May 4 '71 Review With Boxtel

"BIODEGRADATION CONSISTS OF A CHANGE OF A CHEMICAL OR GROUP OF MATERIALS BY LIFE PROCESS AS WITH AN ACCOMPANYING TRANSFER OF ENERGY."

Definition proposed by a sub-committee of the standard methods committee for the W.P.C.F.

"The conversion of chemicals which are potential environmental pollutants to innocuous substances via the biological action of living organisms."

Primary Biodegradation - Minimum alteration of the chemical structure of the material in question to an extent that the characteristic properties of the original material are no longer evident.

Ultimate Biodegradation - Complete conversion of the material in question to CO_2, H_2O, inorganic salts, and products normally associated with the natural metabolic processes of living organisms.

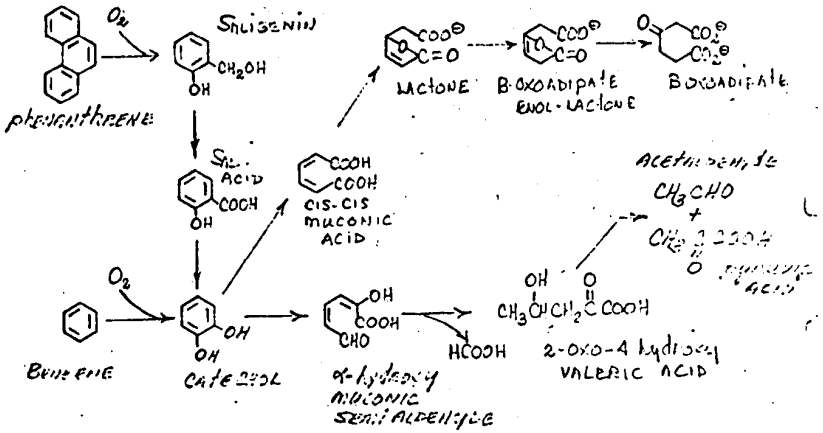
MONS 042280

"Develop a laboratory test procedure which will measure the susceptibility of selected Organic Division Products to primary biodegradation via the biological action of bacteria. Furthermore, the procedure should produce results in a reasonable period of time at a reasonable cost which can be easily extrapolated to the environment."

Potential Environmental Pollutants		Selected Organic Division Products
Living Organisms	-	Bacteria
Conversion	-	Primary Biodegradation
Mixed Bacterial Population	or	Pure Bacterial Culture
Mixed Foods	or	Sole Carbon Source
Aerobic	or	Anaerobic (Slower (Difficult

✓ River Die-Away Test ($\sim 1 \times 10^5$ cells/ml)
Fortified and Inoculated Water Test
Shake Culture Test
Laboratory Trickling Filters
Continuous Flow Activated Sludge
Batch Sludge Die Away
✓ Semi-Continuous Activated Sludge (2×10^8 cells/ml)
✓ Soil Percolation

MONS 042281

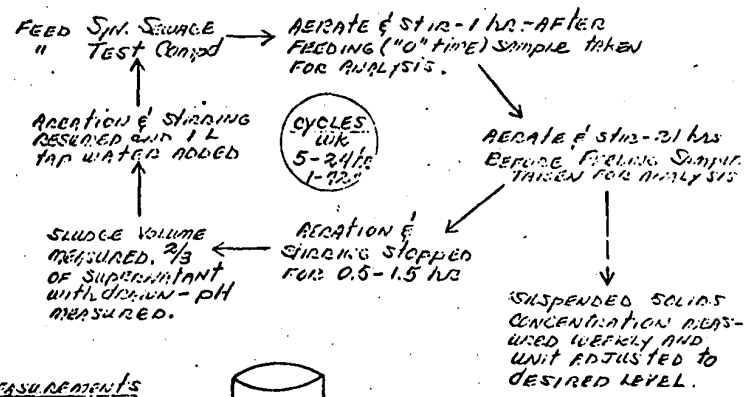

O_2
SALIGENIN
CH_2OH
OH
PHENANTHRENE
LACTONE
B-OXOADIPATE ENOL-LACTONE
B-OXOADIPATE
SAL. ACID
COOH
OH
CIS-CIS MUCONIC ACID
O_2
BENZENE
CATECHOL
OH
OH
COOH
CHO
HCOOH
2-OXO-4 HYDROXY VALERIC ACID
CH_3CHO
+

MONS 042282

Standard Operating Conditions

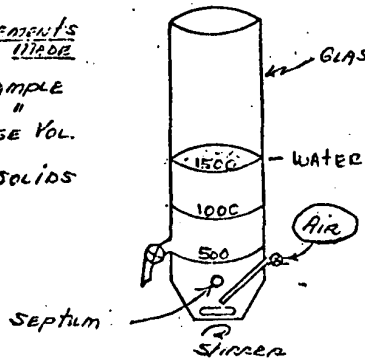
Vol.	1500 mL
SS	3750 mg (2500 ppm) Activitated Sludge
Air	½ ft³/hr
Feed	Synthetic Sewage { Dextrose, Nutrient broth, KH_2PO_4
	Test compd (EtOH)

Mechanical Cycle



Measurements made

1. AF Sample
2. BF "
3. Sludge Vol.
4. pH
5. Sus. Solids

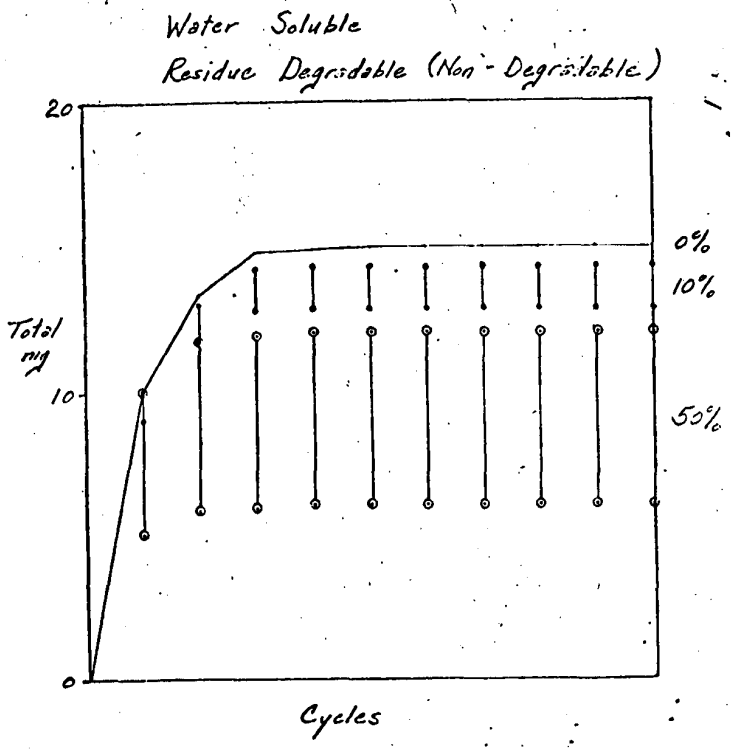


MONS 042283

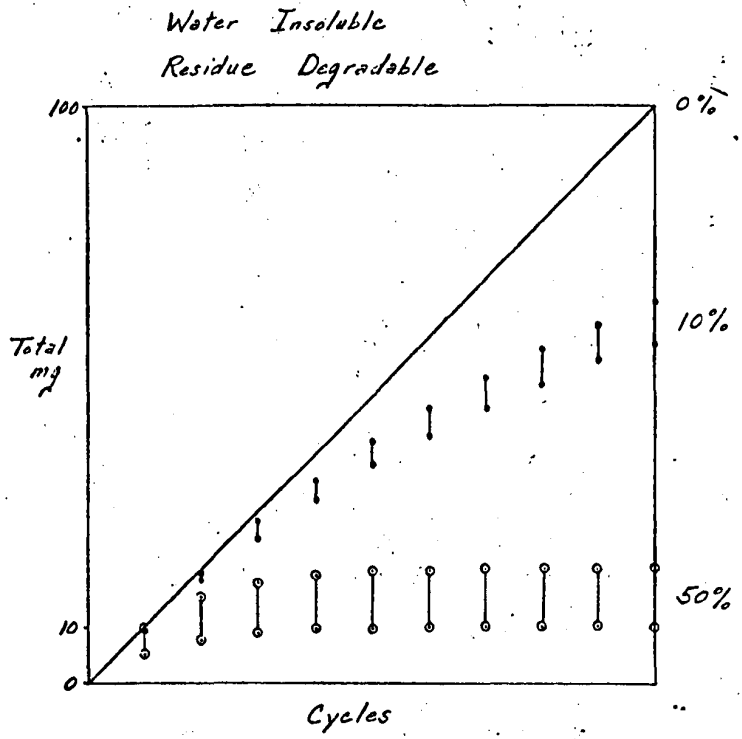
Test Compound	Molecular Structure	Feed Level (mg)	Cycle Time (hr)	Disappearance Rate (%)	Water Solubility %	Vapor Pressure mmHg (25°)	Method of Analysis
LAS (Dodecane-1)		20	24	>99.5	Soluble		Colorimetric
p-Chloroaniline		5	24	97.3±1.6	0.5	0.1	UV
MIPB		10	24	78.2±3.2		0.005	UV, FID-GC
HB-40		10	24	48.6±6.9		N.S.	UV, FID-GC
S-711 M		10	24	48.3±7.7			UV
o-NCB		5	24	37.5±8.8	0.050		UV
Nitrated Orthene		5	24	40.3±3.2			UV
p-NCB		5	24	33.9±2.9	0.005		UV
Aroclor 1221	 21% Cl	1	48	~70.0		0.005	UV
2,2' Dichloro Diphenyl Ether		5	24	12.0±4.8			EC-GC
MCS 1043	 ~30% Cl	1	48	56.2±15.5			UV, EC-GC
Arobromaclor 1232	 21% Cl 11% Br	1	48	54.1±19.3			UV, EC-GC
MCS 1016	 41% Cl	1	48	32.9±13.8			UV, EC-GC
Aroclor 1242	 42% Cl	1	48	26.3±15.3	2×10^{-5}	1.4×10^{-3}	UV, EC-GC
Aroclor 1254	 54% Cl	1	48	15.0±3.7.7	5×10^{-6}	3.4×10^{-4}	EC-GC
TCC		1	24	0	<0.001		UV

* Initial Disappearance Rate 71.3 ± 5.5 %

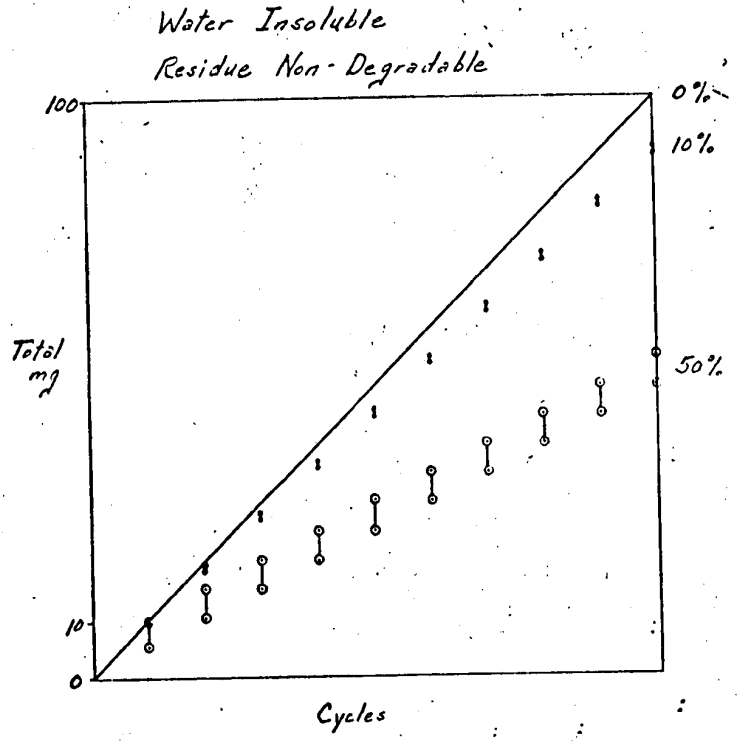
MONS 042284


Water Soluble
Residue Degradable (Non - Degradable)
20
10
0
Total mg
0%
10%
50%
Cycles

