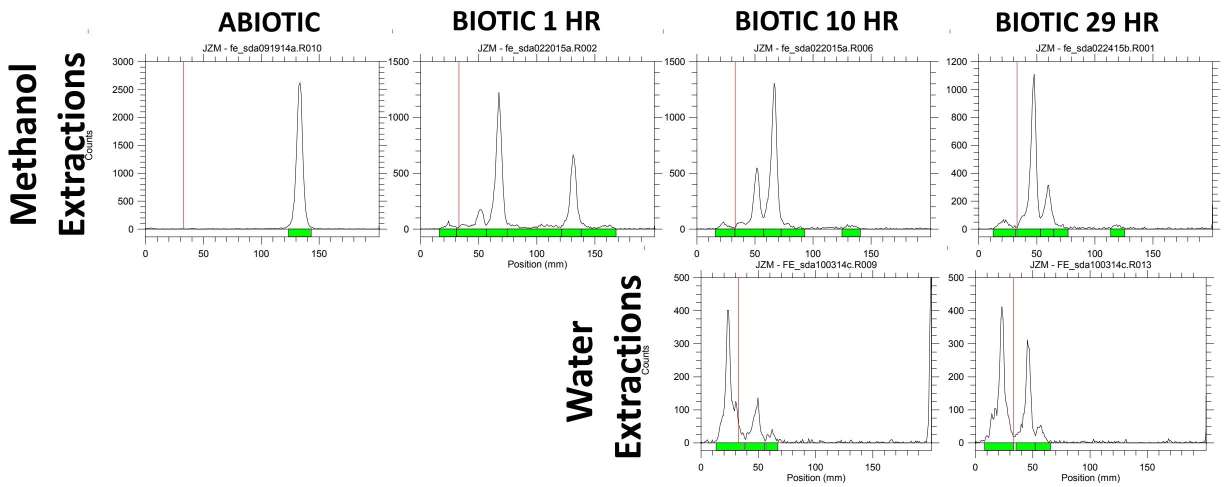


Figure 3.2.4. $C_{12}E_3$-- Representative TLC graphs of the methanol and water extractions.

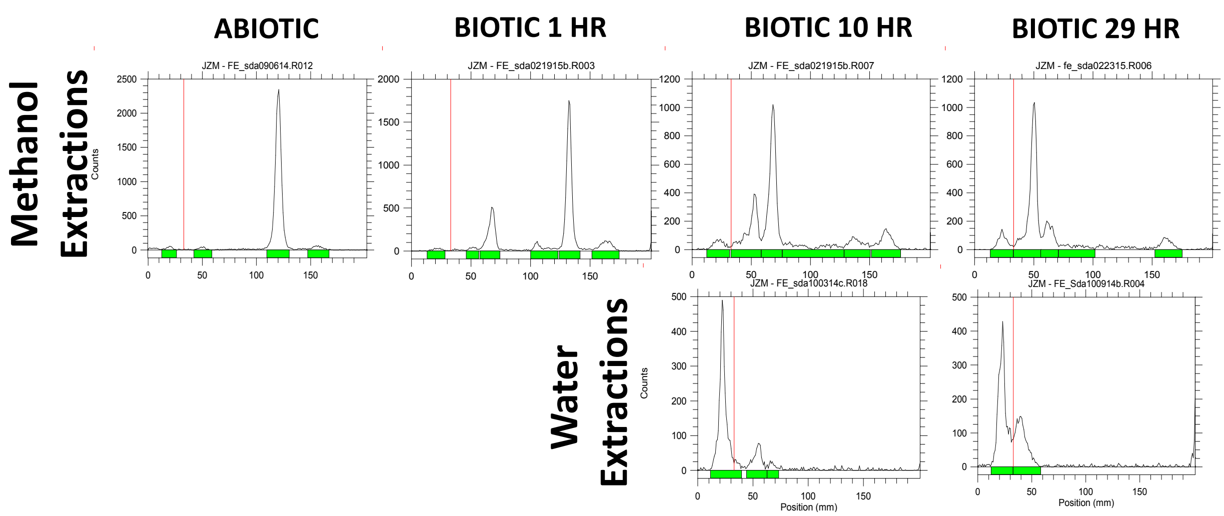


Figure 3.2.5. $C_{14}E_3$-- Representative TLC graphs of the methanol and water extractions.

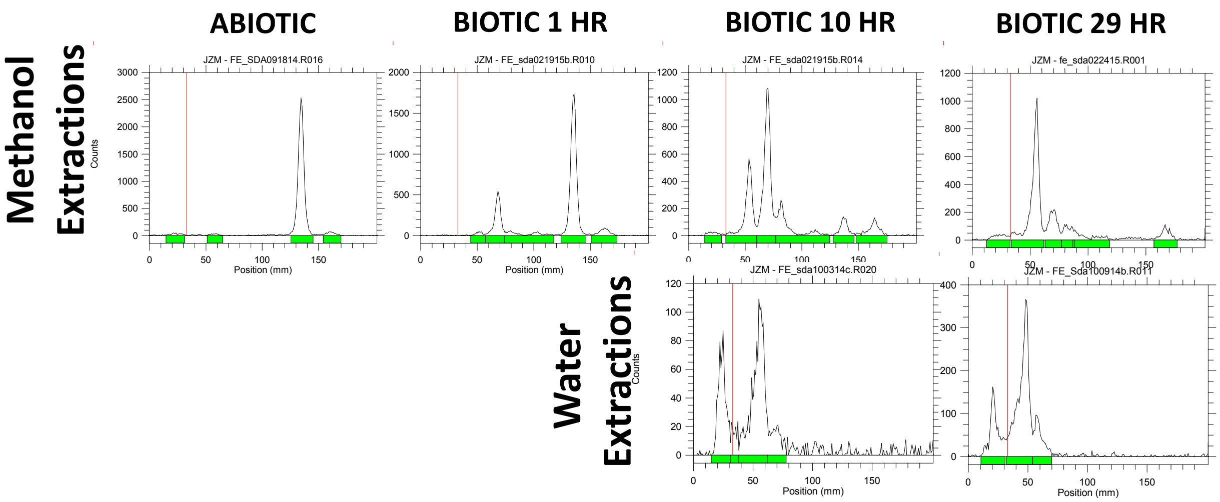


Figure 3.2.6. $C_{16}E_3$-- Representative TLC graphs of the methanol and water extractions.

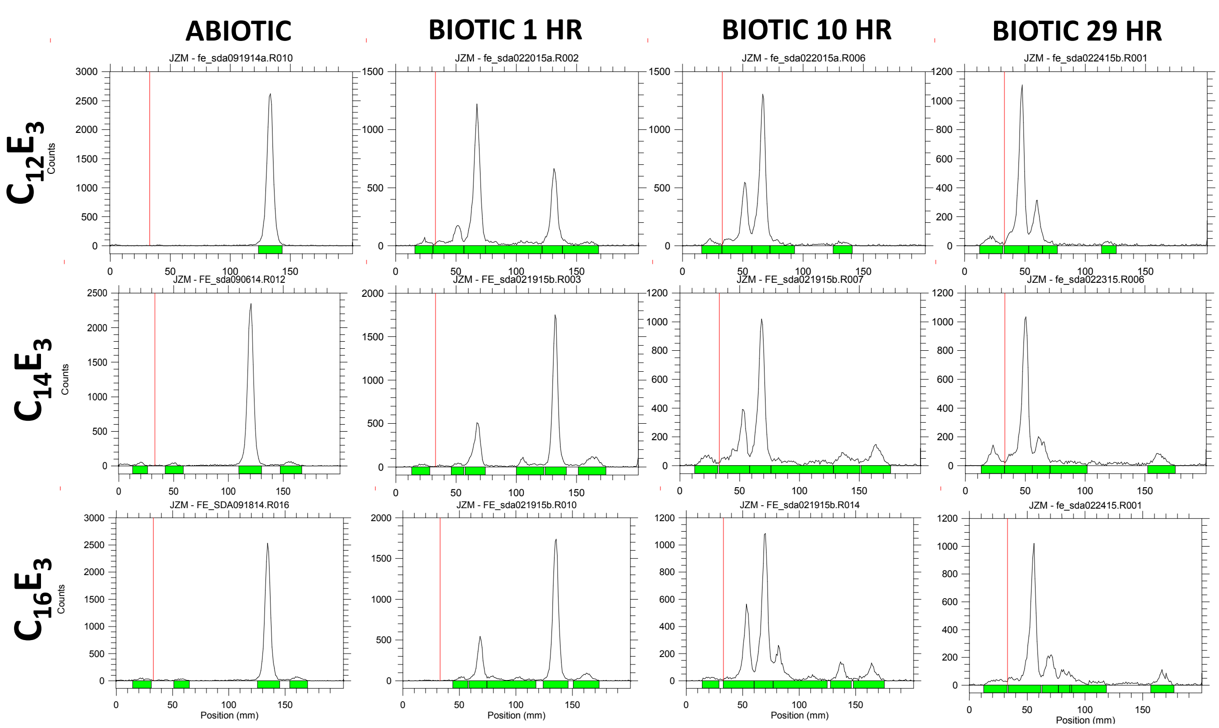


Figure 3.2.7. Comparison of methanol extractions for all Phase 1 test materials.

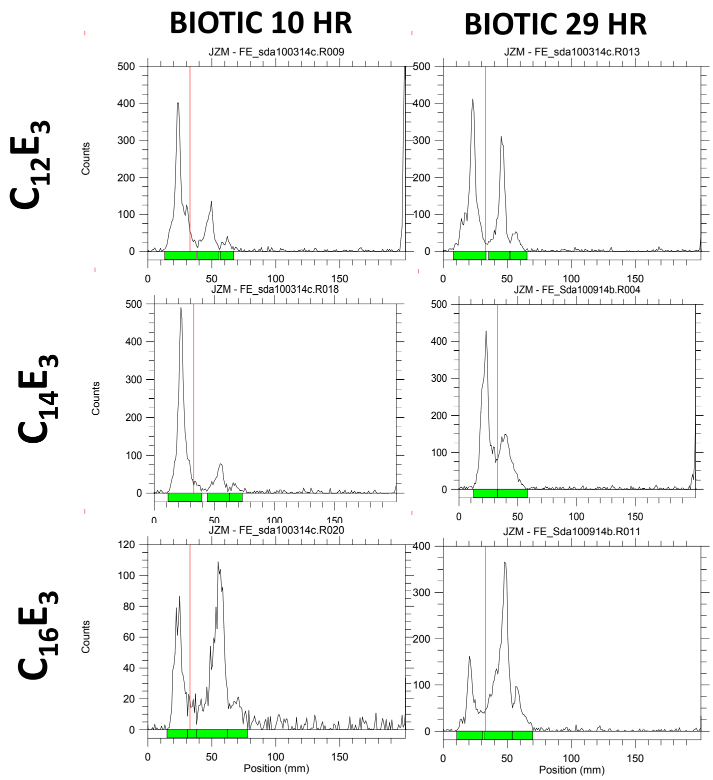


Figure 3.2.8. Comparison of water extractions for all Phase 1 test materials.

3.3 Phase 2 Results—Effect of Ethoxylation on Degradation Rates of AE

The distribution of radioactivity for $C_{14}E_1$ in the biotic treatment and the average for the abiotic treatment over the course of the test are provided in Table 3.3.1. The distribution of radioactivity for all time points is shown graphically in Figure 3.3.1. For this homolog, 32% of the RAM dosed was present as parent after 30 min and less than 10% remained as parent after 1 hour. Both polar and nonpolar metabolites were visualized and at 1 hour the sum of all metabolites increased to 51% of the dosed RAM. Up to 25% of the RAM dosed was not extractable from the solids during the course of the test. This occurred at 24 hours. Mineralization was detectable at 15 min and reached 67% by day 4. Throughout the test, 89 $\pm$ 7% of the dosed RAM was extracted as parent and 9% was extracted as polar metabolites from the abiotic solids. The quantity of the metabolites fluctuated throughout the test and did not trend over time. No mineralization was observed in the abiotic sample. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 91.3% ± 12.1 and 101% ± 5.7 respectively.

Table 3.3.1. Percentage distribution of $C_{14}E_1$ RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0		87.59			
0.017	-2.47	80.11	16.05	0.99	108.50
0.083	-4.43	58.70	26.06	2.13	95.21
0.25	3.49	32.28	35.74	3.07	75.67
0.5	7.93	15.58	43.82	5.28	66.88
1	13.70	6.90	51.01	10.05	68.93
2	16.19	3.57	49.13	8.15	61.35
4	18.79	2.09	47.67	10.99	61.04
6	23.73	2.02	44.30	8.63	55.23
8	30.39	1.07	43.24	8.87	53.34
10	33.00	1.38	37.98	16.57	56.13
12	36.50	1.28	39.69	17.97	59.12
24	44.93	0.74	31.12	25.75	57.71
36	48.80	0.29	31.85	21.76	53.94
48	56.16	0.46	30.58	19.54	50.65
72	60.24	0.26	31.89	11.48	43.66
97	67.27	0.26	23.96	14.73	38.99
Abiotic Avg. (± StDev)	0.0 (± 5.0)	89.7 (± 9.6)	9.08 (± 5.3)	1.32 (± 0.5)	101.4 (± 5.7)

The distribution of radioactivity for $C_{14}E_3$ in the biotic treatment and the average for the abiotic treatment over the course of the test are provided in Table 3.3.2. The distribution of radioactivity for all time points is shown graphically in Figure 3.3.2. For this homolog, 33% of the RAM dosed was present as parent after 15 min and 3% remained as parent after 1 hour. Metabolites formed as parent disappeared and reached 80% at 2 hours. The metabolites remained at this level until 24 hours and then gradually declined. Mineralization was detectable at 30-60 min and reached 35% by day 4. Approximately 18% of the RAM dosed was not extractable from the solids after day 4 of the test. However, for the majority of the test the level on nonextractable radioactivity was around 10%. No metabolites or mineralization was observed in the abiotic sample. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 102% + 6.2 and 103% + 5.1, respectively.

Table 3.3.2. Percentage distribution of $C_{14}E_3$ RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0.0		99.33			
0.02	-2.75	85.01	13.53	2.68	101.27
0.08	4.06	67.96	29.36	2.34	99.70
0.25	1.17	33.46	64.43	6.15	104.07
0.5	5.98	11.86	76.57	9.45	97.89
1	7.73	3.11	66.39	4.56	73.87
2	7.78	2.60	83.72	8.94	95.26
4	10.85	2.20	81.14	10.12	93.46
6	12.72	1.79	83.44	10.13	95.35
8	14.75	1.67	78.42	12.16	92.26
10	14.91	1.63	79.59	10.16	91.39
12	13.84	1.58	80.17	10.01	91.76
24	14.07	1.11	78.39	9.38	88.88
35	22.20	0.70	72.38	9.20	82.28
48	17.69	0.80	66.45	13.29	80.55
72	24.98	0.73	65.22	13.55	79.51
97	34.83	0.67	50.74	18.37	69.78
Abiotic Avg. (± StDev)	0.0 (± 3.8)	101.5 (± 5.5)	0.8 (± 0.6)	1.35 (± 0.5)	103 (± 5.1)

The distribution of radioactivity for $C_{14}E_9$ in the biotic treatment and the average for the abiotic treatment over the course of the test are provided in Table 3.3.3. The distribution of radioactivity for all time points is shown graphically in Figure 3.3.3. For this homolog, 42% of the RAM dosed was present as parent after 15 min and less than 10% remained as parent after 1 hour. The extent of metabolites was 22% at 1 min and increased to 95% of RAM dosed by 2 hours. The amount of RAM dosed that was not extractable from the solids gradually increased throughout the test up to 10%. After 24 hours, 10% of the radioactivity had been mineralized and this level increased to 28% after 4 days. No mineralization or metabolites were observed in the abiotic sample. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 109% ± 5.8 and 107% ± 3.3, respectively.

Table 3.3.3. Percentage distribution of $C_{14}E_9$ RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0		99.17			
0.017	-3.48	81.54	22.58	0.95	108.58
0.083	-7.68	72.13	36.28	1.23	112.57
0.25	-2.78	42.42	62.67	3.61	111.84
0.5	-3.59	17.85	83.99	3.26	106.86
1	11.73	7.04	88.39	6.72	102.51
2	5.40	3.26	95.62	7.01	106.05
4	5.13	2.28	92.46	5.33	100.20
6	7.04	1.64	90.47	7.96	100.16
8	10.80	1.73	92.05	7.20	101.06
10	13.25	1.48	89.21	7.62	98.37
12	8.02	1.49	90.06	7.41	99.04
24	10.20	1.00	88.45	8.85	98.35
36	14.07	0.91	86.75	9.15	96.85
48	14.45	0.42	84.17	7.86	92.48
72	17.18	0.00	81.35	10.69	92.04
97	28.19	0.00	43.65	9.73	92.79
Abiotic Avg. (± StDev)	0.0 (± 4.0)	104 (± 4.7)	1.0 (± 1.0)	1.1 (± 0.8)	107 (± 3.3)

Representative TLC graphs of all three test material in this phase are shown in Figures 3.3.4-3.3.6. Figure 3.3.7 shows a comparison of the TLC chromatographs of methanol extracts of each test material at illustrative time points throughout the test. Figure 3.3.8 is a similar comparison of the water extracts.

The biodegradation of the $C_{14}E_3$ test material produced a metabolite pattern similar to that seen in Phase 1 testing. As parent decreased, two major polar metabolites appeared. Low levels of both polar and nonpolar metabolites were also observed. This pattern was distinct from the metabolite patterns seen either $C_{14}E_1$ or $C_{14}E_9$. The $C_{14}E_9$ chromatographs contained only one major polar metabolite peak. The $C_{14}E_1$ chromatographs contained two distinct nonpolar peaks and two distinct polar peaks. All four peaks were observed as parent disappeared and mineralization occurred. The different number of major polar metabolites in $C_{14}E_3$ and $C_{14}E_9$, was expected based on the different radiolabels of the homolog. In contrast, the nonpolar peaks in the $C_{14}E_1$ chromatographs indicate a distinct biodegradation pathway of this homolog. This is discussed in more detail in Section 4.1.

Figures 3.3.9 and 3.3.10 show the chromatographs of the confirmatory TLC that identified parent peak from the metabolite peaks.

The percent of parent remaining was fit to a first-order decay model and the results of this kinetic analysis are provided in Table 3.3.4. Primary degradation for each material was well described by first order kinetics (Equation 1). The estimated starting concentration was greater than 85% of the dosed RAM (A_o in Table 3.3.4). The r-square values were greater than 0.99 indicating a strong fit of the model to the data. The kinetic rates (k_1) for all three materials were similar. These kinetic rates translate into estimated parent half lives in sewer conditions of 0.16-0.21 hours. Complete mineralization did not occur during the 96 hour test and mineralization kinetics could not be calculated.

Table 3.3.4. First-order kinetics of primary degradation of ^{14}C - $C_{14}E_1$, $C_{14}E_3$ and $C_{14}E_9$ in raw sewage.

Test Material	k_1 (hrs^{-1})	A_o	r^2	Half Life (hours)
$C_{14}E_1$	3.85 $\pm$ 0.19	85.51% $\pm$ 1.4	0.996	0.18
$C_{14}E_3$	4.27 + 0.18	96.19 + 1.3	0.997	0.16
$C_{14}E_9$	3.37 $\pm$ 0.14	97.57 $\pm$ 1.6	0.996	0.21

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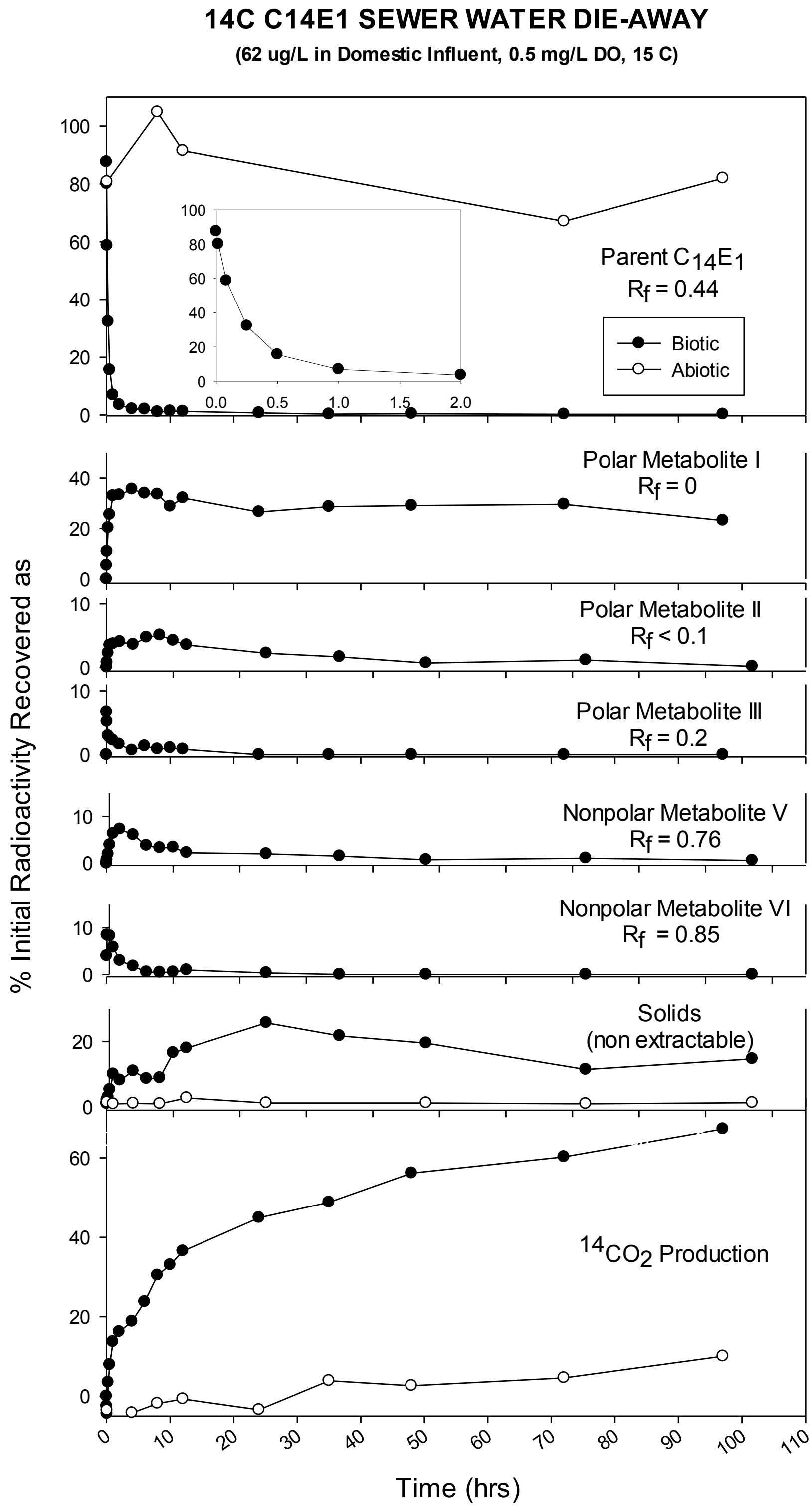

Figure 3.3.1. Radioactivity disposition of $C_{14}E_1$ sewer die away.

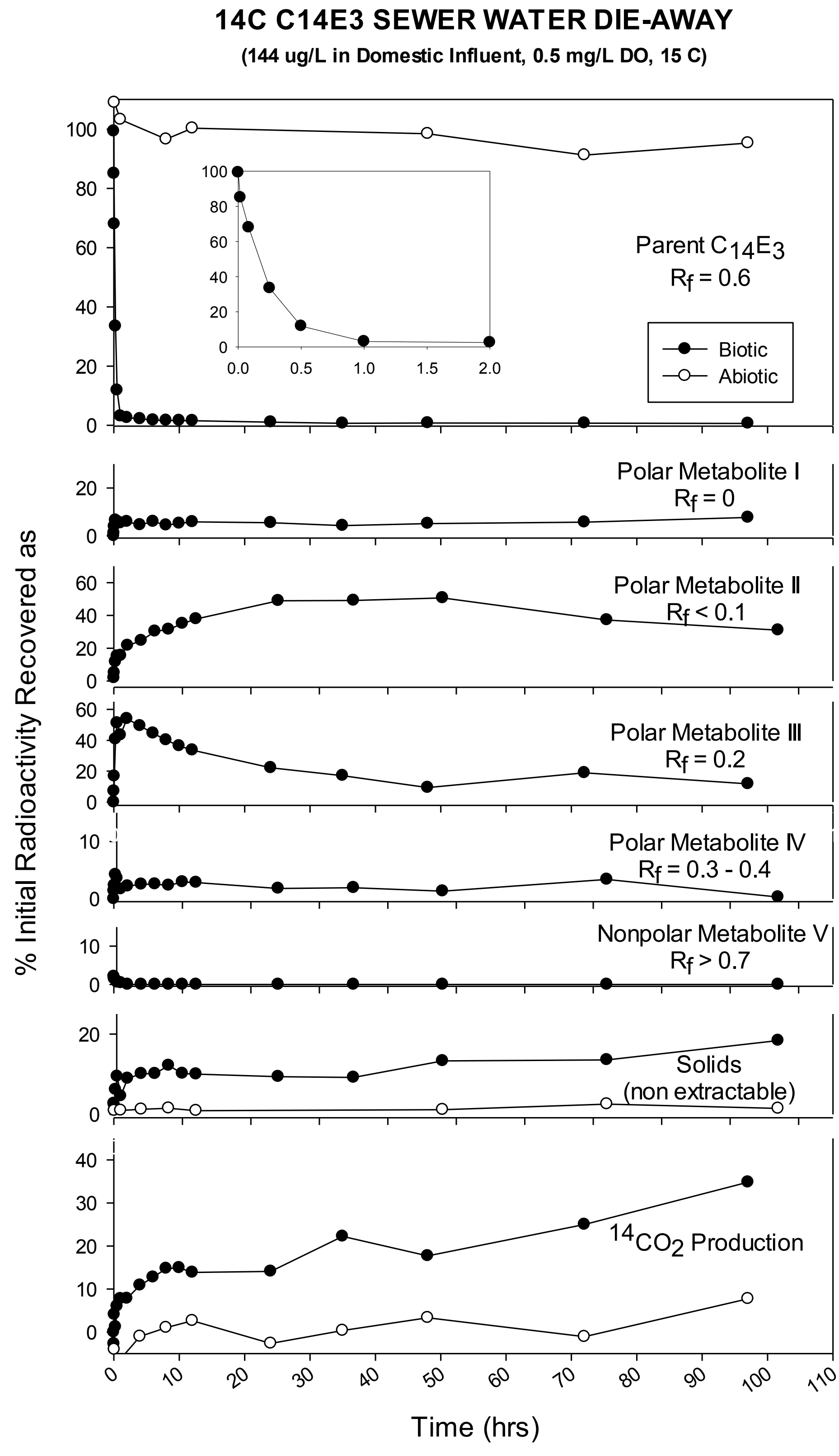

Figure 3.3.2. Radioactivity disposition of $C_{14}E_3$ sewer die away.

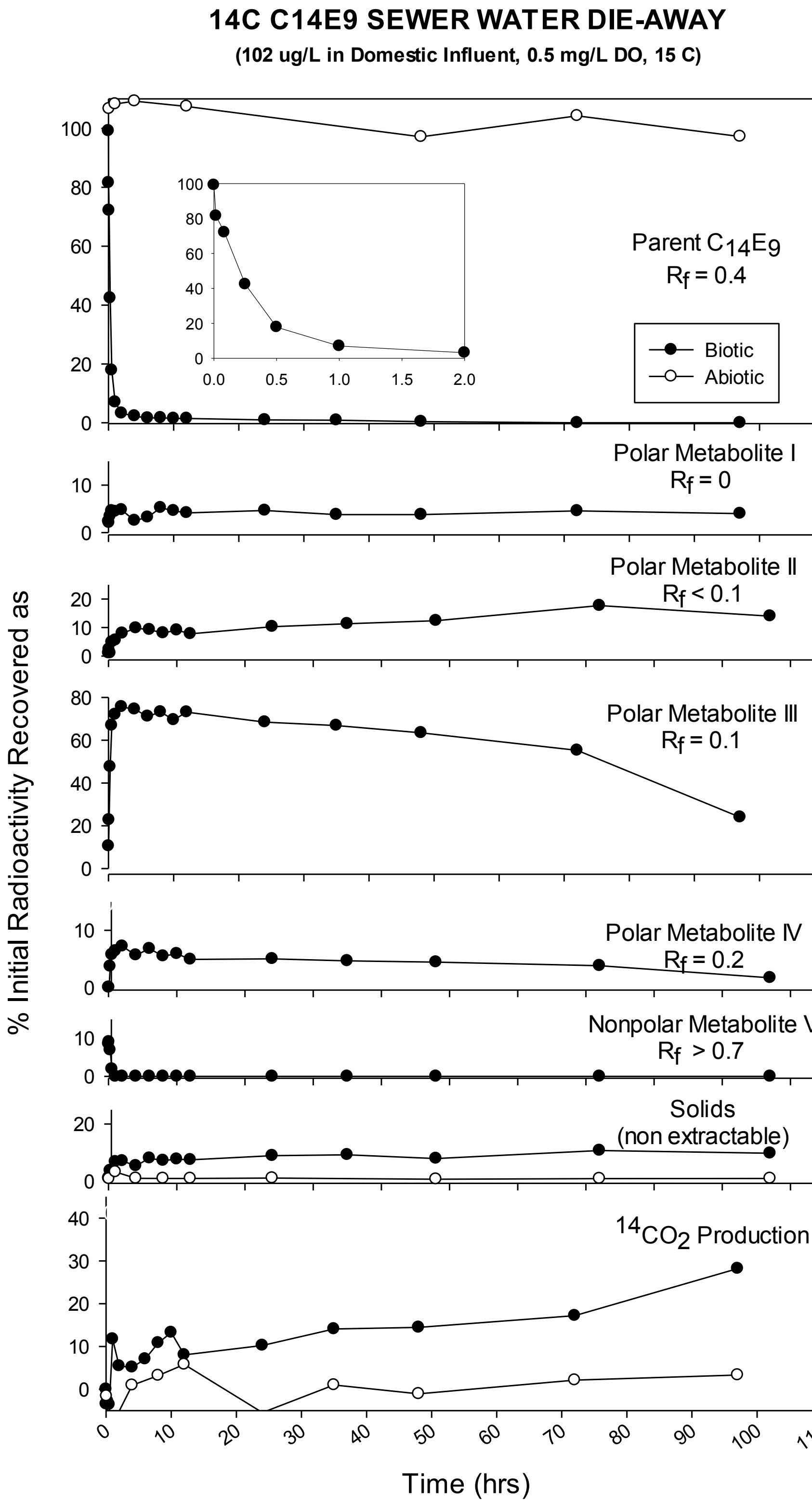

Figure 3.3.3. Radioactivity disposition of $C_{14}E_9$ sewer die away.

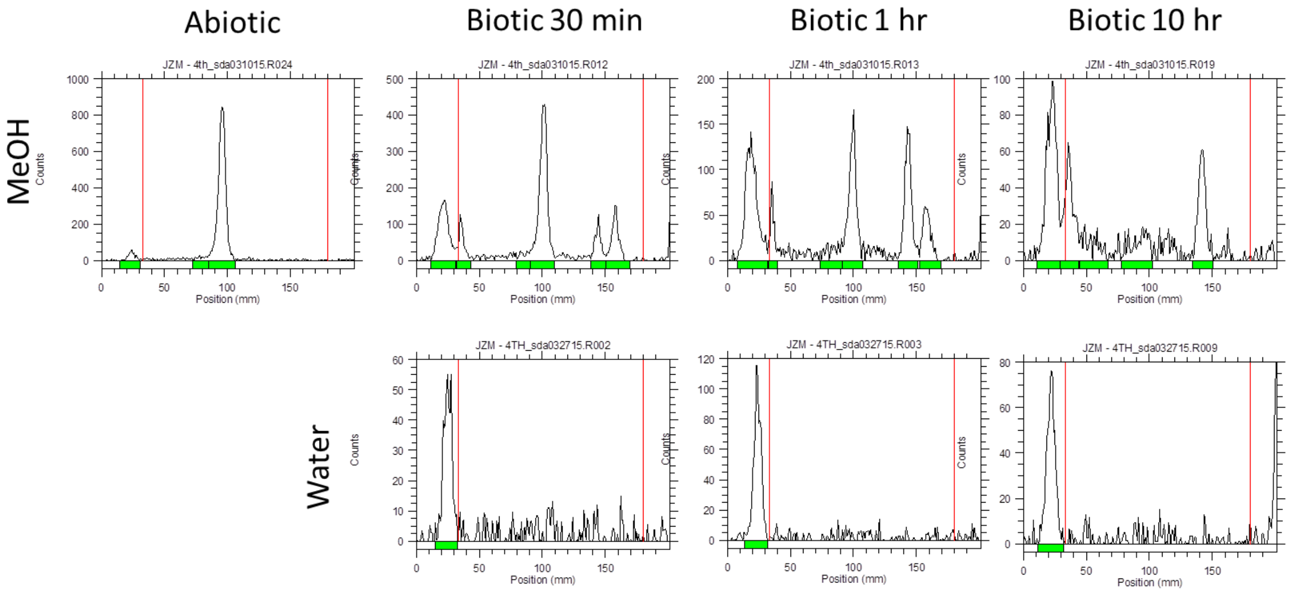

Figure 3.3.4. $C_{14}E_1$-- Representative TLC graphs of the methanol and water extractions.

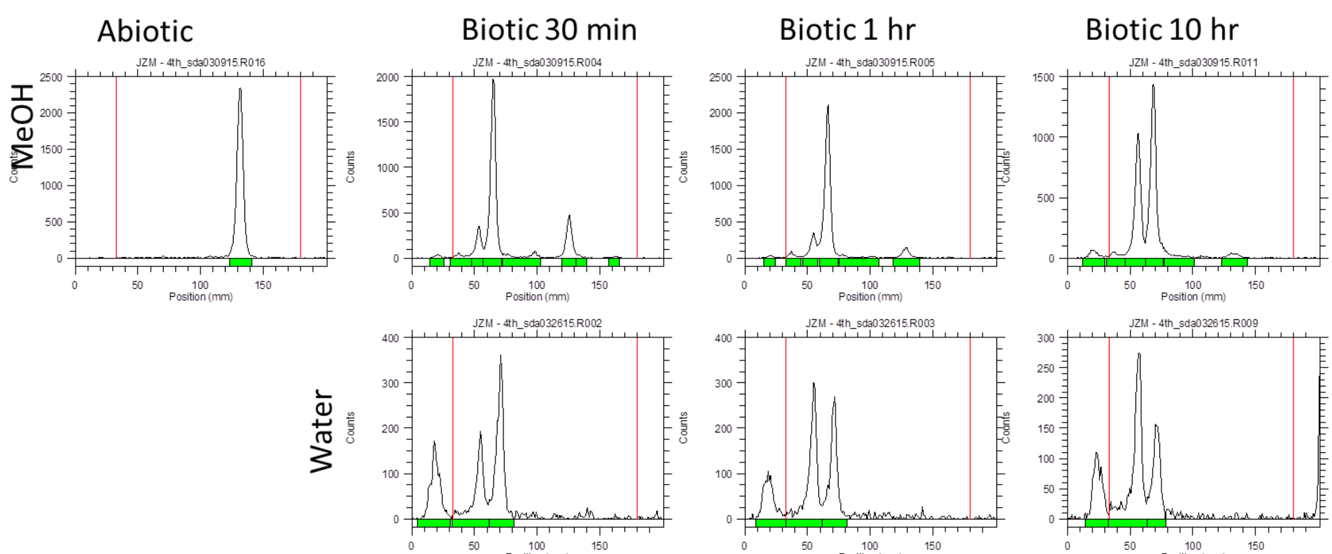

Figure 3.3.5. $C_{14}E_3$-- Representative TLC graphs of the methanol and water extractions.