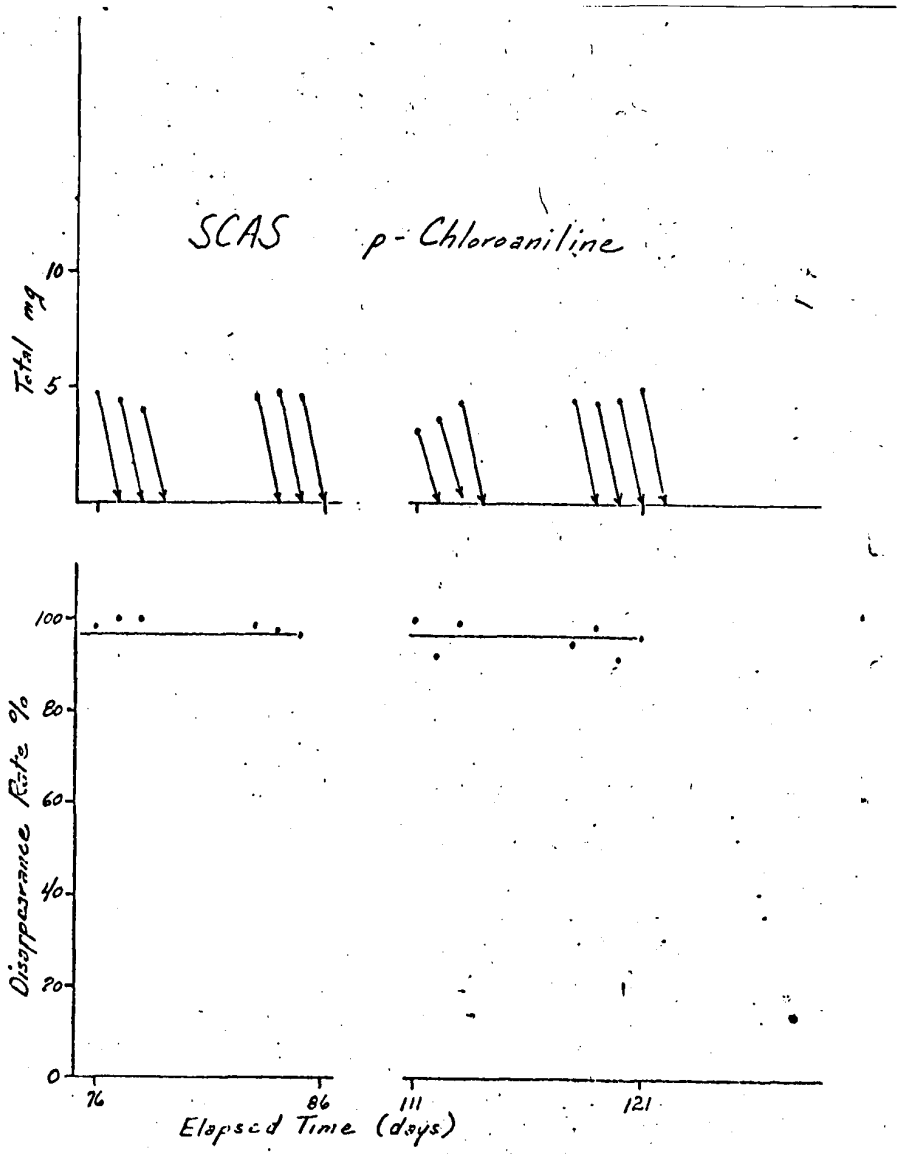
MONS 042285

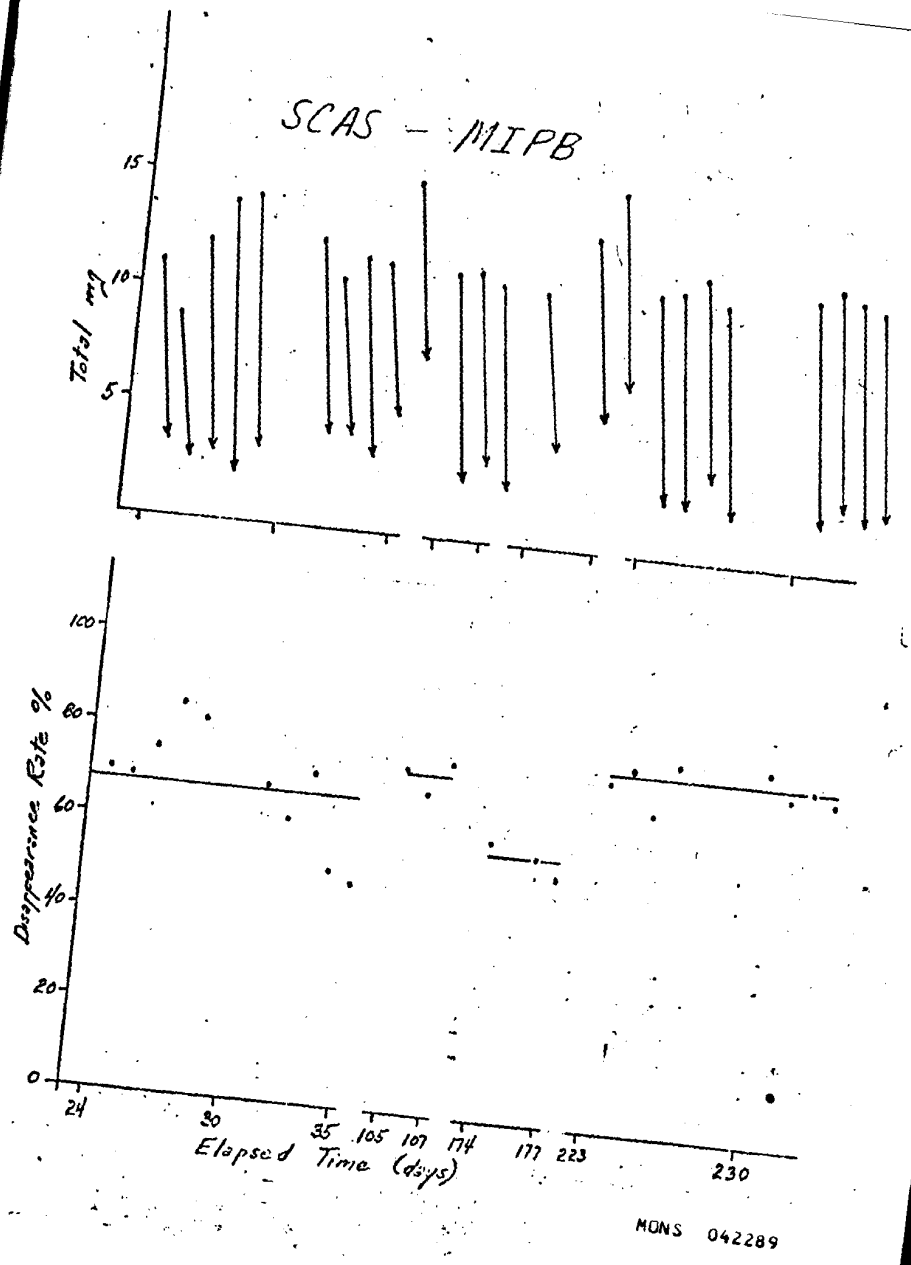

Water Insoluble
Residue Degradable
100
10
0
Total mg
0%
10%
50%
Cycles

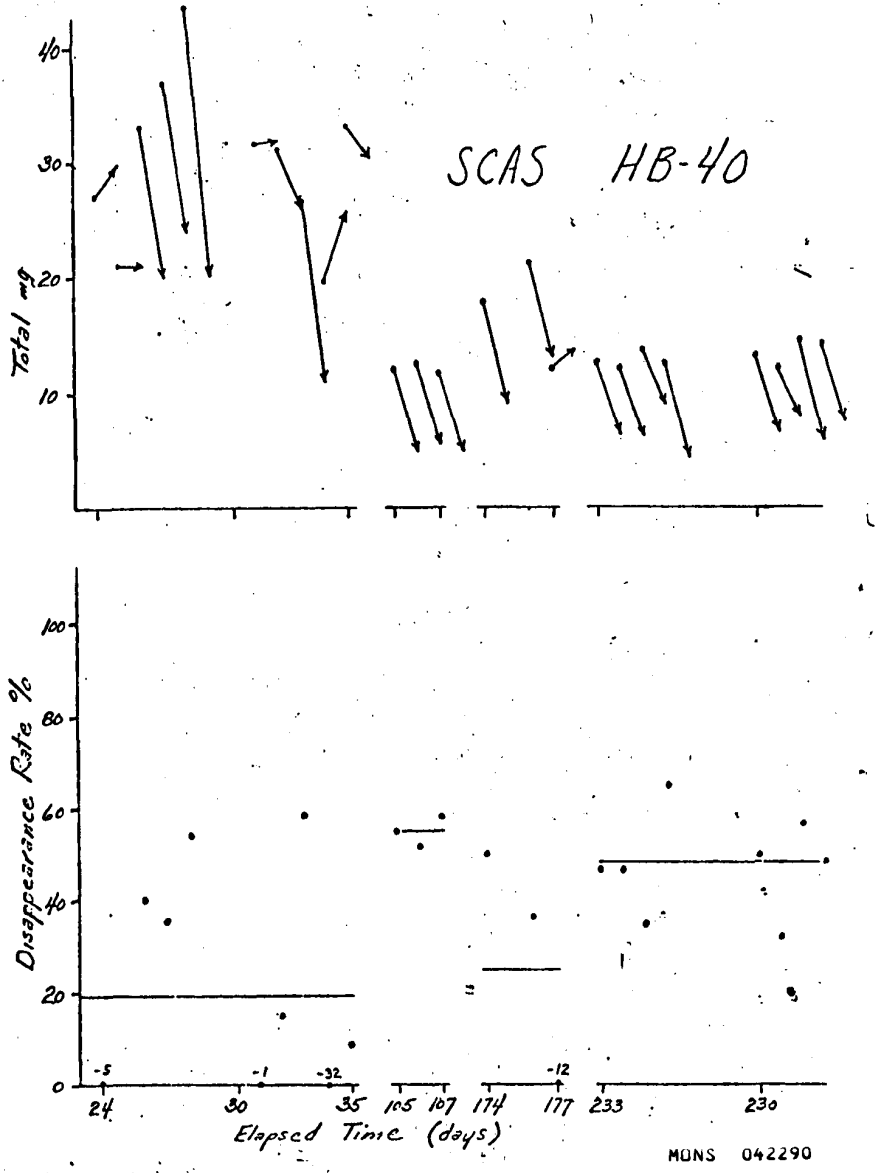
MONS 042286

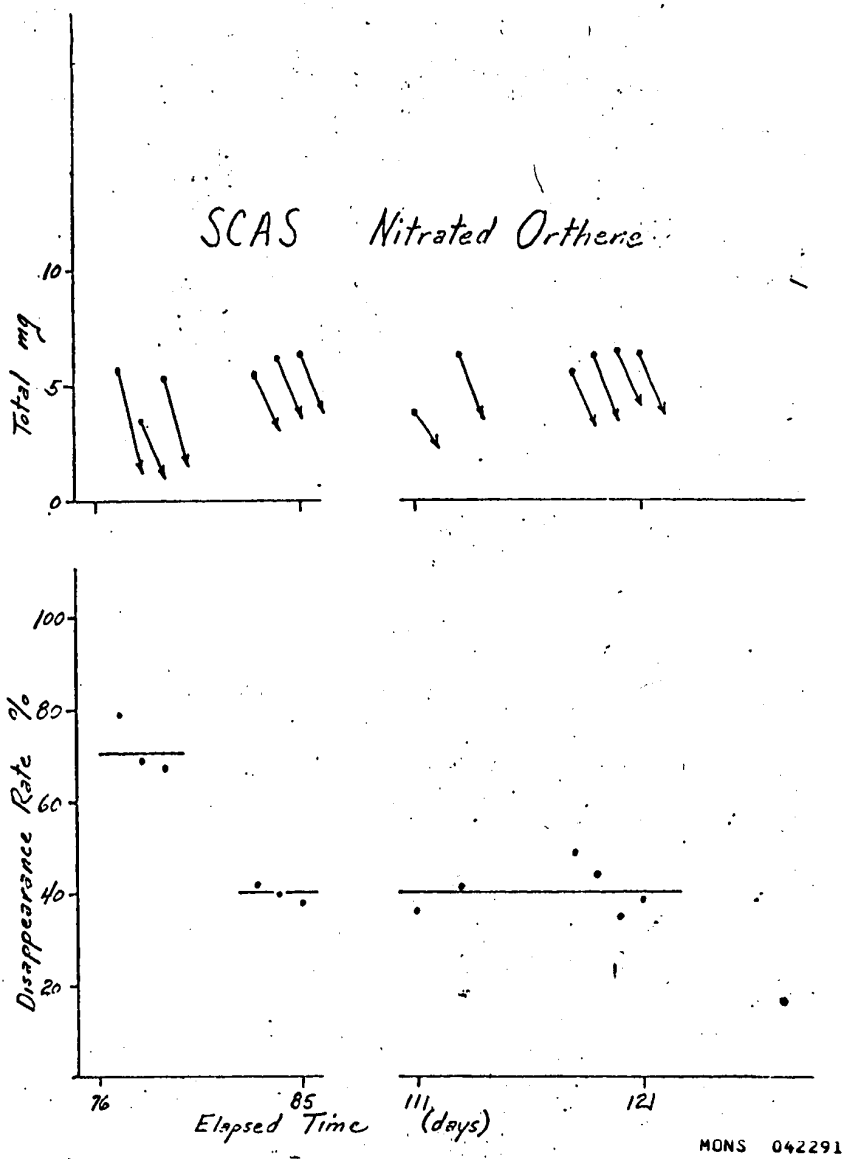

Water Insoluble
Residue Non-Degradable
100
10
0
Total mg
0%
10%
50%
Cycles

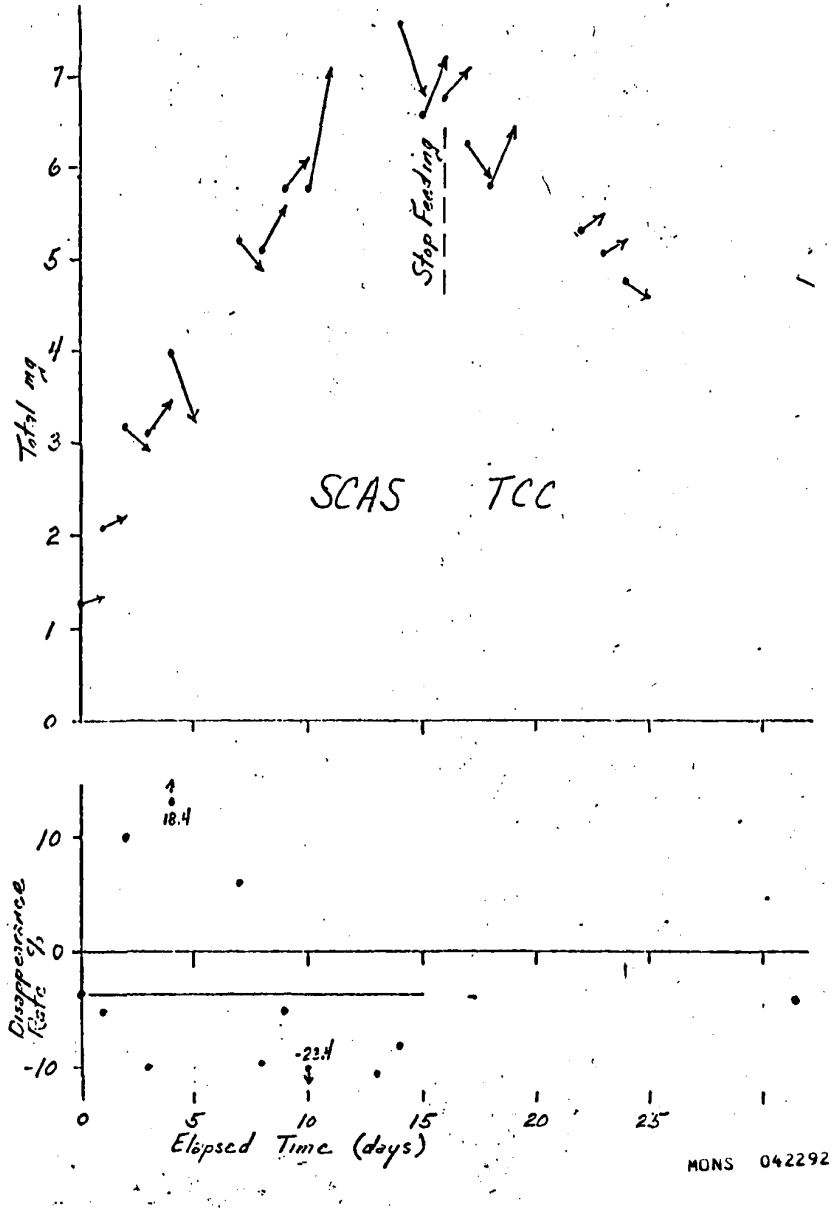
MONS 042287

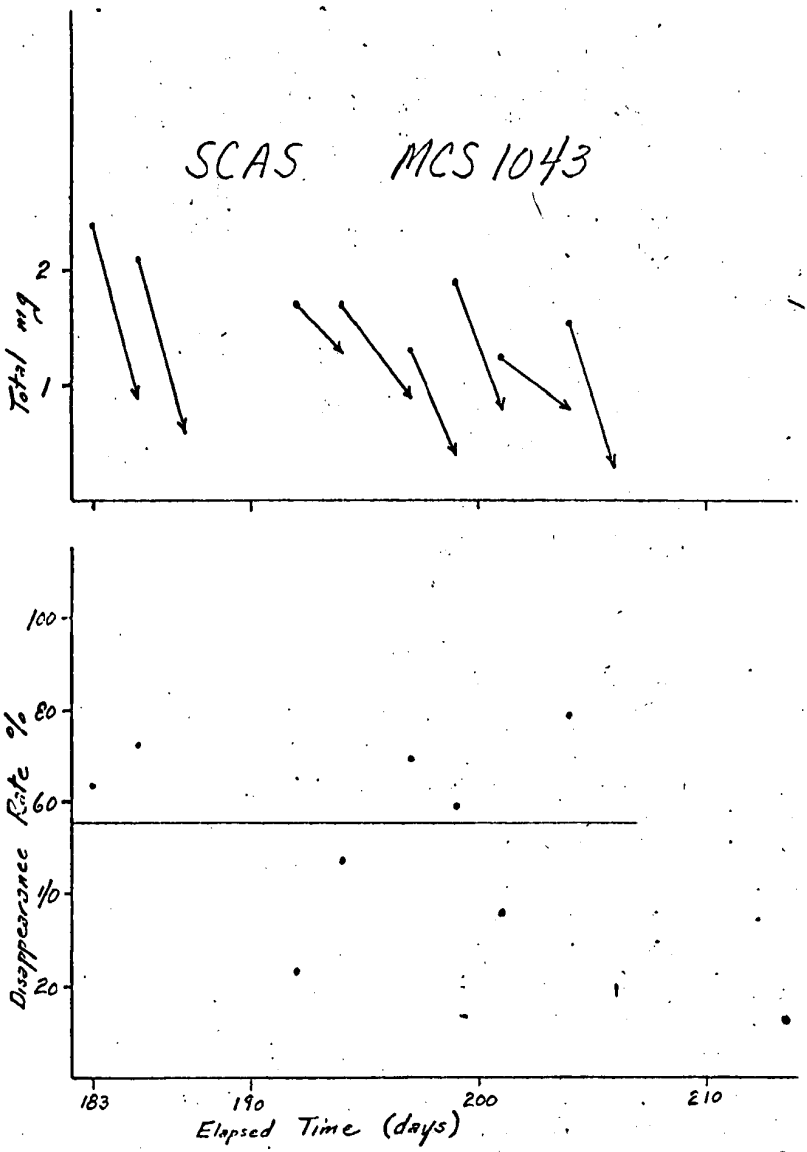

SCAS p-Chloroaniline
Total mg
10
5
Disappearance Rate %
100
80
60
40
20
0
76
86
111
121
Elapsed Time (days)
MONS 042288

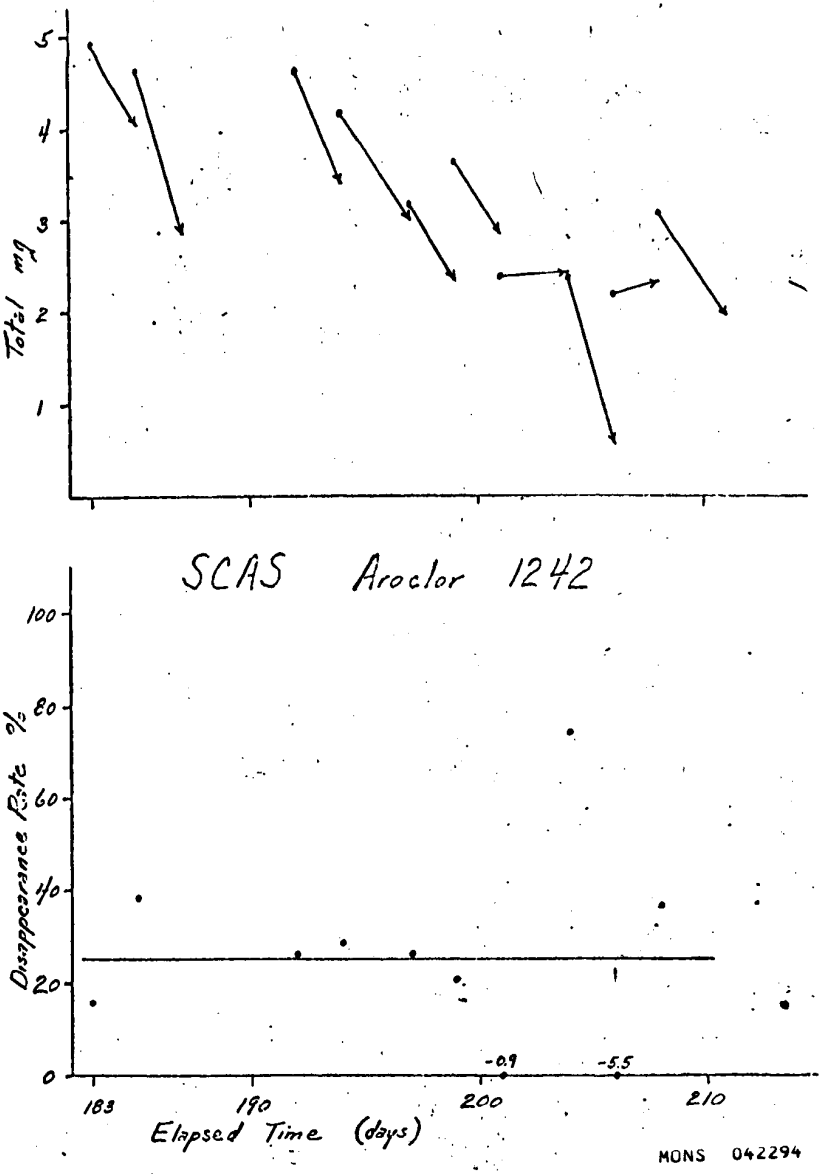

SCAS – MIPB
Total mg
15
10
5
Disappearance Rate %
100
80
60
40
20
0
24
30
35
105
107
174
177
223
230
Elapsed Time (days)
MONS 042289