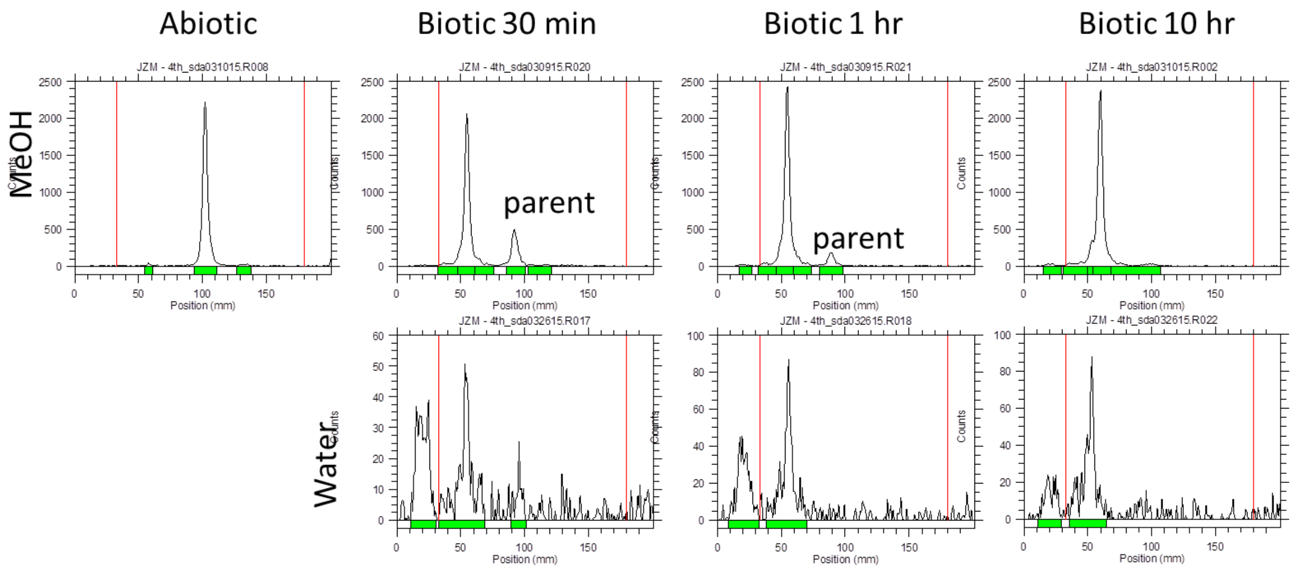

Figure 3.3.6. $C_{14}E_9$-- Representative TLC graphs of the methanol and water extractions.

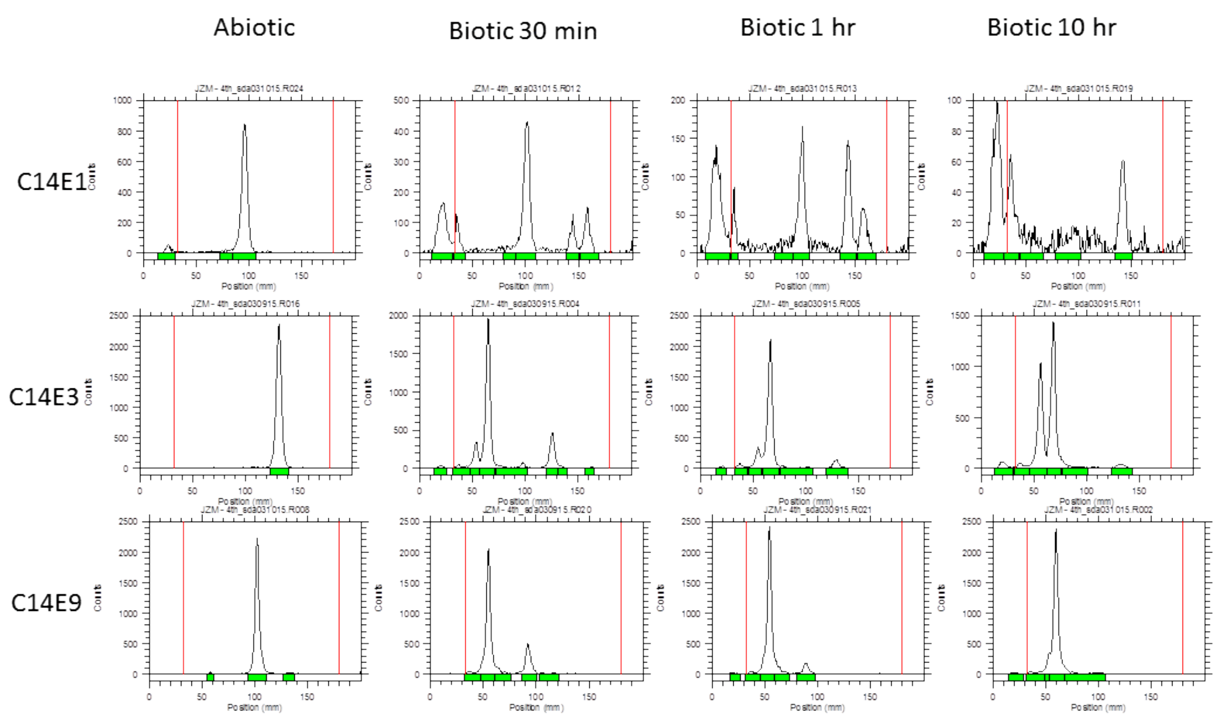

Figure 3.3.7. Comparison of methanol extractions for all Phase 2 test materials.

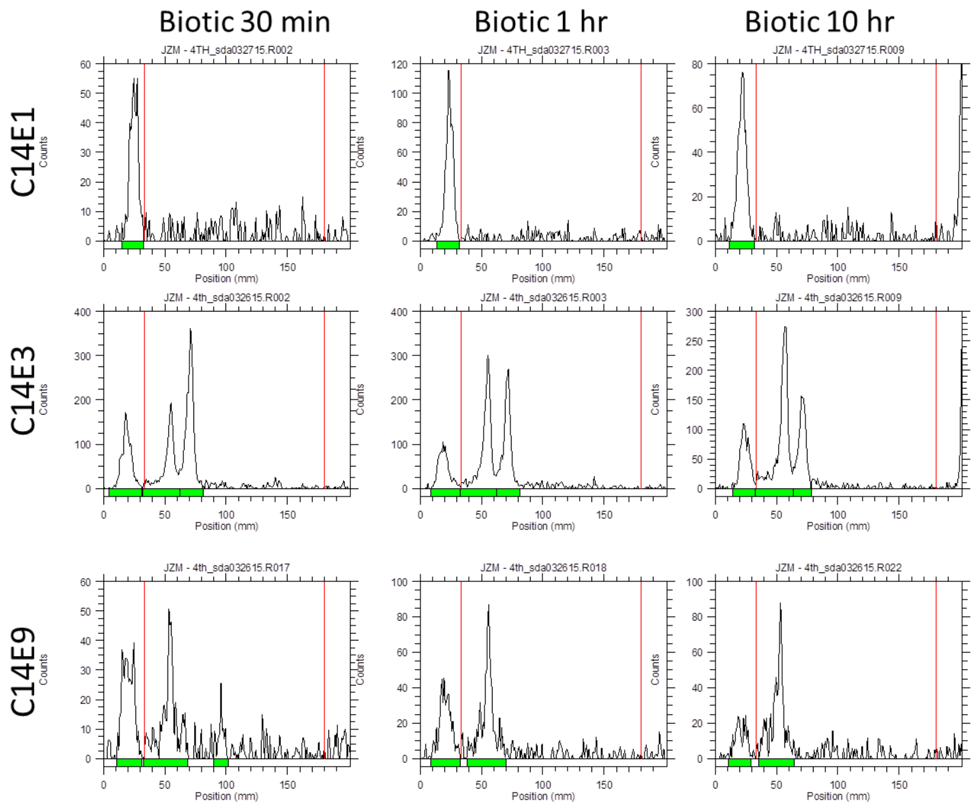

Figure 3.3.8. Comparison of water extractions for all Phase 2 test materials.

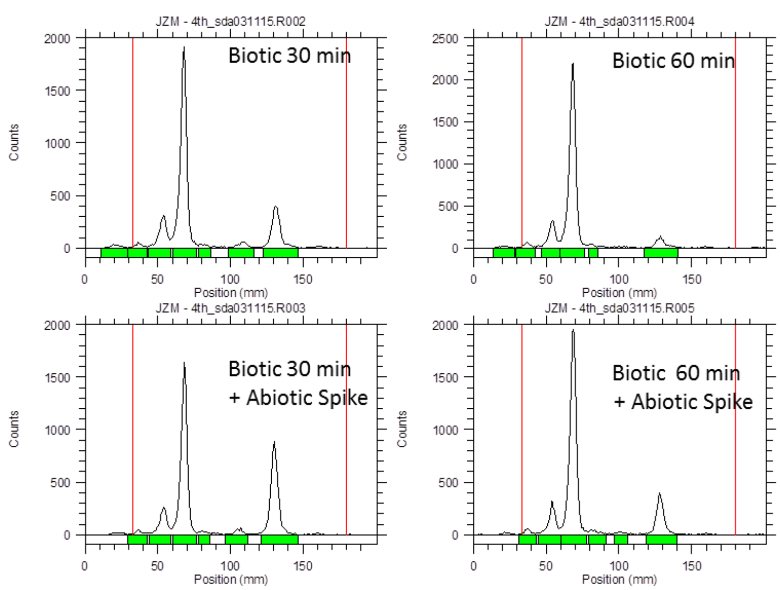

Figure 3.3.9. $C_{14}E_3$-- Parent spike to confirm parent peak.

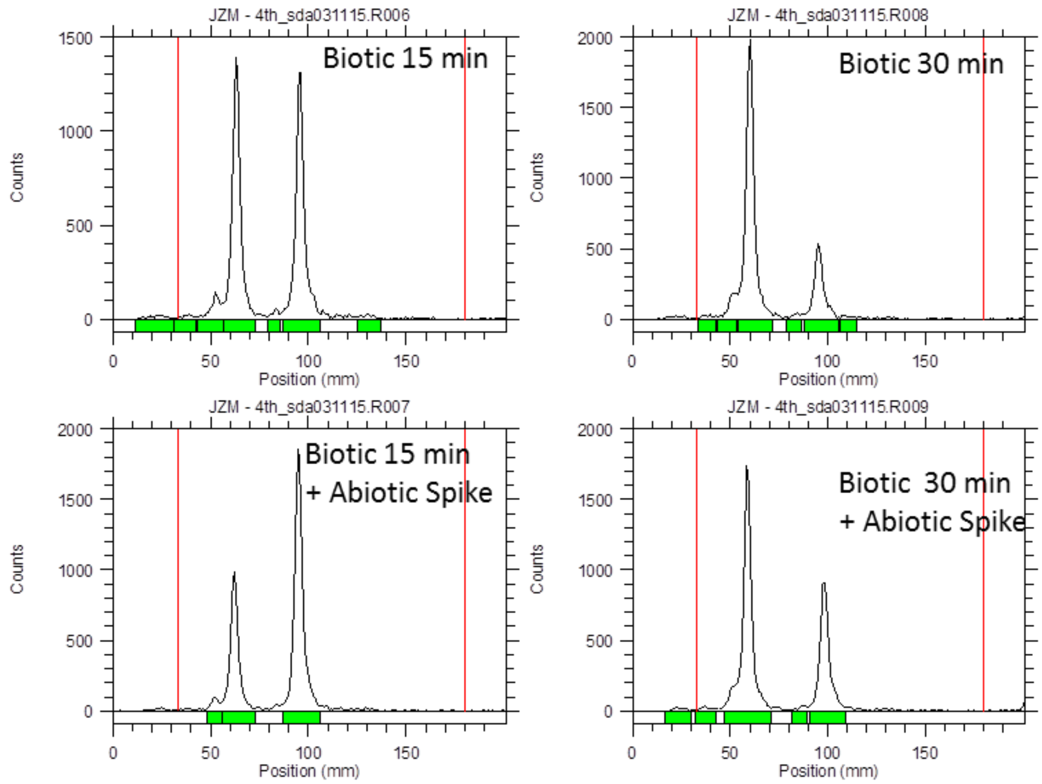

Figure 3.3.10. $C_{14}E_9$-- Parent spike to confirm parent peak.

3.4 Phase 3 Results—Anionic Surfactant Degradation and Rates

The distribution of radioactivity for LAS in the biotic treatment and abiotic treatments are provided in Table 3.4.1. The distribution of radioactivity for all time points is shown graphically in Figure 3.4.1. A significant lag was observed for LAS where greater than 90% of the dosed RAM remained as parent until 2 hours and 50% was still parent LAS after 36 hours. At 48 hours, only 5% remained as parent, 40% was present as metabolites, 10% was not extractable from the solids, and 46% was mineralized to CO_2. On the TLC chromatographs, the majority of the metabolites were visualized as polar metabolites. Two other, minor nonpolar metabolites (less than 2%) were visualized towards then end of the test. The test material in the abiotic treatment remained intact throughout the test and no mineralization was observed. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 106% ± 3.7 and 101% ± 2.5, respectively.

Table 3.4.1. Percentage distribution of LAS RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0		99.27			
1 min	8.14	94.37	1.89	1.17	97.43
5 min	6.96	96.18	1.55	1.05	98.77
15 min	6.96	93.13	2.00	1.09	96.22
0.5	11.36	97.64	1.92	1.25	100.81
1	9.58	93.80	1.91	1.09	96.80
2	11.92	89.13	2.71	1.49	93.34
4	8.31	91.42	3.75	1.02	96.19
6	14.66	87.41	4.42	1.18	93.01
8	14.00	85.76	5.02	1.34	92.12
10	13.88	82.79	6.17	1.57	90.53
12	21.70	81.61	6.85	1.28	89.74
23.5	18.96	67.45	14.55	2.48	84.49
36	24.37	50.59	26.05	4.22	80.86
48	46.35	5.53	40.71	9.50	55.74
72	70.25	2.44	30.24	4.32	36.99
100	73.65	2.62	21.24	10.18	34.04
Abiotic avg. (± StDev)	0.00 (± 4.0)	96.1 (± 2.9)	n/a	2.99 (± 0.78)	101 (± 2.5)

The distribution of radioactivity for $C_{14}E_2S$ in the biotic and abiotic treatments are provided in Table 3.4.2. The distribution of radioactivity for all time points is shown graphically in Figure 3.4.2. At 15 min, less than 40% of the dosed RAM remained as parent and by 2 hours less than 4% remained as parent. Mineralization was quantifiable at 15 min. On the TLC chromatographs, 8 unique metabolite peaks were visualized. The largest metabolite peak was extracted by water and remained at the origin of the TLC plates. This metabolite peak was 14% of the dosed RAM at 15 min and increased to 20% by 24 hr before declining to less than 1% by 4 days. The other metabolite peaks remained less than 3% of the dosed RAM and declined towards the end of the test. A nonpolar peak (Rf 0.92) was presumptively identified as tetradecanol using ^{14}C tetradecanol as a verification standard. The percent of nonextractable radioactivity remaining on the solids increased to 15% at 30 min and remained at that level until 24 hours before declining to 2% by 4 days. The parent in the abiotic treatment remained intact throughout the test. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 109% ± 3.9 and 105% ± 3.8, respectively.

Table 3.4.2. Percentage distribution of $C_{14}E_2S$ RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0		99.77			
1 min	4.27	93.24	2.03	1.84	99.61
5 min	9.30	75.83	12.68	4.65	95.19
15 min	35.28	39.30	22.96	9.71	73.02
0.5	48.73	15.22	26.00	15.76	57.48
1	55.65	5.77	28.20	13.92	48.38
2	60.32	3.78	29.38	14.51	47.97
4	62.35	3.03	26.73	16.70	46.78
6	66.31	2.35	26.54	16.13	45.24
8	69.49	1.61	26.16	14.35	42.32
10	67.08	0.81	27.93	15.29	44.20
12	67.08	0.72	24.81	13.52	39.28
23.5	75.42	0.27	11.65	12.46	36.33
36	81.99	0.45	19.86	11.44	31.92
48	91.38	0.33	15.17	5.36	20.87
72	90.28	0.37	12.94	5.57	18.89
100	93.55	0.26	2.00	2.96	13.41
Abiotic avg. (± StDev)	0.00 (± 3.2)	102 (± 3.1)	n/a	1.1 (± 0.4)	105 (± 3.8)

The distribution of radioactivity for $C_{15}E_3S$ in the biotic and abiotic treatments are provided in Table 3.4.3. The distribution of radioactivity for all time points is shown graphically in Figure

3.4.3. At 30 min, less than 40% of the dosed RAM remained as parent and by 4 hours less than 3% remained as parent. Significant mineralization was not detectable over the course of the test. On the TLC chromatographs, 3 overlapping polar metabolite peaks were visualized. A single polar metabolite at Rf 0.12 increased as parent decreased and then itself decreased as the metabolites associated with it became more polar. The percent of nonextractable radioactivity remained at or below 2% throughout the test. The parent in the abiotic treatment remained intact throughout the test. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 98.6% ± 4.5 and 99.97% ± 4.5, respectively.

Table 3.4.3. Percentage distribution of $C_{15}E_3S$ RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0		98.62			
1 min	7.80	93.85	6.52	0.98	100.50
5 min	0.14	71.37	13.89	0.73	86.09
15 min	1.76	59.90	34.14	0.75	97.06
0.5	2.15	37.87	57.87	0.92	101.55
1	1.94	14.81	86.80	1.10	100.88
2	1.50	5.28	93.78	1.26	100.24
4	-0.02	2.78	95.61	2.62	100.96
6	0.66	2.77	99.01	1.34	103.07
8	1.29	2.20	97.55	1.55	101.27
10	0.14	2.32	99.63	1.62	103.82
12	1.73	2.01	99.12	1.38	102.83
23.5	6.60	1.42	94.32	1.46	97.74
36	4.48	0.44	97.27	1.52	99.71
48	0.08	0.00	92.85	1.74	95.34
72	6.31	0.00	92.30	1.44	94.19
100	16.07	0.00	90.76	1.85	93.00
Abiotic avg. (± StDev)	0.00 (± 3.2)	97.0 (± 5.2)	n/a	0.9 (± 0.4)	99.97 (± 4.5)

Comparison of the $C_{14}E_2S$ metabolite pattern (Figure 3.4.6) to the metabolite pattern in the $C_{15}E_3S$ chromatographs (Figure 3.4.7), supports the hypothesis that the degradation pathway of AES primarily consists of cleavage of the central ether followed by simultaneous alkyl chain shortening from both ends of the alkyl chain through ω and β-oxidation. Comparison of the $C_{15}E_3S$ biotic chromatographs with chromatographs of biotic metabolites of alkyl ethoxylate ($C_{14}E_3$) sewer degradation (also Figure 3.4.7) shows that desulfation of the ethoxy group did not occur during the course of the test.

The distribution of radioactivity for $C_{12}S$ in the biotic treatment and the average for the abiotic treatment over the course of the test are provided in Table 3.4.4. The distribution of radioactivity for all time points is shown graphically in Figure 3.4.4. Less than 50% of the RAM dosed was present as parent after 1 min and 5% remained as parent after 15 min. Correspondingly, metabolite formation was also rapid with 32% of the RAM dosed being present as metabolites at 1 min. The metabolite stayed at this approximate level until 4 hours when it began to gradually decline to 15% by day 4. Mineralization was rapid with 22% of the radioactivity gone from the system at 1 min and at least 50% of the radioactivity gone at 0.5 hour. Up to 20% of the RAM dosed was not extractable from the solids. No mineralization was observed in the abiotic sample. The mass balances of recovered radioactivity for the biotic and abiotic treatments were 114% $\pm$ 6.6 and 106% $\pm$ 2.9, respectively.

Table 3.4.4. Percentage distribution of $C_{12}S$ RAM dose in raw sewage die-away.

Time (hrs)	Mineralized by Difference	Parent Remaining	Metabolites	Nonextractable from Solids	Recovery from Solids
0		93.23			
0.017	21.92	46.84	32.49	4.92	89.88
0.083	38.32	16.80	28.53	12.54	73.70
0.25	52.24	5.04	34.45	13.08	53.15
0.5	58.96	3.22	33.70	16.94	54.23
1	60.47	2.01	25.61	11.16	39.01
2	63.44	2.73	31.20	19.87	54.11
4	64.76	3.00	26.07	17.86	47.27
6	68.78	2.76	27.74	16.98	47.79
8	71.24	2.10	25.20	16.35	43.88
10	73.91	2.16	25.66	18.27	46.34
12	75.07	1.88	26.59	15.82	44.50
24	81.63	1.63	20.64	13.66	36.10
36	84.94	1.01	18.35	13.77	33.25
48	74.94	0.87	20.33	15.76	37.06
72	77.98	0.73	17.21	19.81	38.82
97	82.25	0.43	14.68	15.93	30.66
Abiotic Avg. (± StDev)	0.00 (± 2.3)	96.1 (± 3.48)	7.79 (± 1.58)	0.81 (± 0.29)	106 (± 2.85)

Representative TLC chromatographs for each of the anionic surfactants are shown in Figures 3.4.5-3.4.8. Parent AS and $C_{14}E_2S$ were not well separated from adjacent metabolites. TLC chromatographs of the biotic extracts spiked with parent confirm the location of the parent peak relative to the metabolites (Figure 3.4.6 and 3.4.9). The calculated kinetic rates were minimally affected by different integration estimations. Conservative rates are reported.

The results of the kinetic analysis are shown in Table 3.4.5. With the exception of LAS, primary degradation was well described by the first order model (Equation 1). The estimated starting concentration was greater than 90% of the dosed RAM (A_o in Table 3.4.5). The r-square values were greater than 0.99 indicating a strong fit of the model to the data. The calculated kinetic rates (k_1) for the two AES materials were similar and can be used to estimate parent half lives in sewer conditions of 0.19-0.37 hours. The calculated kinetic rate of AS was an order of magnitude faster than AES and predicts an AS half-life of 0.02 hours in the sewer. The LAS data could be estimated by a first-order model, but due to the lag and shape of the data, the fit was not as strong (r^2 = 0.92). The calculated k_1 was 0.026 hr^{-1} and the estimated half-life in the sewer was greater than 24 hours. LAS, AS, and $C_{14}E_2S$ mineralized extensively during the test, however, the mineralization data did not fit the first order model.

Table 3.4.5. First-order kinetic parameters describing the primary degradation of the anionic surfactants.

Test Material	k_1 (hrs^{-1})	A_o	r^2	Half Life (hours)
LAS (C_{12})	0.026 $\pm$ 0.003	112.5% $\pm$ 3.9	0.922	26.66
$C_{14}E_2S$	3.66 $\pm$ 0.14	100.2% $\pm$ 1.24	0.998	0.37
$C_{15}E_3S$	1.89 $\pm$ 0.13	94.35% $\pm$ 2.14	0.991	0.19
AS (C_{12})	40.53 $\pm$ 3.4	93.3% $\pm$ 2.4	0.991	0.02

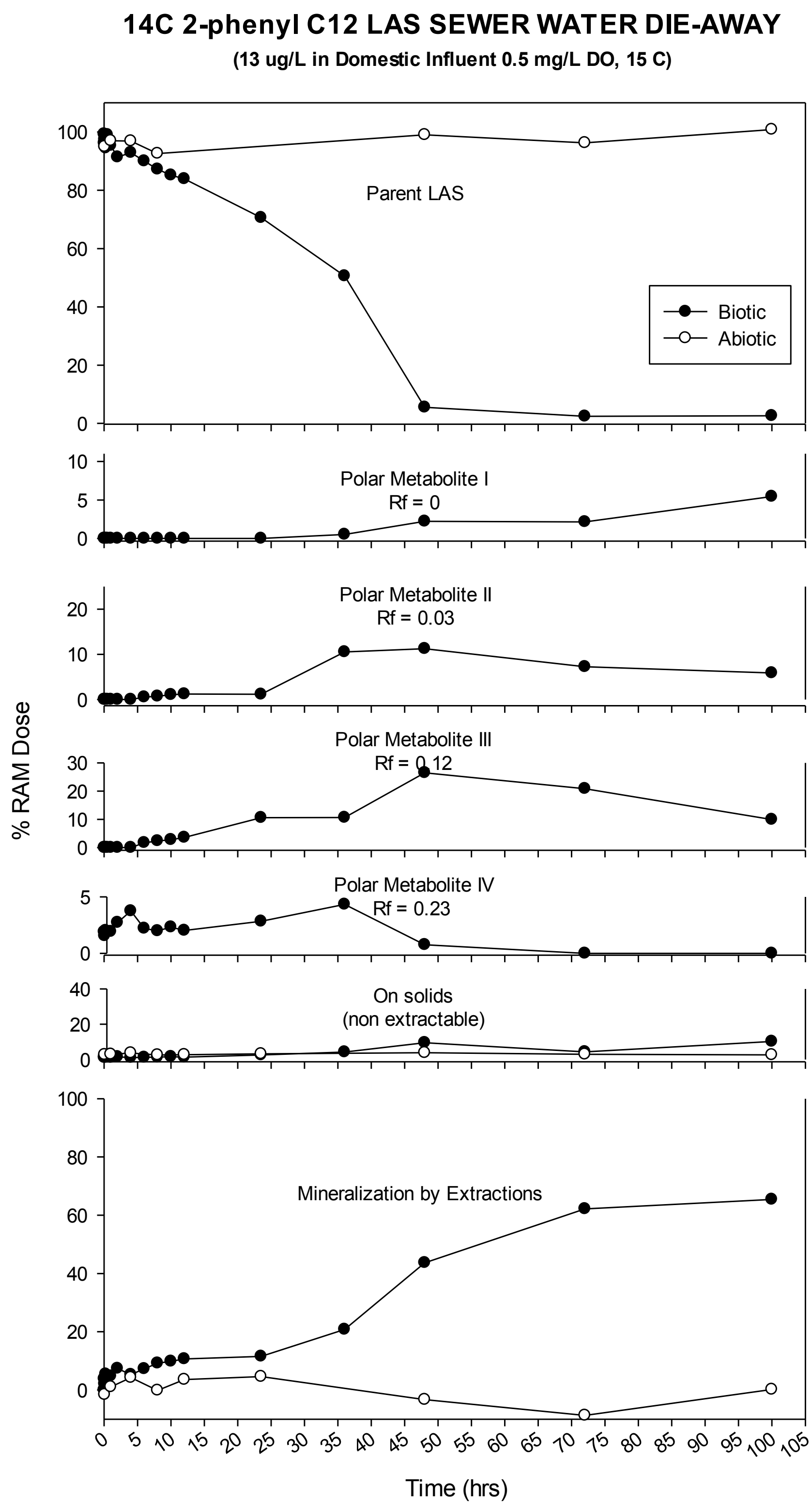

Figure 3.4.1. Radioactivity disposition of LAS sewer die away.

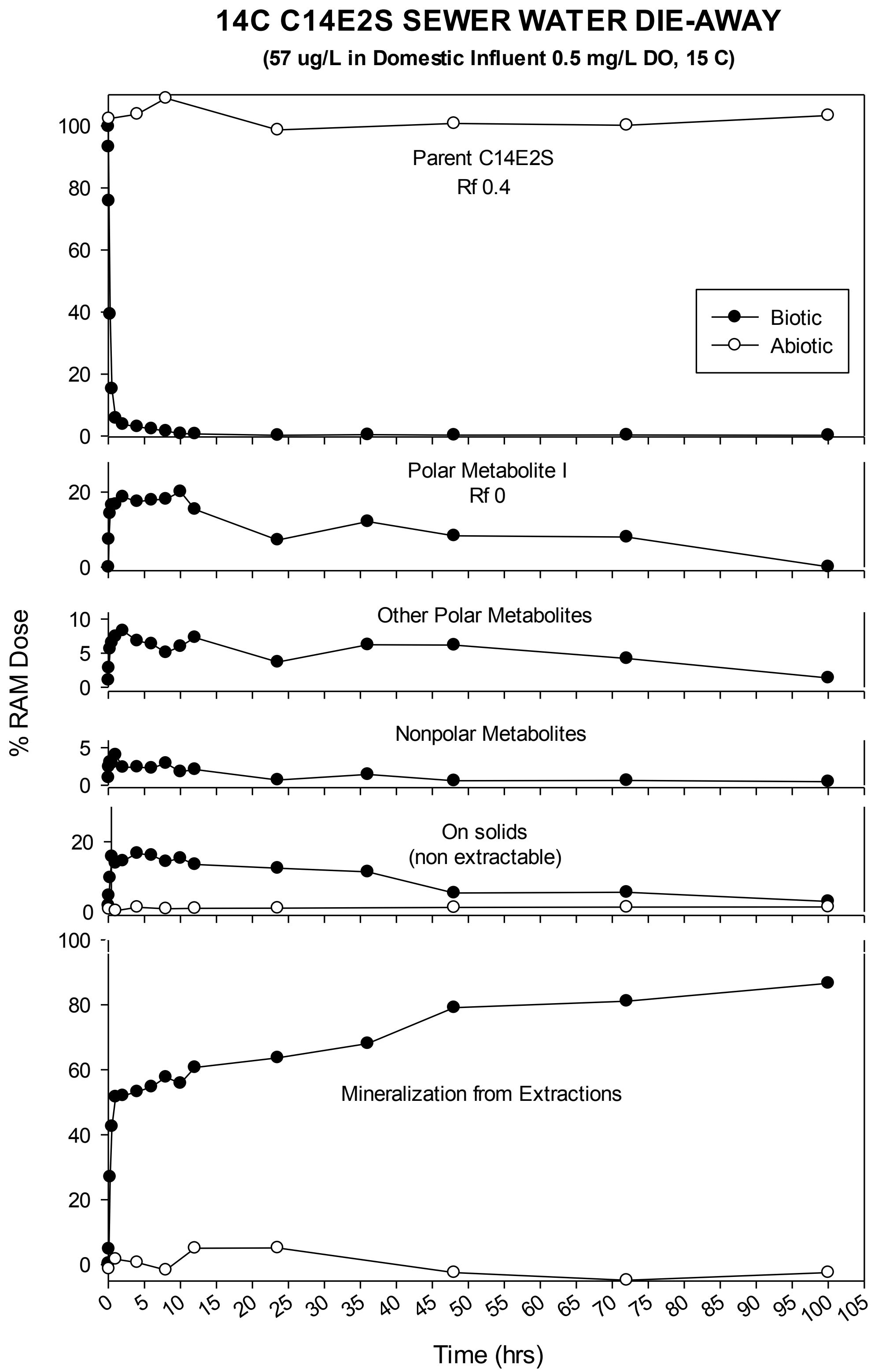

Figure 3.4.2. Radioactivity disposition of $C_{14}E_2S$ sewer die away.

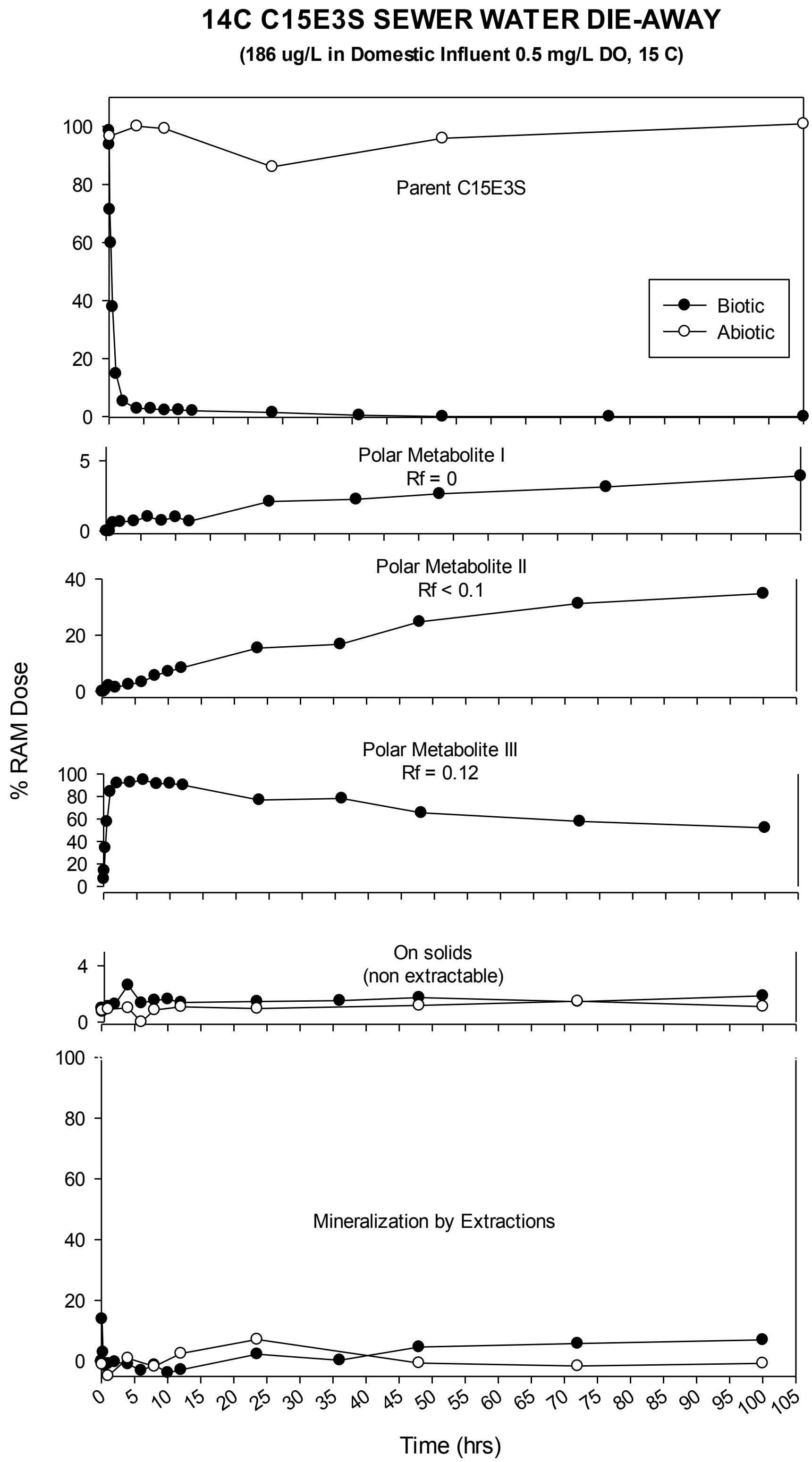

Figure 3.4.3. Radioactivity disposition of $C_{15}E_3S$ sewer die away.