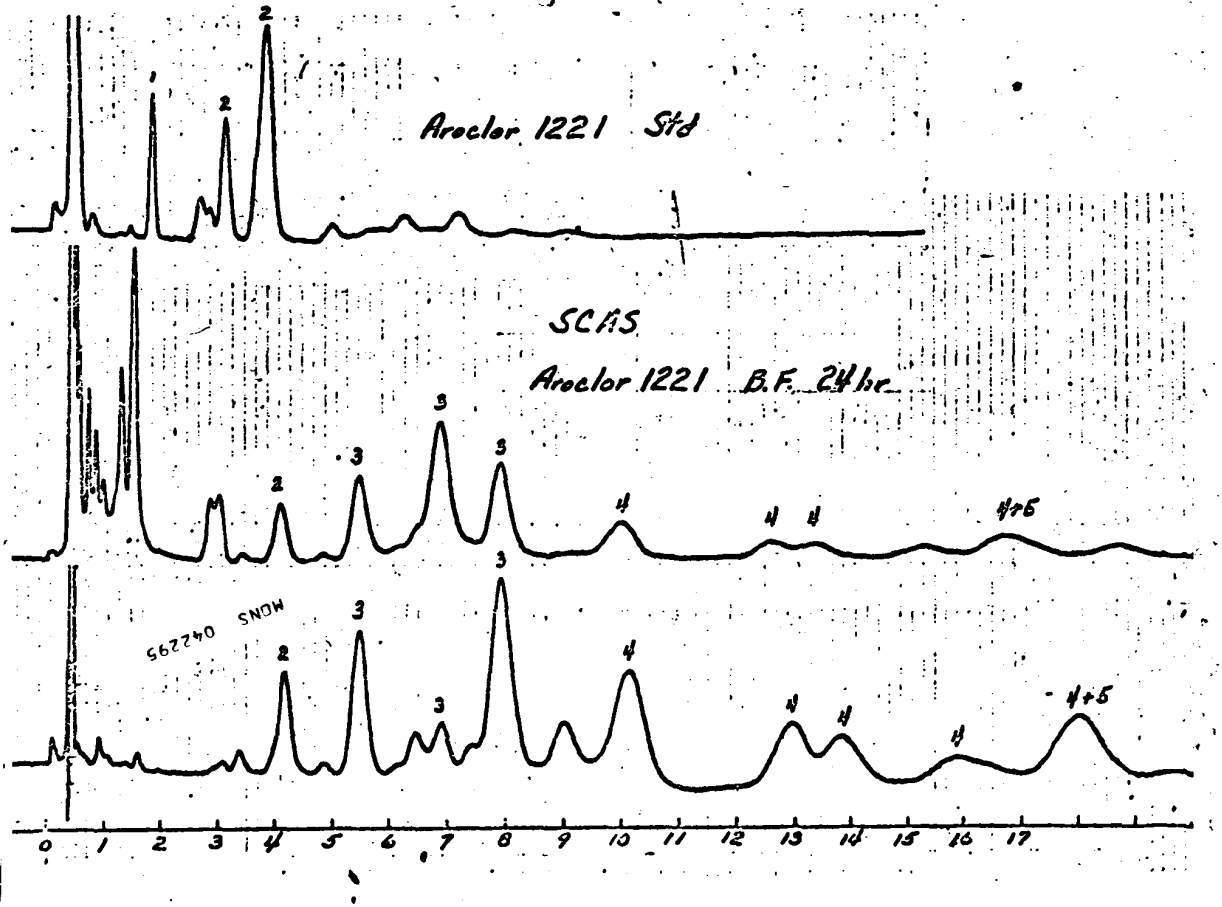

SCAS HB-40
Total mg
40
30
20
10
Disappearance Rate %
100
80
60
40
20
0
-5
-1
-32
-12
24
30
35
105
107
174
177
233
230
Elapsed Time (days)
MONS 042290

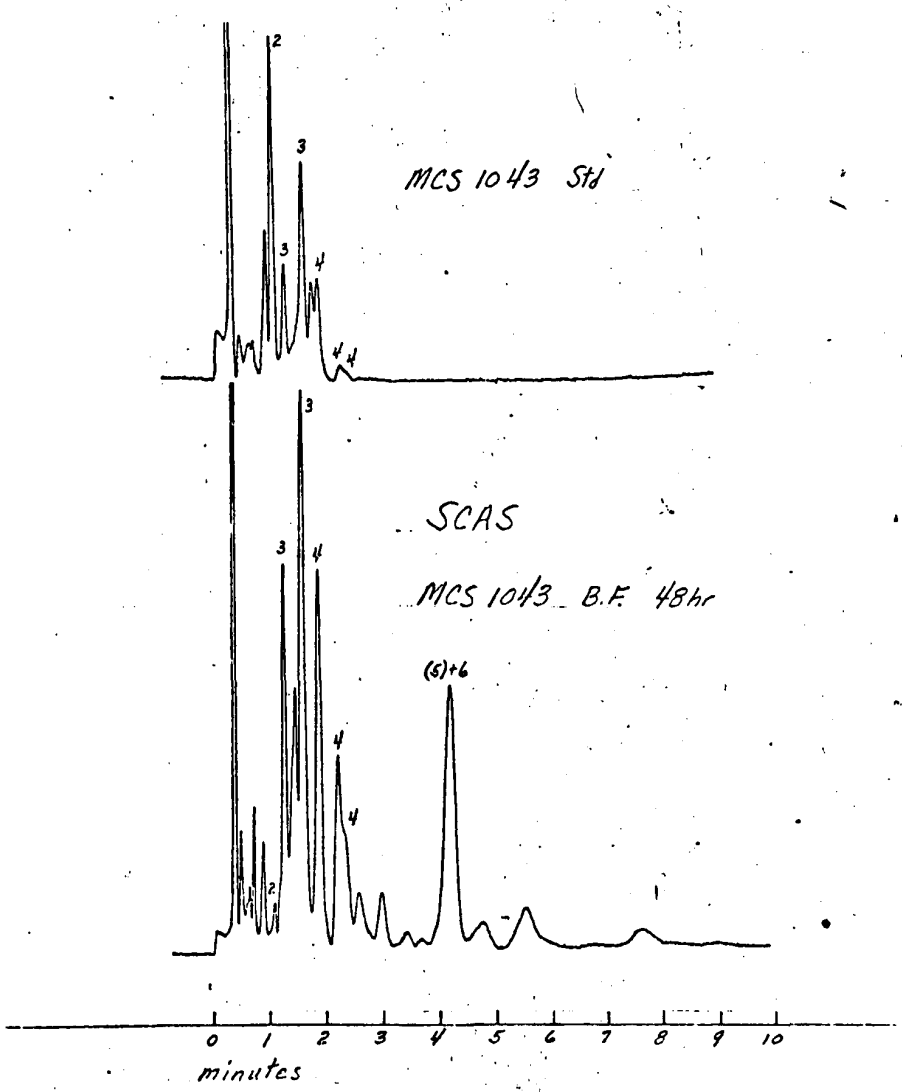

SCAS Nitrated Orthene
Total mg
10
5
0
Disappearance Rate %
100
80
60
40
20
76
85
111
121
Elapsed Time (days)
MONS 042291


Total mg
7
6
5
4
3
2
1
0
Stop Feeding
SCAS TCC
Disappearance Rate %
10
0
-10
18.4
-23.4
0
5
10
15
20
25
Elapsed Time (days)
MONS 042292

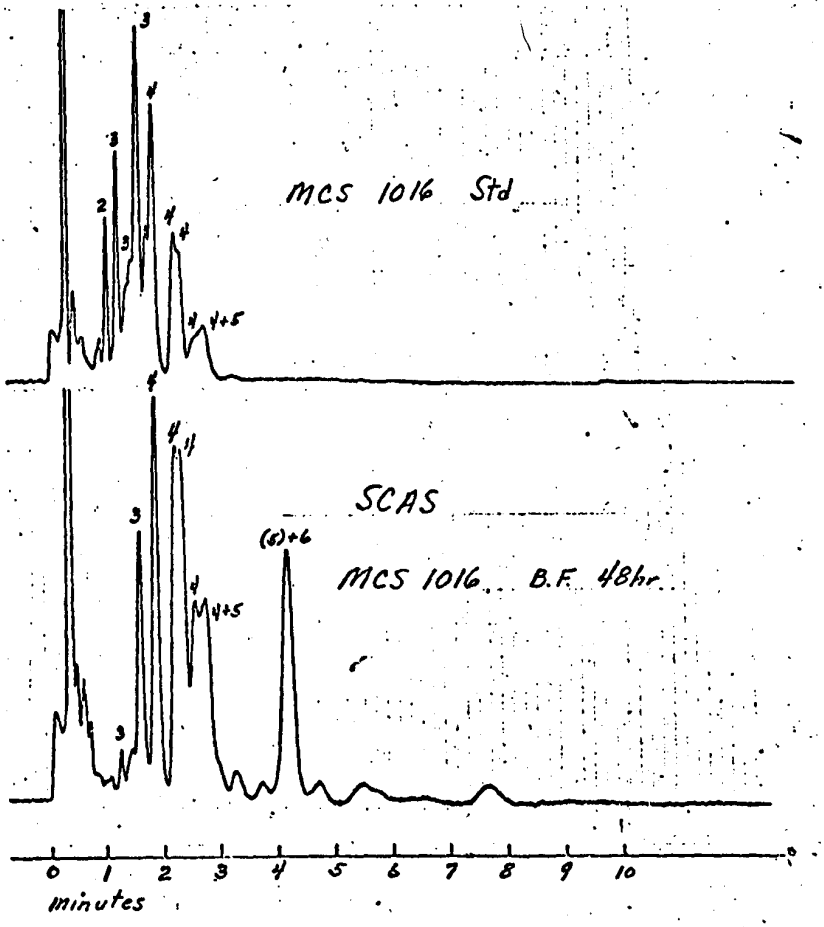

SCAS MCS 1043
Total mg
2
1
100
80
60
40
20
Disappearance Rate %
183
190
200
210
Elapsed Time (days)
MONS 042293


Total mg
5
4
3
2
1
SCAS Aroclor 1242
Disappearance Rate %
100
80
60
40
20
0
-0.9
-5.5
183
190
200
210
Elapsed Time (days)
MONS 042294

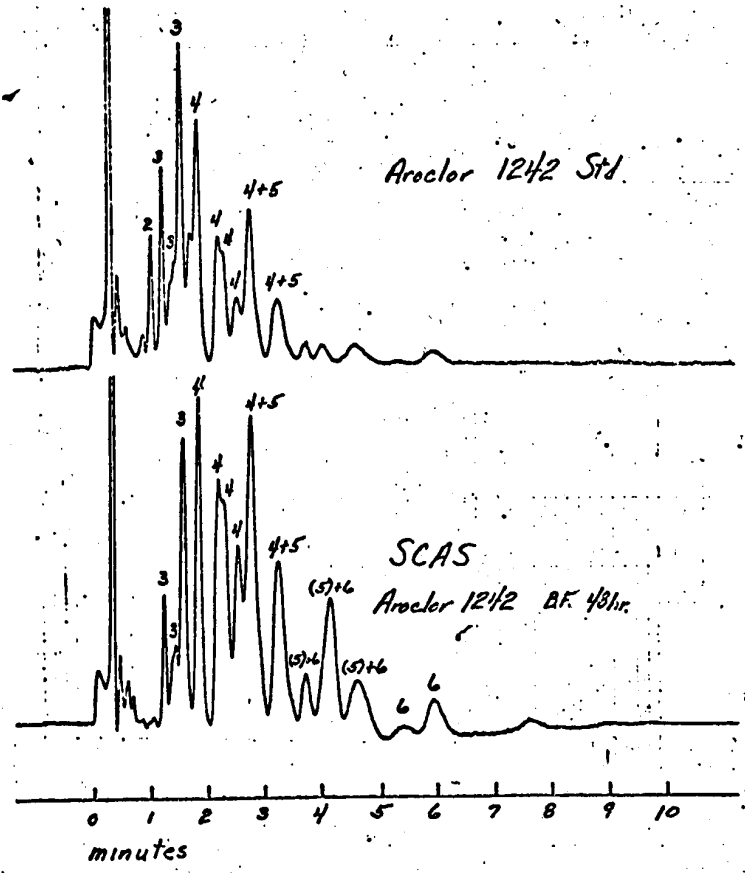

Aroclor 1221 Std
SCAS
Aroclor 1221 B.F. 24 hr
SNOW
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17