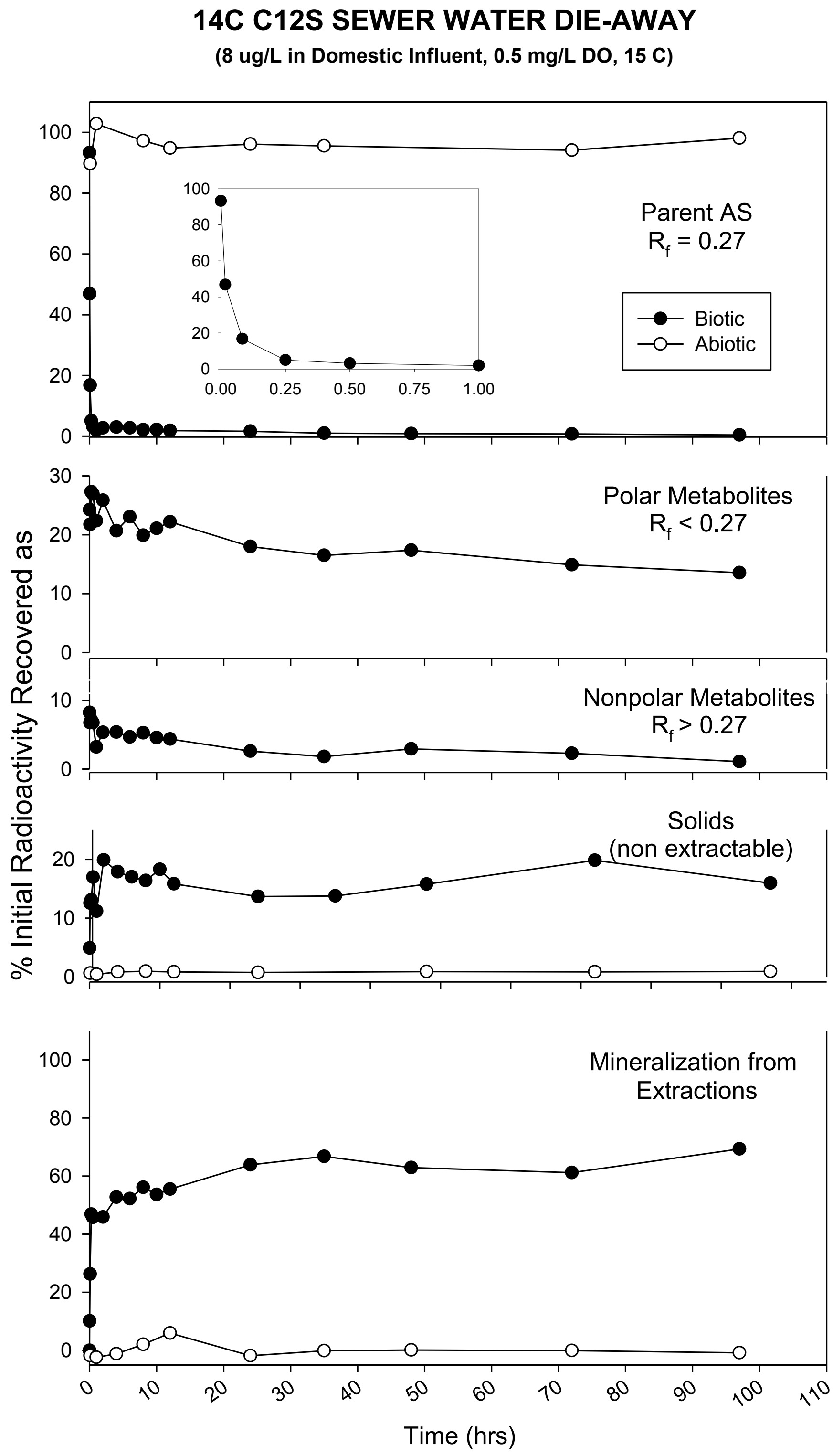

Figure 3.4.4 Radioactivity disposition of $C_{12}S$ sewer die away.

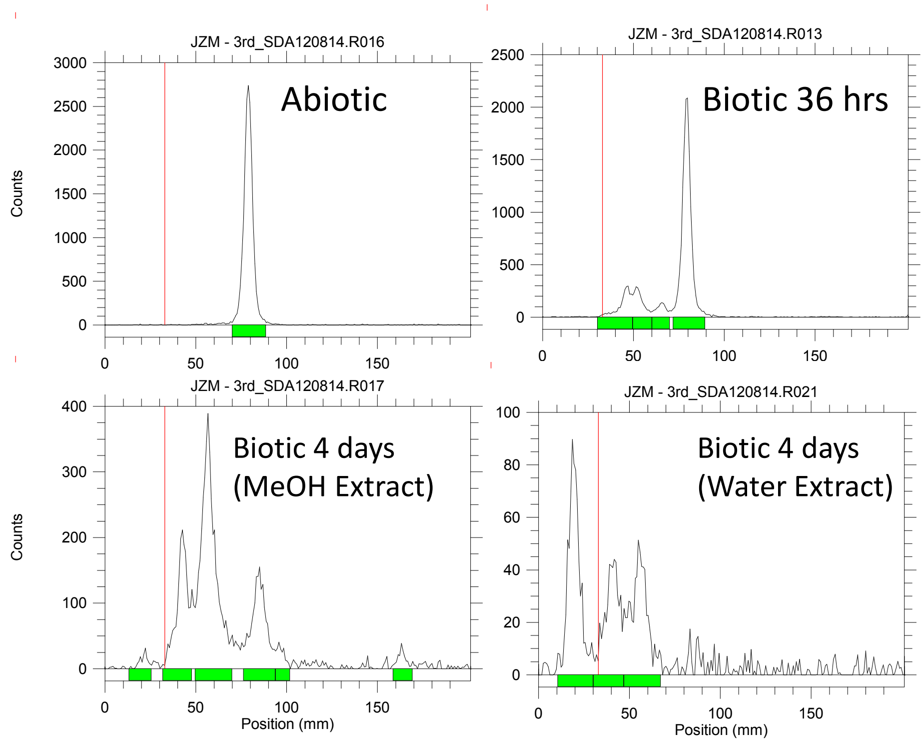

Figure 3.4.5. Representative RAD TLC chromatograms of LAS extracts.

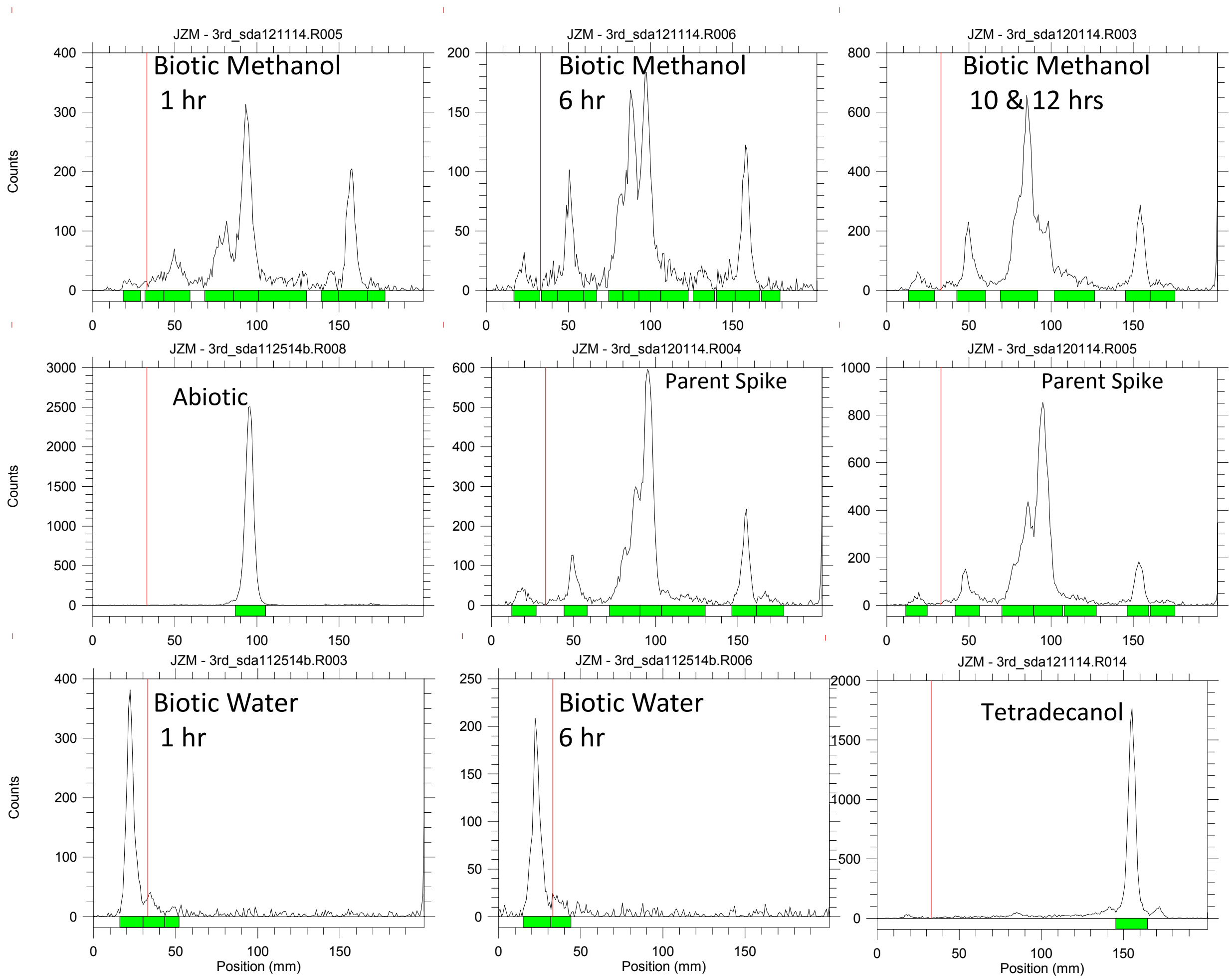

Figure 3.4.6. Representative RAD TLC chromatograms of $C_{14}E_2S$ extracts.

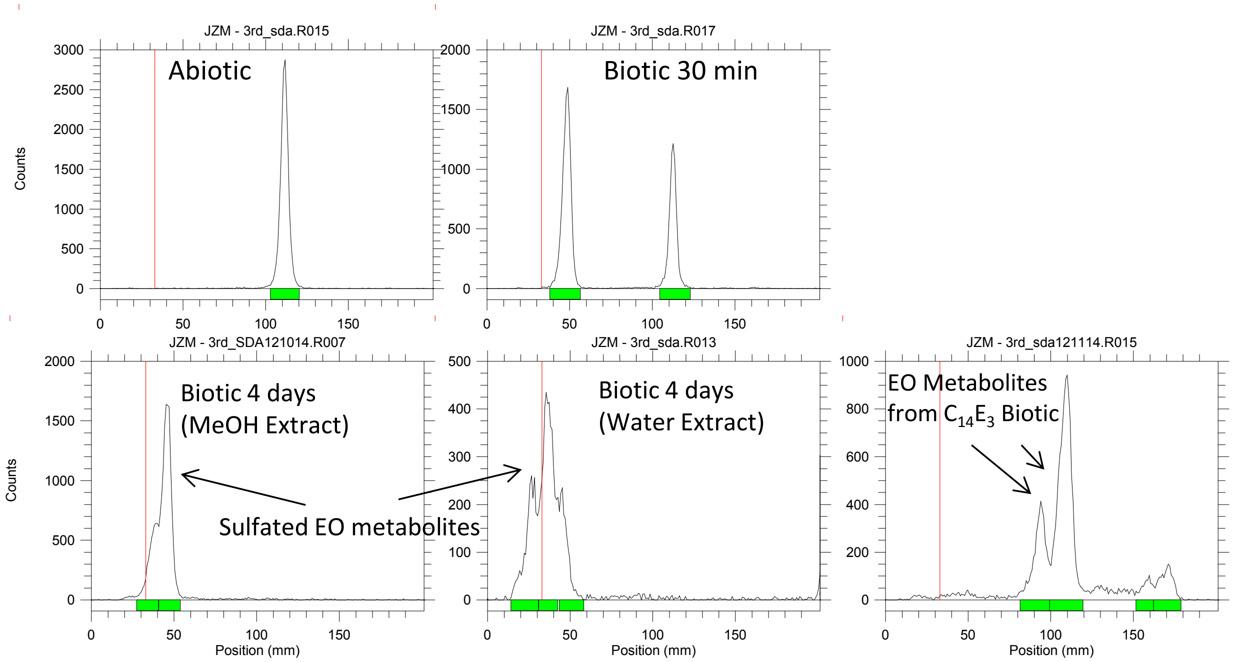

Figure 3.4.7. Representative RAD TLC chromatograms of $C_{15}E_3S$ extracts.

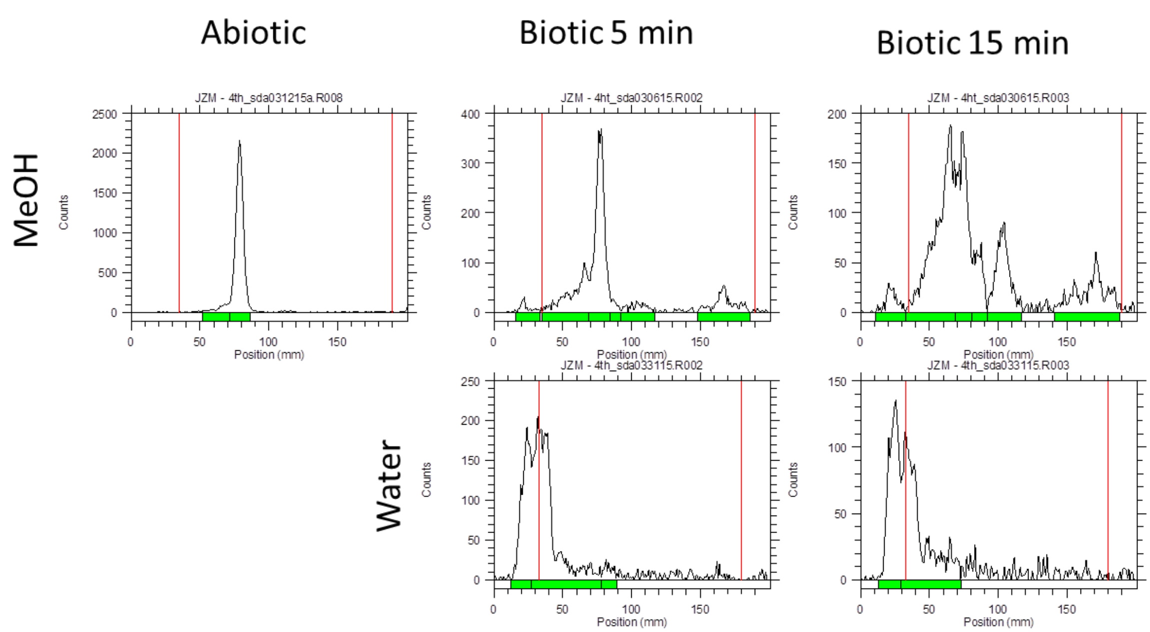

Figure 3.4.8. Representative RAD TLC chromatograms of $C_{12}S$ extracts.

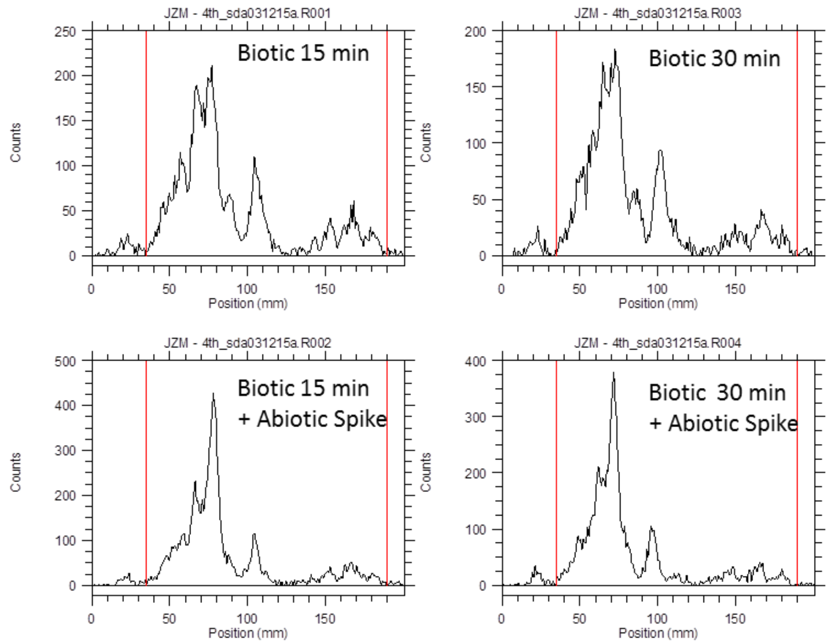

Figure 3.4.9. Alkyl Sulfate-- Parent spike to confirm parent peak.

4. Discussion

4.1 Effect of Structure on Rate of Primary Biotransformation of AE

The calculated rates of primary biotransformation in Phases 1 and 2 show that for AE, the length of the alkyl chain affects the rate of primary biotransformation while the extent of the ethoxylation (EO 1-9) does not. Previous studies have elucidated several different biodegradation pathways for AE[20-24]. In activated sludge at 20 °C, primary biotransformation generally proceeds initially through fission of the central ether with additional biotransformation pathways consisting of oxidation of the alkyl chain or oxidation of the terminal ethoxylate. As the ethoxylation number increases, the prevalence of the central cleavage pathway increases[20]. In anaerobic digestion, biotransformation appears to proceed solely through ethoxylate chain shortening[24].

The chromatograms from Phase 1, shown in Figure 4.1.1, illustrate that in this oxygen limited system the initial step of parent degradation occurs through fission of the central ether to form a polar metabolite (Metabolite IIIs in Figure 4.1.1) with minimal evidence of primary biotransformation through alkyl chain shortening or oxidation of the ethoxylate groups. The incidence of these minor pathways increases with increasing chainlength and decreasing rate of primary degradation.

Chromatographs from early time points from the Phase 2 tests (Figure 4.1.1) illustrate that the preponderance of the central cleavage pathway increases as ethoxylate number increases, while kinetic analysis of the data shows that the rates of primary degradation are similar across test materials. These trends were also seen in activated sludge.[20] The two distinct nonpolar metabolites for $C_{14}E_1$ (V and VI) are hypothesized to be the aldehyde and acid that result from the oxidation of the ethoxylate because of their polarity and the metabolites predicted by the EAWAGG-Pathway Prediction Model (PPS) (see Section 4.3).

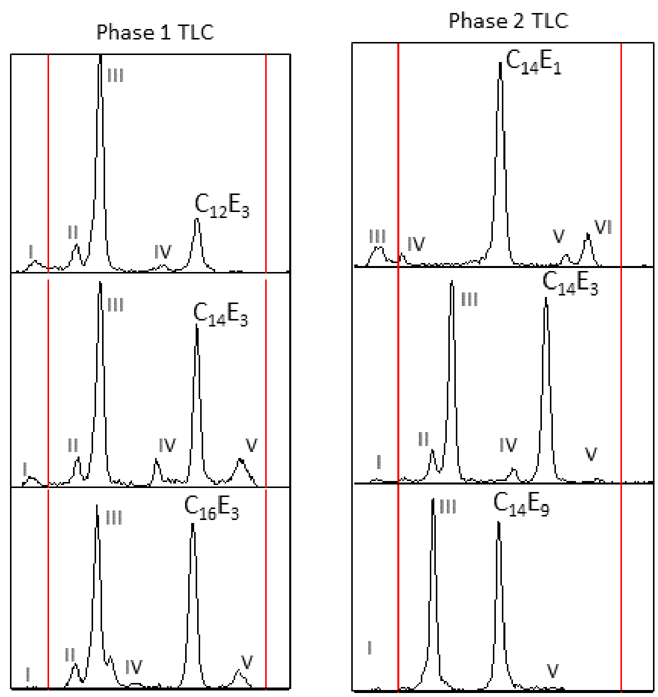

Figure 4.1.1. Early biotic rad-TLC chromatographs from Phases 1 and 2. Parent peaks are identified by test material while metabolite peaks are implicitly identified based on their relative polarities and known biodegradation pathways. They are labeled as a result of their biotransformation. Polar Metabolite III is the resulting polyethylene glycol (PEG) from fission of the central ether that is subsequently degraded to more polar ethoxoylate metabolites I and II. Metabolite IV is hypothesized to be the result of alkyl chain shortening prior to central . Nonpolar Metabolite V, or in the case of $C_{14}E_1$, Metabolites V and VI, is less polar than parent and as such must be the result of oxidation of the ethoxylate chain.

The rate of primary degradation of $C_{14}E_3$ measured in Phase 2 was 4.27 hr^{-1}. This is five times faster than the 0.84 hr^{-1} rate determined in Phase 1 (see Table 3.2.4 and 3.3.4). Additionally, Figure 4.1.2 shows a comparison of the metabolites resulting from the incubation of $C_{14}E_3$ in wastewater collected separately for Phase 1 and Phase 2. Similar metabolite patterns arose, but the metabolites resulting from the minor biotransformation pathways were less prevalent in the faster rate observed in Phase 2. This indicates that the relative abundance or activity of the microbes required for those minor transformation pathways was diminished in comparison to the microbes responsible for central fission.

A comparison of wastewater characteristics between Phase 1 and Phase 2 in Table 3.1.1 reveals that the wastewater used in Phase 2 had a similar suspended solids level, less chemical oxygen demand, less ammonia, and entered the treatment plant at 9 °C cooler. While COD and NH_3 are often used as indicators of strength, it is likely that the lower temperature and correspondingly higher DO during sewer transit allowed for a different community of microbes to develop. This observation implies that factors such as temperature, time of year, diurnal fluctuation, and wastewater composition can influence the rate of these transformations. More work is required to understand the breadth of this variability.

In contrast, Itrich et al. found that the rates of primary degradation in activated sludge were fairly reproducible[20]. This difference can be explained by the variable nature of raw wastewater in contrast to the highly engineered microbial community that is cultured in a wastewater treatment plant.

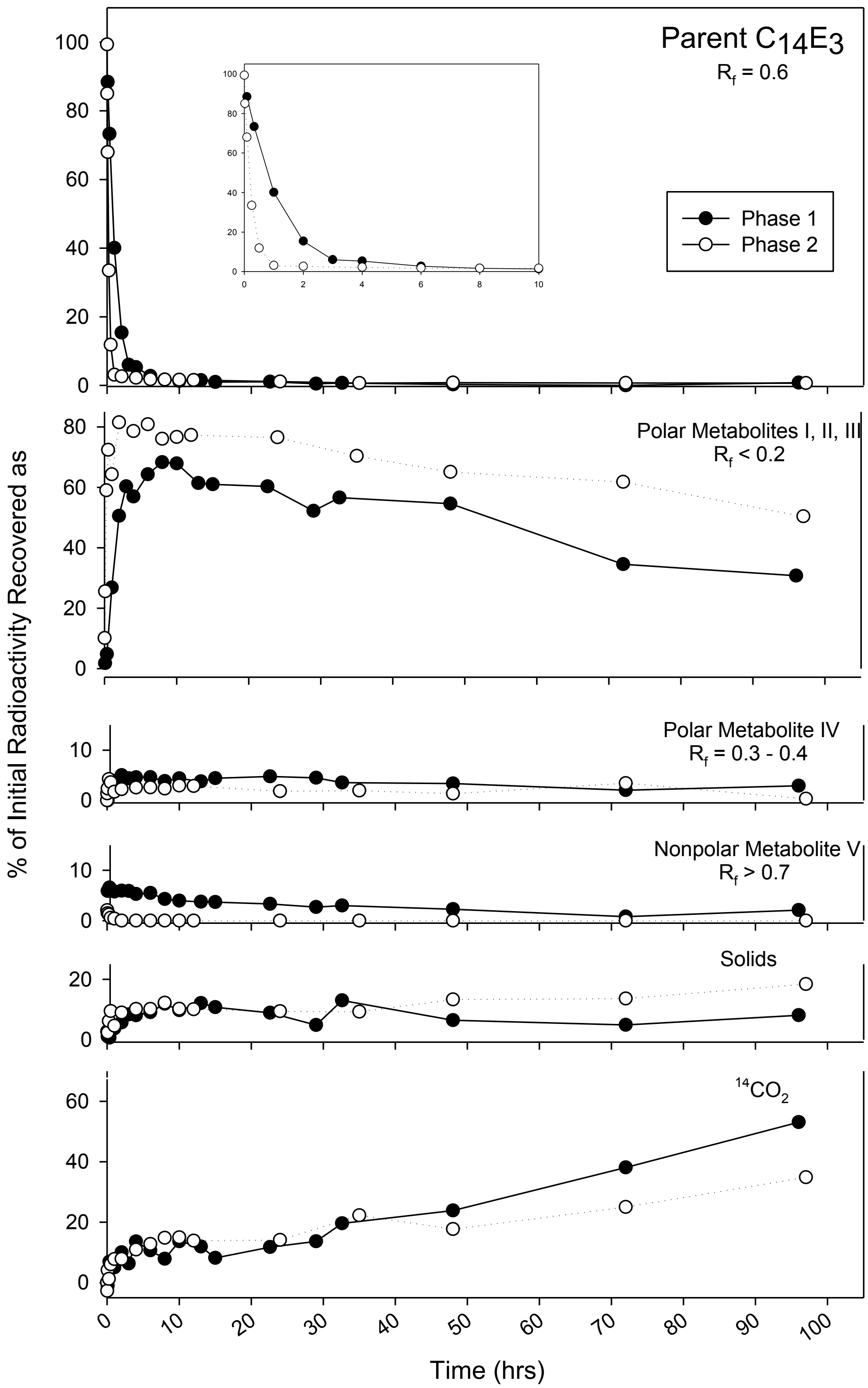

Figure 4.1.2. Phase 1 and 2 comparison distribution of ^{14}C-$C_{14}E_3$ as a function of incubation time in domestic wastewater at 0.5 mg/L DO and 15 °C.

4.2 Biotransformation Pathways of the Anionic Surfactants

The degradation of LAS proceeded after a 12-24 hour lag period (Figure 3.4.1). The metabolite pattern was similar to that seen in activated sludge degradation studies[25] and as such it can be concluded to progress via the previously established pathway of ω and β oxidation of the alkyl chain until the residual, polar sulfonated phenyl carboxylates are attacked at the ring and mineralization occurs[26].

Alkyl sulfate was quickly degraded, with only 46% of the radioactivity remaining as parent at 1 min. (Figure 3.4.4). The low level of radioactivity in the chromatographs reflects the simultaneous rapid mineralization of the radiolabel. The nonpolar peaks are indicative of desulfation that produces a fatty alcohol that is then oxidized through the aldehyde to a carboxylic acid before the two end carbons are cleaved through β-oxidation. This cleavage removes the ^{14}C from the molecule due to its location as the first carbon adjacent to the sulfate. Mineralization of the ^{14}C-acetyl group follows quickly. The visualization of other polar metabolites suggests that simultaneous to the desulfation/ β-oxidation process at one end of the molecule, ω and β oxidation progresses from the other end of the molecule.

The two AES test materials were labeled on either side of the central ether. $C_{15}E_3S$ was labeled in the ethoxylate while $C_{14}E_2S$ was labeled in the first carbon of the alkyl chain. The $C_{15}E_3S$ chromatographs reveal a rapid one to one transformation from parent to a single, polar metabolite (Figures 3.4.7). This polar metabolite slowly progressed to more polar metabolites over time. Comparison to AE extracts showed that the metabolite R_f was distinct from both parent $C_{14}E_3$ and its ethoxylate metabolites (Figure 3.4.7). Thus AES presumably retains its sulfate during primary biotransformation and is cleaved at the central ether. The lack of any minor metabolites indicates cleavage of the central ether as the only primary biotransformation step. Minimal mineralization was detected over the course of the test. However, the sulfated ethoxylate has been shown to degrade in activated sludge and other environmental matrixes[27].

In the $C_{14}E_2S$ chromatographs, the dominant nonpolar peak was presumptively identified as tetradecanol (Figure 3.4.6). Adjacent nonpolar peaks are speculated to be fatty aldehyde and fatty carboxylic acid metabolites based on their relative polarities and the understood pathway of β oxidation. These metabolites were also predicted in the EAWAGG-PPS model (see Section 4.3). Mineralization of the ^{14}C label was rapid. As was observed with AS, the visualization of additional polar metabolites supports the assertion that ω and β oxidation proceeds simultaneously from the other end of the alkyl chain.

4.3 Comparison of Visualized TLC Pathways to EAWAGG-PPS Output

The SMILES structure for each surfactant was entered into the Pathway Prediction System (PPS) in the Biocatalyst/Biodegradation Database run by EAWAGG. The output of the PPS contains all theoretical metabolites based on known biotransformation processes. The PPS output for each of the surfactants contain the metabolites discussed in Sections 4.1 and 4.2. However, the output includes many addition potential metabolites and does not differentiate between likely or known metabolites and unlikely ones in different microbial communities. Thus, while the biotransformation pathways for these surfactants are fairly well understood, the output from the PPS does not differentiate between known or major metabolites and hypothetical ones to highlight these pathways.

For example, the biotransformation rule bt0023 says that a dialiphatic ether will be split into an alcohol and an aldehyde. Thus when this rule is applied to the structure of AE, the hypothetical metabolites of primary biotransformation will include the cleavage of every ether bond in the ethoxylate chain in addition to fission of the central ether. The published biodegradation pathways for AE in activated sludge shows the predominate primary biotransformation step to be fission through the central ether and does not discuss cleavage of any other ether[20,26]. While published pathways include oxidation of the terminal ethoxylate and stepwise cleavage of the ethoxylate chain in anaerobic conditions[24], there is no experimental evidence to support fission of internal ethers as the PPS model predicts.

The situation is similar for AES. The PPS model output includes fission of every ether bond in the molecule. While some microbes can attack internal ethoxylates[22], results in this study show that the dominant primary biotransformation step is fission of the central ether. There is no evidence of any other ether fission occuring.

The SMILES structure for AS was entered into the PPS model. The predicted biotransformation options were: 1) ω and β-oxidation of the alkyl chain and 2) oxidation of the secondary aliphatic carbons along the alkyl chain to secondary alcohols and then to ketones beginning with 2C and proceeding down the chain. Desulfation was not a predicted pathway. TLC chromatographs from this research showed rapid and complex alkyl chain shortening metabolite patterns. One peak on the chromatograph was presumptively identified as tetradecanol. If this identification is accurate, it indicates that desulfation is an additional biotransformation step.

When the SMILES structure for LAS was entered into the PPS model, the predicted preliminary biotransformation metabolites included the products of alkyl chain shortening as well as metabolites resulting from immediate desulfonation and subsequent ring opening. Biodegradation pathways established in the literature begin with alkyl chain shortening and then proceed with digestion of the sulfonated carboxylphenols[25,26,28,29]. LAS is a highly studied molecule and there is no evidence in the literature of desulfonation and ring opening occurring before alkyl chain shortening.

4.4 Comparison to Kinetic Rates in other Environmental Compartments

Kinetic rates of biodegradation are published in the literature for some of the tested surfactants. A comparison between the first-order kinetic rates of the surfactants from this study and published and unpublished rates of similar surfactants in other environmental compartments are provided in Table 4.4.1. The trend in relative rates of primary biotransformation between the different surfactants holds across all three environmental matrices. AS degrades the fastest followed by AE which is slightly faster than AES. LAS degrades an order of magnitude slower than the other surfactants. In both wastewater and activated sludge, there is an inverse relationship between the AE alkyl chainlength and the rate of primary degradation. The extent of AE ethoxylation did not trend with their rates of primary degradation in either environmental compartment for which there are data.

Table 4.4.1. Comparison of first-order kinetic rates in different environmental compartments.

Chemical	Wastewater (hr^{-1})	Activated Sludge (hr^{-1})		River Water (hr^{-1})	
		Rate	Chemical Comparison*	Rate	Chemical Comparison*
$C_{12}E_3$	6.8	69†	$C_{12}E_6$		
$C_{14}E_3$	0.87	71†		1.28^{Φ}	$C_{14}E_3$
$C_{16}E_3$	0.49	18†	$C_{16}E_6$		
$C_{14}E_1$	3.85	62†			
$C_{14}E_3$	4.27	71†		1.28^{Φ}	$C_{14}E_3$
$C_{14}E_9$	3.37	78†			
LAS (C_{12} 2-phenyl)	0.026	6.95‡	C_{12}- LAS	0.059^{Φ}	C_{12}- LAS
$C_{14}E_2S$	3.66	23.4^{Ψ}	$C_{14}E_3S$	0.97^{Φ}	$C_{14}E_3S$
$C_{15}E_3S$	1.89				
AS (C_{12})	40.53	ND		3.17^{Φ}	$C_{14}S$

ND= No data available
*When the data for the specific homolog was not available, a different homolog was used as a comparison.
† Itrich and Federle. (2004) ‡ P&G Unpublished data. Study 51105 $^{\Psi}$ P&G Unpublished data. Study E92-021 $^{\Phi}$ P&G Unpublished data. Study E94-009

4.5 Future Investigation

As with any research project, this work raises as many questions as it answers. The 5x variation in the kinetic rate of $C_{14}E_3$ in two different wastewaters implies that there is a distribution, not a set value, of rates for surfactant degradation in the sewer. This is supported by the wide range of estimated in-sewer removals from monitoring studies. Additional studies should be conducted to investigate the distribution of kinetic rates for each surfactant across a variety of wastewaters, times of day, and seasons. In addition, characterizing the microbial community could help explain differences in kinetic rates throughout the year. Further work could also be done to better characterize and understand the variations in wastewater. Since this study implies that COD does not correlate with microbial activity, other measures of wastewater strength and microbial activity should be considered. Additionally, pairing these laboratory measured rates with a distribution of sewer retention times will enable us to accurately estimate distributions of in-sewer removal to be used in risk assessment. These results can be compared with existing monitoring results and, ideally, lead to field validation.