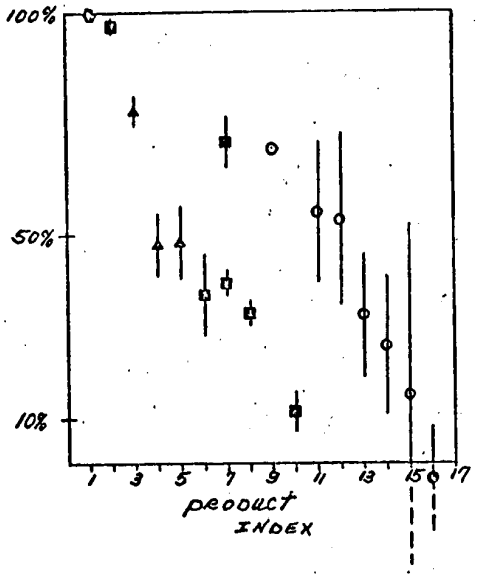
MONS 042296



MONS 042297


Aroclor 1242 Std.
SCAS
Aroclor 1242 BF 48 hr.
minutes
0 1 2 3 4 5 6 7 8 9 10

MUNS 042298

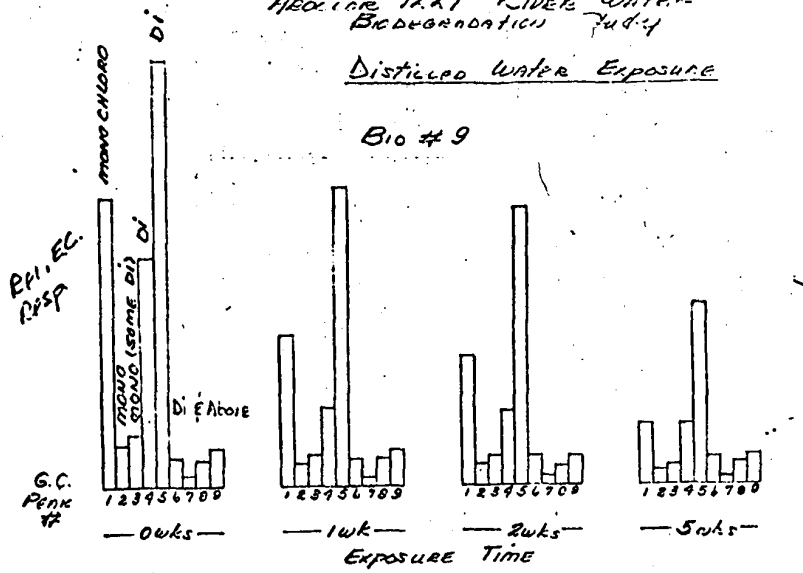


MONS 042299

Aroclor 1221 River Water Biodegradation Study

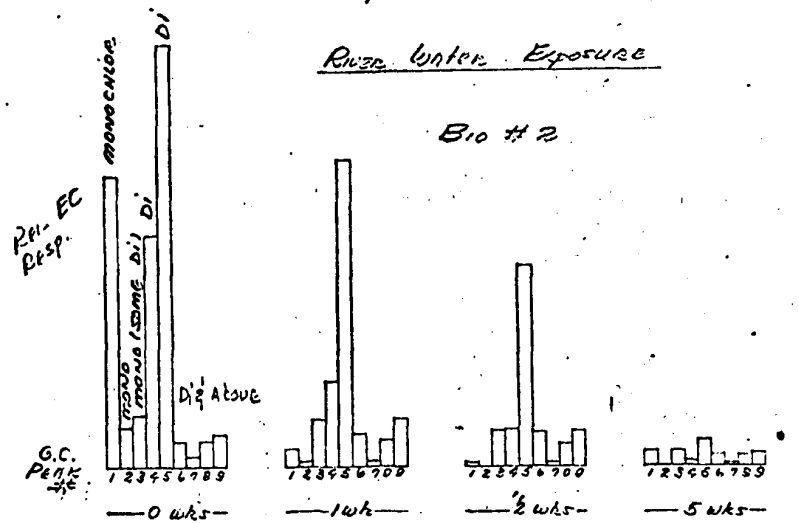
Distilled Water Exposure

Bio #9



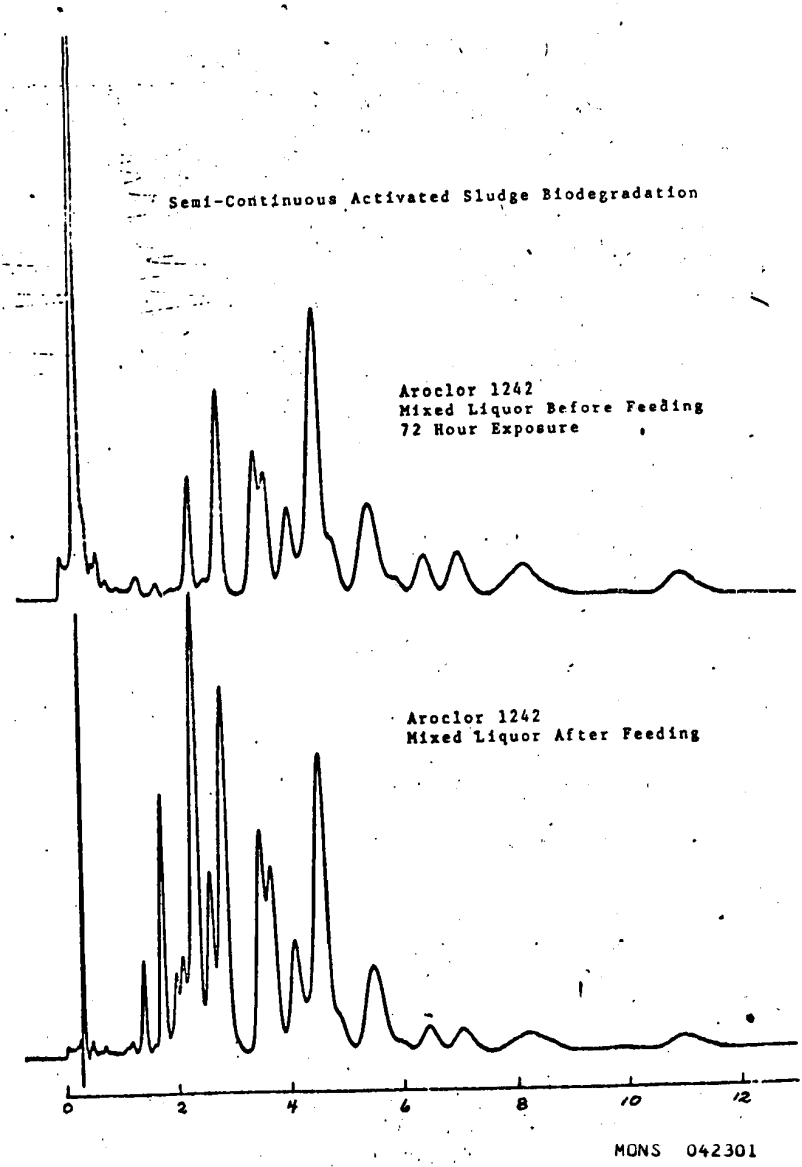
River Water Exposure

Bio #2



MONS 042300

Figure I


Semi-Continuous Activated Sludge Biodegradation
Aroclor 1242
Mixed Liquor Before Feeding
72 Hour Exposure
Aroclor 1242
Mixed Liquor After Feeding
0
2
4
6
8
10
12

MONS 042301

Biotest Studies	Sept 1968 Protocol #	Biotest Study #	Report Date	Status
Chicken Residue		J7010	4-69	Tissues used to verify procedures
Rat Tissue Study	I	J7009	4-69	Analysis of Aroclor tissues complete
Chicken Toxicity, Reproduction and Meat and Egg Residue Study	II	J7300	6-70	~ 220 Samples at Biotest
Mallard Duck Study	III			Deleted
3-Generation Reproduction Study in Rats	IV	P7297	9-70 (15 mo)	No tissues collected?
2-Year Chronic Toxicity Study in Rats	V	B7298	9-70 (12 mo.)	~ 320 Samples 204 at So 2nd St.
2-Year Chronic Toxicity Study in Dogs	VI	C7299	12-70 (18 mo.)	No tissues collected?
Subacute Fish Toxicity Study	VII	A7204	6-69	Deleted.
		A8570		69 Fish samples ~ 200 Water samples at So. 2nd St.

MDNS 042302

#B7298 Two Year Chronic Oral Toxicity Study Of Aroclor In Albino Rats Adverse Effects

		3 Mo. Sacrifice						6 Mo. Sacrifice						12 Mo. Sacrifice						24 Mo. Sacrifice					
		1 ppm		10 ppm		100 ppm		1 ppm		10 ppm		100 ppm		1 ppm		10 ppm		100 ppm		1 ppm		10 ppm		100 ppm	
		♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀
Aroclor 1242	Muscle														x	x		x							
	Kidney														x	x		x							
	Liver														x	x		x							
	Fat														x	x		x							
Aroclor 1254	Muscle														x	x									
	Kidney														x	x									
	Liver														x	x									
	Fat														x	x									
Aroclor 1260	Muscle													x		x									
	Kidney													x		x									
	Liver													x		x									
	Fat													x		x									
Control	Muscle			*						*				x		*						*			
	Kidney													x											
	Liver													x											
	Fat													x											

* Common control, one per sacrifice.

1) Enlarged liver

x) Samples at So 2nd St (12 mo. sacrifice)
(Samples of all tissues through 6 mo. sacrifice of So 2nd St.)

MONS 042303

#17300 Toxicity, Reproduction and Residue Study on Aroclor in White Leghorn Chickens, Adverse Effects

		Test Sacrifice (1)						1st wk	2nd wk	3rd wk	4th wk	1st wk	2nd wk	3rd wk	4th wk	1st wk	2nd wk	3rd wk	4th wk	Recovery Sacrifice (2)					
		1 ppm		10 ppm		100 ppm		1 ppm				10 ppm				100 ppm				1 ppm		10 ppm		100 ppm	
		♂	♀	♂	♀	♂	♀	Recovery Period												♂	♀	♂	♀	♂	♀
Aroclor 1242	Muscle	m(1)	m(1)		m(1)																				
	Kidney	m(1)	m(1)		m(1)																				
	Liver	m(1)	m(1)		m(1)																				
	Fat	m(1)	m(1)		m(1)																				
	Egg																								
Aroclor 1254	Muscle	m(1)	m(1)	m(1)	m(1)																	m(1)	m(1)		
	Kidney	m(1)	m(1)	m(1)	m(1)																	m(1)	m(1)		
	Liver	m(1)	m(1)	m(1)	m(1)																	m(1)	m(1)		
	Fat	m(1)	m(1)	m(1)	m(1)																	m(1)	m(1)		
	Egg																								
Aroclor 1260	Muscle	m(1)	m(1)	m(1)	m(1)	m(1)	m(1)																		
	Kidney	m(1)	m(1)	m(1)	m(1)	m(1)	m(1)																		
	Liver	m(1)	m(1)	m(1)	m(1)	m(1)	m(1)																		
	Fat	m(1)	m(1)	m(1)	m(1)	m(1)	m(1)																		
	Egg																								
Control	Muscle	m(a)	m(a)	m*								m*								m*					
	Kidney	m(a)	m(a)																						
	Liver	m(a)	m(a)																						
	Fat	m(a)	m(a)																						
	Egg																								

* Common control
(1) Between 22nd and 26th week
(2) Between 26th and 30th week

a) Lower body weights
b) Lower feed consumption
c) Lower egg production

d) Poor hatchability of eggs
e) Thinner egg shells
m) Mortalities (number)

Please note: Results above are for the test groups rather than individual birds or tissues.

MONS 042304

ppm Wet Weight Basis

		3 Mo. Sacrifice					
		1 ppm		10 ppm		100 ppm	
		♂	♀	♂	♀	♂	♀
1) Aroclor 1242	Muscle	0.4	1	0.7	2	4	6
	Kidney	0.2	(2) 2	0.5	0.3	0.8	0.9
	Liver	0.1	0.1	0.2	0.3	0.9	0.9
	Fat	5	6	12	12	44	75
Aroclor 1254	Muscle	2	1	8	8	31	62
	Kidney	0.2	0.2	1	1	5	8
	Liver	0.1	0.1	1	1	18	12
	Fat	8	3	84	66	306	211
Aroclor 1260	Muscle	9	2	6	11	11	10
	Kidney	0.1	0.5	1	4	14	18
	Liver	0.2	—	1	2	18	19
	Fat	—	28	110	724	366	599

1) Calculated as Aroclor 1254

2) Aroclor 1260

ppm Lipid Weight Basis

		3 Mo. Sacrifice					
		1 ppm		10 ppm		100 ppm	
		♂	♀	♂	♀	♂	♀
1.) Aroclor 1242	Muscle	5	10	13	18	64	89
	Kidney	7	72 (2)	21	16	41	40
	Liver	10	12	20	31	82	72
	Fat	6	6	13	13	53	81
Aroclor 1254	Muscle	9	12	85	107	486	1026
	Kidney	11	12	54	41	233	323
	Liver	15	20	108	122	3239	1137
	Fat	8	3	86	66	329	2362
Aroclor 1260	Muscle	98	16	44	76	137	156
	Kidney	7	22	66	142	606	831
	Liver	20	—	112	93	1539	1815
	Fat	—	32	138	760	1060	672

1.) Calculated as Aroclor-1254

2.) Aroclor 1260

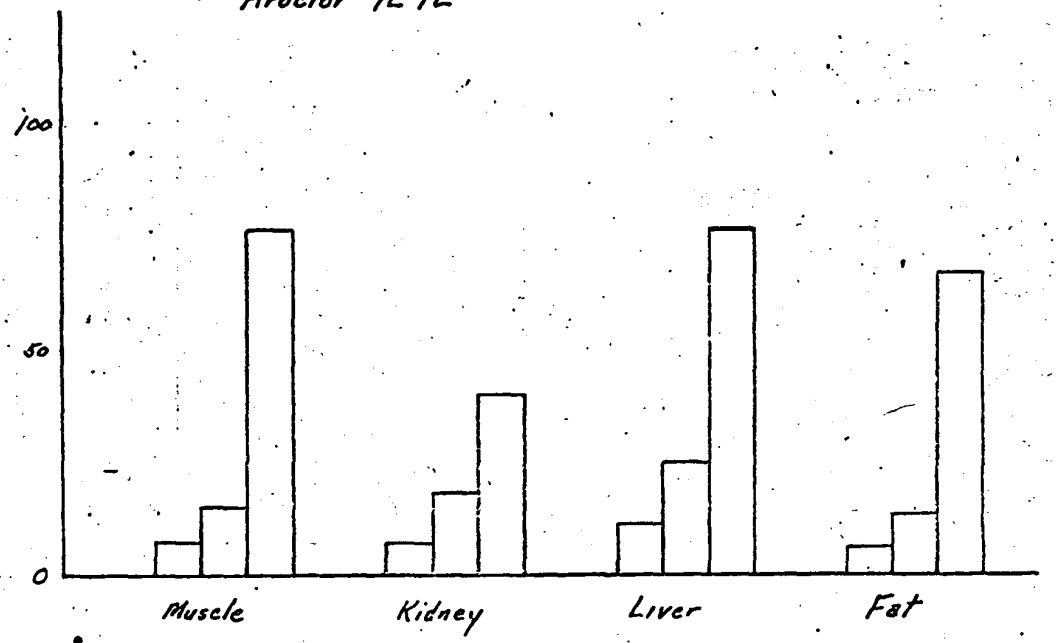
		3 Mo. Sacrifice					
		1 ppm		10 ppm		100 ppm	
		♂	♀	♂	♀	♂	♀
1) Aroclor 1242	Muscle	5	10	13	18	64	89
	Kidney	7	72 [2]	21	16	41	40
	Liver	10	12	20	31	82	72
	Fat	6	6	13	13	53	81

1) Calculated as Aroclor 1254

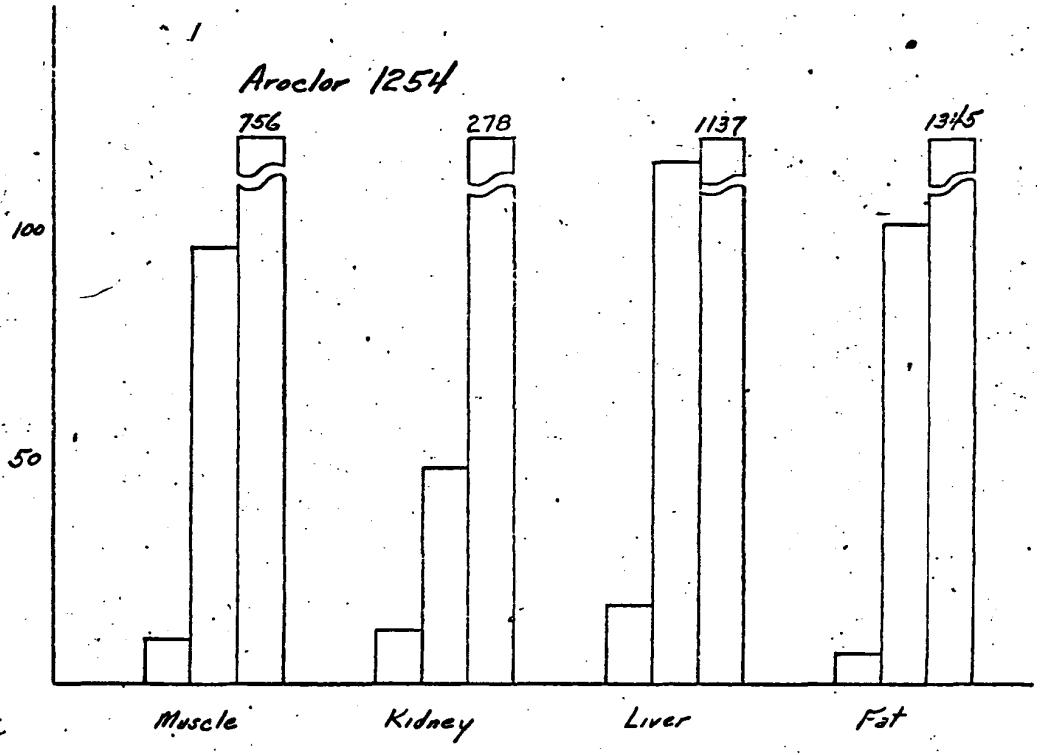
2) Aroclor 1260

		3 Mo. Sacrifice					
		1 ppm		10 ppm		100 ppm	
		♂	♀	♂	♀	♂	♀
Aroclor 1254	Muscle	9	12	65	107	486	1026
	Kidney	11	12	54	41	233	323
	Liver	15	20	108	122	3239	1137
	Fat	8	3	86	66	329	2362

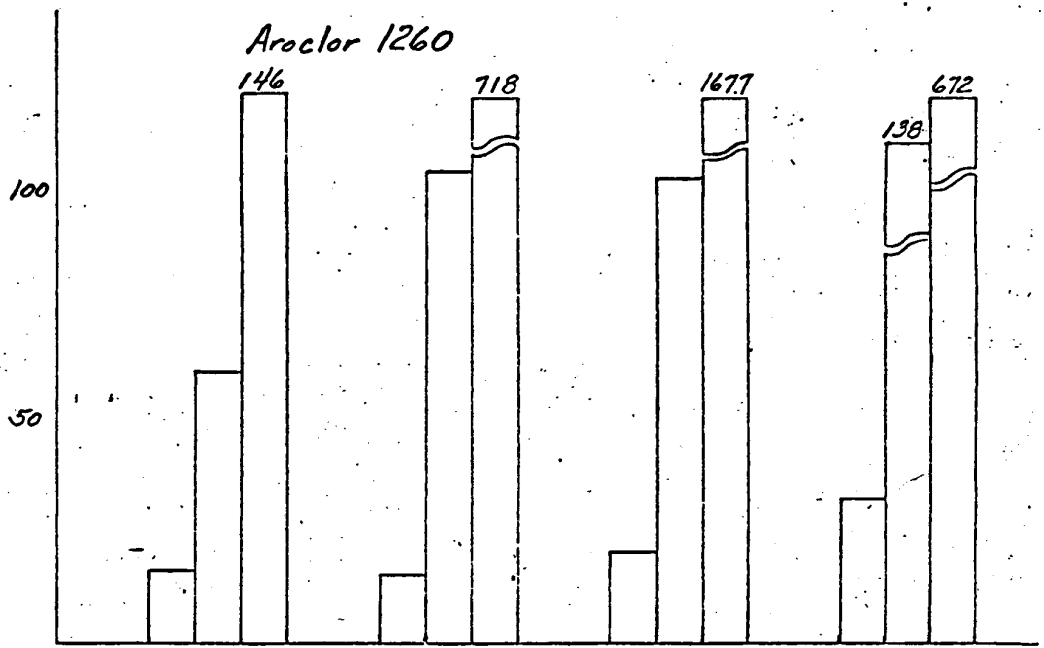
		3 Mo. Sacrifice					
		1 ppm		10 ppm		100 ppm	
		♂	♀	♂	♀	♂	♀
Aroclor 1260	Muscle	98	16	44	76	137	156
	Kidney	7	22	66	142	606	831
	Liver	20	—	112	93	1539	1815
	Fat	—	32	138	760	1060	672