5. Conclusions

This research shows that organisms in domestic wastewater can degrade surfactants. Data from the OECD 314A was used to calculate rates of primary biodegradation and provided information on biodegradation pathways of AE, AS, LAS, and AES. These surfactants were degraded to more polar metabolites and in many cases were completely mineralized over the course of the four day test. With the exception of LAS, the kinetic rates observed in this study imply that these transformations can happen at a meaningful rate during sewer transit and thus support the monitoring studies that suggest in-sewer removal of the surfactants. The half-life for LAS was 27 hours. The half-lives for AE ranged from 0.1-1.4 hours. The half-life for AS was 0.02 hours and the half-life for AES was 0.2-0.4 hours. These short half-lives imply that with as little as a 2 hour transit time in the sewer, at least 50% of each of these surfactants could be removed prior to wastewater treatment. These rates can be combined with transit data to better evaluate the fate of surfactants during down-the-drain disposal.

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Appendix 1 - Phase 1 Raw Data

Table A1-1. $C_{12}E_3$ Raw Data.

Test Chemical:	^{14}C $C_{12}E_3$
RC info:	RC 95061; omega ethoxylate labeled with ^{14}C; specific activity: 17 µCi/mg
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 °C, 0.5 mg/L DO
Date:	8/12/2014
Test Chem purity:	96.78% by Rad-TLC
Test Chemical Rad-TLC Analysis:	90:10:1 Chloroform:Methanol:formic acid on Analtech Cat # 43011
Test Concentration (mg/L):	0.103

Mineralization		***Abiotic***			***Biotic***			
Sample #	Time (hrs)	Direct	Acidified	% CO2	Direct	Acidified	% CO2	% CO2 From Recovery
		dpm/mL	dpm/mL	%	dpm/mL	dpm/mL	%	%
B1	0.08	3872.00	3864.00	-0.04	3729	3445	10.81	13.87
B2	0.33				4051	3803	1.54	13.47
B3	1	4018	3990	-3.30	3778	3317	14.12	16.63
B4	2				3872	3220	16.64	19.26
B5	3				3787	3116	19.33	23.03
B6	4				3876	3243	16.04	20.71
B7	6	3835	3930	-1.75	3661	3165	18.06	21.79
B8	8	3933	3766	2.50	3790	3274	15.24	27.52
B9	10				3722	3064	20.67	24.96
B10	13	3957	3978	-2.99	3779	3113	19.41	25.03
B11	15				3769	3191	17.39	23.85
B12	22.6				3486	2965	23.24	26.96
B13	29	3846	3807	1.44	3580	2926	24.25	29.43
B14	32.6				3245	2806	27.35	32.41
B15	48	3822	3843	0.51	3580	2786	27.87	36.86
B16	72	3825	3830	0.84	2902	2289	40.74	48.33
B17	96	3851	3755	2.78	2545	1772	54.12	57.73
average		3884	3863	0	3801			
stdev		65	81	2.1	367			

Average RAM Distribution for Graphs

Sample #	Time (hrs)	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.64 parent	>0.7	time (hrs) 0	adjusted parent 99.05	Biotic Metabolites 0.95	Biotic Solids	Recovery from Solids	Total Biotic Recovery	Abiotic Parent	Abiotic Metabolites	Abiotic Solids
B1	0.08	3.69	5.40	12.96	2.52	57.55	2.55	0.08	58.37	27.12	1.45	86.13	96.94	102.83	0.00	0.15
B2	0.33	22.22	10.18	33.57	4.02	9.74	0.00	0.33	9.88	69.98	7.63	86.53	88.07			
B3	1	26.77	12.34	29.03	3.36	9.06	0.92	1.00	9.19	72.42	9.35	83.37	97.49	97.72	0.92	
B4	2	25.82	12.62	33.89	1.89	2.37	0.00	2.00	2.41	74.22	9.17	80.74	97.38			
B5	3	23.39	13.97	28.99	1.78	2.94	0.00	3.00	2.98	68.12	9.90	76.97	96.30			
B6	4	25.22	14.63	32.27	1.82	1.47	0.00	4.00	1.49	73.94	10.50	79.29	95.33			
B7	6	19.37	19.18	29.79	1.67	1.65	0.00	6.00	1.67	70.01	11.36	78.21	96.27			0.21
B8	8	22.47	19.55	24.83	1.64	1.50	0.00	8.00	1.52	68.50	7.60	72.48	87.72	99.34		0.25
B9	10	20.94	21.44	22.94	1.84	1.40	0.00	10.00	1.42	67.16	10.47	75.04	95.72			
B10	13	21.61	22.77	21.41	1.74	1.51	0.16	13.00	1.53	67.70	10.53	74.97	94.38			0.25
B11	15	20.33	24.45	18.78	1.71	1.88	0.18	15.00	1.91	65.45	10.38	76.15	93.54			3.40
B12	22.6	17.91	30.25	12.25	2.83	1.07	0.00	22.60	1.09	63.24	10.44	73.04	96.28			
B13	29	10.31	37.59	7.96	3.23	1.49	0.00	29.00	1.51	59.08	10.00	70.57	94.81	92.70	4.31	0.32
B14	32.6	16.54	31.39	7.79	1.75	2.07	0.00	32.60	2.10	57.46	10.23	67.59	94.94			
B15	48	19.05	26.53	5.64	1.01	1.47	0.00	48.00	1.49	52.23	9.60	63.14	91.02	100.50	0.00	0.37
B16	72	10.22	18.45	2.29	0.80	1.13	0.48	72.00	1.15	32.24	18.92	51.67	92.41	98.90	1.68	0.29
B17	96	12.79	8.75	1.68	1.09	1.39	0.00	96.00	1.41	24.32	18.11	42.27	96.39	99.03	0.00	0.33
												average	94.41	98.72	1.15	0.62
												stdev	2.90	2.87	1.54	0.99

Table A1-1 (continued)

BIOTIC TLC Extracts

REP 1

Sample #	Time (hrs)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery +CO2
B1	0.08	823	129	486	63.92	13.36	1.26	78.54	89.35
B2	0.33	683	246	2034	53.05	25.48	5.27	83.79	85.33
B3	1	637	306	450	49.48	31.69	1.17	82.33	96.45
B4	2	594	289	1375	46.14	29.93	3.56	79.62	96.26
B5	3	546	253	1797	42.41	26.20	4.65	73.26	92.59
B6	4	598	285	863	46.45	29.51	2.23	78.19	94.23
B7	6	574	236	1499	44.58	24.44	3.88	72.90	90.96
B8	8	557	253	435	43.26	26.20	1.13	70.59	85.83
B9	10	502	260	2141	38.99	26.93	5.54	71.46	92.13
B10	13	554	240	1832	43.03	24.85	4.74	72.63	92.03
B11	15	575	214	1707	44.66	22.16	4.42	71.24	88.63
B12	22.6	538	222	1631	41.79	22.99	4.22	69.00	92.24
B13	29	459	230	4090	35.65	23.82	10.59	70.06	94.30
B14	32.6	463	231	2096	35.96	23.92	5.43	65.31	92.66
B15	48	409	214	1905	31.77	22.16	4.93	58.86	86.73
B16	72	224	152	6402	17.40	15.74	16.57	49.71	90.45
B17	96	90	177	4829	6.99	18.33	12.50	37.82	91.95

Relative Abundance in Methanol extacts on TLC Plates

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.64 parent	>0.7
B1	2.48	1.9	17.11	3.72	71.59	3.2
B2	4.93	10.29	61.13	5.58	18.07	
B3	2.48	10.43	48.42	7.19	27.77	3.73
B4	2.24	16.38	73.31	2.77	5.30	
B5	3.05	20.1	65.06	3.1	8.69	
B6	2.4	23.09	68.89	2.6	3.02	
B7	3.31	29.34	61.77	3.24	2.34	
B8	3.22	36.3	53.76	3.72	2.99	
B9	3.58	35.02	54.15	4.11	3.15	
B10	3.47	43.37	47.64	2.63	2.9	
B11	4.4	48.18	41.86	2.81	2.74	
B12	4.09	59.79	28.14	5.91	2.07	
B13	3.9	68.81	20.78	3.47	3.04	
B14	3.43	60.85	21.89	6.63	7.19	
B15	2.88	73.34	15.17	4.21	4.41	
B16	5.89	74.08	6.13	2.4	9.04	2.46
B17	14.25	21.75	30.3	14.8	18.88	

% of Initial Radioactivity

Sample #	origin	Rf 0.02 monoethoxy	0.14 ethoxy+ shoulder	Rf 0.41 oxidation of c-chain	0.64 Parent	0.87 oxidation of ethoxy
B1	1.59	1.21	10.94	2.38	45.76	2.05
B2	2.62	5.46	32.43	2.96	9.59	0.00
B3	1.23	5.16	23.96	3.56	13.74	1.85
B4	1.03	7.56	33.82	1.28	2.45	0.00
B5	1.29	8.52	27.59	1.31	3.69	0.00
B6	1.11	10.72	32.00	1.21	1.40	0.00
B7	1.48	13.08	27.54	1.44	1.04	0.00
B8	1.39	15.70	23.26	1.61	1.29	0.00
B9	1.40	13.65	21.11	1.60	1.23	0.00
B10	1.49	18.66	20.50	1.13	1.25	0.00
B11	1.97	21.52	18.69	1.25	1.22	0.00
B12	1.71	24.98	11.76	2.47	0.86	0.00
B13	1.39	24.53	7.41	1.24	1.08	0.00
B14	1.23	21.88	7.87	2.38	2.59	0.00
B15	0.91	23.30	4.82	1.34	1.40	0.00
B16	1.02	12.89	1.07	0.42	1.57	0.43
B17	1.00	1.52	2.12	1.03	1.32	0.00

BIOTIC TLC Extracts

REP 2

Sample #	Time (hrs)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery +CO2
B1	0.08	928	193	638	72.08	19.99	1.65	93.72	104.53
B2	0.33	714	230	3859	55.46	23.82	9.99	89.26	90.81
B3	1	645	162	6774	50.10	16.78	17.54	84.41	98.53
B4	2	633	173	5709	49.16	17.92	14.78	81.86	98.50
B5	3	609	176	5851	47.30	18.23	15.15	80.67	100.00
B6	4	584	157	7247	45.36	16.26	18.76	80.38	96.42
B7	6	642	143	7278	49.86	14.81	18.84	83.51	101.57
B8	8	571	154	5436	44.35	15.95	14.07	74.37	89.61
B9	10	570	183	5949	44.27	18.95	15.40	78.62	99.30
B10	13	588	148	6304	45.67	15.33	16.32	77.32	96.72
B11	15	588	184	6310	45.67	19.05	16.34	81.06	98.45
B12	22.6	526	189	6432	40.85	19.57	16.65	77.08	100.32
B13	29	358	327	3634	27.81	33.86	9.41	71.08	95.32
B14	32.6	454	189	5805	35.26	19.57	15.03	69.86	97.22
B15	48	403	211	5514	31.30	21.85	14.28	67.43	95.30
B16	72	230	140	8216	17.86	14.50	21.27	53.63	94.37
B17	96	100	147	9164	7.77	15.22	23.73	46.72	100.84
				average	41.05	21.45	10.33		94.41
				stdev	13.46	5.22	6.62		4.63

Relative Abundance in Methanol extacts on TLC Plates

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.64 parent	>0.7
B1	1.42	1.94	15.98	3.68	74.37	2.61
B2	6.01	12.16	60.6	6.8	14.42	
B3	4.08	14.94	68.07	6.31	6.6	
B4	4.15	13	69.07	5.1	4.68	
B5	4	22.41	64.24	4.73	4.62	
B6	3.82	25.43	61.96	5.38	3.38	
B7	5.48	30.93	55.27	3.79	4.53	
B8	6.36	35.07	50.93	3.78	3.86	
B9	4.21	38.33	49.24	4.68	3.55	
B10	4.18	43.62	42.45	5.16	3.87	0.72
B11	6	47.07	35.87	4.74	5.56	0.77
B12	4.04	58.22	26.81	7.79	3.13	
B13	2.94	46.98	24.48	18.77	6.83	
B14	6.4	67.07	18.96	3.15	4.41	
B15	6.58	71.58	14.81	2.15	4.89	
B16	11.84	68.86	5.89	6.59	3.84	2.99
B17	16.6	42.44	7.42	14.85	18.69	

% of Initial Radioactivity

Sample #	origin	Rf 0.02 xy	0.14 ethoxy+	Rf 0.41 of c-chain	0.64 Parent	0.87 ethoxy
B1	1.02	1.40	11.52	2.65	53.60	1.88
B2	3.33	6.74	33.61	3.77	8.00	0.00
B3	2.04	7.48	34.10	3.16	3.31	0.00
B4	2.04	6.39	33.96	2.51	2.30	0.00
B5	1.89	10.60	30.39	2.24	2.19	0.00
B6	1.73	11.53	28.10	2.44	1.53	0.00
B7	2.73	15.42	27.56	1.89	2.26	0.00
B8	2.82	15.55	22.59	1.68	1.71	0.00
B9	1.86	16.97	21.80	2.07	1.57	0.00
B10	1.91	19.92	19.39	2.36	1.77	0.33
B11	2.74	21.50	16.38	2.16	2.54	0.35
B12	1.65	23.79	10.95	3.18	1.28	0.00
B13	0.82	13.06	6.81	5.22	1.90	0.00
B14	2.26	23.65	6.69	1.11	1.56	0.00
B15	2.06	22.40	4.64	0.67	1.53	0.00
B16	2.12	12.30	1.05	1.18	0.69	0.53
B17	1.29	3.30	0.58	1.15	1.45	0.00

BIOTIC TLC Extracts

REP 1 — **Relative Abundance in Water extacts on TLC Plates** / **% of Initial Radioactivity**

Sample #	Time (hrs)	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.64 parent	>0.7	origin	Rf 0.02 monoethoxy	0.14 shoulder	Rf 0.41 chain	0.64 Parent	0.87 ethoxy
B1	0.08	26.9	51.82	6.1		15.18		3.59	6.92	0.81	0.00	2.03	0.00
B2	0.33	75.53	16.02	2.16	2.56	3.73		19.24	4.08	0.55	0.65	0.95	0.00
B3	1	79.31	18.99	0		1.7		25.13	6.02	0.00	0.00	0.54	0.00
B4	2	81.14	18.86	0				24.28	5.64	0.00	0.00	0.00	0.00
B5	3	83.19	16.81	0				21.80	4.40	0.00	0.00	0.00	0.00
B6	4	80.63	11.85	7.53				23.80	3.50	2.22	0.00	0.00	0.00
B7	6	70.65	20.16	9.19				17.27	4.93	2.25	0.00	0.00	0.00
B8	8	77.72	14.98	7.3				20.36	3.92	1.91	0.00	0.00	0.00
B9	10	71.73	22.76	5.51				19.31	6.13	1.48	0.00	0.00	0.00
B10	13	80.1	14	5.9				19.91	3.48	1.47	0.00	0.00	0.00
B11	15	81.11	13.3	5.6				17.98	2.95	1.24	0.00	0.00	0.00
B12	22.6	70.6	25.53	3.89				16.23	5.87	0.89	0.00	0.00	0.00
B13	29	57.51	35.36	7.13				13.70	8.42	1.70	0.00	0.00	0.00
B14	32.6	61.83	36.03	2.14				14.79	8.62	0.51	0.00	0.00	0.00
B15	48	79.27	16.6	4.13				17.57	3.68	0.92	0.00	0.00	0.00
B16	72	54.96	37.23	7.81				8.65	5.86	1.23	0.00	0.00	0.00
B17	96	63.55	34.62	1.83				11.65	6.35	0.34	0.00	0.00	0.00

BIOTIC TLC Extracts

REP 2 — **Relative Abundance in Water extacts on TLC Plates** / **% of Initial Radioactivity**

Sample #	Time (hrs)	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.64 parent	>0.7	origin	Rf 0.02 monoethoxy	0.14 shoulder	Rf 0.41 chain	0.64 Parent	0.87 ethoxy
B1	0.08	5.86	6.32	13.31		68.61	5.9	1.17	1.26	2.66	0.00	13.71	1.18
B2	0.33												
B3	1												
B4	2												
B5	3												
B6	4												
B7	6												
B8	8												
B9	10												
B10	13												
B11	15												
B12	22.6												
B13	29	13.9	86.1					4.71	29.16	0.00	0.00	0.00	0.00
B14	32.6												
B15	48												
B16	72												
B17	96												

Table A1-1 (continued)

Average MEOH Extract Disribution / **Average Water Extract Disribution**

Sample #	Time (hrs)	MeOH: origin	MeOH: Rf <0.1	MeOH: 0.20	MeOH: Rf 0.3-0.4	MeOH: 0.64 parent	MeOH: >0.7	Water: origin	Water: Rf <0.1	Water: 0.20	Water: Rf 0.3-0.4	Water: 0.64 parent	Water: >0.7
B1	0.08	1.30	1.31	11.23	2.52	49.68	1.96	2.38	4.09	1.74	0.00	7.87	0.59
B2	0.33	2.97	6.10	33.02	3.37	8.79	0.00	19.24	4.08	0.55	0.65	0.95	0.00
B3	1	1.64	6.32	29.03	3.36	8.52	0.92	25.13	6.02	0.00	0.00	0.54	0.00
B4	2	1.54	6.97	33.89	1.89	2.37	0.00	24.28	5.64	0.00	0.00	0.00	0.00
B5	3	1.59	9.56	28.99	1.78	2.94	0.00	21.80	4.40	0.00	0.00	0.00	0.00
B6	4	1.42	11.13	30.05	1.82	1.47	0.00	23.80	3.50	2.22	0.00	0.00	0.00
B7	6	2.10	14.25	27.55	1.67	1.65	0.00	17.27	4.93	2.25	0.00	0.00	0.00
B8	8	2.11	15.63	22.92	1.64	1.50	0.00	20.36	3.92	1.91	0.00	0.00	0.00
B9	10	1.63	15.31	21.46	1.84	1.40	0.00	19.31	6.13	1.48	0.00	0.00	0.00
B10	13	1.70	19.29	19.94	1.74	1.51	0.16	19.91	3.48	1.47	0.00	0.00	0.00
B11	15	2.35	21.51	17.54	1.71	1.88	0.18	17.98	2.95	1.24	0.00	0.00	0.00
B12	22.6	1.68	24.38	11.36	2.83	1.07	0.00	16.23	5.87	0.89	0.00	0.00	0.00
B13	29	1.10	18.80	7.11	3.23	1.49	0.00	9.20	18.79	0.85	0.00	0.00	0.00
B14	32.6	1.75	22.77	7.28	1.75	2.07	0.00	14.79	8.62	0.51	0.00	0.00	0.00
B15	48	1.49	22.85	4.73	1.01	1.47	0.00	17.57	3.68	0.92	0.00	0.00	0.00
B16	72	1.57	12.59	1.06	0.80	1.13	0.48	8.65	5.86	1.23	0.00	0.00	0.00
B17	96	1.14	2.41	1.35	1.09	1.39	0.00	11.65	6.35	0.34	0.00	0.00	0.00

ABIOTIC TLC Extracts:

REP 1

Sample #	Time (hrs)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.08333	1330	5	22	103.30		0.06	103.36
A2	0.33333							
A3	1	1232	12		95.69	1.24		96.93
A4	2							
A5	3							
A6	4							
A7	6	1271		85	98.72		0.22	98.94
A8	8	1314	4	68	102.06		0.18	102.23
A9	10							
A10	13	1292		85	100.35		0.22	100.57
A11	15	1261	8	2532	97.94		6.56	104.50
A12	22.6							
A13	29	1249	9	137	97.01	0.93	0.35	98.30
A14	32.6							
A15	48	1294	11	144	100.50		0.37	100.88
A16	72	1232		106	95.69		0.27	95.96
A17	96	1163		143	90.33		0.37	90.70

Sample #	Relative Abundance in MeOH Extacts: origin	Rf 0.02 y	0.64 Parent	% of Initial Radioactivity: origin	Rf 0.02 monoethoxy	0.64 Parent
A1						
A2						
A3			100	0.00	0.00	95.69
A4						
A5						
A6						
A7	47.58	2.85	49.58	46.97	2.81	48.94
A8						
A9						
A10	22.29	1.24	76.47	22.37	1.24	76.74
A11						
A12						
A13	4.44		95.56	4.31	0.00	92.70
A14						
A15			100			100.50
A16						
A17						

ABIOTIC TLC Extracts:

REP 2

Sample #	Time (hrs)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.08333	1324	3	92	102.83	0.31	0.24	103.38
A2	0.33333							
A3	1	1308	11		101.59	1.14		102.73
A4	2							
A5	3							
A6	4							
A7	6	1335	10	80	103.69	1.04	0.21	104.93
A8	8	1279	3	125	99.34	0.31	0.32	99.97
A9	10							
A10	13	1297	13	109	100.74	1.35	0.28	102.36
A11	15	1331	13	98	103.38	1.35	0.25	104.98
A12	22.6							
A13	29	1252	9	114	97.24	0.93	0.30	98.47
A14	32.6							
A15	48	1298	15	142	100.81	1.55	0.37	102.74
A16	72	1295	11	118	100.58	1.14	0.31	102.03
A17	96	1275	0	114	99.03	0.00	0.30	99.32
				average	99.54	0.94	0.62	100.66
				stdev	3.19	0.46	1.44	3.41

Sample #	Relative Abundance in MeOH Extacts: origin	Rf 0.02 monoethoxy	0.64 Parent	% of Initial Radioactivity: origin	Rf 0.02 monoethoxy	0.64 Parent
A1			100	0.00	0.00	102.83
A2						
A3		1.81	98.19	0.00	1.84	99.75
A4						
A5						
A6						
A7						
A8			100			99.34
A9						
A10						
A11						
A12						
A13	63.78		36.22	62.02	0.00	35.22
A14						
A15						
A16		1.67	98.33	0.00	1.68	98.90
A17			100			99.03
average						98.59
stdev						3.09

Table A1-2. $C_{14}E_3$ Raw Data.

Test Chemical:	^{14}C $C_{14}E_3$
RC info:	RC 95062; omega ethoxylate labeled with 14C; specific activity: 13.7 µCi/mg
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 °C, 0.5 mg/L DO
Date:	8/12/2014
Test Chem purity:	91.94% by Rad-TLC
Test Chemical Rad-TLC Analysis:	90:10:1 Chloroform:Methanol:formic acid on Analtech Cat# 43011
Test Concentration (mg/L):	0.121

Mineralization		***Abiotic***			***Biotic***			
Sample #	Time (hrs)	Direct dpm/mL	Acidified dpm/mL	% CO2 %	Direct dpm/mL	Acidified dpm/mL	% CO2 %	% CO2 From Recovery %
B1	0.08	4033	3983	0.78	4056	4058	-1.09	3.97
B2	0.33				4068	3739	6.86	10.88
B3	1	4034	4046	-0.79	3990	3815	4.96	20.98
B4	2				4011	3614	9.97	17.84
B5	3				4041	3762	6.28	14.77
B6	4				3885	3469	13.58	17.27
B7	6	3998	4073	-1.46	3762	3587	10.64	15.73
B8	8	4001	4051	-0.92	3965	3700	7.83	14.87
B9	10				3962	3467	13.63	15.77
B10	13	4039	4047	-0.82	3870	3538	11.86	17.95
B11	15				4044	3688	8.13	19.46
B12	22.6				3773	3544	11.71	22.62
B13	29	4008	4060	-1.14	3827	3468	13.61	27.10
B14	32.6				3515	3228	19.59	23.44
B15	48	4074	4131	-2.91	3576	3057	23.85	34.76
B16	72	3810	3834	4.49	3122	2485	38.10	43.37
B17	96	3890	3903	2.77	2575	1883	53.09	57.31
avg		3987	4014	0.0	3943			
stdev		79	87	2.2	382			

Distribution of Radioactivity for Graphs

Sample #	Time (hrs)	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.68 parent	>0.7	Adjusted parent 90.71	Biotic Metabolites	Biotic Solids	Abiotic Parent	Abiotic Metabolites	Abiotic Solids
B1	0.08	0.00	0.00	1.81	0.00	86.44	5.86	91.01	7.67	1.03	86.00	11.01	0.23
B2	0.33	0.00	0.00	4.81	1.67	71.67	6.57	75.46	13.05	0.63			
B3	1	9.11	3.08	14.64	3.81	38.04	5.73	40.05	36.38	3.66	85.58	9.33	0.44
B4	2	22.24	4.60	23.71	5.02	14.62	5.92	15.40	61.49	5.66			
B5	3	26.99	6.01	27.32	4.37	5.72	5.87	6.02	70.56	8.35			
B6	4	18.45	18.56	19.95	4.61	5.06	5.26	5.32	66.83	8.01			
B7	6	28.13	12.84	23.34	4.65	2.60	5.47	2.74	74.42	9.05			1.69
B8	8	27.77	18.49	22.06	3.84	1.59	4.26	1.68	76.42	11.85			0.21
B9	10	27.04	20.66	20.24	4.38	1.24	3.94	1.31	76.26	9.72			
B10	13	18.42	29.14	13.85	3.77	1.43	3.73	1.50	68.92	12.10	81.52	14.07	0.21
B11	15	23.23	24.63	13.19	4.40	0.94	3.67	0.99	69.11	10.74	89.46	5.01	0.36
B12	22.6	25.70	24.76	9.84	4.77	1.01	3.31	1.07	68.39	8.87			
B13	29	19.72	27.57	4.92	4.51	0.49	2.70	0.52	59.42	4.81	84.42	7.21	0.64
B14	32.6	21.30	29.34	5.99	3.53	0.68	2.99	0.71	63.15	12.98	91.12	8.96	0.40
B15	48	16.99	33.87	3.76	3.35	0.25	2.28	0.27	60.24	6.44			0.05
B16	72	12.30	20.70	1.57	2.02	0.00	0.80	0.00	37.38	4.89			2.35
B17	96	16.08	13.58	1.14	2.91	0.76	2.09	0.81	35.79	8.07	85.01	9.83	0.58
										avg	86.16	9.34	0.65
										stdev	2.97	2.64	0.68

Table A1-2 (continued)

BIOTIC TLC Extracts

REP 1

Sample #	Time (days)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Extract 3 MeOH 0.25ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	Extract 3 Recovery	% on Solids	Total Extract Recovery	Total Recovery +CO2
B1	0.083	1185	41		669	88.56	4.09		1.67	94.31	93.22
B2	0.333	1102	69		82	82.36	6.88		0.20	89.44	96.29
B3	1	841	122		1235	62.85	12.16		3.08	78.08	83.05
B4	2	705	235		1517	52.69	23.42		3.78	79.88	89.85
B5	3	627	281		3136	46.86	28.00		7.81	82.67	88.95
B6	4	599	320		2615	44.77	31.89		6.51	83.17	96.75
B7	6	625	312		2510	46.71	31.09		6.25	84.05	94.69
B8	8	553	352		3785	41.33	35.08		9.43	85.83	93.66
B9	10	580	339		2900	43.35	33.78		7.22	84.35	97.98
B10	13	533	301		3489	39.83	29.99		8.69	78.52	90.38
B11	15	554	301		3677	41.40	29.99		9.16	80.56	88.68
B12	22.6	514	293		2634	38.41	29.20		6.56	74.17	85.89
B13	29	513	269	37	2904	38.34	26.80	1.84	7.23	74.22	87.83
B14	32.6	501	270		2830	37.44	26.90		7.05	71.40	90.98
B15	48	463	255		896	34.60	25.41		2.23	62.24	86.09
B16	72	225	191	238	2635	16.82	19.03	11.86	6.56	54.27	92.36
B17	96	199	218		1194	14.87	21.72		2.97	39.57	92.66

TLC Relative Abundance in Methanol Extract on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.68 parent	>0.7	origin	Rf <0.1	0.13	Rf 0.3-0.4	0.59 parent	0.80
B1			2.1		89.98	7.92	0.00	0.00	1.86	0.00	79.69	7.01
B2			5.19	1.2	85.01	8.61	0.00	0.00	4.27	0.99	70.01	7.09
B3		2.19	21.49	6.22	60.64	9.46	0.00	1.38	13.51	3.91	38.11	5.95
B4	2.85	4.62	45.44	9.25	28.03	9.81	1.50	2.43	23.94	4.87	14.77	5.17
B5	3.53	9.24	54.65	9.17	12.42	10.98	1.65	4.33	25.61	4.30	5.82	5.15
B6	6.03	8.91	34.65	12.38	27.77	10.26	2.70	3.99	15.51	5.54	12.43	4.59
B7	5.34	16.05	49.95	11.74	5.57	11.36	2.49	7.50	23.33	5.48	2.60	5.31
B8	3.04	23.78	50.85	8.6	4.41	9.32	1.26	9.83	21.02	3.55	1.82	3.85
B9	3.77	29.62	45.42	9.37	3.41	8.4	1.63	12.84	19.69	4.06	1.48	3.64
B10	4.55	35.22	38.63	9.1	3.12	9.39	1.81	14.03	15.39	3.62	1.24	3.74
B11	4.63	38.31	34.07	11.71	2.48	8.79	1.92	15.86	14.11	4.85	1.03	3.64
B12	7.23	48.93	21.22	12.41	3.62	6.58	2.78	18.80	8.15	4.77	1.39	2.53
B13	9.07	59.69	15.51	7.85		7.88	3.48	22.88	5.95	3.01	0.00	3.02
B14	4.98	60.53	16.33	9.03	1.43	7.69	1.86	22.66	6.11	3.38	0.54	2.88
B15	4.33	66.73	10.68	9.72	1.46	7.08	1.50	23.09	3.70	3.36	0.51	2.45
B16	5.81	80.44	5.71	8.05			0.98	13.53	0.96	1.35	0.00	0.00
B17	7.86	45.36	9.88	17.64	4.94	14.33	1.17	6.75	1.47	2.62	0.73	2.13

BIOTIC TLC Extracts

REP 2

Sample #	Time (days)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Extract 3 MeOH 0.25ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	Extract 3 Recovery	% on Solids	Total Recovery	Total Recovery +CO2
B1	0.083	1268	26		156	94.76	2.59		0.39	97.74	96.65
B2	0.333	1110	48		426	82.96	4.78		1.06	88.80	95.66
B3	1	825	141		1703	61.66	14.05		4.24	79.95	84.91
B4	2	705	243		3026	52.69	24.21		7.54	84.44	94.41
B5	3	665	293		3567	49.70	29.20		8.89	87.78	94.06
B6	4	594	285		3816	44.39	28.40		9.51	82.30	95.88
B7	6	604	276		4756	45.14	27.50		11.85	84.49	95.13
B8	8	596	257		5731	44.54	25.61		14.28	84.43	92.26
B9	10	590	279		4900	44.09	27.80		12.21	84.10	97.73
B10	13	547	293		6225	40.88	29.20		15.51	85.58	97.45
B11	15	518	296		4948	38.71	29.50		12.33	80.53	88.66
B12	22.6	562	275		4490	42.00	27.40		11.19	80.59	92.30
B13	29	373	384.5	52	956	27.88	38.31	3.00	2.38	71.57	85.18
B14	32.6	487	265		7592	36.40	26.41		18.91	81.71	101.30
B15	48	476	221		4275	35.57	22.02		10.65	68.24	92.09
B16	72	266	168	119	1293	19.88	16.74	19.15	3.22	59.00	97.09
B17	96	198	179		5288	14.80	17.84		13.17	45.81	98.90
Avg										Avg	92.62
Stdev										Stdev	4.46

Relative Abundance in Methanol Extract on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.68 parent	>0.7	origin	Rf <0.1	0.13	Rf 0.3-0.4	0.59 parent	0.80
B1			1.86		93.17	4.97	0.00	0.00	1.76	0.00	88.29	4.71
B2			6.45	2.83	83.43	7.29	0.00	0.00	5.35	2.35	69.21	6.05
B3	1.74	2.95	21.7	6.02	58.65	8.95	1.07	1.82	13.38	3.71	36.16	5.52
B4	2.79	7.54	39.75	9.8	27.48	12.66	1.47	3.97	20.94	5.16	14.48	6.67
B5	2.81	12.17	51.48	8.94	11.31	13.29	1.40	6.05	25.58	4.44	5.62	6.60
B6	3.54	15.68	51.83	8.27	7.33	13.36	1.57	6.96	23.01	3.67	3.25	5.93
B7	5	22.7	45.64	8.44	5.76	12.46	2.26	10.25	20.60	3.81	2.60	5.62
B8	4.59	29.21	43.43	9.25	3.07	10.49	2.04	13.01	19.34	4.12	1.37	4.67
B9	4.19	33.59	39.68	10.65	2.28	9.61	1.85	14.81	17.50	4.70	1.01	4.24
B10	4.62	42.61	30.13	9.56	3.95	9.12	1.89	17.42	12.32	3.91	1.61	3.73
B11	4.22	42.12	31.68	10.19	2.22	9.56	1.63	16.31	12.26	3.94	0.86	3.70
B12	4.34	51.46	21.56	11.38	1.52	9.74	1.82	21.61	9.06	4.78	0.64	4.09
B13	5.81	46.62	13.94	21.56	3.55	8.53	1.62	13.00	3.89	6.01	0.99	2.38
B14	3.56	59.45	16.1	10.12	2.25	8.53	1.30	21.64	5.86	3.68	0.82	3.10
B15	4.52	69.45	10.75	9.36		5.91	1.61	24.71	3.82	3.33	0.00	2.10
B16	6.43	61.07	10.97	13.5		8.03	1.28	12.14	2.18	2.68	0.00	1.60
B17	9.93	43.81	5.49	21.56	5.37	13.83	1.47	6.48	0.81	3.19	0.79	2.05

Relative Abundance in Water Extract / **% of Initial Radioactivity**

Sample #	Time (days)	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.68 parent	origin	Rf <0.1	0.13	Rf 0.3-0.4	0.59 parent
B1	0.083					60	0.00	0.00	0.00	0.00	2.45
B2	0.333					30	0.00	0.00	0.00	0.00	2.06
B3	1	70.53	12.22	9.86		7.39	8.57	1.49	1.20	0.00	0.90
B4	2	88.62	5.97	5.42			20.75	1.40	1.27	0.00	0.00
B5	3	90.94	2.92	6.15			25.46	0.82	1.72	0.00	0.00
B6	4	51.16	41.03	2.17		5.65	16.31	13.08	0.69	0.00	1.80
B7	6	82.84	12.76	4.41			25.75	3.97	1.37	0.00	0.00
B8	8	74.46	20.17	5.37			26.12	7.07	1.88	0.00	0.00
B9	10	74.89	20.22	4.89			25.30	6.83	1.65	0.00	0.00
B10	13	55.26	44.74				16.57	13.42	0.00	0.00	0.00
B11	15	71.53	28.48				21.45	8.54	0.00	0.00	0.00
B12	22.6	80.15	15.6	4.25			23.40	4.55	1.24	0.00	0.00
B13	29	64.06	35.94				17.17	9.63	0.00	0.00	0.00
B14	32.6	73.28	26.72				19.72	7.19	0.00	0.00	0.00
B15	48	60.75	39.25				15.44	9.97	0.00	0.00	0.00
B16	72	58.68	41.32				11.17	7.86	0.00	0.00	0.00
B17	96	67.93	32.07				14.76	6.97	0.00	0.00	0.00

Table A1-2 (continued)

		Methanol Extract Average RAM Distribution						Water Extract Average RAM Distribution				
Sample #	Time (days)	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.68 parent	>0.7	origin	Rf <0.1	0.13	Rf 0.3-0.4	0.59 parent
B1	0.083	0.00	0.00	1.81	0.00	83.99	5.86	0.00	0.00	0.00	0.00	2.45
B2	0.333	0.00	0.00	4.81	1.67	69.61	6.57	0.00	0.00	0.00	0.00	2.06
B3	1	0.54	1.60	13.44	3.81	37.14	5.73	8.57	1.49	1.20	0.00	0.90
B4	2	1.49	3.20	22.44	5.02	14.62	5.92	20.75	1.40	1.27	0.00	0.00
B5	3	1.53	5.19	25.60	4.37	5.72	5.87	25.46	0.82	1.72	0.00	0.00
B6	4	2.14	5.47	19.26	4.61	3.25	5.26	16.31	13.08	0.69	0.00	1.80
B7	6	2.38	8.87	21.97	4.65	2.60	5.47	25.75	3.97	1.37	0.00	0.00
B8	8	1.65	11.42	20.18	3.84	1.59	4.26	26.12	7.07	1.88	0.00	0.00
B9	10	1.74	13.82	18.59	4.38	1.24	3.94	25.30	6.83	1.65	0.00	0.00
B10	13	1.85	15.72	13.85	3.77	1.43	3.73	16.57	13.42	0.00	0.00	0.00
B11	15	1.78	16.08	13.19	4.40	0.94	3.67	21.45	8.54	0.00	0.00	0.00
B12	22.6	2.30	20.20	8.60	4.77	1.01	3.31	23.40	4.55	1.24	0.00	0.00
B13	29	2.55	17.94	4.92	4.51	0.49	2.70	17.17	9.63	0.00	0.00	0.00
B14	32.6	1.58	22.15	5.99	3.53	0.68	2.99	19.72	7.19	0.00	0.00	0.00
B15	48	1.55	23.90	3.76	3.35	0.25	2.28	15.44	9.97	0.00	0.00	0.00
B16	72	1.13	12.83	1.57	2.02	0.00	0.80	11.17	7.86	0.00	0.00	0.00
B17	96	1.32	6.61	1.14	2.91	0.76	2.09	14.76	6.97	0.00	0.00	0.00

ABIOTIC TLC Extracts:
REP 1

									Relative Abundance in Methanol Extract					% of Initial Radioactivity				
Sample #	Time (days)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	origin	Rf <0.1	0.13	0.59 parent	0.80	origin	Rf <0.1	0.13	0.59 parent	0.80
A1	0.083	1298	7	131	97.01		0.33	97.33	5.92		1.21	88.65	4.22	5.74	0.00	1.17	86.00	4.09
A2	0.333																	
A3	1	1270	6	177	94.91	0.60	0.44	95.95	4.45		1.24	90.17	4.14	4.22	0.00	1.18	85.58	3.93
A4	2																	
A5	3																	
A6	4																	
A7	6	1251	7.5	1254	93.49	0.75	3.12	97.36	13.69	5.56	1.36	74.89	4.23					
A8	8	1255	6	64	93.79	0.60	0.16	94.55										
A9	10																	
A10	13	1279	10	52	95.59	1.00	0.13	96.71	9.36		1.6	85.28	3.76	8.95	0.00	1.53	81.52	3.59
A11	15	1264	6.5	167	94.46	0.65	0.42	95.53			1.49	94.7	3.81	0.00	0.00	1.41	89.46	3.60
A12	22.6																	
A13	29	1226		251	91.62		0.63	92.25	2.14		1.96	92.14	3.77	1.96	0.00	1.80	84.42	3.45
A14	32.6																	
A15	48																	
A16	72	1192		793	89.08		1.98	91.06										
A17	96	1203	10.5	269	89.91	1.05	0.67	91.62										

ABIOTIC TLC Extracts:
REP 2

									Relative Abundance in Methanol Extract					% of Initial Radioactivity				
Sample #	Time (days)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	origin	Rf <0.1	0.13	0.59 parent	0.80	origin	Rf <0.1	0.13	0.59 parent	0.80
A1	0.083	1305	3	53	97.53		0.13	97.66										
A2	0.333																	
A3	1	1294	11	174	96.71	1.10	0.43	98.24										
A4	2																	
A5	3																	
A6	4																	
A7	6	1328	6	104	99.25		0.26	99.51										
A8	8	1333	14	104	99.62		0.26	99.88										
A9	10																	
A10	13	1342	22	116	100.29		0.29	100.58										
A11	15	1231	12	123	92.00		0.31	92.30										
A12	22.6																	
A13	29	1286	15	264	96.11	1.49	0.66	98.26										
A14	32.6	1339	18	161	100.07		0.40	100.47	4.83		1.36	91.06	2.76	4.83	0.00	1.36	91.12	2.76
A15	48	1345	16	20	100.52		0.05	100.57										
A16	72	1108		1096	82.81		2.73	85.54										
A17	96	1269	20	200	94.84		0.50	95.34	4.18		2.15	89.64	4.03	3.96	0.00	2.04	85.01	3.82
Average					94.98	0.90	0.69	96.04										
Std Dev					4.32	0.29	0.84	3.81										

Table A1-3. $C_{16}E_3$ Raw Data.

Test Chemical:	^{14}C $C_{16}E_3$
RC info:	RC 95063; omega ethoxylate labeled with 14C; specific activity: 14 µCi/mg
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 °C, 0.5 mg/L DO
Date:	8/12/2014
Test Chem purity:	92.43% by Rad-TLC
Test Chemical Rad-TLC Analysis:	90:10:1 Chloroform:Methanol:formic acid on Analtech Cat# 43011
Test Concentration (mg/L):	0.165

Mineralization		***Abiotic***			***Biotic***				
Sample #	Time (hrs)	Direct dpm/mL	Acidified dpm/mL	% CO2 %	Direct dpm/mL	Acidified dpm/mL	% CO2 %	% CO2 from Recovery %	
B1	0.083	4531	4581	11.21	4894	5196	-0.71	12.93	0.083
B2	0.333				5563	5207	-0.92	0.09	0.333
B3	1	4952	5378	-4.23	5521	5384	-4.35	4.39	1
B4	2				5609	5458	-5.78	3.48	2
B5	3				5745	5401	-4.68	6.83	3
B6	4				5558	5204	-0.86	8.06	4
B7	6	5195	5035	2.41	5532	5102	1.12	5.58	6
B8	8	6057	5944	-15.20	5834	5311	-2.94	8.19	8
B9	10				5500	5129	0.59	8.30	10
B10	13	5217	5406	-4.78	5602	5302	-2.76	10.33	13
B11	15				5680	5273	-2.20	12.18	15
B12	22.6				5527	5151	0.17	12.59	22.6
B13	29	5167	5044	2.24	5473	5005	3.00	12.50	29
B14	32.6				5202	4854	5.92	13.63	32.6
B15	48	5244	5243	-1.62	5232	4557	11.68	18.12	48
B16	72	5003	5053	2.07	4275	3734	27.63	20.50	72
B17	96	4963	4752	7.90	3908	2944	42.94	41.83	96
avg		5148	5160	0.0	5547				
stdev		383	376	7.3	504				

Distribution of Radioactivity for Graphs

Time	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.70 parent	>0.8	adjusted parent	% on Biotic Solids	Average Biotic Extract	% Biotic Metabolites	% on Abiotic Solids	% Abiotic Recovery	% Abiotic Metabolites	% Abiotic Parent
0							89.09							
0.08	0.00	0.00	1.71	1.34	76.43	5.30	81.26	0.15	87.07	8.35	0.15	94.48	6.13	77.16
0.33	0.00	0.00	5.58	0.95	88.42	4.58	94.01	0.61	99.91	11.12				
1.00	0.00	1.99	17.47	5.38	63.31	5.05	67.32	1.71	95.61	29.88	0.22	98.65	11.76	85.40
2.00	5.76	6.93	34.79	8.76	32.82	5.11	34.90	3.64	96.52	61.34				
3.00	6.29	9.45	42.05	9.87	16.97	4.85	18.04	4.78	93.17	72.51				
4.00	6.61	14.50	39.18	10.80	9.69	4.95	10.30	5.72	91.94	76.04				
6.00	10.98	21.45	35.09	11.04	5.41	5.17	5.75	6.24	94.42	83.73	0.11	92.87		
8.00	7.88	24.87	31.11	12.85	2.73	4.65	2.90	5.89	91.81	81.36	0.17	90.70		
10.00	7.43	34.09	26.83	11.97	2.10	4.58	2.24	5.69	91.70	84.90				
13.00	7.15	37.34	20.02	9.93	1.81	4.83	1.93	6.10	89.67	79.27	0.17	101.65	15.98	84.73
15.00	8.48	38.39	17.94	10.07	1.71	4.52	1.81	3.83	87.82	79.40	0.18	99.12		
22.60	8.53	47.63	11.83	9.26	1.18	3.84	1.25	4.95	87.41	81.08				
29.00	6.21	42.98	10.88	7.33	0.68	3.92	0.72	5.23	87.50	71.31	0.38	97.55		
32.60	5.21	51.76	8.97	7.03	1.00	3.90	1.07	6.13	86.37	76.87				
48.00	8.68	50.54	7.94	5.33	0.50	2.83	0.54	6.97	81.88	75.32	0.25	94.99	5.10	83.12
72.00	15.33	24.57	2.53	3.62	0.52	2.51	0.56	19.64	79.50	48.57	3.60	90.29		
96.00	11.31	13.45	0.49	3.84	1.14	3.50	1.21	17.15	58.17	32.59	0.28	92.04	3.71	85.96
										Average	0.55	95.23	8.54	83.27
										Stdev	1.0	3.7	4.6	3.2

Table A1-3 (continued)

BIOTIC TLC Extracts

REP 1

Relative Abundance in Methanol extacts on TLC Plates; % of Initial Radioactivity

Sample #	Time (days)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Extract 3	Solids	Extract 1 Recovery	Extract 2 Recovery	Extract 3 Recovery	% on Solids	Total Extract Recovery	Total Recovery +CO2	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.70 parent	>0.8	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.68 parent	>0.7
B1	0.083	1443	22	71	88	86.63	1.71	2.75	0.17	91.26	90.55			2.48	3.09	88.24	6.2	0.00	0.00	2.15	2.68	76.44	5.37
B2	0.333	1679	23		374	97.62	1.78		0.72	100.13	99.21			5.92	0	89.1	4.97	0.00	0.00	5.78	0.00	86.98	4.85
B3	1	1570	50		1003	91.29	3.88		1.94	97.11	92.76		2.12	19.8	6.46	65.67	5.94	0.00	1.94	18.07	5.90	59.95	5.42
B4	2	1464	124		2201	85.12	9.61		4.27	99.00	93.22	0.7	5.98	38.19	11.08	37.75	6.29	0.60	5.09	32.51	9.43	32.13	5.35
B5	3	1362	166		2477	79.19	12.87		4.80	96.86	92.18	0.88	10.61	49.07	13.28	20.35	5.8	0.70	8.40	38.86	10.52	16.12	4.59
B6	4	1263	150		3439	73.44	11.63		6.67	91.73	90.87	1.79	14.93	49.58	15.7	11.28	6.72	1.31	10.96	36.41	11.53	8.28	4.93
B7	6	1261	195		3258	73.32	15.12		6.31	94.75	95.87	1.97	23.61	45.9	15.09	6.39	7.03	1.44	17.31	33.65	11.06	4.69	5.15
B8	8	1258	179		3149	73.15	13.88		6.10	93.13	90.19	1.26	28.61	41.68	17.59	3.46	7.4	0.92	20.93	30.49	12.87	2.53	5.41
B9	10	1139	227		3989	66.23	17.60		7.73	91.56	92.15	1.45	33.48	34.44	18.99	3.51	8.13	0.96	22.17	22.81	12.58	2.32	5.38
B10	13	1123	191		3560	65.30	14.81		6.90	87.00	84.24	1.99	44.41	25.96	16.67	2.49	8.47	1.30	29.00	16.95	10.88	1.63	5.53
B11	15	1137	229		2588	66.11	17.75		5.02	88.88	86.68	2.17	47.18	23.97	16.69	2.18	7.81	1.43	31.19	15.85	11.03	1.44	5.16
B12	22.6	1075	236		3134	62.51	18.30		6.07	86.88	87.04	2.22	57.33	16.92	15.56	1.78	6.22	1.39	35.83	10.58	9.73	1.11	3.89
B13	29	975	265	16	2612	56.69	20.54	0.62	5.06	82.92	85.91	2.16	60.95	14.42	12.99	1.39	8.07	1.22	34.55	8.17	7.36	0.79	4.57
B14	32.6	953	276		2202	55.41	21.40		4.27	81.08	87.00	1.36	64.22	12.29	12.73	1.9	7.5	0.75	35.59	6.81	7.05	1.05	4.16
B15	48	807	284		4049	46.92	22.02		7.85	76.79	88.47	5.8	64.53	12.56	11.13		5.97	2.72	30.28	5.89	5.22	0.00	2.80
B16	72	261	267	153	8064	28.50	20.70	11.86	15.63	76.69	104.32	11.7	58.76	5.91	15.14		8.49	3.33	16.75	1.68	4.32	0.00	2.42
B17	96	238	196		10760	13.84	15.20		20.85	49.89	92.83	21.88	34.17		17.29	6.12	20.54	3.03	4.73	0.00	2.39	0.85	2.84

BIOTIC TLC Extracts

REP 2

Relative Abundance in Methanol extacts on TLC Plates; % of Initial Radioactivity

Sample #	Time (days)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Extract 3	Solids	Extract 1 Recovery	Extract 2 Recovery	Extract 3 Recovery	% on Solids	Total Extract Recovery	Total Recovery +CO2	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.70 parent	>0.8	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.68 parent	>0.7
B1	0.083	1353	24	36	66	79.51	1.85	1.40	0.13	82.88	82.17			1.61		91.81	6.57	0.00	0.00	1.28	0.00	73.00	5.22
B2	0.333	1699	17		256	97.89	1.31		0.49	99.69	98.77			5.5	1.94	88.15	4.41	0.00	0.00	5.38	1.90	86.29	4.32
B3	1	1568	71		790	87.37	5.27		1.47	94.11	89.76		2.34	19.3	5.56	67.44	5.35	0.00	2.04	16.86	4.86	58.92	4.67
B4	2	1528	96		1647	83.99	7.04		3.02	94.04	88.26		5.55	39.14	9.62	39.9	5.79	0.00	4.66	32.87	8.08	33.51	4.86
B5	3	1333	144		2572	74.04	10.66		4.76	89.47	84.79		8.61	47.95	12.46	24.06	6.91	0.00	6.38	35.50	9.23	17.81	5.12
B6	4	1297	164		2486	74.77	12.61		4.78	92.15	91.29		14.23	50.82	13.46	14.84	6.65	0.00	10.64	38.00	10.06	11.10	4.97
B7	6	1271	168		3149	74.74	13.17		6.17	94.08	95.19	3.37	22.01	44.75	14.73	8.2	6.93	2.52	16.45	33.44	11.01	6.13	5.18
B8	8	1191	233		3015	67.28	17.55		5.68	90.50	87.57	3.05	28.81	40.92	17.09	4.34	5.78	2.05	19.38	27.53	11.50	2.92	3.89
B9	10	1241	200		1874	72.59	15.60		3.65	91.84	92.43	2.62	35.12	38.81	15.66	2.59	5.2	1.90	25.49	28.17	11.37	1.88	3.77
B10	13	1189	262		2808	67.28	19.77		5.30	92.34	89.58	5.04	42.31	30.23	13.33	2.97	6.13	3.39	28.46	20.34	8.97	2.00	4.12
B11	15	1065	310		1396	60.59	23.52		2.65	86.76	84.56	5.12	43.62	26.58	15.02	3.25	6.4	3.10	26.43	16.11	9.10	1.97	3.88
B12	22.6	1123	241		1973	65.40	18.71		3.83	87.95	88.12	4.08	57.19	17.6	13.44	1.9	5.79	2.67	37.40	11.51	8.79	1.24	3.79
B13	29	771	492	29	2697	46.21	39.32	1.16	5.39	92.08	95.08	6.04	53.14	16.74	15.8	1.22	7.05	2.79	24.56	7.74	7.30	0.56	3.26
B14	32.6	931	317		3881	57.54	26.12		8.00	91.66	97.58	4.34	62.24	13.26	12.17	1.66	6.33	2.50	35.81	7.63	7.00	0.96	3.64
B15	48	922	230		2775	60.70	20.19		6.09	86.98	98.65	7.07	65.15	12.46	8.96	1.66	4.7	4.29	39.54	7.56	5.44	1.01	2.85
B16	72	352	172	223	8830	28.28	18.43	11.94	23.65	82.30	109.93	12.71	57.07	6.98	10.37	3.7	9.17	3.59	16.14	1.97	2.93	1.05	2.59
B17	96	228	219		3961	23.23	29.76		13.45	66.44	109.38	16.93	32.08	4.18	22.76	6.16	17.89	3.93	7.45	0.97	5.29	1.43	4.16
										avg	92.25												
										stdev	6.51												

Relative Abundance in Water Extacts; % of Initial Radioactivity

Sample #	Time (days)	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.70 parent	origin	Rf <0.1	0.20	Rf 0.3-0.4	Rf 0.70 parent
B1	0.083					100	0	0	0	0.00	1.71
B2	0.333					100	0	0	0	0.00	1.78
B3	1					100	0	0	0	0.00	3.88
B4	2	56.8	21.33	21.86			5.46	2.05	2.10	0.00	0.00
B5	3	46.16	16	37.84			5.94	2.06	4.87	0.00	0.00
B6	4	51.18	31.8	17.02			5.95	3.70	1.98	0.00	0.00
B7	6	59.51	30.26	10.22			9.00	4.57	1.55	0.00	0.00
B8	8	46.07	34	15.14	4.8		6.39	4.72	2.10	0.67	0.00
B9	10	34.06	58.31	7.63			5.99	10.26	1.34	0.00	0.00
B10	13	32.46	58.15	9.29			4.81	8.61	1.38	0.00	0.00
B11	15	34.96	53.96	11.08			6.21	9.58	1.97	0.00	0.00
B12	22.6	35.52	60.16	4.32			6.50	11.01	0.79	0.00	0.00
B13	29	20.43	65.33	14.24			4.20	13.42	2.93	0.00	0.00
B14	32.6	16.77	75.07	8.16			3.59	16.06	1.75	0.00	0.00
B15	48	23.51	70.96	5.52			5.18	15.62	1.22	0.00	0.00
B16	72	57.33	39.27	3.4			11.87	8.13	0.70	0.00	0.00
B17	96	51.55	48.45	0			7.83	7.36	0.00	0.00	0.00

MeOH Extract Average RAM Distribution

Sample #	Time (hrs)	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.70 parent	>0.8
B1	0.083	0.00	0.00	1.71	1.34	74.72	5.30
B2	0.333	0.00	0.00	5.58	0.95	86.64	4.58
B3	1	0.00	1.99	17.47	5.38	59.44	5.05
B4	2	0.30	4.88	32.69	8.76	32.82	5.11
B5	3	0.35	7.39	37.18	9.87	16.97	4.85
B6	4	0.66	10.80	37.20	10.80	9.69	4.95
B7	6	1.98	16.88	33.55	11.04	5.41	5.17
B8	8	1.49	20.15	29.01	12.18	2.73	4.65
B9	10	1.43	23.83	25.49	11.97	2.10	4.58
B10	13	2.35	28.73	18.64	9.93	1.81	4.83
B11	15	2.27	28.81	15.98	10.07	1.71	4.52
B12	22.6	2.03	36.62	11.04	9.26	1.18	3.84
B13	29	2.01	29.56	7.96	7.33	0.68	3.92
B14	32.6	1.63	35.70	7.22	7.03	1.00	3.90
B15	48	3.51	34.91	6.73	5.33	0.50	2.83
B16	72	3.46	16.44	1.83	3.62	0.52	2.51
B17	96	3.48	6.09	0.49	3.84	1.14	3.50

Table A1-3 (continued)

ABIOTIC TLC Extracts:
REP 1

Sample #	Time (days)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.083	1538	11	39	89.43	0.85	0.08	90.35
A2	0.333							
A3	1	1809	19	127	105.18	1.47	0.25	106.90
A4	2							
A5	3							
A6	4							
A7	6	1641	5	68	95.42	0.39	0.13	95.93
A8	8	1484	18	74	86.29	1.40	0.14	87.83
A9	10							
A10	13	1740	20	57	101.17	1.55	0.11	102.83
A11	15	1624	10	121	94.43	0.78	0.23	95.44
A12	22.6							
A13	29	1639	16	200	95.30	1.24	0.39	96.93
A14	32.6							
A15	48	1639	11	109	95.30	0.85	0.21	96.36
A16	72	1386		3310	80.59		6.42	87.00
A17	96	1522	21	133	88.50	1.63	0.26	90.38

	Relative Abundance in Methanol extacts on TLC Plates					% of Initial Radioactivity				
			Rf					Rf		
Sample #	origin	0.14 shoulder	0.41 chain	0.70 Parent	>.8 ethoxy	origin	0.14 ethoxy+	0.41 c-chain	0.70 Parent	>.8 of ethoxy
A1	5.67	1.38	1.59	86.28	5.08	5.07	1.23	1.42	77.16	4.54
A2										
A3	5.41	1.56	1.18	87.59	4.26	5.69	1.64	1.24	92.13	4.48
A4										
A5										
A6										
A7										
A8										
A9										
A10	5.45	1.66		88.27	4.62	5.51	1.68		89.30	4.67
A11										
A12										
A13										
A14										
A15										
A16										
A17										

ABIOTIC TLC Extracts:
REP 2

Sample #	Time (days)	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.083	1692	15	111	98.38		0.22	98.60
A2	0.333							
A3	1	1533	14	95	89.14	1.09	0.18	90.41
A4	2							
A5	3							
A6	4							
A7	6	1543	16	49	89.72		0.09	89.81
A8	8	1606	5	98	93.38		0.19	93.57
A9	10							
A10	13	1724	14	119	100.24		0.23	100.47
A11	15	1766	10	64	102.68		0.12	102.81
A12	22.6							
A13	29	1658	18	197	96.40	1.40	0.38	98.18
A14	32.6							
A15	48	1605	19	149	93.32		0.29	93.61
A16	72	1596		405	92.80		0.78	93.58
A17	96	1606	19	161	93.38		0.31	93.69
Average					94.05	1.15	0.55	95.23
Std Dev					5.73	0.37	1.35	5.18

	Relative Abundance in Methanol extacts on TLC Plates					% of Initial Radioactivity				
			Rf					Rf		
Sample #	origin	0.14 2-3 ethoxy+ shoulder	0.41 oxidation of c-chain	0.64 Parent	0.87 oxidation of ethoxy	origin	0.14 2-3 ethoxy+ shoulder	0.41 oxidation of c-chain	0.70 Parent	>.8 oxidation of ethoxy
A1										
A2										
A3	5.29	2.03		88.26	4.43	4.72	1.81	0.00	78.67	3.95
A4										
A5										
A6										
A7										
A8										
A9										
A10	16.87	1.31		79.97	1.86	16.91	1.31	0.00	80.16	1.86
A11										
A12										
A13										
A14										
A15	4.31	1.33	1.78	89.07	3.51	4.02	1.24	1.66	83.12	3.28
A16										
A17	2.39	1.67		92.05	3.89	2.23	1.56		85.96	3.63
Average						6.31	1.50	0.86	83.79	3.77
Std Dev						4.46	0.22	0.72	5.19	0.91

Appendix 2 – Phase 2 Raw Data

Table A2-1. $C_{14}E_1$ Raw Data.

Test Chemical:	^{14}C C14E1
RC info:	RC 94357-04; C- in ethoxylates; specific activity: 23.2 mCi/g
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 C, 0.5 mg/L DO
Date:	2/3/2015
Test Chem purity:	97.65% by Rad-TLC
Test Chemical Rad-TLC Analysis:	50:50 Ethylacetate:hexanes on Analtech 43011
Test Concentration (mg/L):	0.062

Mineralization		***Abiotic***			***Biotic***			**meOH + water**		**% of Initial Radioactivity**						
Sample #	Time (hrs)	Direct dpm/mL	Acidified dpm/mL	% CO2 %	Direct dpm/mL	Acidified dpm/mL	% CO2 %	origin	Rf <0.1	0.20	0.44 parent	0.76	0.85	adjusted parent	% solids	% abiotic solids
B1	0.01666667				3415	3340	-2.47							86.33		
B2	0.08333333	3406	3376	-3.57	3461	3404	-4.43	5.43	0.00	6.67	91.46	0.00	3.95	78.96	0.99	
B3	0.25				3473	3146	3.49	10.89	0.81	5.20	67.01	0.74	8.42	57.85	2.13	1.37
B4	0.5				3392	3001	7.93	20.33	2.25	2.99	36.86	1.94	8.23	31.82	3.07	
B5	1	3420	3500	-7.38	3446	2813	13.70	25.48	3.49	2.69	17.79	3.92	8.24	15.35	5.28	
B6	2				3273	2732	16.19	32.96	3.69	2.26	7.88	6.30	5.80	6.80	10.05	0.91
B7	4	3560	3397	-4.22	3534	2647	18.79	33.28	4.03	1.62	4.07	7.28	2.92	3.52	8.15	
B8	6					2486	23.73	35.57	3.59	0.68	2.38	6.07	1.76	2.06	10.99	1.10
B9	8		3320	-1.85		2269	30.39	33.96	4.72	1.36	2.30	3.76	0.50	1.99	8.63	
B10	10					2184	33.00	33.57	5.06	0.87	1.23	3.30	0.44	1.06	8.87	0.98
B11	12		3283	-0.72		2070	36.50	28.79	4.24	1.06	1.58	3.41	0.49	1.36	16.57	
B12	24		3372	-3.45		1795	44.93	32.14	3.53	0.86	1.47	2.22	0.94	1.26	17.97	2.78
B13	36		3133	3.88		1669	48.80	26.56	2.24	0.00	0.85	2.01	0.31	0.73	25.75	1.22
B14	48		3174	2.63		1429	56.16	28.67	1.66	0.00	0.34	1.51	0.00	0.29	21.76	
B15	72		3110	4.59		1296	60.24	29.14	0.70	0.00	0.52	0.74	0.00	0.45	19.54	1.22
B16	97		2931	10.08		1067	67.27	29.66	1.15	0.00	0.29	1.07	0.00	0.25	11.48	0.96
term								23.20	0.17	0.00	0.29	0.59	0.00	0.25	14.73	1.30
	avg	3462.00	3259.60	0.00	3427.71											
				4.954024792												

Table A2-1 (continued)

BIOTIC TLC Extracts

REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery with CO2
B1	0.02	1096	43	372	100.87	5.28	1.14	107.29	104.82
B2	0.08	910	86	607	83.75	10.55	1.86	96.17	91.74
B3	0.25	605	98	1001	55.68	12.03	3.07	70.78	74.26
B4	0.50	515	100	1538	47.40	12.27	4.72	64.39	72.32
B5	1.00	447	154	1548	41.14	18.90	4.75	64.79	78.49
B6	2.00	431	156	1762	39.67	19.14	5.41	64.22	80.40
B7	4.00	375	80	3058	34.51	9.82	9.38	53.71	72.51
B8	6.00	329	159	3600	30.28	19.51	11.04	60.84	84.57
B9	8.00	290	176	3680	26.69	21.60	11.29	59.58	89.97
B10	10.00	244	112	5679	22.46	13.74	17.42	53.62	86.62
B11	12.00	235	171	5152	21.63	20.98	15.81	58.42	94.91
B12	24.00	141	139	7960	12.98	17.06	24.42	54.45	99.39
B13	36.00	136	158	7256	12.52	19.39	22.26	54.17	102.96
B14	48.00	90	223	4485	8.28	27.37	13.76	49.41	105.57
B15/16	72.00	142	196	2879	13.07	24.05	8.83	45.95	106.19
B17	97.00	61	184	3058	5.61	22.58	9.38	37.58	104.84

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

origin	Rf <0.1	0.20	0.44 parent	0.76	0.85	origin	Rf <0.1	0.20	0.44 parent	0.76	0.85
2.81		8.29	86.35		2.56	2.83	0.00	8.36	87.10	0.00	2.58
3.57		8.09	75.85	0.94	11.55	2.99	0.00	6.78	63.53	0.79	9.67
9.98	3.22	6.34	66.57	3.58	10.31	5.56	1.79	3.53	37.07	1.99	5.74
21.97	8.24	8.71	40.77	8.07	12.25	10.41	3.91	4.13	19.32	3.83	5.81
31.42	6.28	8.72	22.8	20.79	9.99	12.93	2.58	3.59	9.38	8.55	4.11
40.67	10.7	5.51	13.29	22.3	7.54	16.13	4.24	2.19	5.27	8.85	2.99
53.68	11.56		9.59	19.96	5.21	18.53	3.99	0.00	3.31	6.89	1.80
46.63	20.05	8.98	9.9	14.44		14.12	6.07	2.72	3.00	4.37	0.00
47.99	26.06	6.51	5.21	14.21		12.81	6.96	1.74	1.39	3.79	0.00
39.96	22.31	9.46	8.23	20.04		8.97	5.01	2.12	1.85	4.50	0.00
47.39	21.81	7.92	7.28	11.41	4.19	10.25	4.72	1.71	1.57	2.47	0.91
54.43	17.29		9.05	19.23		7.06	2.24	0.00	1.17	2.50	0.00
69.76	15.97			14.27		8.73	2.00	0.00	0.00	1.79	0.00
79.12	11.51			9.37		6.55	0.95	0.00	0.00	0.78	0.00
75.54	12.98			11.48		9.87	1.70	0.00	0.00	1.50	0.00
91.93				8.07		5.16	0.00	0.00	0.00	0.45	0.00

Relative Abundance in Water extacts on TLC Plates / **% of Initial Radioactivity**

origin	Rf <0.1	0.20	0.44 parent	0.76	0.85		origin	Rf <0.1	0.20	0.44 parent
34.83			65.17				1.84	0.00	0.00	3.44
71.97			28.03				7.60	0.00	0.00	2.96
87.42			12.58				10.51	0.00	0.00	1.51
88.44			11.56				10.85	0.00	0.00	1.42
93.65			6.35				17.70	0.00	0.00	1.20
100							19.14	0.00	0.00	0.00
100							9.82	0.00	0.00	0.00
100							19.51	0.00	0.00	0.00
100							21.60	0.00	0.00	0.00
100							13.74	0.00	0.00	0.00
100							20.98	0.00	0.00	0.00
100							17.06	0.00	0.00	0.00
100							19.39	0.00	0.00	0.00
100							27.37	0.00	0.00	0.00
100							24.05	0.00	0.00	0.00
100							22.58	0.00	0.00	0.00
							0.00	0.00	0.00	0.00

BIOTIC TLC Extracts

REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery with CO2
B1	0.02	1139	33	274	104.83	4.05	0.84	109.72	107.25
B2	0.08	918	60	783	84.49	7.36	2.40	94.25	89.82
B3	0.25	658	138	1002	60.56	16.93	3.07	80.57	84.05
B4	0.50	525	124	1901	48.32	15.22	5.83	69.37	77.30
B5	1.00	442	139	5001	40.68	17.06	15.34	73.08	86.78
B6	2.00	373	108	3551	34.33	13.25	10.89	58.48	74.66
B7	4.00	398	156	4105	36.63	19.14	12.59	68.37	87.16
B8	6.00	301	128	2026	27.70	15.71	6.22	49.63	73.36
B9	8.00	255	140	2104	23.47	17.18	6.45	47.10	77.49
B10	10.00	233	175	5121	21.44	21.48	15.71	58.63	91.63
B11	12.00	198	175	6562	18.22	21.48	20.13	59.83	96.32
B12	24.00	147	166	8824	13.53	20.37	27.07	60.97	105.90
B13	36.00	122	173	6928	11.23	21.23	21.25	53.71	102.51
B14	48.00	86	152	8253	7.92	18.65	25.32	51.89	108.05
B15/16	72.00	64	174	4605	5.89	21.35	14.13	41.37	101.61
B17	97.00	50	128	6548	4.60	15.71	20.09	40.40	107.66
					0	0	0	0.00	
									91.29
									12.14

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

origin	Rf <0.1	0.20	0.44 parent	0.76	0.85	origin	Rf <0.1	0.20	0.44 parent	0.76	0.85
4.73		4.75	85.44		5.08	4.96	0.00	4.98	89.57	0.00	5.33
8.03	1.92	4.3	76.45	0.82	8.48	6.78	1.62	3.63	64.59	0.69	7.16
16.55	4.48	4.05	54.11	3.11	17.71	10.02	2.71	2.45	32.77	1.88	10.73
32.62	6.35	2.58	28.04	8.3	22.11	15.76	3.07	1.25	13.55	4.01	10.68
47.54	11.79	2.28	10.01	9.97	18.41	19.34	4.80	0.93	4.07	4.06	7.49
52.51	11.14	3.05	8.38	16.65	8.28	18.03	3.82	1.05	2.88	5.72	2.84
64.57	8.73	3.69	3.98	14.32	4.72	23.65	3.20	1.35	1.46	5.25	1.73
67.09	12.14		5.81	11.36	3.6	18.59	3.36	0.00	1.61	3.15	1.00
66.23	13.51		4.52	11.94	3.79	15.54	3.17	0.00	1.06	2.80	0.89
62.39	16.16		6.09	10.83	4.53	13.38	3.47	0.00	1.31	2.32	0.97
63.51	12.88		7.44	10.86	5.31	11.57	2.35	0.00	1.36	1.98	0.97
63.73	16.53		3.87	11.25	4.62	8.62	2.24	0.00	0.52	1.52	0.63
71.22	11.77		5.99	11.02		8.00	1.32	0.00	0.67	1.24	0.00
72.19	5.73		13.22	8.86		5.71	0.45	0.00	1.05	0.70	0.00
68.58	10.4		10.01	11.02		4.04	0.61	0.00	0.59	0.65	0.00
64.3	7.3		12.76	15.65		2.96	0.34	0.00	0.59	0.72	0.00
						0.00	0.00	0.00	0.00	0.00	0.00

% of Initial Radioactivity

origin	Rf <0.1	0.20	0.44 parent	0.76	0.85		origin	Rf <0.1	0.20	0.44 parent
30.34			69.66				1.23	0.00	0.00	2.82
59.91			40.09				4.41	0.00	0.00	2.95
86.01			13.99				14.57	0.00	0.00	2.37
91.59			8.41				13.94	0.00	0.00	1.28
93.49			6.51				15.95	0.00	0.00	1.11
100			0				13.25	0.00	0.00	0.00
100							19.14	0.00	0.00	0.00
100							15.71	0.00	0.00	0.00
100							17.18	0.00	0.00	0.00
100							21.48	0.00	0.00	0.00
100							21.48	0.00	0.00	0.00
100							20.37	0.00	0.00	0.00
100							21.23	0.00	0.00	0.00
100							18.65	0.00	0.00	0.00
100							21.35	0.00	0.00	0.00
100							15.71	0.00	0.00	0.00

meoh average / **% of Initial Radioactivity**

Time hrs	origin	Rf <0.1	0.20	0.44 parent	0.76	0.85
0.02	3.90	0.00	6.67	88.33	0.00	3.95
0.08	4.89	0.81	5.20	64.06	0.74	8.42
0.25	7.79	2.25	2.99	34.92	1.94	8.23
0.50	13.09	3.49	2.69	16.44	3.92	8.24
1.00	16.13	3.69	2.26	6.73	6.30	5.80
2.00	17.08	4.03	1.62	4.07	7.28	2.92
4.00	21.09	3.59	0.68	2.38	6.07	1.76
6.00	16.35	4.72	1.36	2.30	3.76	0.50
8.00	14.18	5.06	0.87	1.23	3.30	0.44
10.00	11.18	4.24	1.06	1.58	3.41	0.49
12.00	10.91	3.53	0.86	1.47	2.22	0.94
24.00	7.84	2.24	0.00	0.85	2.01	0.31
36.00	8.36	1.66	0.00	0.34	1.51	0.00
48.00	6.13	0.70	0.00	0.52	0.74	0.00
72.00	6.96	1.15	0.00	0.29	1.07	0.00
97.00	4.06	0.17	0.00	0.29	0.59	0.00

water average / **% of Initial Radioactivity**

origin	Rf <0.1	0.20	0.44 parent	0.76	0.85
1.53			3.13		
6.00			2.95		
12.54			1.94		
12.39			1.35		
16.82			1.16		
16.20			0.00		
14.48			0.00		
17.61			0.00		
19.39			0.00		
17.61			0.00		
21.23			0.00		
18.71			0.00		
20.31			0.00		
23.01			0.00		
22.70			0.00		
19.14			0.00		

Table A2-1 (continued)

ABIOTIC TLC Extracts:																	
REP 1											**Relative Abundance in Methanol extacts on TLC Plates**					**% of Initial Radioactivity**	
Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery				polar	parent			polar	parent
A1	0.02																
A2	0.08	1062	11	339	97.74	1.35		99.09				17.37	82.63			16.98	80.76
A3	0.25			401													
A4	0.50																
A5	1.00	1157	17	259	106.49	2.09	0.79	109.37									
A6	2.00																
A7	4.00	1062	14	372	97.74	1.72	1.14	100.60									
A8	6.00																
A9	8.00	1170	14	213	107.68	1.72	0.65	110.05				2.63	97.37			2.83	104.85
A10	10.00																
A11	12.00	1062	17	1416	97.74	2.09	4.34	104.17				6.38	93.61			6.24	91.50
A12	24.00	1099	26	320	101.15	3.19	0.98	105.32									
A13	36.00	999	39	1469	91.94	4.79	4.51	101.24									
A14	48.00	1023	18	473	94.15	2.21	1.45	97.81									
A15/16	72.00	937	18	241	86.24	2.21	0.74	89.19				15.82	77.79	6.39		13.64	67.08
A17	97.00	1002	8	533	92.22	0.98	1.64	94.84				11.13	88.87			10.26	81.96
																9.99	85.23
																5.05	12.52

ABIOTIC TLC Extracts:

REP 2								
Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.02							
A2	0.08	1105	1	448	101.70	0.12	1.37	103.20
A3	0.25	1084	4	296				
A4	0.50							
A5	1.00	1152	4	336	106.03	0.49	1.03	107.55
A6	2.00							
A7	4.00	1097	6	347	100.96	0.74	1.06	102.76
A8	6.00							
A9	8.00	1149	0	425	105.75	0.00	1.30	107.05
A10	10.00							
A11	12.00	1124	6	396	103.45	0.74	1.21	105.40
A12	24.00	1125	0	473	103.54	0.00	1.45	
A13	36.00	1064	7	430				
A14	48.00	1066	3	321	98.11	0.37	0.98	99.46
A15/16	72.00	1028	9	385	94.61	1.10	1.18	96.90
A17	97.00	963	15	314	88.63	1.84	0.96	91.43
A17								
Average					98.7		1.5	101.4
Std Dev					6.05		1.07	5.71

Table A2-2. $C_{14}E_3$ Raw Data.