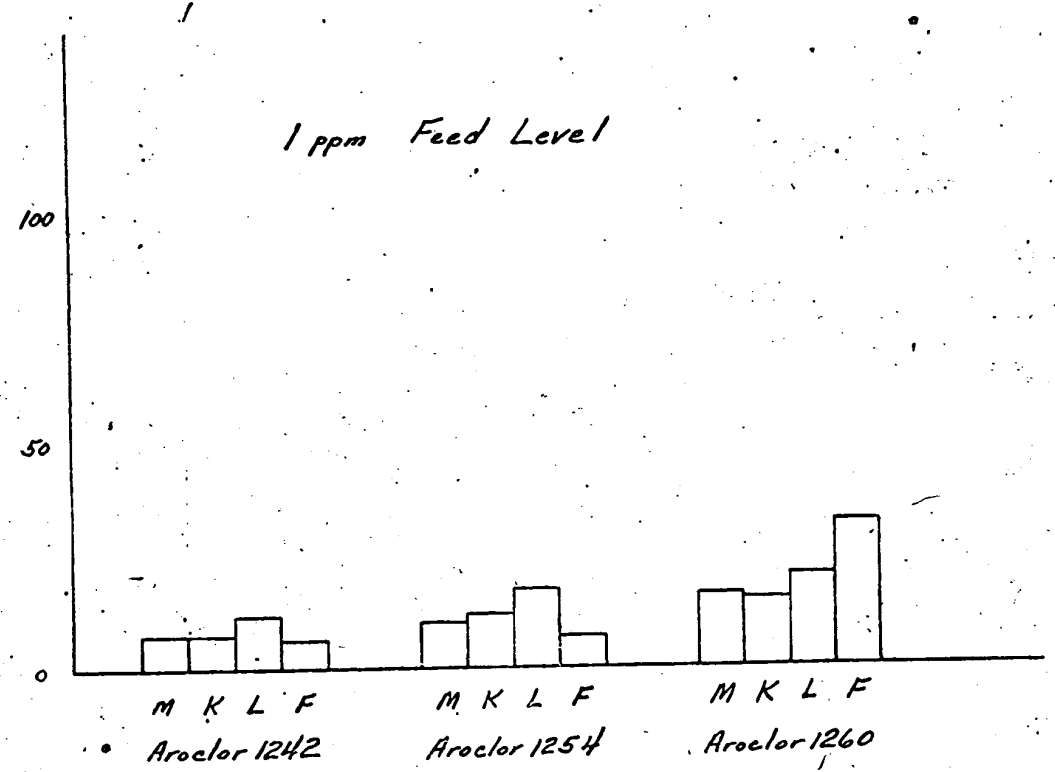
MONS 042310


Aroclor 1254
756
278
1137
1345
100
50
Muscle
Kidney
Liver
Fat

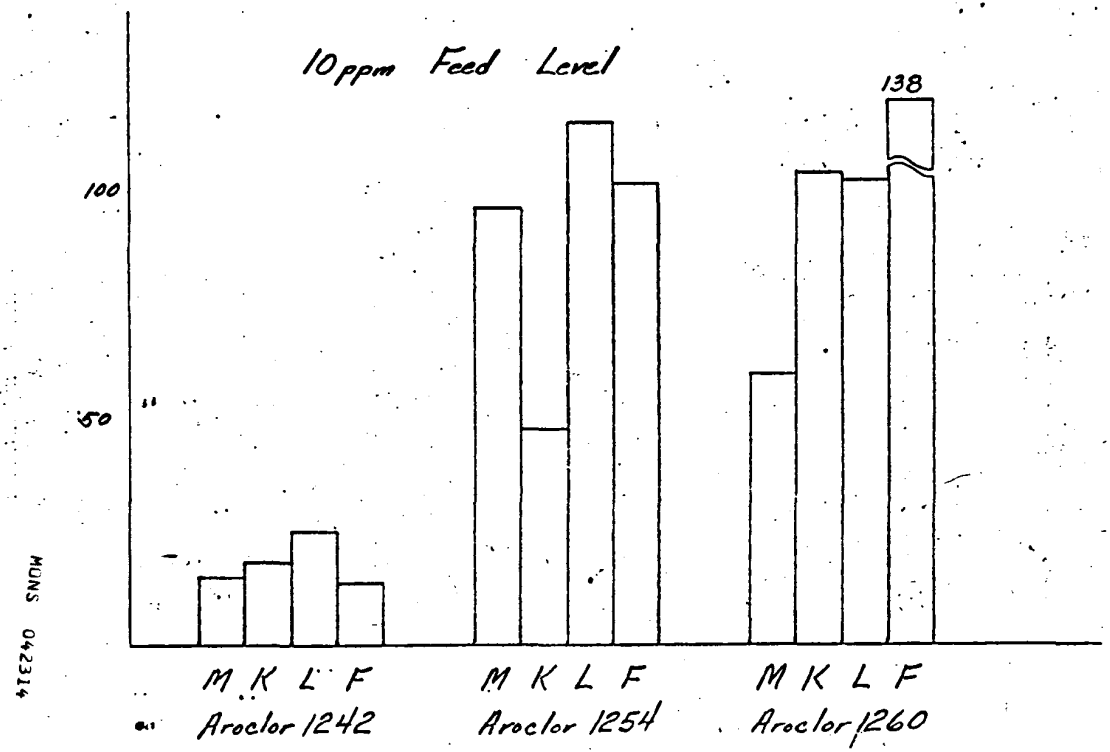
MDNS 042311


Aroclor 1260
146
718
1677
672
198
100
50
Muscle
Kidney
Liver
Fat

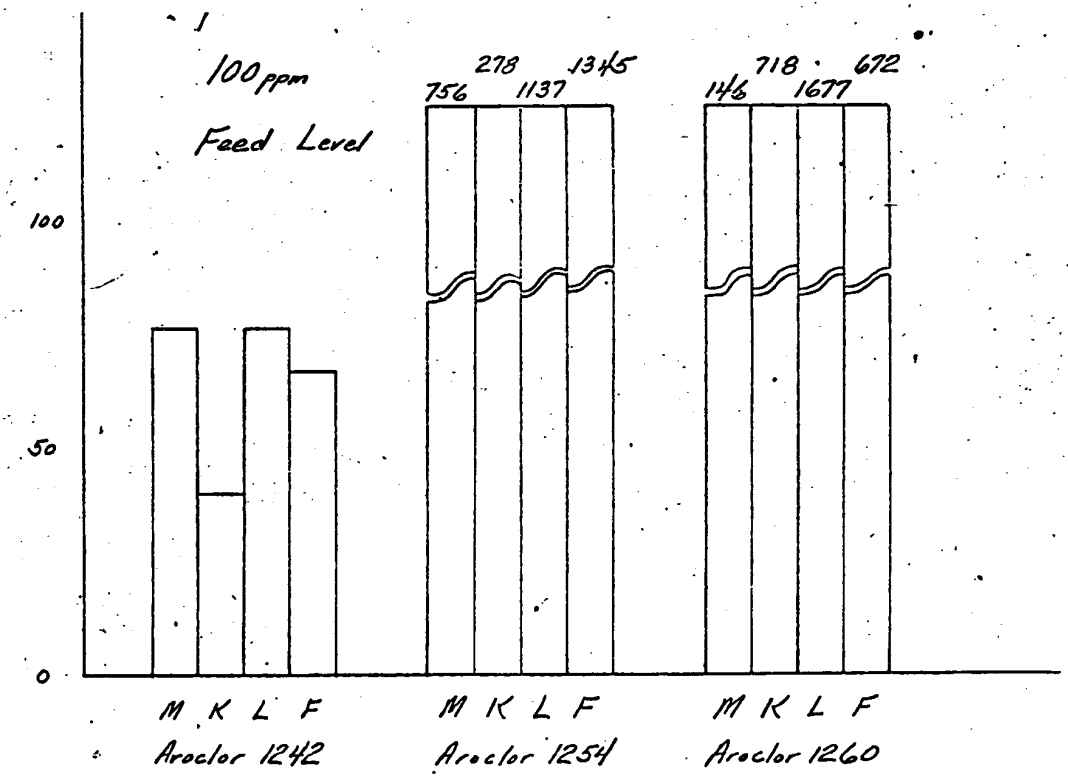
MUNS 042312


1 ppm Feed Level
100
50
0
M K L F
M K L F
M K L F
Aroclor 1242
Aroclor 1254
Aroclor 1260

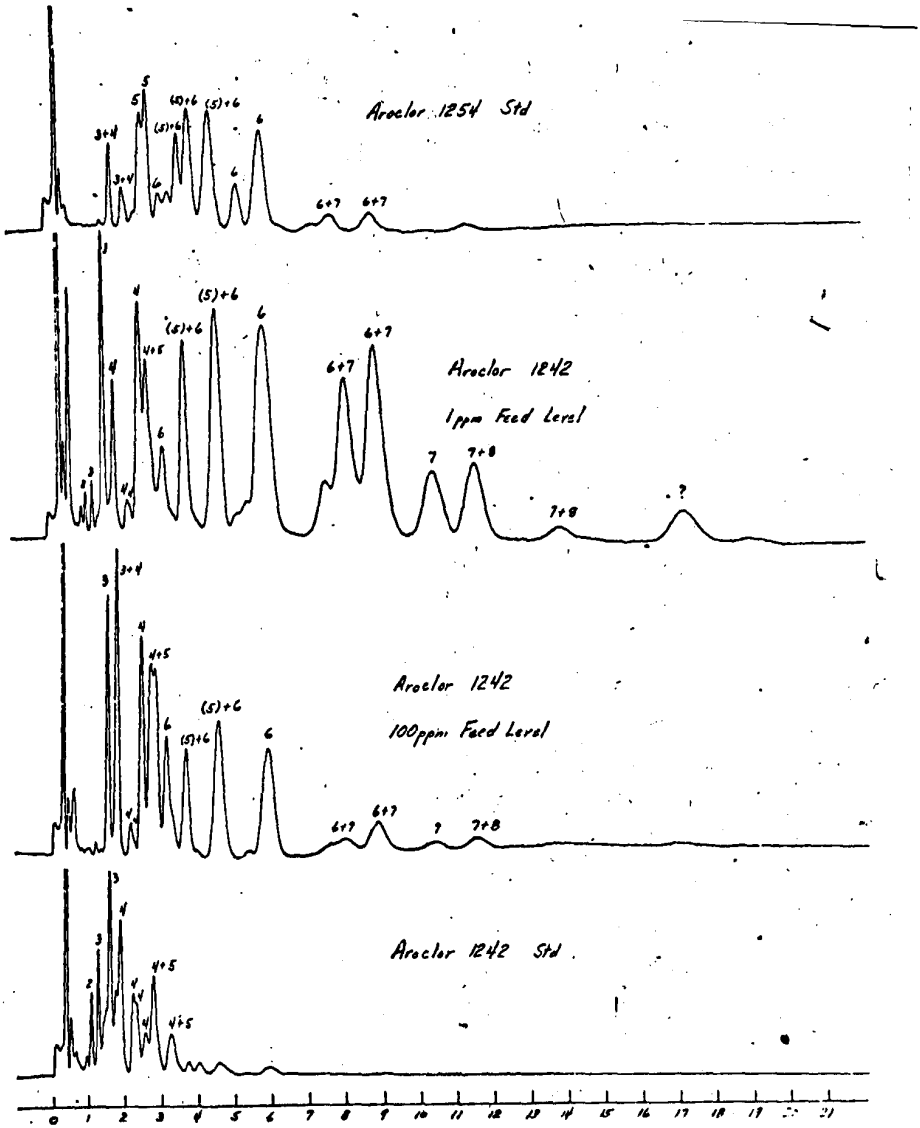
MONS 042313


10 ppm Feed Level
138
100
50
M K L F
Aroclor 1242
M K L F
Aroclor 1254
M K L F
Aroclor 1260