RC info:	95062-00A2; 1st and 3rd ethoxylates; specific activity: 13.7 mCi/g
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 C, 0.5 mg/L DO
Date:	2/3/2015
Test Chem purity:	100% by Rad-TLC
Test Chemical Rad-TLC Analysis:	90:10:1 Chlor:Meth:Formic on Analtech Cat 43011
Test Concentration (mg/L):	0.144

Mineralization		***Abiotic***			***Biotic***		
Sample #	Time (hrs)	Direct	Acidified	% CO2	Direct	Acidified	% CO2
		dpm/mL	dpm/mL	%	dpm/mL	dpm/mL	%
B1	0.02				4251	4509	-2.75
B2	0.08	2696	2669	-4.04	4195	4210	4.06
B3	0.25				4426	4337	1.17
B4	0.5				4506	4126	5.98
B5	1	2775	2727	-6.30	4400	4049	7.73
B6	2				4498	4047	7.78
B7	4	2777	2591	-1.00	4442	3912	10.85
B8	6					3830	12.72
B9	8		2539	1.03		3741	14.75
B10	10					3734	14.91
B11	12		2498	2.62		3781	13.84
B12	24		2633	-2.64		3771	14.07
B13	35		2556	0.36		3414	22.20
B14	48		2480	3.33		3612	17.69
B15	72		2593	-1.08		3292	24.98
B16	97		2367	7.73		2860	34.83
term							
	avg	2749.33	2565.30	0.00	4388.29		
				3.795836199			

meOH + water	**% of Initial Radioactivity**									
origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7	time	adjusted parent	% solids	abiotic solids	Sum Metabolites
						0.0	99.10			
1.14	1.93	7.03	1.38	85.07	2.05	0.02	85.01	2.68		13.53
3.87	5.06	16.65	2.34	68.00	1.45	0.08	67.96	2.34	0.96	29.36
6.53	11.75	40.74	4.22	33.48	1.20	0.25	33.46	6.15		64.43
6.05	15.15	51.23	3.59	11.87	0.55	0.5	11.86	9.45		76.57
5.35	15.53	43.44	1.68	3.11	0.38	1	3.11	4.56	0.97	66.39
5.92	21.64	53.95	2.21	2.60	0.00	2	2.60	8.94		83.72
4.62	24.67	49.32	2.53	2.20	0.00	4	2.20	10.12	1.26	81.14
5.96	30.28	44.65	2.55	1.79	0.00	6	1.79	10.13		83.44
4.46	31.48	40.16	2.33	1.67	0.00	8	1.67	12.16	1.51	78.42
5.22	35.07	36.37	2.94	1.63	0.00	10	1.63	10.16		79.59
5.85	37.83	33.67	2.83	1.58	0.00	12	1.58	10.01	0.93	80.17
5.47	48.94	22.16	1.82	1.11	0.00	24	1.11	9.38		78.39
4.30	49.09	17.06	1.93	0.70	0.00	35	0.70	9.20		72.38
5.11	50.68	9.34	1.33	0.80	0.00	48	0.80	13.29	1.15	66.45
5.71	37.19	18.92	3.39	0.73	0.00	72	0.73	13.55	2.55	65.22
7.74	30.98	11.71	0.32	0.67	0.00	97	0.67	18.37	1.49	50.74
									1.35	
									0.50	

Table A2-2 (continued)

BIOTIC TLC Extracts

REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery +CO2
B1	0.01666667	1368	66	1398	93.52	6.02	3.19	102.72	99.97
B2	0.08333333	1323	100	1078	90.45	9.12	2.46	102.02	106.08
B3	0.25	1135	220	2890	77.59	20.05	6.59	104.23	105.40
B4	0.5	1013	207	4206	69.25	18.87	9.58	97.71	103.68
B5	1	713	213	2253	48.74	19.42	5.13	73.29	81.02
B6	2	960	230	3821	65.63	20.96	8.71	95.30	103.08
B7	4	895	243	4442	61.19	22.15	10.12	93.46	104.31
B8	6	941	238	4072	64.33	21.69	9.28	95.30	108.03
B9	8	900	221	4885	61.53	20.14	11.13	92.80	107.55
B10	10	940	193	3793	64.26	17.59	8.64	90.50	105.41
B11	12	815	254	4759	55.72	23.15	10.84	89.71	103.55
B12	24	762	280	4758	52.09	25.52	10.84	88.46	102.52
B13	36	746	236	3283	51.00	21.51	7.48	79.99	102.19
B14	48	584	290	6392	39.92	26.43	14.57	80.92	98.61
B15	72	615	250	6486	42.04	22.79	14.78	79.61	104.59
B16	97	414	245	8521	28.30	22.33	19.42	70.05	104.88
B17									

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7
B1		0.68	4.79		93.04	1.5	0.00	0.64	4.48	0.00	87.01	1.40
B2	1.16	4.25	13.99	2.28	76.58	1.74	1.05	3.84	12.65	2.06	69.26	1.57
B3	0.78	8.22	41.99	5.54	41.46	2	0.61	6.38	32.58	4.30	32.17	1.55
B4	1.27	13.45	62.49	5.11	16.1	1.59	0.88	9.31	43.28	3.54	11.15	1.10
B5	1.54	15.22	73.54	2.81	6.88		0.75	7.42	35.85	1.37	3.35	
B6	1.57	21.09	69.22	3.82	4.29		1.03	13.84	45.43	2.51	2.82	
B7	1.65	25.44	65.19	4.13	3.6		1.01	15.57	39.89	2.53	2.20	
B8	2	32.2	58.29	4.59	2.92		1.29	20.71	37.50	2.95	1.88	
B9	2.55	36.04	54.46	4.01	2.93		1.57	22.17	33.51	2.47	1.80	
B10	3.34	39.9	48.75	5.4	2.61		2.15	25.64	31.33	3.47	1.68	
B11	3.06	43.4	45.93	4.75	2.86		1.70	24.18	25.59	2.65	1.59	
B12	2.74	55.46	35.82	3.6	2.38		1.43	28.89	18.66	1.88	1.24	
B13	3.11	62.79	28.02	4.36	1.72		1.59	32.02	14.29	2.22	0.88	
B14	3.69	70.71	19.87	3.9	1.82		1.47	28.23	7.93	1.56	0.73	
B15	6.18	5.93	72.84	12.77	2.28		2.60	2.49	30.62	5.37	0.96	
B16	7.37	6.95	80.79	2.24	2.65		2.09	1.97	22.87	0.63	0.75	
B17												

Relative Abundance in Water extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7
B1	19.36	15.19	13.33		52.12		1.16	0.91	0.80		3.14	
B2	30.52	19.96	25.29		24.23		2.78	1.82	2.31		2.21	
B3	27.93	26.11	39.95		6		5.60	5.24	8.01		1.20	
B4	26.78	27.74	45.47				5.05	5.23	8.58		0.00	
B5	20.08	43.92	36				3.90	8.53	6.99			
B6	20.37	28.66	50.97				4.27	6.01	10.69			
B7	16.28	41.12	42.6				3.61	9.11	9.44			
B8	20.23	39.7	40.07				4.39	8.61	8.69			
B9	17.27	49.84	32.89				3.48	10.04	6.63			
B10	21.76	50.87	27.36				3.83	8.95	4.81			
B11	16.79	52.83	30.37				3.89	12.23	7.03			
B12	15.72	69.97	14.31				4.01	17.86	3.65			
B13	14.96	70.97	14.06				3.22	15.27	3.02			
B14	15.46	79	5.55				4.09	20.88	1.47			
B15	16.52	78.72	4.75				3.76	17.94	1.08			
B16	24.92	75.09	0				5.57	16.77	0.00			
B17												

BIOTIC TLC Extracts

REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery +CO2
B1	0.01666667	1335	70	955	91.27	6.38	2.18	99.82	97.07
B2	0.08333333	1212	135	972	82.86	12.31	2.21	97.38	101.44
B3	0.25	1163	205	2507	79.51	18.69	5.71	103.91	105.07
B4	0.5	1005	220	4085	68.71	20.05	9.31	98.07	104.04
B5	1	620	308	1747	42.39	28.07	3.98	74.44	82.17
B6	2	928	248	4027	63.44	22.61	9.18	95.22	103.00
B7	4								
B8	6	883	264	4816	60.37	24.06	10.97	95.40	108.13
B9	8	838	233	5791	57.29	21.24	13.20	91.72	106.47
B10	10	827	264	5123	56.54	24.06	11.67	92.28	107.19
B11	12	874	273	4024	59.75	24.88	9.17	93.80	107.64
B12	24	728	347	3471	49.77	31.63	7.91	89.31	103.37
B13	36	648	322	4794	44.30	29.35	10.92	84.58	106.78
B14	48	601	297	5273	41.09	27.07	12.02	80.17	97.86
B15	72	586.5	296	5409	40.10	26.98	12.33	79.40	104.38
B16	97	398	274	7599	27.21	24.98	17.32	69.50	104.33
B17							0		
									102.49
									6.17

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7
B1		1.84	7.4	3.02	84.79	2.95	0.00	1.68	6.75	2.76	77.38	2.69
B2	0.81	2.55	17.4	3.15	74.51	1.59	0.67	2.11	14.42	2.61	61.74	1.32
B3	1.93	8.34	41.9	5.21	41.56	1.06	1.53	6.63	33.31	4.14	33.04	0.84
B4	1.64	12.22	63.25	5.29	17.6		1.13	8.40	43.46	3.63	12.09	0.00
B5	2.7	17.63	67.32	4.71	6.75	0.89	1.14	7.47	28.53	2.00	2.86	0.38
B6	3.46	19.74	70.04	3	3.77		2.20	12.52	44.43	1.90	2.39	0.00
B7												
B8	4.13	28.81	60.68	3.56	2.82		2.49	17.39	36.63	2.15	1.70	0.00
B9	2.11	31.16	60.19	3.84	2.7		1.21	17.85	34.48	2.20	1.55	0.00
B10	3.06	36.01	53.87	4.25	2.8		1.73	20.36	30.46	2.40	1.58	0.00
B11	3.55	40.44	48.36	5.04	2.62		2.12	24.16	28.90	3.01	1.57	0.00
B12	3.33	57.13	34.01	3.55	1.98		1.66	28.43	16.93	1.77	0.99	0.00
B13	3.33	58.17	33.61	3.71	1.19		1.48	25.77	14.89	1.64	0.53	0.00
B14	3.78	72.34	19.07	2.67	2.15		1.55	29.72	7.84	1.10	0.88	0.00
B15	3.89	79.13	12.2	3.54	1.25		1.56	31.73	4.89	1.42	0.50	0.00
B16	8.26	87.54	2.05	0	2.14		2.25	23.82	0.56	0.00	0.58	0.00
B17												

% of Initial Radioactivity

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7
B1	17.56	10.04	31.62		40.78		1.12	0.64	2.02		2.60	
B2	26.39	18.98	31.9		22.73		3.25	2.34	3.93		2.80	
B3	28.45	28.12	40.52		2.91		5.32	5.25	7.57		0.54	
B4	25.17	36.64	35.68		2.51		5.05	7.35	7.16		0.50	
B5	17.48	27.25	55.27				4.91	7.65	15.52			
B6	19.21	48.26	32.52				4.34	10.91	7.35			
B7												
B8	15.55	57.52	26.93				3.74	13.84	6.48			
B9	12.5	60.68	26.82				2.65	12.89	5.70			
B10	11.36	63.12	25.52				2.73	15.19	6.14			
B11	16.04	60.6	23.36				3.99	15.08	5.81			
B12	12.13	71.78	16.09				3.84	22.70	5.09			
B13	7.9	85.6	6.5				2.32	25.12	1.91			
B14	11.46	83.23	5.31				3.10	22.53	1.44			
B15	12.98	82.39	4.62				3.50	22.23	1.25			
B16	22.31	77.69	0				5.57	19.40	0.00			
B17												

Meoh average **% of Initial Radioactivity**

Time hrs	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7
0.016667	0.00	1.16	5.62	1.38	82.20	2.05
0.083333	0.86	2.98	13.54	2.34	65.50	1.45
0.25	1.07	6.50	32.95	4.22	32.61	1.20
0.5	1.00	8.86	43.37	3.59	11.62	0.55
1	0.95	7.45	32.19	1.68	3.11	0.38
2	1.61	13.18	44.93	2.21	2.60	0.00
4	1.01	15.57	39.89	2.53	2.20	0.00
6	1.89	19.05	37.06	2.55	1.79	0.00
8	1.39	20.01	34.00	2.33	1.67	0.00
10	1.94	23.00	30.89	2.94	1.63	0.00
12	1.91	24.17	27.24	2.83	1.58	0.00
24	1.54	28.66	17.79	1.82	1.11	0.00
36	1.53	28.90	14.59	1.93	0.70	0.00
48	1.51	28.98	7.88	1.33	0.80	0.00
72	2.08	17.11	17.76	3.39	0.73	0.00
97	2.17	12.89	11.71	0.32	0.67	0.00

water average

origin	Rf <0.1	0.20	Rf 0.3-0.4	0.66 parent	>0.7
1.14	0.78	1.41		2.87	
3.01	2.08	3.12		2.50	
5.46	5.25	7.79		0.87	
5.05	6.29	7.87		0.25	
4.40	8.09	11.25			
4.31	8.46	9.02			
3.61	9.11	9.44			
4.07	11.23	7.59			
3.07	11.46	6.16			
3.28	12.07	5.48			
3.94	13.66	6.42			
3.92	20.28	4.37			
2.77	20.20	2.47			
3.59	21.71	1.45			
3.63	20.08	1.16			
5.57	18.09	0.00			

Table A2-2 (continued)

ABIOTIC TLC Extracts:
REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	**Relative Abundance** polar metabolites	parent	nonpolar metabolites	**% of Initial Radioactivity** polar metabolites	parent	
A1	0.01666667													
A2	0.08333333	943	13	242	110.28	2.03	0.94	113.25	1.15	98.85		1.15	109.01	
A3	0.25													
A4	0.5													
A5	1	883	1	260	103.26	0.16	1.01	104.43		100		0	103.26	
A6	2													
A7	4	872	8	287	101.98	1.25	1.12	104.34						
A8	6													
A9	8	852	11	442	99.64	1.72	1.72	103.08	1.75	96.95	1.3	1.75	96.60	0.02
A10	10													
A11	12	858	3	244	100.34	0.47	0.95	101.76		100			100.34	
A12	24													
A13	36	836	9	462										
A14	48	842	19	303	98.47	2.96	1.18	102.61		100			98.47	
A15	72	780	41	882	91.22	6.39	3.44	101.05		100			91.22	
A16	97	815	4	357	95.31	0.62	1.39	97.33		100			95.31	
A17														
													99.17	

ABIOTIC TLC Extracts:
REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.01666667							
A2	0.08333333	924	6	248	108.06	0.94	0.97	109.96
A3	0.25							
A4	0.5							
A5	1	911	8	237	106.54	1.25	0.92	108.71
A6	2							
A7	4	871	6	357	101.86	0.94	1.39	104.19
A8	6							
A9	8	888	10	335	103.85	1.56	1.31	106.71
A10	10							
A11	12	782	5	232	91.45	0.78	0.90	93.14
A12	24							
A13	36	853	9	304				
A14	48	860	5	285	100.57	0.78	1.11	102.46
A15	72	825	12	424	96.48	1.87	1.65	100.00
A16	100	785	11	409	91.80	1.72	1.59	95.11
A17								
Average					100.1		1.4	103.0
Std Dev					5.59		0.60	5.07

Table A2-3. $C_{14}E_9$ Raw Data.

Test Chemical:	^{14}C C14E9
RC info:	RC 96455-03E; ethoxy; specific activity: 8 mCi/g
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 C, 0.5 mg/L DO
Date:	2/3/2015
Test Chem purity:	98.4% by Rad-TLC
Test Chemical Rad-TLC Analysis:	90:10:1 Chloroform:Methanol:formic acid on Analtech Cat 43011
Test Concentration (mg/L):	0.102

Mineralization		***Abiotic***			***Biotic***		
Sample #	Time (hrs)	Direct	Acidified	% CO2	Direct	Acidified	% CO2
		dpm/mL	dpm/mL	%	dpm/mL	dpm/mL	%
B1	0.017				1958	1898	-3.48
B2	0.083	1878	1864	-1.63	1974	1975	-7.68
B3	0.25				2021	1885	-2.78
B4	0.5				2147	1900	-3.59
B5	1	2010	1984	-8.17	2028	1619	11.73
B6	2				2152	1735	5.40
B7	4	1884	1817	0.93	1982	1740	5.13
B8	6					1705	7.04
B9	8		1776	3.17		1636	10.80
B10	10					1591	13.25
B11	12		1728	5.78		1687	8.02
B12	24		1934	-5.45		1647	10.20
B13	36		1816	0.99		1576	14.07
B14	48		1854	-1.09		1569	14.45
B15	72		1795	2.13		1519	17.18
B16	97		1773	3.33		1317	28.19
term							
	avg	1924.00	1834.10	0.00	2037.43		
				4.02			

meOH + water		**% of Initial Radioactivity**							
origin	Rf <0.1	0.20	Rf 0.3-0.4	0.53 parent	>0.7	time	adjusted pa	solids	abiotic solids
						0.00	#REF!		
2.36	1.15	10.53	0.00	85.62	8.54	0.02	81.54	0.95	
2.12	2.40	22.71	0.00	75.75	9.06	0.08	72.13	1.23	0.72
3.45	1.10	47.55	3.67	44.55	6.90	0.25	42.42	3.61	
4.58	4.90	66.93	5.70	18.75	1.88	0.50	17.85	3.26	
4.41	5.60	72.02	6.36	7.39	0.00	1.00	7.04	6.72	3.20
4.78	8.03	75.62	7.18	3.42	0.00	2.00	3.26	7.01	
2.53	9.82	74.48	5.63	2.40	0.00	4.00	2.28	5.33	0.92
3.24	9.29	71.18	6.77	1.72	0.00	6.00	1.64	7.96	
5.24	8.15	73.21	5.46	1.81	0.00	8.00	1.73	7.20	0.87
4.61	9.14	69.57	5.89	1.55	0.00	10.00	1.48	7.62	
4.15	7.83	73.19	4.90	1.56	0.00	12.00	1.49	7.41	0.87
4.65	10.32	68.49	4.99	1.05	0.00	23.50	1.00	8.85	1.04
3.79	11.40	66.91	4.65	0.95	0.00	36.00	0.91	9.15	
3.81	12.44	63.52	4.40	0.45	0.00	48.00	0.42	7.86	0.61
4.58	17.70	55.27	3.80	0.00	0.00	72.00	0.00	10.69	0.82
3.99	14.02	23.97	1.66	0.00	0.00	100.00	0.00	9.73	0.86
									1.10
									0.749076

Table A2-3 (continued)

BIOTIC TLC Extracts

REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery (+CO2)
B1	0.017	581	27	194	95.03	5.89	1.06	101.98	98
B2	0.083	643	25	226	105.17	5.45	1.23	111.86	104
B3	0.25	615	35	662	100.59	7.63	3.61	111.84	109
B4	0.5	562	54	122	91.93	11.78	0.67	104.37	101
B5	1	532	60	1277	87.02	13.09	6.96	107.07	119
B6	2	535	57	1261	87.51	12.43	6.88	106.82	112
B7	4								
B8	6	463	56	1378	75.73	12.21	7.51	95.46	102
B9	8	484	72	1317	79.17	15.70	7.18	102.05	113
B10	10	479	71	1437	78.35	15.48	7.83	101.67	115
B11	12	470	55	1369	76.88	11.99	7.46	96.34	104
B12	24	460	67	1561	75.24	14.61	8.51	98.36	109
B13	36	463	53	1642	75.73	11.56	8.95	96.24	110
B14	48	468	59	1402	76.55	12.87	7.64	97.06	112
B15	72	429	62	2424	70.17	13.52	13.22	96.91	114
B16	97	375	59	1936	61.34	12.87	10.56	84.76	113
B17							0.00		

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.53 parent	>0.7	% origin	% Rf <0.1	% 0.20	% Rf 0.3-0.4	% 0.53 parent	% >0.7
B1	1.88	2.42	8.79		77.92	8.99	1.79	2.30	8.35	0.00	74.05	8.54
B2	0.78	0.92	19.9		69.78	8.61	0.82	0.97	20.93	0.00	73.39	9.06
B3	1	0.66	44.14	3.65	42.69	6.86	1.01	0.66	44.40	3.67	42.94	6.90
B4		2.69	69.61	5.59	20.07	2.04	0.00	2.47	63.99	5.14	18.45	1.88
B5	1.38	1.7	81.99	6.12	8.8		1.20	1.48	71.35	5.33	7.66	0.00
B6	2.2	7.43	79.52	6.55	4.29		1.93	6.50	69.59	5.73	3.75	0.00
B7												
B8	1.46	10.55	78.73	7.73	1.52		1.11	7.99	59.62	5.85	1.15	0.00
B9	2.68	12.45	77.21	5.27	2.4		2.12	9.86	61.12	4.17	1.90	0.00
B10	2	12.51	76.61	6.28	2.61		1.57	9.80	60.02	4.92	2.04	0.00
B11	1.81	11.39	78.56	5.96	2.28		1.39	8.76	60.39	4.58	1.75	0.00
B12	1.62	15.73	75.27	6.01	1.36		1.22	11.84	56.63	4.52	1.02	0.00
B13	1.47	17.06	75.22	5.33	0.92		1.11	12.92	56.97	4.04	0.70	0.00
B14	1.87	19.08	73.08	5.97			1.43	14.61	55.94	4.57	0.00	0.00
B15	1.77	23.44	69.83	4.96			1.24	16.45	49.00	3.48	0.00	0.00
B16	2.78	30.62	62.02	4.58			1.71	18.78	38.04	2.81	0.00	0.00
B17												

Relative Abundance in Water extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.53 parent	>0.7	% origin	% Rf <0.1	% 0.20	% Rf 0.3-0.4	% 0.53 parent
B1	20.59		19.3		60.11		1.21	0.00	1.14	0.00	3.54
B2	21.46	9.05	24.91		44.59		1.17	0.49	1.36	0.00	2.43
B3	31.96	5.77	41.26		21		2.44	0.44	3.15	0.00	1.60
B4	43.26	6.2	33.65	7.36	9.53		5.09	0.73	3.96	0.87	1.12
B5	39.35	13.04	40.23	7.38			5.15	1.71	5.26	0.97	0.00
B6	38.98	10.96	39.5	10.56			4.85	1.36	4.91	1.31	0.00
B7											
B8	15.98	6.93	66.71	10.38			1.95	0.85	8.15	1.27	0.00
B9	25.96	7.96	57.02	9.06			4.08	1.25	8.95	1.42	0.00
B10	24.21	17.15	49.55	9.09			3.75	2.66	7.67	1.41	0.00
B11	28.19	12.08	52.29	7.44			3.38	1.45	6.27	0.89	0.00
B12	26.6	13.34	47.24	12.82			3.89	1.95	6.90	1.87	0.00
B13	22.67	15.65	50.15	11.52			2.62	1.81	5.80	1.33	0.00
B14	24.47	19.17	52.33	4.04			3.15	2.47	6.73	0.52	0.00
B15	29.4	22.47	42.22	5.9			3.98	3.04	5.71	0.80	0.00
B16	26.38	21.42	48.2	4.01			3.39	2.76	6.20	0.52	0.00
B17											

BIOTIC TLC Extracts

REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery (+CO2)
B1	0.017	675	18	154	110.41	3.93	0.84	115.17	112
B2	0.083	645	30	227	105.50	6.54	1.24	113.28	106
B3	0.25								
B4	0.5	558	56	1075	91.27	12.21	5.86	109.35	106
B5	1	486	55	1187	79.49	11.99	6.47	97.96	110
B6	2	504	72	1311	82.44	15.70	7.15	105.29	111
B7	4	484	72	978	79.17	15.70	5.33	100.20	105
B8	6	487	77	1542	79.66	16.79	8.41	104.86	112
B9	8	485	62	1323	79.33	13.52	7.21	100.07	111
B10	10	444	69	1357	72.62	15.05	7.40	95.07	108
B11	12	501	57	1350	81.95	12.43	7.36	101.74	110
B12	24	469	57	1686	76.71	12.43	9.19	98.34	109
B13	36	472	50	1716	77.20	10.90	9.36	97.46	112
B14	48	436	39	1480	71.32	8.51	8.07	87.89	102
B15/16	72	411	54	1497	67.23	11.78	8.16	87.17	104
B17	97	482	60	1632	78.84	13.09	8.90	100.82	129
							0		
									109
									5.77

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.53 parent	>0.7	% origin	% Rf <0.1	% 0.20	% Rf 0.3-0.4	% 0.53 parent	% >0.7
B1	1.56		9.53		82.22	6.69	1.72	0.00	10.52	0.00	90.78	
B2	1.32	2.35	20.42		68.65	7.26	1.39	2.48	21.54	0.00	72.43	
B3												
B4	1.25	5.24	64.62	5.92	19.04	3.93	1.14	4.78	58.98	5.40	17.38	
B5	0.62	7.52	75.62	8.09	8.14		0.49	5.98	60.11	6.43	6.47	
B6		5.49	82.56	8.88	3.07		0.00	4.53	68.06	7.32	2.53	
B7	0.81	8.1	81.7	7.11	2.27		0.64	6.41	64.68	5.63	1.80	
B8	0.95	8.3	80.49	8.05	2.2		0.76	6.61	64.12	6.41	1.75	
B9	1.36	2.65	88.54	5.27	2.18		1.08	2.10	70.24	4.18	1.73	
B10	1.28	3.78	86.96	6.53	1.46		0.93	2.75	63.15	4.74	1.06	
B11	1.31	2.83	89.82	4.37	1.67		1.07	2.32	73.61	3.58	1.37	
B12	1.72	4.46	87.76	4.66	1.4		1.32	3.42	67.32	3.57	1.07	
B13	1.49	5.05	86.82	5.09	1.56		1.15	3.90	67.03	3.93	1.20	
B14	1.7	6.21	85.63	5.21	1.25		1.21	4.43	61.07	3.72	0.89	0.00
B15/16	2.61	16.57	75.9	4.93	0		1.75	11.14	51.02	3.31	0.00	0.00
B17							0.00	0.00	0.00	0.00	0.00	0.00

Relative Abundance in Water extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.53 parent	>0.7	% origin	% Rf <0.1	% 0.20	% Rf 0.3-0.4	% 0.53 parent
B1			26.7		73.3		0.00	0.00	1.05	0.00	2.88
B2	13.02	13.05	24.25		49.67		0.85	0.85	1.59	0.00	3.25
B3											
B4	23.98	14.79	56.79		4.44		2.93	1.81	6.94	0.00	0.54
B5	16.53	16.95	61.02		5.49		1.98	2.03	7.32	0.00	0.66
B6	17.7	23.41	55.35		3.54		2.78	3.68	8.69	0.00	0.56
B7	12	21.73	62.44		3.82		1.88	3.41	9.80	0.00	0.60
B8	15.82	18.67	62.35		3.16		2.66	3.14	10.47	0.00	0.53
B9	23.64	22.81	45.08	8.48			3.20	3.08	6.10	1.15	0.00
B10	19.83	20.41	55.1	4.66			2.98	3.07	8.29	0.70	0.00
B11	19.81	25.22	49.06	5.91			2.46	3.14	6.10	0.73	0.00
B12	23.14	27.58	49.28				2.88	3.43	6.13	0.00	0.00
B13	24.73	38.25	37.02				2.70	4.17	4.04	0.00	0.00
B14	21.47	39.68	38.85				1.83	3.37	3.30	0.00	0.00
B15/16	18.57	40.58	40.85				2.19	4.78	4.81	0.00	0.00
B17	22.04	49.65	28.3				2.88	6.50	3.70	0.00	0.00

meoh average — **% of Initial Radioactivity**

Time hrs	origin	Rf <0.1	0.20	Rf 0.3-0.4	0.53 parent	>0.7
0.017	1.75	1.15	9.44	0.00	82.41	8.54
0.083	1.11	1.72	21.24	0.00	72.91	9.06
0.25	1.01	0.66	44.40	3.67	42.94	6.90
0.5	0.57	3.63	61.48	5.27	17.91	1.88
1	0.85	3.73	65.73	5.88	7.06	0.00
2	0.96	5.51	68.82	6.53	3.14	0.00
4	0.64	6.41	64.68	5.63	1.80	0.00
6	0.93	7.30	61.87	6.13	1.45	0.00
8	1.60	5.98	65.68	4.18	1.81	0.00
10	1.25	6.27	61.59	4.83	1.55	0.00
12	1.23	5.54	67.00	4.08	1.56	0.00
24	1.27	7.63	61.98	4.05	1.05	0.00
36	1.13	8.41	62.00	3.98	0.95	0.00
48	1.32	9.52	58.51	4.14	0.45	0.00
72	1.50	13.79	50.01	3.40	0.00	0.00
97	0.85	9.39	19.02	1.40	0.00	0.00

water average — **% of Initial Radioactivity**

origin	Rf <0.1	0.20	Rf 0.3-0.4	0.53 parent	>0.7	
0.61	0.00	1.09	0.00	3.21		
1.01	0.67	1.47	0.00	2.84		
2.44	0.44	3.15	0.00	1.60		
4.01	1.27	5.45	0.43	0.83		
3.57	1.87	6.29	0.48	0.33		
3.81	2.52	6.80	0.66	0.28		
1.88	3.41	9.80	0.00	0.60		
2.30	1.99	9.31	0.63	0.27		
3.64	2.17	7.52	1.28	0.00		
3.37	2.86	7.98	1.05	0.00		
2.92	2.29	6.19	0.81	0.00		
3.38	2.69	6.51	0.94	0.00		
2.66	2.99	4.92	0.67	0.00		
2.49	2.92	5.02	0.26	0.00		
3.08	3.91	5.26	0.40	0.00		
3.14	4.63	4.95	0.26	0.00		

Table A2-3 (continued)

ABIOTIC TLC Extracts:
REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Relative Abundance Polar	Relative Abundance Parent	Relative Abundance Nonpolar	% of Initial Radioactivity Polar	% of Initial Radioactivity Parent	% of Initial Radioactivity Nonpolar
A1	0.017													
A2	0.083	652	7	137	106.65	1.53	0.75	108.92		100		0.00	106.65	0.00
A3	0.25			133										
A4	0.5													
A5	1	662	4	1059	108.28	0.87	5.77	114.93		100		0.00	108.28	0.00
A6	2													
A7	4	668	0	228	109.26	0.00	1.24	110.51		100		0.00	109.26	0.00
A8	6													
A9	8	637	1	185	104.19	0.22	1.01	105.42		100				
A10	10													
A11	12	657	2	204	107.46	0.44	1.11	109.01		100		0.00	107.46	0.00
A12	24	643	7	192	105.17	1.53	1.05	107.75						
A13	36	649	5	170										
A14	48	594	9	212	97.16	1.96	1.16	100.28		100		0.00	97.16	0.00
A15	72	646	6	170	105.66	1.31	0.93	107.90		98.69	1.31	0.00	104.28	1.38
A16	97	630	6	143	103.05	1.31	0.78	105.14	0.88	94.36	4.76	0.91	97.24	4.91
A17														
												0.13	104.33	0.90
												0.32	4.73	1.70

ABIOTIC TLC Extracts:
REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.02							
A2	0.08	625	0	127	102.23	0.00	0.69	102.92
A3	0.25	677	3	130				
A4	0.50							
A5	1.00	632	6	113	103.37	1.31	0.62	105.30
A6	2.00							
A7	4.00	648	2	111	105.99	0.44	0.61	107.03
A8	6.00							
A9	8.00	637	4	134	104.19	0.87	0.73	
A10	10.00							
A11	12.00	659	7	116	107.79	1.53	0.63	109.95
A12	24.00	641	10	191	104.85	2.18	1.04	108.07
A13	36.00	661	1	135				
A14	48.00	628	8	11	102.72	1.74	0.06	104.53
A15	72.00	630	11	130	103.05	2.40	0.71	106.16
A16	97.00	667	2	174	109.10	0.44	0.95	110.48
A17								
Average					105.01		1.10	107.31
Std Dev					2.88		1.16	3.27

Appendix 3 — Phase 3 Raw Data

Table A3-1. LAS Raw Data.

Test Chemical:	^{14}C 2-phenyl dodecylbenzene sulfate
RC info:	RC 05030-03; ring ^{14}C; specific activity: 146 mCi/g
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 C, 0.5 mg/L DO
Date:	11/11/2014
Test Chem purity:	100% by Rad-TLC
Test Chemical Rad-TLC Analysis:	75:25:3:2 Chloroform:Methanol:water:formic acid on Analtech Cat 43011
Test Concentration (mg/L):	0.013

Mineralization		***Abiotic***			***Biotic***			
	Time	Direct	Acidified	% CO2	Direct	Acidified	% CO2	% CO2 from
Sample #	(hrs)							Recovery
		dpm/mL	dpm/mL	%	dpm/mL	dpm/mL	%	%
B1	0.01666667				3764	3890	-3.49	-8.37
B2	0.08333333	4297	4302	-1.58	3920	3940	-4.82	-10.32
B3	0.25				3731	3940	-4.82	-6.52
B4	0.5				3853	3754	0.13	-11.53
B5	1	4251	4190	1.06	3655	3829	-1.87	-7.31
B6	2				3819	3730	0.77	-4.37
B7	4	4301	4053	4.30	3754	3883	-3.30	-6.71
B8	6				3761	3614	3.85	-4.49
B9	8	4098	4241	-0.14	3647	3642	3.11	-2.27
B10	10				3684	3647	2.97	-1.59
B11	12		4083	3.59		3316	11.78	-0.64
B12	23.5		4039	4.63		3432	8.69	0.34
B13	36					3203	14.79	15.41
B14	48		4375	-3.31		2272	39.56	40.13
B15	72		4604	-8.72		1260	66.48	59.73
B16	100		4227	0.19		1116	70.31	63.22
term								
	Avg	4237	4235	0.00	3759			
	Stdev	82	169	3.99	87			

Average RAM Distribution for Graphs

origin	0.03	0.12	tail 0.23	Parent 0.33	0.40	0.90	time	adjusted pa	% on Biotic Solids	% on Abiotic Solids
							0.00	99.27		
0.00	0.00	0.00	2.13	104.92			0.02	104.16	1.32	
0.00	0.00	0.00	1.74	106.31			0.08	105.54	1.18	2.64
0.00	0.00	0.00	2.25	103.03			0.25	102.29	1.23	
0.00	0.00	0.00	2.16	107.95			0.50	107.17	1.41	
0.00	0.00	0.00	2.16	103.93			1.00	103.17	1.23	2.92
0.00	0.00	0.00	3.06	99.64			2.00	98.91	1.68	
0.00	0.00	0.00	4.22	101.34			4.00	100.60	1.15	3.64
0.00	0.61	1.88	2.48	98.18			6.00	97.47	1.33	
0.00	0.82	2.60	2.23	95.11			8.00	94.42	1.50	2.55
0.00	1.23	3.11	2.61	92.88			10.00	92.20	1.77	
0.00	1.40	4.04	2.28	91.48			12.00	90.82	1.45	2.71
0.00	1.31	11.89	3.19	77.00			23.50	76.44	2.80	3.18
0.55	11.20	11.40	4.62	52.27			36.00	51.90	4.55	
2.40	12.06	28.01	0.85	5.69	1.48	0.11	48.00	5.64	10.10	3.78
2.28	7.77	22.16	0.00	2.47	0.90	0.07	72.00	2.45	4.61	2.88
5.79	6.26	10.54	0.00	2.69	0.46	0.33	100.00	2.67	10.87	2.61

Table A3-1 (continued)

BIOTIC TLC Extracts

REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery (+CO2)
B1	0.01666667	1310	16	424	104.55	1.70	1.13	107.39	104
B2	0.08333333	1328	17	424	105.99	1.81	1.13	108.93	104
B3	0.25	1344	15	453	107.27	1.60	1.21	110.07	105
B4	0.5	1303	8	562	104.00	0.85	1.50	106.34	106
B5	1	1348	15	453	107.59	1.60	1.21	110.39	109
B6	2	1285	22	670	102.56	2.34	1.78	106.68	107
B7	4	1300	14	433	103.76	1.49	1.15	106.40	103
B8	6	1294	27	542	103.28	2.87	1.44	107.59	111
B9	8	1265	12	557	100.96	1.28	1.48	103.72	107
B10	10	1262	24	732	100.72	2.55	1.95	105.23	108
B11	12	1209	24	465	96.49	2.55	1.24	100.28	112
B12	23.5	1127	65	1052	89.95	6.92	2.80	99.66	108
B13	36	810	85	1429	62.35	8.03	3.37	73.76	89
B14	48	469	148	4023	33.22	13.98	9.50	56.70	96
B15	72	284	137	1727	20.12	12.94	4.08	37.14	104
B16	100	172	132	4010	12.18	12.47	9.47	34.12	104

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

0.03	0.12	tail 0.23	Parent 0.33	0.40	0.90	0.03	0.12	tail 0.23	Parent 0.33	0.40	0.90
		1.88	98.12			0.00	0.00	1.97	102.59		
		1.85	98.15			0.00	0.00	1.96	104.03		
		1.92	98.08			0.00	0.00	2.06	105.21		
		1.89	98.11			0.00	0.00	1.97	102.03		
		1.6	98.4			0.00	0.00	1.72	105.87		
		3.22	96.78			0.00	0.00	3.30	99.26		
		4.14	95.85			0.00	0.00	4.30	99.45		
	2.79	2.73	94.48			0.00	2.88	2.82	97.58		
	3.7	2.03	94.27			0.00	3.74	2.05	95.18		
	4.47	2.84	92.7			0.00	4.50	2.86	93.37		
	6.16	2	91.84			0.00	5.94	1.93	88.62		
1.46	13.22	3.55	81.76			1.31	11.89	3.19	73.54		
12.85	10.64	5.26	71.26			8.01	6.63	3.28	44.43		
13.03	66.57		15.36	4.39	0.65	4.33	22.12	0.00	5.10	1.46	0.22
19.17	66.27		10.38	3.46	0.72	3.86	13.33	0.00	2.09	0.70	0.14
20	51.98		21.72	3.95	2.35	2.44	6.33	0.00	2.65	0.48	0.29

BIOTIC TLC Extracts

REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery (+CO2)
B1	0.01666667	1322	22	565	105.51	2.34	1.50	109.36	106
B2	0.08333333	1371	10	463	109.42	1.06	1.23	111.72	107
B3	0.25	1257	13	474	100.32	1.38	1.26	102.97	98
B4	0.5	1419	20	499	113.25	2.13	1.33	116.71	117
B5	1	1269	16	469	101.28	1.70	1.25	104.23	102
B6	2	1227	24	594	97.93	2.55	1.58	102.06	103
B7	4	1304	17	429	104.08	1.81	1.14	107.03	104
B8	6	1219	27	457	97.29	2.87	1.22	101.38	105
B9	8	1220	18	574	97.37	1.92	1.53	100.81	104
B10	10	1174	25	599	93.70	2.66	1.59	97.95	101
B11	12	1214	23	622	96.89	2.45	1.65	100.99	113
B12	23.5								
B13	36	1012	84	2149	80.77	8.94	5.72	95.43	110
B14	48	453	152	4021	36.16	16.18	10.70	63.03	103
B15	72	278	151	1931	22.19	16.07	5.14	43.39	110
B16	100	179	121	4613	14.29	12.88	12.27	39.44	110
					0				
								Average	105
								Stdev	5.23

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

0	0.03	0.12	tail 0.23	Parent 0.33	0.40	0.90	0	0.03	0.12	tail 0.23	Parent 0.33	0.40	0.90
			2.18	97.82			0.00	0.00	0.00	2.30	103.21		
			1.39	96.61			0.00	0.00	0.00	1.52	105.71		
			2.44	97.56			0.00	0.00	0.00	2.45	97.88		
			2.08	97.92			0.00	0.00	0.00	2.36	110.90		
			2.56	97.44			0.00	0.00	0.00	2.59	98.69		
			2.87	97.13			0.00	0.00	0.00	2.81	95.12		
			3.99	96.01			0.00	0.00	0.00	4.15	99.92		
	1.26	0.91	2.2	95.63			0.00	1.23	0.89	2.14	93.04		
	1.69	1.5	2.48	94.33			0.00	1.65	1.46	2.41	91.85		
	2.62	1.83	2.52	93.03			0.00	2.45	1.71	2.36	87.17		
	2.89	2.2	2.71	92.21			0.00	2.80	2.13	2.63	89.34		
							0.00						
	12.05	10.89	5.5	71.56			0.00	9.73	8.80	4.44	57.80		
1.04	28.43	45.46	4.71	16.2	4.16		0.38	10.28	16.44	1.70	5.86	1.50	0.00
	17.97	67.88		9.15	5		0.00	3.99	15.06	0.00	2.03	1.11	0.00
2.33	23.93	48.87		19.07	3.12	2.67	0.33	3.42	6.98	0.00	2.72	0.45	0.38

TLC Relative Abundance in Water extacts / **% of Initial Radioactivity**

Sample #	Time hrs	origin	0.03	0.12	tail 0.23	Parent Rf.41	origin	0.03	0.12	tail 0.23	Parent Rf.41
B1	0.017					100					1.70
B2	0.083					100					1.81
B3	0.25					100					1.60
B4	0.5					100					0.85
B5	1					100					1.60
B6	2					100					2.34
B7	4					100					1.49
B8	6					100					2.87
B9	8					100					1.28
B10	10					100					2.55
B11	12					100					2.55
B12	23.5					100					6.92
B13	36	7.93	35.82	33.41	9.16	13.69	0.64	2.88	2.68	0.74	1.10
B14	48	11.81	39.91	45.34		2.94	1.65	5.58	6.34	0.00	0.41
B15	72	18.04	21.03	54.56		6.37	2.33	2.72	7.06	0.00	0.82
B16	100	45.74	25.53	28.73			5.70	3.18	3.58	0.00	0.00

BIOTIC TLC Extracts

REP 2 — **Relative Abundance in Water extacts** / **% of Initial Radioactivity**

Sample #	Time hrs	0	0.03	0.12	tail 0.23	Parent Rf.41	origin	0.03	0.12	tail 0.23	Parent Rf.41
B1	0.017					100					2.34
B2	0.083					100					1.06
B3	0.25					100					1.38
B4	0.5					100					2.13
B5	1					100					1.70
B6	2					100					2.55
B7	4					100					1.81
B8	6					100					2.87
B9	8					100					1.92
B10	10					100					2.66
B11	12					100					2.45
B12	23.5					100					0.00
B13	36	5.24	20	52.34	8.79	13.62	0.47	1.79	4.68	0.79	1.22
B14	48	14.87	24.29	68.84			2.41	3.93	11.14	0.00	0.00
B15	72	13.88	30.98	55.14			2.23	4.98	8.86	0.00	0.00
B16	100	40.42	27.11	32.47			5.20	3.49	4.18	0.00	0.00

Methanol Extracts Average RAM Distribution / **Water Extracts Average RAM Distribution**

Time hrs	0.00	0.03	0.12	tail 0.23	Parent 0.33	0.40	0.90	0.00	0.03	0.12	tail 0.23	Parent 0.33
0.016667	0.00	0.00	0.00	2.13	102.90							2.02
0.083333	0.00	0.00	0.00	1.74	104.87							1.44
0.25	0.00	0.00	0.00	2.25	101.54							1.49
0.5	0.00	0.00	0.00	2.16	106.46							1.49
1	0.00	0.00	0.00	2.16	102.28							1.65
2	0.00	0.00	0.00	3.06	97.19							2.45
4	0.00	0.00	0.00	4.22	99.69							1.65
6	0.00	0.61	1.88	2.48	95.31							2.87
8	0.00	0.82	2.60	2.23	93.51							1.60
10	0.00	1.23	3.11	2.61	90.27							2.61
12	0.00	1.40	4.04	2.28	88.98							2.50
23.5	0.00	1.31	11.89	3.19	73.54							3.46
36	0.00	8.87	7.72	3.86	51.12			0.55	2.33	3.68	0.76	1.16
48	0.38	7.30	19.28	0.85	5.48	1.48	0.11	2.03	4.75	8.74	0.00	0.21
72	0.00	3.92	14.20	0.00	2.06	0.90	0.07	2.28	3.85	7.96	0.00	0.41
100	0.33	2.93	6.66	0.00	2.69	0.46	0.33	5.45	3.34	3.88	0.00	0.00

Table A3-1 (continued)

ABIOTIC TLC Extracts:

REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	TLC Relative Abundance in Methanol extacts: tail polar	TLC Relative Abundance in Methanol extacts: Parent Rf.41	TLC Relative Abundance in Methanol extacts: tail nonpolar	% of Initial Radioactivity: tail polar	% of Initial Radioactivity: Parent Rf.41	% of Initial Radioactivity: tail nonpolar
A1	0.017													
A2	0.083	1357	6	835	96.13	0.57	1.97	98.67	2.51	95.53	2.26	2.41	91.83	2.17
A3	0.25													
A4	0.5													
A5	1	1360	5	827	96.34	0.47	1.95	98.77						
A6	2													
A7	4	1461	11	1220	103.50	1.04	2.88	107.42	1.35	96.97	1.69	1.40	100.36	1.75
A8	6													
A9	8	1307	19	1026	92.59	1.79	2.42	96.81		100			92.59	
A10	10													
A11	12	1347	20	1182	95.42	1.89	2.79	100.10						
A12	23.5	1394	11	1270	98.75	1.04	3.00	102.79						
A13	36													
A14	48	1398	17	1090	99.03	1.61	2.57	103.21		100			99.03	
A15	72	1359	16	954	96.27	1.51	2.25	100.04		100			96.27	
A16	100	1423	7	1058	100.81	0.66	2.50	103.96		100			100.81	
A17														

ABIOTIC TLC Extracts:

REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	TLC Relative Abundance: Parent Rf.41		% of Initial Radioactivity: Parent Rf.41		average Parent
A1	0.017												
A2	0.083	1385	9	1402	98.11	0.85	3.31	102.27	100		98.11		94.97
A3	0.25												
A4	0.5												
A5	1	1369	13	1644	96.98	1.23	3.88	102.09	100		96.98		96.98
A6	2												
A7	4	1319	11	1860	93.44	1.04	4.39	98.87	100		93.44		96.90
A8	6												
A9	8	1357	12	1137	96.13	1.13	2.68						92.59
A10	10												
A11	12	1350	13	1110	95.63	1.23	2.62	99.48					
A12	23.5	1390	14	1420	98.47	1.32	3.35	103.14					
A13	36												
A14	48	1305	16	2115	92.45	1.51	4.99	98.95					
A15	72	1345	22	1488	95.28	2.08	3.51	100.87					96.27
A16	100	1365	18	1156	96.70	1.70	2.73	101.13					
A17													
Average					96.78	1.26	2.99	101.09				average	96.08
Std Dev					2.68	0.45	0.78	2.50				Stdev	2.93

Table A3-2. $C_{14}E_2S$ Raw Data.

Test Chemical:	^{14}C C14E2S
RC info:	RC 03031-07A; ^{14}C- adjacent to ethoxylates; specific activity: 28.98 mCi/g
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 C, 0.5 mg/L DO
Date:	11/11/2014
Test Chem purity:	100% by Rad-TLC
Test Chemical Rad-TLC Analysis:	75:25:3:2 Chloroform:Methanol:water:formic acid on Analtech Cat 43011
Test Concentration (mg/L):	0.057

Mineralization

		Abiotic			***Biotic***			
Sample #	Time (hrs)	Direct	Acidified	% CO2	Direct	Acidified	% CO2 by difference	% CO2 from Recovery
		dpm/mL	dpm/mL	%	dpm/mL	dpm/mL	%	%
B1	0.02				3605	3486	4.27	0.39
B2	0.08	3759	3688	-1.27	3464	3303	9.30	4.81
B3	0.25				3412	2357	35.28	26.98
B4	0.50				3399	1867	48.73	42.52
B5	1.0	3721	3581	1.67	3287	1615	55.65	51.62
B6	2.0				3072	1445	60.32	52.03
B7	4.0	3650	3618	0.65	2913	1371	62.35	53.22
B8	6.0				3122	1227	66.31	54.76
B9	8.0	3618	3701	-1.63	2723	1111	69.49	57.68
B10	10.0				2900	1199	67.08	55.80
B11	12.0		3457	5.07		1199	67.08	60.72
B12	23.5		3454	5.15		895	75.42	63.67
B13	36.0					656	81.99	68.08
B14	48.0		3730	-2.43		314	91.38	79.13
B15	72.0		3816	-4.79		354	90.28	81.11
B16	100.0		3730	-2.43		235	93.55	86.59
	avg	3687	3642	0.00	3470			
	std dev	56	118	3.24	248			

Distribution of Radioactivity for Graphs

		Minor Polar Metabolites				Minor Nonpolar Metabolites										
Time (hrs)	origin	0.04	0.12	0.32	Parent 0.40	0.54	0.75	0.83	0.92	adjusted parent 99.77	% on Biotic Solids	% on Abiotic Solids	Average total extract recovery	Polar Metabolites	Minor Nonpolar Metabolites	All metabolites
0.02	0.00	0.00	0.00	1.05	95.74	0.00	0.00	0.98	0.00	93.24	1.84		99.61	1.05	0.98	2.03
0.08	7.40	0.95	0.26	1.63	77.86	0.52	0.00	1.78	0.14	75.83	4.65	0.78	95.19	10.24	2.43	12.68
0.25	14.28	2.48	1.41	1.73	40.35	0.25	0.19	2.36	0.24	39.30	9.71		73.02	19.91	3.04	22.96
0.5	16.56	2.26	2.34	1.93	15.63	0.93	0.32	1.43	0.22	15.22	15.76		57.48	23.09	2.90	26.00
1	16.74	3.06	2.70	1.68	5.93	0.76	0.38	2.55	0.34	5.77	13.92	0.31	48.38	24.18	4.02	28.20
2	18.73	3.66	2.65	1.99	3.89	0.51	0.21	1.52	0.12	3.78	14.51		47.97	27.02	2.36	29.38
4	17.52	2.84	1.68	2.28	3.11	0.60	0.20	1.28	0.35	3.03	16.70	1.31	46.78	24.31	2.42	26.73
6	17.91	3.34	1.52	1.50	2.42	0.50	0.07	1.53	0.16	2.35	16.13		45.24	24.27	2.26	26.54
8	18.15	2.76	0.84	1.50	1.65	0.55	0.12	2.02	0.22	1.61	14.35	0.88	42.32	23.24	2.92	26.16
10	20.15	2.16	0.81	3.03	0.83	0.35	0.05	1.26	0.12	0.81	15.29		44.20	26.15	1.78	27.93
12	15.41	1.53	3.01	2.76	0.74	0.32	0.06	1.57	0.14	0.72	13.52	1.01	39.28	22.72	2.09	24.81
24	7.26	0.87	1.59	1.24	0.28	0.17	0.00	0.40	0.12	0.27	12.46	1.05	36.33	10.96	0.69	11.65
36	12.19	1.48	3.23	1.53	0.47	0.24	0.00	1.05	0.14	0.45	11.44		31.92	18.44	1.43	19.86
48	8.38	0.47	4.91	0.82	0.34	0.24	0.00	0.29	0.05	0.33	5.36	1.25	20.87	14.58	0.59	15.17
72	8.07	0.96	2.44	0.83	0.38	0.26	0.00	0.32	0.05	0.37	5.57	1.35	18.89	12.31	0.63	12.94
100.00	0.15	0.12	0.61	0.66	0.26	0.19	0.00	0.23	0.05	0.26	2.96	1.41	13.41	1.54	0.47	2.00

Table A3-2 (continued)

BIOTIC TLC Extracts

REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery with CO2
B1	0.017	1125	64	720	92.68	7.03	1.98	101.68	105.96
B2	0.083	1011	119	1712	83.29	13.07	4.70	101.06	110.36
B3	0.25	527	177	3761	43.41	19.44	10.33	73.18	108.46
B4	0.5	256	219	5378	21.09	24.05	14.77	59.91	108.64
B5	1	165	232	4972	13.59	25.48	13.65	52.73	108.38
B6	2	131	245	4635	10.79	26.91	12.73	50.43	110.75
B7	4	120	192	5205	9.89	21.09	14.29	45.27	107.62
B8	6	106	206	5661	8.73	22.63	15.55	46.90	113.21
B9	8	107	202	4519	8.81	22.19	12.41	43.41	112.90
B10	10	91	220	5383	7.50	24.16	14.78	46.44	113.52
B11	12	98	186	5002	8.07	20.43	13.74	42.24	109.31
B12	23.5	71	164	4539	5.85	18.01	12.46	36.33	111.75
B13	36	55	140	4327	4.53	15.38	11.88	31.79	113.78
B14	48	43	111	2319	3.54	12.19	6.37	22.10	113.48
B15	72	40	99	2124	3.30	10.87	5.83	20.00	110.28
B16	100	36	70	1066	2.97	7.69	2.93	13.58	107.13

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf 0.04	0.12	0.32	Parent 0.40	0.54	0.75	0.83	0.92	origin	Rf 0.04	0.12	0.32	Parent 0.40	0.54	0.75	0.83	0.92
B1					99.19			0.81		0.00	0.00	0.00	0.00	91.93	0.00	0.00	0.75	0.00
B2				1.73	96.09			1.86	0.33	0.00	0.00	0.00	1.44	80.03	0.00	0.00	1.55	0.27
B3	1.09	0.74	1.44	3.23	87.09	1.15	0.89	3.26	1.11	0.47	0.32	0.63	1.40	37.81	0.50	0.39	1.42	0.48
B4	1.82	1.16	3.88	6.49	75.27	3.14	1.68	5.25	1.3	0.38	0.24	0.82	1.37	15.87	0.66	0.35	1.11	0.27
B5	3.69	2.25	8.23	7.01	49.83	6.01	2.22	17.11	3.65	0.50	0.31	1.12	0.95	6.77	0.82	0.30	2.33	0.50
B6	3.82	1.53	10.11	10.6	48.32	6.55	1.57	15.82	1.67	0.41	0.17	1.09	1.14	5.21	0.71	0.17	1.71	0.18
B7	2.75	2.38	10.43	12.77	45.86	7.92	1.45	10.73	5.7	0.27	0.24	1.03	1.26	4.53	0.78	0.14	1.06	0.56
B8	3.99	1.97	10.78	11.34	40.89	7.85	1.61	19	2.58	0.35	0.17	0.94	0.99	3.57	0.69	0.14	1.66	0.23
B9	4.33	5.37	10.16	7.4	27.3	8.42	2.66	30.27	4.09	0.38	0.47	0.90	0.65	2.41	0.74	0.23	2.67	0.36
B10	3.62	1.29	11.24	50.11	10.36	4.65	1.43	15.3	1.98	0.27	0.10	0.84	3.76	0.78	0.35	0.11	1.15	0.15
B11	3.62	1.29	11.24	50.11	10.36	4.65	1.43	15.3	1.98	0.29	0.10	0.91	4.05	0.84	0.38	0.12	1.24	0.16
B12	4.35	3.8	16.27	42.29	9.55	5.85	0	13.74	4.16	0.25	0.22	0.95	2.47	0.56	0.34	0.00	0.80	0.24
B13	4.35	3.8	16.27	42.29	9.55	5.85	0	13.74	4.16	0.20	0.17	0.74	1.92	0.43	0.27	0.00	0.62	0.19
B14	7.02	4.2	25.45	33.99	9.04	8.5	0	8.73	3.07	0.25	0.15	0.90	1.20	0.32	0.30	0.00	0.31	0.11
B15	7.02	4.2	25.45	33.99	9.04	8.5	0	8.73	3.07	0.23	0.14	0.84	1.12	0.30	0.28	0.00	0.29	0.10
B16	7.02	4.2	25.45	33.99	9.04	8.5	0	8.73	3.07	0.21	0.12	0.75	1.01	0.27	0.25	0.00	0.26	0.09

BIOTIC TLC Extracts

REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery with CO2
B1	0.017	1094	52	622	90.12	5.71	1.71	97.54	101.82
B2	0.083	891	103	1675	73.40	11.31	4.60	89.31	98.61
B3	0.25	562	159	3310	46.30	17.46	9.09	72.85	108.13
B4	0.5	241	168	6097	19.85	18.45	16.74	55.05	103.78
B5	1	173	142	5166	14.25	15.60	14.19	44.03	99.69
B6	2	112	182	5931	9.23	19.99	16.29	45.50	105.82
B7	4	113	181	6956	9.31	19.88	19.10	48.29	110.64
B8	6	82	183	6090	6.76	20.10	16.72	43.58	109.89
B9	8	80	167	5936	6.59	18.34	16.30	41.23	110.73
B10	10	79	179	5751	6.51	19.66	15.79	41.96	109.04
B11	12	74	154	4846	6.10	16.92	13.31	36.32	103.39
B12	23.5								
B13	36	57	149	4003	4.70	16.37	10.99	32.05	114.04
B14	48	27	119	1583	2.22	13.07	4.35	19.64	111.02
B15	72	34	88	1936	2.80	9.67	5.32	17.78	108.06
B16	100	19	79	1089	1.57	8.68	2.99	13.23	106.78
								Average	108.61
								Std Dev	3.93

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	Rf 0.04	0.12	0.32	Parent 0.40	0.54	0.75	0.83	0.92	origin	Rf 0.04	0.12	0.32	Parent 0.40	0.54	0.75	0.83	0.92
B1				2.33	96.32			1.35		0.00	0.00	0.00	2.10	86.81	0.00	0.00	1.22	0.00
B2				2.47	94.8			2.73		0.00	0.00	0.00	1.81	69.58	0.00	0.00	2.00	0.00
B3	0.97	0.82	2	4.46	84.61			7.14		0.45	0.38	0.93	2.06	39.17	0.00	0.00	3.31	0.00
B4	2.2		4.45	12.54	66.93	1.76	1.45	8.83	0.87	0.44	0.00	0.88	2.49	13.29	0.35	0.29	1.75	0.17
B5	2.36	3.33	8.23	16.9	35.65	4.92	3.21	19.4	1.25	0.34	0.47	1.17	2.41	5.08	0.70	0.46	2.76	0.18
B6	3.31	1.64	11.31	30.65	27.72	3.49	2.61	14.39	0.75	0.31	0.15	1.04	2.83	2.56	0.32	0.24	1.33	0.07
B7	3.92	1.8	10.97	35.5	18.19	4.45	2.66	16.04	1.36	0.36	0.17	1.02	3.30	1.69	0.41	0.25	1.49	0.13
B8	4.58	2.35	13	29.88	18.71	4.73		20.69	1.48	0.31	0.16	0.88	2.02	1.26	0.32	0.00	1.40	0.10
B9	4.31	2.25	11.86	35.5	13.52	5.54		20.96	1.3	0.28	0.15	0.78	2.34	0.89	0.37	0.00	1.38	0.09
B10	4.31	2.25	11.86	35.5	13.52	5.54		20.96	1.3	0.28	0.15	0.77	2.31	0.88	0.36	0.00	1.36	0.08
B11	4.99	3.01	12.61	24.34	10.65	4.49		31.4	1.89	0.30	0.18	0.77	1.48	0.65	0.27	0.00	1.91	0.12
B12										0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
B13	4.99	3.01	12.61	24.34	10.65	4.49		31.4	1.89	0.23	0.14	0.59	1.14	0.50	0.21	0.00	1.47	0.09
B14	6.14	7.51	29.23	19.51	16.52	8.49		12.6		0.14	0.17	0.65	0.43	0.37	0.19	0.00	0.28	0.00
B15	6.14	7.51	29.23	19.51	16.52	8.49		12.6		0.17	0.21	0.82	0.55	0.46	0.24	0.00	0.35	0.00
B16	6.14	7.51	29.23	19.51	16.52	8.49		12.6		0.10	0.12	0.46	0.31	0.26	0.13	0.00	0.20	0.00

Relative Abundance in Water extacts / **% of Initial Radioactivity**

Sample #	Time hrs	origin	Rf 0.04	0.12	Parent 0.40	0.54	origin	Rf 0.04	0.12	Parent 0.40	0.54
B1	0.017				100		0	0	0	7.03	0
B2	0.083	67.44	7.34	3.99	21.22		8.82	0.96	0.52	2.77	0
B3	0.25	75.22	10.25	6.56	7.98		14.62	1.99	1.28	1.55	0
B4	0.5	81.63	7.24	6.05	5.08		19.64	1.74	1.46	1.22	0.00
B5	1	83.85	12.79	3.36			21.37	3.26	0.86	0.00	0.00
B6	2	82.89	13.49	3.63			22.31	3.63	0.98	0.00	0.00
B7	4	82.8	11.01	6.19			17.46	2.32	1.31	0.00	0.00
B8	6	83.01	11.61	5.38			18.78	2.63	1.22	0.00	0.00
B9	8	89.4	10.6				19.84	2.35	0.00	0.00	0.00
B10	10	93.05	6.95				22.49	1.68	0.00	0.00	0.00
B11	12	85.85	3.95	10.2			17.54	0.81	2.08	0.00	0.00
B12	23.5	79.19	8.46	12.34			14.27	1.52	2.22	0.00	0.00
B13	36	80.92	7.05	12.03			12.44	1.08	1.85	0.00	0.00
B14	48	72.53		27.47			8.84	0.00	3.35	0.00	0.00
B15	72	55.9	14.49	29.61			6.08	1.58	3.22	0.00	0.00
B16	100						0.00	0.00	0.00	0.00	0.00
							0.00	0.00	0.00	0.00	0.00

Relative Abundance in Water extacts / **% of Initial Radioactivity**

Sample #	Time hrs	origin	Rf 0.04	0.12	Parent 0.40	0.54	origin	Rf 0.04	0.12	Parent 0.40	0.54
B1	0.017				100		0	0	0	5.71	0
B2	0.083	52.96	8.36		29.47	9.21	5.99	0.95	0.00	3.33	1.04
B3	0.25	74.55	13.01		12.43		13.02	2.27	0.00	2.17	0
B4	0.5	68.6	13.78	8.27	4.75	4.6	12.66	2.54	1.53	0.88	0.85
B5	1	72.26	13.36	14.38			11.27	2.08	2.24	0.00	0.00
B6	2	72.22	16.87	10.91			14.44	3.37	2.18	0.00	0.00
B7	4	85.17	14.83				16.93	2.95	0.00	0.00	0.00
B8	6	81.5	18.5				16.38	3.72	0.00	0.00	0.00
B9	8	86.15	13.85				15.80	2.54	0.00	0.00	0.00
B10	10	87.76	12.24				17.25	2.41	0.00	0.00	0.00
B11	12	75.03	11.64	13.32			12.69	1.97	2.25	0.00	0.00
B12	23.5						0.00	0.00	0.00	0.00	0.00
B13	36	70.33	9.6	20.06			11.51	1.57	3.28	0.00	0.00
B14	48	57.58	4.79	37.63			7.53	0.63	4.92	0.00	0.00
B15	72	100					9.67	0.00	0.00	0.00	0.00
B16	100						0.00	0.00	0.00	0.00	0.00
							0.00	0.00	0.00	0.00	0.00

MEOH Extract Average RAM Distribution

Sample #	Time (hrs)	origin	Rf 0.04	0.12	0.32	Parent 0.40	0.54	0.75	0.83	0.92
	0									
B1	0.01666667	0.00	0.00	0.00	1.05	89.37	0.00	0.00	0.98	0.00
B2	0.08333333	0.00	0.00	0.00	1.63	74.81	0.00	0.00	1.78	0.14
B3	0.25	0.46	0.35	0.78	1.73	38.49	0.25	0.19	2.36	0.24
B4	0.5	0.41	0.12	0.85	1.93	14.58	0.51	0.32	1.43	0.22
B5	1	0.42	0.39	1.15	1.68	5.93	0.76	0.38	2.55	0.34
B6	2	0.36	0.16	1.07	1.99	3.89	0.51	0.21	1.52	0.12
B7	4	0.32	0.20	1.03	2.28	3.11	0.60	0.20	1.28	0.35
B8	6	0.33	0.17	0.91	1.50	2.42	0.50	0.07	1.53	0.16
B9	8	0.33	0.31	0.84	1.50	1.65	0.55	0.12	2.02	0.22
B10	10	0.28	0.12	0.81	3.03	0.83	0.35	0.05	1.26	0.12
B11	12	0.30	0.14	0.84	2.76	0.74	0.32	0.06	1.57	0.14
B12	23.5	0.13	0.11	0.48	1.24	0.28	0.17	0.00	0.40	0.12
B13	36	0.22	0.16	0.66	1.53	0.47	0.24	0.00	1.05	0.14
B14	48	0.19	0.16	0.78	0.82	0.34	0.24	0.00	0.29	0.05
B15	72	0.20	0.17	0.83	0.83	0.38	0.26	0.00	0.32	0.05
B16	100	0.15	0.12	0.61	0.66	0.26	0.19	0.00	0.23	0.05

Water Extract Average RAM Distribution

origin	Rf 0.04	0.12	Parent 0.40	0.54
0.00	0.00	0.00	6.37	0.00
7.40	0.95	0.26	3.05	0.52
13.82	2.13	0.64	1.86	0.00
16.15	2.14	1.49	1.05	0.42
16.32	2.67	1.55	0.00	0.00
18.37	3.50	1.58	0.00	0.00
17.20	2.64	0.65	0.00	0.00
17.58	3.17	0.61	0.00	0.00
17.82	2.45	0.00	0.00	0.00
19.87	2.04	0.00	0.00	0.00
15.12	1.39	2.17	0.00	0.00
7.13	0.76	1.11	0.00	0.00
11.98	1.33	2.57	0.00	0.00
8.18	0.31	4.13	0.00	0.00
7.87	0.79	1.61	0.00	0.00
0.00	0.00	0.00	0.00	0.00

Table A3-2 (continued)

ABIOTIC TLC Extracts:
REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	**TLC**	**Abundance** Parent 0.40	**% of Initial Radioactivity** Parent 0.40
A1	0.017										
A2	0.083	1242	2		102.32	0.22		102.54		100	102.32
A3	0.25										
A4	0.5										
A5	1	1304	12	227	107.42	1.32	0.62	109.36			
A6	2										
A7	4	1260	8	450	103.80	0.88	1.24	105.91		100	103.80
A8	6										
A9	8	1288	5	324	106.11	0.55	0.89	107.54			
A10	10										
A11	12	1202	1	244	99.02	0.11	0.67	99.80			
A12	23.5	1198	10	579	98.69	1.10	1.59	101.38		100	98.69
A13	36										
A14	48	1233	11	488	101.57	1.21	1.34	104.12		100	101.57
A15	72	1316	5	457	108.41	0.55	1.25	110.22			
A16	100	1284	13	390	105.78	1.43	1.07	108.27		100	105.78

ABIOTIC TLC Extracts:
REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	**TLC** Parent 0.40	**Abundance** 0.83	**% of Initial Radioactivity** Parent 0.40	0.83
A1	0.017											
A2	0.083	1187	0	284	97.78	0.00	0.78	98.56				
A3	0.25											
A4	0.5											
A5	1	1293	7		106.52	0.77	0.00	107.29				
A6	2											
A7	4	1321	7	501	108.82	0.77	1.38	110.97				
A8	6											
A9	8	1322	10	320	108.91	1.10	0.88	110.88	100		108.91	
A10	10											
A11	12	1253	4	489	103.22	0.44	1.34	105.00				
A12	23.5	1060	5	186	87.32	0.55	0.51					
A13	36											
A14	48	1213	8	420	99.93	0.88	1.15	101.96	100		99.93	
A15	72	1216	4	526	100.17	0.44	1.44	102.06	100		100.17	
A16	100	1244	7	634	102.48	0.77	1.74	104.99	98.44	1.56	100.88	1.60
Total average					102.7	0.7	1.1	105.3			102.4	
Total StDev					5.13	0.40	0.43	3.79			3.05	

Table A3-3. $C_{15}E_3S$ Raw Data.