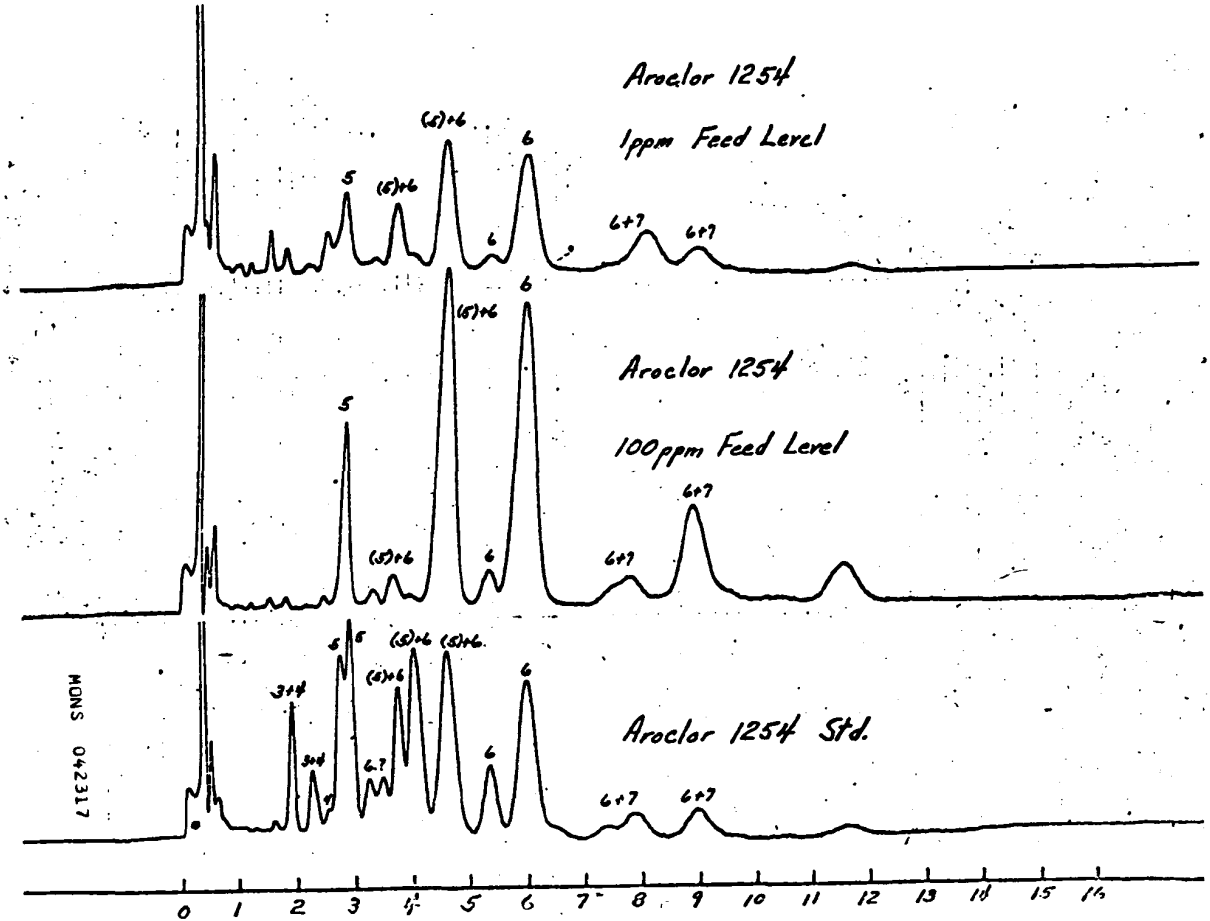
MONS 04314

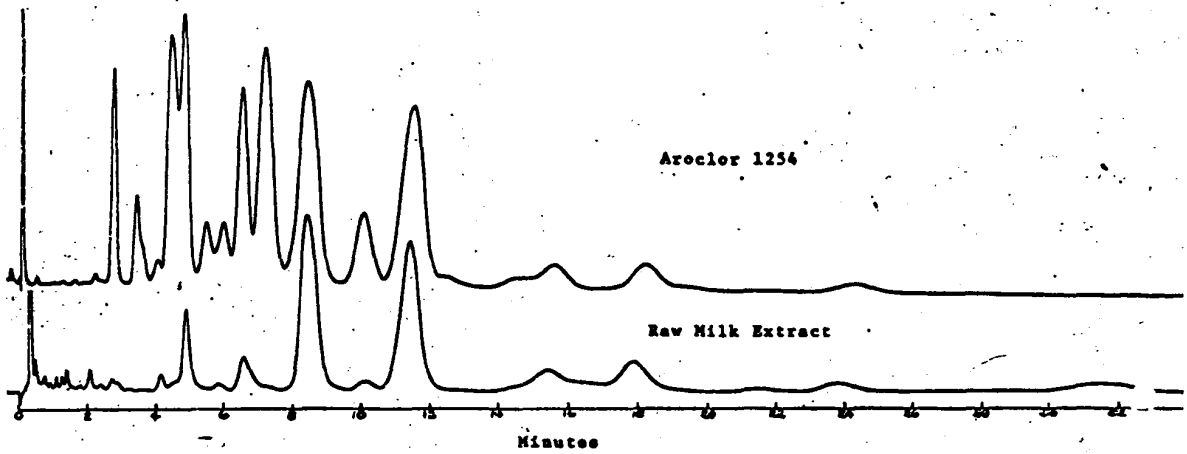

100 ppm
Feed Level
100
50
0
756
278
1137
1345
146
718
1677
672
M K L F
Aroclor 1242
M K L F
Aroclor 1254
M K L F
Aroclor 1260

MUNS 042315

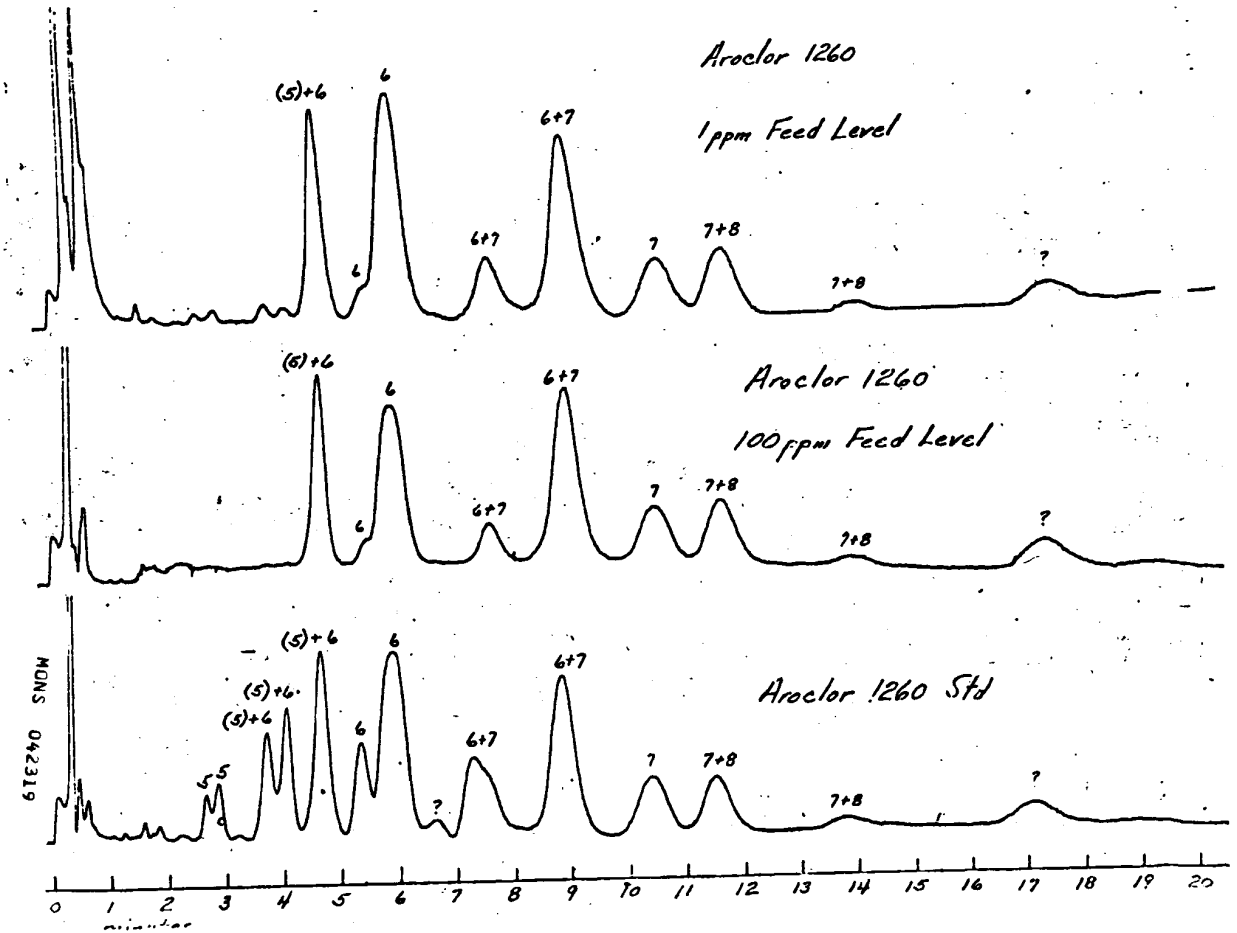

Aroclor 1254 Std
Aroclor 1242
1ppm Feed Level
Aroclor 1242
100ppm Feed Level
Aroclor 1242 Std.
minutes

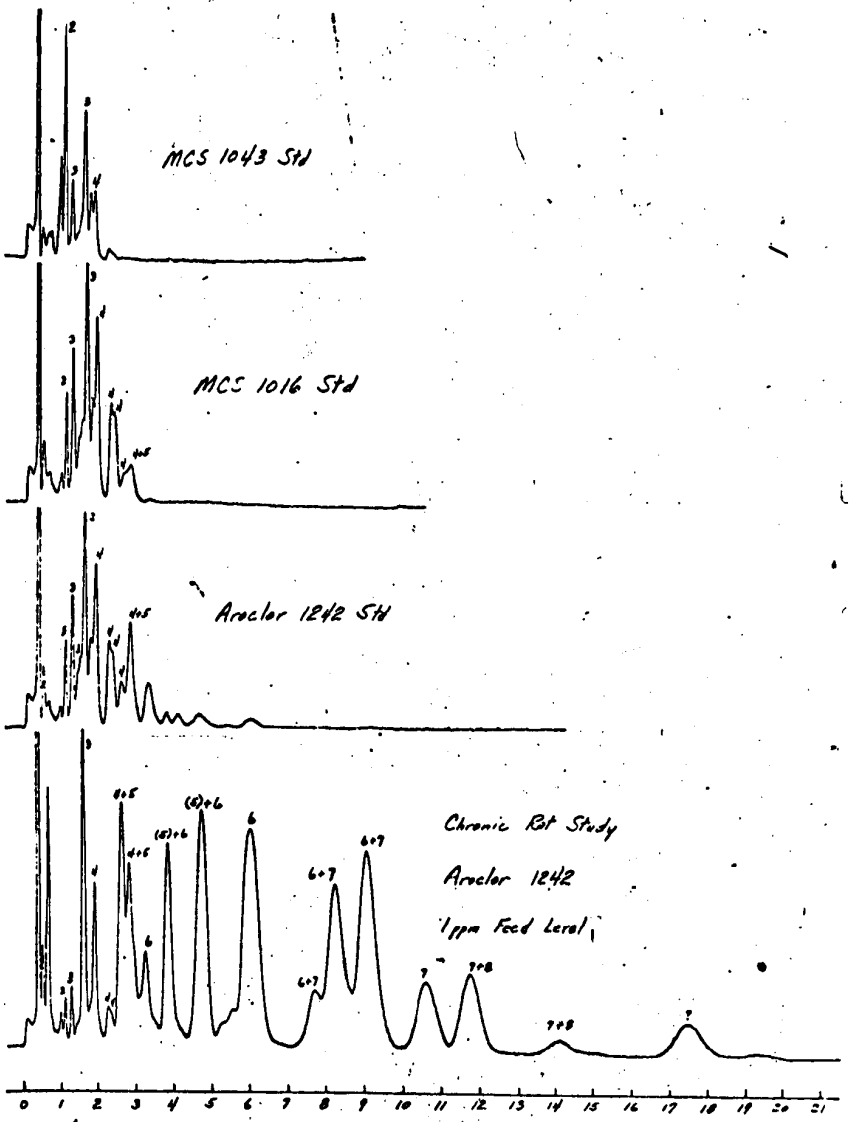
MONS 042316


Aroclor 1254
1ppm Feed Level
5
(5)+6
(5)+6
6
6+7
6+7
Aroclor 1254
100ppm Feed Level
5
(5)+6
(5)+6
6
6
6+7
6+7
Aroclor 1254 Std.
3+4
3+4
5
6
(5)+6
(5)+6
(5)+6
6+7
6
6
6+7
6+7
MONS 042317
0
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16


Aroclor 1254
Raw Milk Extract
0
2
4
6
8
10
12
14
16
18
20
22
24
26
28
Minutes

MONS 042318


Aroclor 1260
1 ppm Feed Level
(5)+6
6
6+7
7
7+8
?
Aroclor 1260
100 ppm Feed Level
Aroclor 1260 Std
5
MONS 042319
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20


MCS 1043 Std
MCS 1016 Std
Aroclor 1242 Std
Chronic Rat Study
Aroclor 1242
1 ppm Feed Level
minutes
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21

MONS 042320