Test Chemical:	[14]C C15E3S
RC info:	B9612-05; [14]C in 1st and 3rd ethoxylates; specific activity: 9.3 mCi/g
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 C, 0.5 mg/L DO
Date:	11/11/2014
Test Chem purity:	100% by Rad-TLC
Test Chemical Rad-TLC Analysis:	75:25:3:2 Chloroform:Methanol:water:formic acid on Analtech Cat 43011
Test Concentration (mg/L):	0.185

Mineralization		***Abiotic***			***Biotic***			
	Time	Direct	Acidified	% CO2	Direct	Acidified	% CO2	% CO2
Sample #	(hrs)						by difference	from Recovery
		dpm/mL	dpm/mL	%	dpm/mL	dpm/mL	%	%
B1	0.01666667				3666	3524	7.80	-0.50
B2	0.08333333	3893	3862	-1.04	3789	3817	0.14	13.91
B3	0.25				3874	3755	1.76	2.94
B4	0.5				3866	3740	2.15	-1.55
B5	1	3897	4007	-4.83	3799	3748	1.94	-0.88
B6	2				3754	3765	1.50	-0.24
B7	4	3780	3786	0.95	3918	3823	-0.02	-0.96
B8	6				3942	3797	0.66	-3.07
B9	8	3787	3889	-1.75	3901	3773	1.29	-1.27
B10	10				3855	3817	0.14	-3.82
B11	12		3726	2.52		3756	1.73	-2.83
B12	23.5		3549	7.15		3570	6.60	2.26
B13	36					3651	4.48	0.29
B14	48		3847	-0.65		3819	0.08	4.66
B15	72		3884	-1.62		3581	6.31	5.81
B16	100		3850	-0.73		3208	16.07	7.00
	avg	3839	3822	0	3836			
	stdev	56	120	3	59			

Distribution of Radioactivity for Graphs

Time (hrs)	origin	Shoulder	Polar 0.10	Parent RF 0.51	adjusted parent	% biotic Solids	% abiotic solids	Total Metabolites
					98.62			
0.02	0.00	0.00	6.87	92.28	93.85	0.98		6.87
0.08	0.00	0.00	14.70	70.18	71.37	0.73	0.80	14.70
0.25	0.00	0.00	34.14	58.90	59.90	0.75		34.14
0.5	0.61	0.46	61.70	37.24	37.87	0.92		62.77
1	0.60	2.06	84.14	14.56	14.81	1.10	0.90	86.80
2	0.66	1.44	91.67	5.19	5.28	1.26		93.78
4	0.72	2.46	92.44	2.74	2.78	2.62	0.99	95.61
6	1.02	3.33	94.66	2.72	2.77	1.34	0.00	99.01
8	0.74	5.62	91.19	2.17	2.20	1.55	0.85	97.55
10	1.00	7.07	91.55	2.29	2.32	1.62		99.63
12	0.71	8.36	90.05	1.97	2.01	1.38	1.08	99.12
24	2.10	15.42	76.80	1.39	1.42	1.46	0.95	94.32
36	2.26	16.79	78.22	0.43	0.44	1.52		97.27
48	2.66	24.77	65.43	0.00	0.00	1.74	1.17	92.85
72	3.14	31.30	57.86	0.00	0.00	1.44	1.48	92.30
100	3.93	34.82	52.02	0.00	0.00	1.85	1.10	90.76

Table A3-3 (continued)

BIOTIC TLC Extracts

REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery +CO2
B1	0.02	1274	17	349	99.99	1.78	0.91	102.69	110.49
B2	0.08	1077	24	333	84.53	2.51	0.87	87.91	88.05
B3	0.25	1179	36	288	92.54	3.77	0.75	97.06	98.82
B4	0.5	1206	62	334	94.66	6.49	0.87	102.02	104.17
B5	1	1188	90	402	93.24	9.42	1.05	103.71	105.66
B6	2	1155	87	490	90.65	9.10	1.28	101.04	102.54
B7	4	1121	88	478	87.99	9.21	1.25	98.45	98.43
B8	6	1191	93	449	93.48	9.73	1.17	104.39	105.05
B9	8	1086	126	583	85.24	13.19	1.53	99.95	101.24
B10	10	1136	128	537	89.16	13.40	1.40	103.96	104.10
B11	12	1136	110	551	89.16	11.51	1.44	102.12	103.85
B12	23.5	977	161	521	76.68	16.85	1.36	94.90	101.49
B13	36	1064	155	519	83.51	16.22	1.36	101.09	105.57
B14	48	976	220	651	76.60	23.02	1.70	101.33	101.42
B15	72	816	291	450	64.05	30.45	1.18	95.68	101.99
B16	100	863	253	723	67.74	26.48	1.89	96.10	112.17

TLC Relative % in Methanol Exacts / **% RAM Distribution** (REP 1)

Sample #	TLC Origin	TLC Shoulder >0.1	TLC Polar 0.10	TLC Parent RF 0.51	RAM Origin	RAM Shoulder	RAM Polar 0.10	RAM Parent RF 0.51
B1			6.94	93.06		0.00	6.94	93.05
B2			16.39	83.61		0.00	13.85	70.68
B3			36.89	63.11		0.00	34.14	58.40
B4			60.07	39.93		0.00	56.86	37.80
B5			82.92	17.08		0.00	77.32	15.93
B6		1.58	92.63	5.79		1.43	83.97	5.25
B7		2.32	94.69	2.99		2.04	83.31	2.63
B8		2.54	94.36	3.1		2.37	88.21	2.90
B9		2.77	94.47	2.76		2.36	80.52	2.35
B10		3.97	93.47	2.56		3.54	83.34	2.28
B11		6.54	91.21	2.26		5.83	81.33	2.02
B12		8.62	89.38	2		6.61	68.54	1.53
B13		13.22	86.78	0		11.04	72.47	0.00
B14		18.76	81.24	0		14.37	62.23	0.00
B15		27.86	72.14	0		17.84	46.20	0.00
B16		30.6	69.4	0		20.73	47.01	0.00

TLC Relative % in Water Exacts / **% RAM Distribution** (REP 1)

Sample #	TLC Origin	TLC Shoulder	TLC Polar 0.10	TLC Parent RF 0.51	RAM Origin	RAM Shoulder	RAM Polar 0.10	RAM Parent RF 0.51
B1								
B2								
B3								
B4								
B5	8.29		91.71		0.78	0.00	8.64	
B6	8.29		91.71		0.75	0.00	8.35	
B7	7.98		92.02		0.73	0.00	8.47	
B8	7.98		92.02		0.78	0.00	8.96	
B9	4.46	26.16	69.38		0.59	3.45	9.15	
B10	7.13	30.09	62.78		0.96	4.03	8.41	
B11	4.99	31.99	63.02		0.57	3.68	7.25	
B12	5.44	51.6	42.96		0.92	8.69	7.24	
B13	4.17	48.27	47.56		0.68	7.83	7.71	
B14	1.67	67.45	30.88		0.38	15.53	7.11	
B15	1.62	55.77	42.61		0.49	16.98	12.98	
B16	1.76	73.52	24.72		0.47	19.47	6.55	

BIOTIC TLC Extracts

REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery +CO2
B1	0.02	1226	10	400	96.23	1.05	1.05	98.32	106.12
B2	0.08	1046	15	228	82.10	1.57	0.60	84.27	84.40
B3	0.25								
B4	0.5	1209	50	367	94.89	5.23	0.96	101.09	103.24
B5	1	1132	77	436	88.85	8.06	1.14	98.05	99.99
B6	2	1139	84	477	89.40	8.79	1.25	99.44	100.93
B7	4	1121	110	1522	87.99	11.51	3.98	103.48	103.46
B8	6	1132	109	576	88.85	11.41	1.51	101.76	102.42
B9	8	1107	135	601	86.89	14.13	1.57	102.59	103.87
B10	10	1135	122	698	89.08	12.77	1.83	103.68	103.81
B11	12	1157	109	506	90.81	11.41	1.32	103.54	105.27
B12	23.5	1011	188	593	79.35	19.67	1.55	100.58	107.18
B13	36	950	211	640	74.56	22.08	1.67	98.32	102.80
B14	48	781	251	679	61.30	26.27	1.78	89.34	89.43
B15	72	822	253	651	64.52	26.48	1.70	92.70	99.01
B16	100	769	265	692	60.36	27.73	1.81	89.90	105.97
								Avg	102.03
								Stdev	5.70

TLC Relative % in Methanol Extacts / % RAM Distribution (REP 2)

Sample #	TLC Origin	TLC Shoulder	TLC Polar 0.10	TLC Parent RF 0.51	RAM Origin	RAM Shoulder	RAM Polar 0.10	RAM Parent RF 0.51
B1			6.34	93.66		0.00	6.10	90.13
B2			16.97	83.03		0.00	13.93	68.17
B3								
B4		0.97	61.07	37.97		0.92	57.95	36.03
B5		0.72	84.43	14.85		0.64	75.02	13.19
B6		0.99	93.26	5.75		0.89	83.37	5.14
B7		2.06	94.72	3.23		1.81	83.34	2.84
B8		1.82	95.32	2.86		1.62	84.69	2.54
B9		2.96	94.76	2.28		2.57	82.33	1.98
B10	0.65	3.77	93.01	2.57		3.36	82.86	2.29
B11	0.8	5.01	92.07	2.13	0.73	4.55	83.61	1.93
B12	1.42	7.65	89.35	1.58	1.13	6.07	70.90	1.25
B13	1.3	7.49	90.05	1.16	0.97	5.58	67.14	0.86
B14	1.14	13.54	85.32		0.70	8.30	52.30	0.00
B15	1.4	21.04	77.56		0.90	13.57	50.04	0.00
B16	1.29	25.55	73.16		0.78	15.42	44.16	0.00
						0.00	0.00	0.00

TLC Relative % in Water Extracts / **% RAM Distribution** (REP 2)

Sample #	TLC Origin	TLC Shoulder	TLC Polar 0.10	TLC Parent RF 0.51	RAM Origin	RAM Shoulder	RAM Polar 0.10	RAM Parent RF 0.51
B1			33.79	66.21			0.35	0.69
B2			51.44	48.56			0.81	0.76
B3							0.00	0.00
B4	11.73		81.98	6.29	0.61		4.29	0.33
B5	5.09	43.26	90.65		0.41	3.49	7.30	0.00
B6	6.52	6.46	87.03		0.57	0.57	7.65	
B7	6.1	9.2	84.7		0.70	1.06	9.75	
B8	11.13	23.45	65.41		1.27	2.67	7.46	
B9	6.38	20.25	73.37		0.90	2.86	10.37	
B10	8.23	25.19	66.58		1.05	3.22	8.50	
B11	7.35	23.28	69.37		0.84	2.66	7.91	
B12	16.73	48.09	35.19		3.29	9.46	6.92	
B13	17.43	41.3	41.28		3.85	9.12	9.12	
B14	18.77	43.14	35.09		4.93	11.33	9.22	
B15	21.86	53.61	24.53		5.79	14.19	6.49	
B16	26.64	50.57	22.79		7.39	14.02	6.32	

MeOH Extract Average RAM Distribution / **Water Extract Average RAM Distribution**

Sample #	Time hrs	MeOH Shoulder	MeOH Polar 0.10	MeOH Parent RF 0.51	Water origin	Water Shoulder	Water Polar 0.10	Water Parent RF 0.51
B1	0.017	0.00	6.52	91.59			0.35	0.69
B2	0.083	0.00	13.89	69.42			0.81	0.76
B3	0.25	0.00	34.14	58.40				0.50
B4	0.5	0.46	57.41	36.91	0.61		4.29	0.33
B5	1	0.32	76.17	14.56	0.60	1.74	7.97	0.00
B6	2	1.16	83.67	5.19	0.66	0.28	8.00	
B7	4	1.93	83.33	2.74	0.72	0.53	9.11	
B8	6	2.00	86.45	2.72	1.02	1.34	8.21	
B9	8	2.47	81.43	2.17	0.74	3.16	9.76	
B10	10	3.45	83.10	2.29	1.00	3.62	8.46	
B11	12	5.19	82.47	1.97	0.71	3.17	7.58	
B12	23.5	6.34	69.72	1.39	2.10	9.08	7.08	
B13	36	8.31	69.81	0.43	2.26	8.47	8.41	
B14	48	11.34	57.27	0.00	2.66	13.43	8.16	
B15	72	15.71	48.12	0.00	3.14	15.59	9.74	
B16	100	18.07	45.58	0.00	3.93	16.75	6.43	

Table A3-3 (continued)

ABIOTIC TLC Extracts:
REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.02							
A2	0.08	1295	1	367	101.64	0.10	0.96	102.71
A3	0.25							
A4	0.5							
A5	1	1235	7	374	96.93	0.73	0.98	98.64
A6	2							
A7	4	1299	5	438	101.96	0.52	1.15	103.63
A8	6							
A9	8	1265	3	326	99.29	0.31	0.85	100.45
A10	10							
A11	12	1250	6	442	98.11	0.63	1.16	99.89
A12	23.5	1305	6	447	102.43	0.63	1.17	104.22
A13	36							
A14	48	1359	2	508	106.67	0.21	1.33	108.20
A15	72	1260	11	616	98.90	1.15	1.61	101.66
A16	100	1286	7	490	100.94	0.73	1.28	102.95

TLC Relative Abundance in Methanol Extacts and % Initial RAM

Sample #	Parent RF 0.51	Parent RF 0.51
A2	100	101.64
A7	100	101.96
A9	100	99.29
A16	100	100.94

ABIOTIC TLC Extracts:
REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.02							
A2	0.08	1168	5	248	91.67	0.52	0.65	92.85
A3	0.25							
A4	0.5							
A5	1	1255	2	316	98.50	0.21	0.83	99.54
A6	2							
A7	4	1252	11	319	98.27	1.15	0.83	100.25
A8	6						0.00	
A9	8	1317	0	191				
A10	10							
A11	12	1254	1	383	98.42	0.10	1.00	99.53
A12	23.5	1097	3	281	86.10	0.31	0.74	87.15
A13	36							
A14	48	1222	13	388	95.91	1.36	1.02	98.29
A15	72	1258	2	515	98.74	0.21	1.35	100.30
A16	100	1238	11	348	97.17	1.15	0.91	99.23
Average					98.33	0.59	0.99	99.97
Std Dev					4.35	0.39	0.34	4.47

Relative Abundance in Methanol Extacts and % Initial RAM

Sample #	Parent RF 0.51	Parent RF 0.51
A2	100	91.67
A7	100	98.27
A12	100	86.10
A14	100	95.91
	Avg	96.97
	Stdev	5.21

Table A3-4. $C_{12}S$ Raw Data.

Test Chemical:	^{14}C C12S
RC info:	RC 14525.00; 1C labeled specific activity: 191 mCi/g
Study Number:	
Test:	Sewer Water Die Away
Test System:	100% Influent from FFWWTP, 15 C, 0.5 mg/L DO
Date:	2/3/2015
Test Chem purity:	93.22% by Rad-TLC
Test Chemical Rad-TLC Analysis:	75:25:3:2 Chlor:Meth:Water:formic on Analtech G Catalog 16011
Test Concentration (mg/L):	0.008

Mineralization		***Abiotic***			***Biotic***		
Sample #	Time (hrs)	Direct	Acidified	% CO2	Direct	Acidified	% CO2
		dpm/mL	dpm/mL	%	dpm/mL	dpm/mL	%
B1	0.01666667				3675	2894	21.92
B2	0.08333333	3763	3774	-1.82	3706	2286	38.32
B3	0.25				3730	1770	52.24
B4	0.5				3236	1521	58.96
B5	1	3868	3795	-2.39	3478	1465	60.47
B6	2				3464	1355	63.44
B7	4	3789	3750	-1.18	3328	1306	64.76
B8	6				2945	1157	68.78
B9	8		3630	2.06		1066	71.24
B10	10					967	73.91
B11	12		3485	5.97		924	75.07
B12	24		3774	-1.82		681	81.63
B13	36		3710	-0.10		558	84.94
B14	48		3702	0.12		929	74.94
B15	72		3708	-0.04		816	77.98
B16	97		3736	-0.80		658	82.25
term							
	avg	3806.67	3706.40	0.00	3703.67		
				2.33			

meOH + water		**% of Initial Radioactivity**										
origin	polar	parent	0.33	0.45	FAC/FAL	adjusted parent	% solids	% abiotic solids	average total extract recovery	polar metabolties	Nonpolar Metabolite	All metabolites
						94.12						
5.50	18.75	52.15	0.00	0.65	7.59	47.29	4.92		89.88	24.25	8.24	32.49
5.43	16.34	18.70	0.95	1.94	3.87	16.96	12.54	0.63	73.70	21.77	6.76	28.53
9.56	17.76	5.61	1.46	2.49	3.18	5.09	13.08		53.15	27.32	7.14	34.45
12.15	14.79	3.58	1.64	2.58	2.53	3.25	16.94		54.23	26.95	6.76	33.70
10.94	11.44	2.24	0.50	1.48	1.24	2.03	11.16	0.43	39.01	22.38	3.23	25.61
12.88	12.98	3.04	1.55	1.02	2.77	2.76	19.87		54.11	25.86	5.34	31.20
11.78	8.90	3.34	1.13	2.34	1.91	3.03	17.86	0.86	47.27	20.68	5.38	26.07
14.21	8.86	3.07	0.90	1.73	2.04	2.78	16.98		47.79	23.07	4.67	27.74
12.07	7.84	2.34	0.96	1.50	2.83	2.12	16.35	0.96	43.88	19.91	5.29	25.20
13.46	7.63	2.41	0.78	1.34	2.45	2.18	18.27		46.34	21.09	4.57	25.66
13.89	8.33	2.09	0.83	1.61	1.94	1.89	15.82	0.86	44.50	22.22	4.38	26.59
11.00	7.01	1.81	0.45	0.66	1.52	1.64	13.66	0.75	36.10	18.00	2.63	20.64
11.36	5.16	1.13	0.31	0.46	1.06	1.02	13.77		33.25	16.52	1.83	18.35
12.75	4.64	0.97	0.30	1.01	1.64	0.88	15.76	0.90	37.06	17.39	2.94	20.33
11.59	3.32	0.81	0.71	0.49	1.10	0.73	19.81	0.86	38.82	14.91	2.30	17.21
10.76	2.82	0.48	0.56	0.56	0.00	0.43	15.93	0.94	30.66	13.57	1.11	14.68

Table A3-4 (continued)

BIOTIC TLC Extracts

REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery with CO2
B1	0.01666667	835	159	1577	67.59	17.16	4.25	89.00	110.92
B2	0.08333333	462	252	4361	37.39	27.20	11.77	76.36	114.68
B3	0.25	251	190	4008	20.32	20.51	10.81	51.64	103.88
B4	0.5	217	194	6555	17.56	20.94	17.69	56.19	115.15
B5	1	173	203	6641	14.00	21.91	17.92	53.83	114.30
B6	2	180	169	7291	14.57	18.24	19.67	52.48	115.92
B7	4	159	168	6029	12.87	18.13	16.27	47.27	112.03
B8	6	150	187	6265	12.14	20.18	16.90	49.23	118.01
B9	8	145	150	6273	11.74	16.19	16.92	44.85	116.09
B10	10	138	170	7550	11.17	18.35	20.37	49.89	123.80
B11	12	137	186	5822	11.09	20.07	15.71	46.87	121.94
B12	24	102	130	5249	8.26	14.03	14.16	36.45	118.07
B13	36	72	129	4890	5.83	13.92	13.19	32.94	117.89
B14	48	79	139	5504	6.39	15.00	14.85	36.25	111.18
B15/16	72	60	141	6992	4.86	15.22	18.86	38.94	116.92
B17	97	54	92	5618	4.37	9.93	15.16	29.46	111.70
					0.00	0.00			

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	polar	parent 0.29	0.33	0.45	FAC/FAL	origin	polar	parent	0.33	0.45	FAC/FAL
B1		11.45	78.59			8.99	0.00	7.74	53.12	0.00	0.00	6.08
B2	2.98	23.52	53.3	2.49	5.35	12.35	1.11	8.80	19.93	0.93	2.00	4.62
B3	3.34	37.87	21.5	9.14	14.36	13.78	0.68	7.69	4.37	1.86	2.92	2.80
B4	3.73	36.64	21.05	9.38	15.68	13.52	0.66	6.44	3.70	1.65	2.75	2.37
B5	2.61	40.19	27.24		18.32	11.63	0.37	5.63	3.81	0.00	2.57	1.63
B6	3.31	25.51	33.42	7.1	12.13	18.52	0.48	3.72	4.87	1.03	1.77	2.70
B7	2.53	26.47	27.75	9.84	19.26	14.15	0.33	3.41	3.57	1.27	2.48	1.82
B8	3.2	31.07	27.79	7.86	15.19	14.89	0.39	3.77	3.37	0.95	1.84	1.81
B9	3.79	32.17	15.88	10.32	12.04	25.8	0.44	3.78	1.86	1.21	1.41	3.03
B10		35.47	22.58	7.94	12.58	21.42	0.00	3.96	2.52	0.89	1.41	2.39
B11	4.31	39.57	15.54	7.28	13.33	19.98	0.48	4.39	1.72	0.81	1.48	2.22
B12	5.45	37.44	22.98	4.53	7.61	22.01	0.45	3.09	1.90	0.37	0.63	1.82
B13	6.78	38.81	19.22	6.16	9.87	19.15	0.40	2.26	1.12	0.36	0.58	1.12
B14	6.75	28.58	20.67		16.63	27.37	0.43	1.83	1.32	0.00	1.06	1.75
B15/16	9.73	22.42	22.42	21.23		24.19	0.47	1.09	1.09	1.03	0.00	1.17
B17												

Relative Abundance in Water extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	polar	parent	0.33	0.45	FAC/FAL	origin	polar	parent	0.33	0.45	FAC/FAL
B1	23.81	61.78	7.83		4.44	2.15	4.09	10.60	1.34	0.00	0.76	0.37
B2	32.48	59.44	3.62				8.83	16.17	0.98	0.00	0.00	0.00
B3	39.03	57.1	3.86				8.00	11.71	0.79	0.00	0.00	0.00
B4	56.49	43.51					11.83	9.11	0.00	0.00	0.00	0.00
B5	58.76	41.24					12.87	9.03	0.00	0.00	0.00	0.00
B6	60.1	39.9					10.96	7.28	0.00	0.00	0.00	0.00
B7	61.56	38.44					11.16	6.97	0.00	0.00	0.00	0.00
B8	67.64	32.36					13.65	6.53	0.00	0.00	0.00	0.00
B9	62.38	37.62					10.10	6.09	0.00	0.00	0.00	0.00
B10	75.53	24.47					13.86	4.49	0.00	0.00	0.00	0.00
B11	69.16	30.84					13.88	6.19	0.00	0.00	0.00	0.00
B12	62.48	37.52					8.77	5.26	0.00	0.00	0.00	0.00
B13	72.56	27.44					10.10	3.82	0.00	0.00	0.00	0.00
B14	80.21	19.79					12.03	2.97	0.00	0.00	0.00	0.00
B15/16									0.00	0.00	0.00	0.00
B17									0.00	0.00	0.00	0.00
									0.00	0.00	0.00	0.00

BIOTIC TLC Extracts

REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Total Recovery with CO2
B1	0.01666667	791	196	2070	64.02	21.15	5.58	90.76	112.68
B2	0.08333333	384	247	4931	31.08	26.66	13.30	71.04	109.36
B3	0.25	239	185	5691	19.34	19.97	15.35	54.66	106.91
B4	0.5	211	176	6002	17.08	18.99	16.19	52.27	111.23
B5	1	75	127	1635	6.07	13.71	4.41	24.19	84.66
B6	2	182	194	7441	14.73	20.94	20.08	55.74	119.19
B7	4	141	152	7209	11.41	16.40	19.45	47.27	112.03
B8	6	134	171	6323	10.85	18.45	17.06	46.36	115.14
B9	8	142	145	5848	11.49	15.65	15.78	42.92	114.16
B10	10	133	147	5993	10.77	15.86	16.17	42.80	116.71
B11	12	121	152	5902	9.79	16.40	15.92	42.12	117.19
B12	24	82	148	4874	6.64	15.97	13.15	35.76	117.39
B13	36	60	133	5321	4.86	14.35	14.36	33.57	118.51
B14	48	74	141	6181	5.99	15.22	16.68	37.88	112.82
B15/16	72	58.5	123	7690	4.74	13.22	20.75	38.70	116.69
B17	97	46	106	6188	3.72	11.44	16.70	31.86	114.11
									113.79
									6.58

Relative Abundance in Methanol extacts on TLC Plates / **% of Initial Radioactivity**

Sample #	origin	polar	parent	0.33	0.45	FAC/FAL	origin	polar	parent	0.33	0.45	FAC/FAL
B1		15.07	74.31			10.62	0.00	9.65	47.58	0.00	0.00	6.80
B2	2.93	24.81	53.04	3.13	6.04	10.05	0.91	7.71	16.49	0.97	1.88	3.12
B3	3.19	35.17	27.02	5.5	10.71	18.42	0.62	6.80	5.23	1.06	2.07	3.56
B4	4.22	36.02	20.3	9.58	14.1	15.77	0.72	6.15	3.47	1.64	2.41	2.69
B5	3.14	48.84	10.81	16.6	6.42	14.18	0.19	2.96	0.66	1.01	0.39	0.86
B6	3.06	53.56	8.23	14	1.85	19.29	0.45	7.89	1.21	2.06	0.27	2.84
B7	2.95	24.21	27.29	8.64	19.33	17.59	0.34	2.76	3.11	0.99	2.21	2.01
B8	3.95	26.87	25.48	7.83	14.94	20.94	0.43	2.91	2.76	0.85	1.62	2.27
B9	4.43	28.25	24.42	6.08	13.86	22.95	0.51	3.25	2.81	0.70	1.59	2.64
B10	5.46	31.8	21.31	6.2	11.85	23.38	0.59	3.42	2.29	0.67	1.28	2.52
B11	4.09	27.45	25.04	8.62	17.82	16.98	0.40	2.69	2.45	0.84	1.75	1.66
B12	4.1	32.98	26.02	7.97	10.55	18.38	0.27	2.19	1.73	0.53	0.70	1.22
B13	6.3	37.23	23.44	5.39	6.92	20.72	0.31	1.81	1.14	0.26	0.34	1.01
B14	8.25	30.19	10.31	9.86	15.96	25.42	0.49	1.81	0.62	0.59	0.96	1.52
B15/16	8.79	29.42	11.22	8.25	20.83	21.49	0.42	1.39	0.53	0.39	0.99	1.02
B17	12.49	44.79	12.84	14.92	14.96		0.47	1.67	0.48	0.56	0.56	0.00

% of Initial Radioactivity

Sample #	origin	polar	parent	0.33	0.45	FAC/FAL	origin	polar	parent	0.33	0.45	FAC/FAL
B1	32.7	44.96	10.68		2.52	9.14	6.92	9.51	2.26	0.00	0.53	1.93
B2							0.00	0.00	0.00			
B3	49.15	46.67	4.18				9.81	9.32	0.83			
B4	58.46	41.54					11.10	7.89	0.00			
B5	61.7	38.3					8.46	5.25	0.00			
B6	66.23	33.77					13.87	7.07	0.00			
B7	71.6	28.41					11.75	4.66	0.00			
B8	75.65	24.36					13.96	4.50	0.00			
B9	83.59	16.41					13.08	2.57	0.00			
B10	78.65	21.35					12.48	3.39	0.00			
B11	79.37	20.63					13.02	3.38	0.00			
B12	78.28	21.72					12.50	3.47	0.00			
B13	83.09	16.91					11.93	2.43	0.00			
B14	82.46	17.54					12.55	2.67	0.00			
B15/16	84.3	15.7					11.14	2.08	0.00			
B17	89.95	10.05					10.29	1.15	0.00			
							0.00	0.00	0.00			

meoh average / **% of Initial Radioactivity**

	origin	polar	parent	0.33	0.45	FAC/FAL
0.016667	0.00	8.69	50.35	0.00	0.00	6.44
0.083333	1.01	8.25	18.21	0.95	1.94	3.87
0.25	0.65	7.25	4.80	1.46	2.49	3.18
0.5	0.69	6.29	3.58	1.64	2.58	2.53
1	0.28	4.30	2.24	0.50	1.48	1.24
2	0.47	5.80	3.04	1.55	1.02	2.77
4	0.33	3.08	3.34	1.13	2.34	1.91
6	0.41	3.34	3.07	0.90	1.73	2.04
8	0.48	3.51	2.34	0.96	1.50	2.83
10	0.29	3.69	2.41	0.78	1.34	2.45
12	0.44	3.54	2.09	0.83	1.61	1.94
24	0.36	2.64	1.81	0.45	0.66	1.52
36	0.35	2.03	1.13	0.31	0.46	1.06
48	0.46	1.82	0.97	0.30	1.01	1.64
72	0.44	1.24	0.81	0.71	0.49	1.10
97	0.47	1.67	0.48	0.56	0.56	0.00

water average / **% of Initial Radioactivity**

origin	polar	parent	0.33	0.45	FAC/FAL
5.50	10.06	1.80	0.00	0.65	1.15
4.42	8.08	0.49	0.00	0.00	0.00
8.91	10.51	0.81	0.00	0.00	0.00
11.47	8.50	0.00	0.00	0.00	0.00
10.66	7.14	0.00	0.00	0.00	0.00
12.41	7.17	0.00	0.00	0.00	0.00
11.45	5.81	0.00	0.00	0.00	0.00
13.81	5.51	0.00	0.00	0.00	0.00
11.59	4.33	0.00	0.00	0.00	0.00
13.17	3.94	0.00	0.00	0.00	0.00
13.45	4.79	0.00	0.00	0.00	0.00
10.63	4.37	0.00	0.00	0.00	0.00
11.01	3.12	0.00	0.00	0.00	0.00
12.29	2.82	0.00	0.00	0.00	0.00
11.14	2.08	0.00	0.00	0.00	0.00
10.29	1.15	0.00	0.00	0.00	0.00

Table A3-4 (continued)

ABIOTIC TLC Extracts:
REP 1

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery	Relative Abundance polar	Relative Abundance parent	Relative Abundance nonpolar	% of Initial Radioactivity polar	% of Initial Radioactivity parent	% of Initial Radioactivity nonpolar
A1	0.01666667													
A2	0.08333333	1249	32	175	101.10	3.45		104.55	9.12	88.74	2.12	9.22	89.71	2.14
A3	0.25													
A4	0.5													
A5	1	1368	22	19	110.75	2.37	0.05	113.18	7.16	92.84		7.93	102.82	0.00
A6	2													
A7	4	1242	33	240	100.53	3.56	0.65	104.74	6.51	93.49		6.54		0.00
A8	6													
A9	8	1313	29	312	106.28	3.13	0.84	110.25	7.05	91.5	1.44	7.49	97.24	1.53
A10	10													
A11	12	1255	29	306	101.58	3.13	0.83	105.54	6.68	93.32		6.79	94.80	0.00
A12	24	1268	25	216	102.63	2.70	0.58	105.91	6.35	93.65		6.52	96.12	0.00
A13	36	1241	31	266					6.91	93.09			95.54	
A14	48	1212	33	282	98.10	3.56	0.76	102.42				0.00		0.00
A15	72	1256	39	318	101.66	4.21	0.86	106.73	7.42	92.58		7.54	94.12	0.00
A16	97	1293	21	236	104.68	2.27	0.64	107.58	6.3	93.7		6.59	98.09	0.00
A17														
										92.55			96.05	
										1.49			3.48	

ABIOTIC TLC Extracts:
REP 2

Sample #	Time hrs	Extract 1 MeOH 0.5 ml/15	Extract 2 H2O 0.25 ml/10	Solids	Extract 1 Recovery	Extract 2 Recovery	% on Solids	Total Extract Recovery
A1	0.01666667							
A2	0.08333333	1205	37	235	97.53	3.99	0.63	102.16
A3	0.25							
A4	0.5							
A5	1	1295	26	297	104.82	2.81	0.80	108.43
A6	2							
A7	4	1280	35	400	103.60	3.78	1.08	108.46
A8	6							
A9	8	1275	37	398	103.20	3.99	1.07	108.27
A10	10							
A11	12	1209	28	331	97.86	3.02	0.89	101.77
A12	24	1229	25	340	99.48	2.70	0.92	
A13	36	1276	28	355				
A14	48	1259	29	386	101.90	3.13	1.04	106.08
A15/16	72	1259	21	322	101.90	2.27	0.87	105.04
A17	97	1228	45	458	99.40	4.86	1.24	105.49
Average					102.1		0.81	106.3
Std Dev					3.19		0.26	2.86