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**FINAL REPORT:
BIOLOGICAL BREAKDOWN OF PESTICIDES IN
LAKE APOPKA NORTH SHORE RESTORATION AREA
SOIL IN A MESOCOSM EXPERIMENT**



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Introduction:

Design and implementation of *in situ* remediation of organochlorine pesticide (OCP) typically involves a series of pilot tests of increasing scale to optimize and evaluate conditions for efficient removal of the contaminant. Bench scale microcosm studies are conducted initially because of the feasibility of testing a broader range of remediation strategies than would be possible with field scale tests. Information obtained from bench scale microcosms is subsequently used to design tests of larger scale, such as mesocosm or field scale studies.

As a prelude to larger scale tests, bench scale microcosm studies were conducted to investigate processes likely to affect biological degradation of toxaphene and DDT in Lake Apopka soils under anaerobic conditions. After one month of incubation, the greatest disappearance was observed with H₂, acetate or lactate serving as electron donors. Laboratory microcosms such as those described above provide valuable information, but are not intended to mirror conditions in the field. High concentrations of substrates, including toxaphene, were used to maximize the response of the indigenous community so that processes leading to degradation could be identified after short term incubations. Larger-scale mesocosms are more representative of the field site in both environmental and experimental respects to evaluate the efficacy of relatively inexpensive strategies for decontamination of OCPs in soil.

Concepts developed from the microcosm studies were tested in a series of replicated tanks containing soil from the Lake Apopka North Shore Restoration Area (NSRA) using selected carbon and energy sources (Table 1). Based on prior results, lactate and plant materials were tested as sources of electron donors. Readily available plant material was used in these experiments as a source of fermentable carbon that would likely lead to the production of a variety of electron donors including H₂; H₂ cannot be safely or practically injected into these mesocosms, but indigenous fermentative microorganisms will convert available cellulose to H₂ and other fermentation products until N becomes limiting.

Bioremediation strategies for detoxification of persistent OCPs frequently rely on cycling between anoxic and oxic systems (Gavrilescu 2005); anoxic conditions promote dechlorination reactions, which serve as a preparatory stage for oxidative attack of the remaining compound. The ability to control hydrology at NSRA makes such cycling feasible, and one cycle of anoxic/oxic incubations were included in these studies.

Study Methods: Experimental details

Soil

Prior to determining the site for soil collection for the mesocosm studies, soil samples were taken from five separate sites at NSRA. Candidate sites were selected, marked, and cleared of vegetation by district personnel. Samples from the individual sites were composited from five randomly selected spots at each site. The soil from each site was initially mixed on site, transported to the laboratory in Gainesville, where soil was thoroughly mixed by hand in the laboratory. Approximately half the soil in each sample was shipped to Pace Analytical Services, Inc. for analysis of OCP residues, and the other half was analyzed at the University of Florida using accelerated solvent extraction (ASE) methods as described below.

The site chosen for soil collection for use in the mesocosms was ZSS0750, located in a field south of Lust Rd (UTM coordinates, X 44411, Y 3171355), and was selected on the basis of OCP concentration and ease of access (Fig. 1). The OCP values reported by Pace for this site were: toxaphene, 9200 µg/kg; DDT, 130 µg/kg; and DDTx, 1620 µg/kg (including DDT, DDE, and DDD).

Approximately 6 cubic yards of soil were collected from a 13 ft x 13 ft plot at this site on December 12, 2005. The soil was mixed at the site using a trackhoe by a hazardous materials contractor under the direction of District personnel (Fig. 2). The soil was loaded onto a dump truck and transported to the Soil and Water Science Department's greenhouses on the University of Florida campus in Gainesville.

Mesocosms

Once at the University of Florida greenhouses, soil was initially manually mixed with shovels, and added to the tanks sequentially, one wheel barrow at a time. Mixing was deemed to be satisfactory, as indicated by the relatively low variability within OCP concentrations within and between tanks, as reported by Pace Analytical Services.

Mesocosms were based on a design used by the Wetland Biogeochemistry Laboratory at the University of Florida. Mesocosms were constructed from 150 gallon Rubbermaid "Farm Tough" stock tanks; these are constructed of high density polyethylene and are approximately 35 inches (88.9 cm) wide at the top, 54 inches (137.16 cm) long at top, and 23 inches (58.42 cm) deep, with drains built into the sides of the tanks. Prior to addition of soil, a layer of gravel approximately five inches (12.5 cm) thick was added to the bottom of each mesocosm to facilitate drainage.

Nine mesocosm tanks were set on tables reinforced with cement block supports within the greenhouse (Fig. 3). The order of the mesocosms, from north to south, is Control-1, Cattail-1, Lactate-1; Control-2, Cattail-2, Lactate-2; Control-3, Cattail-3, Lactate-3. This design is intended to randomize any effects that location in the greenhouse might have, such as temperature. Soil was added to the mesocosm tanks to a depth of approximately 26 cm.

The top 10 cm of soil was removed from each of the cattail tanks and mixed with 1 kg dried, ground cattail in a small electric cement mixer. Cattail for this study was harvested from Lake Alice on the University of Florida campus. This level of plant material is not related to standing crop on site (which has little or no cattail), but rather

that amount of cattail that could be easily obtained and plowed into the soil. The mixture was then added back to the appropriate tanks. On January 10, 2006, tap water was added to all of the mesocosm tanks to a height of 10 cm above soil surface. For the lactate mesocosms, lactic acid was mixed with the tap water in 20 L containers to form a final concentration of 10 mM lactate. Approximately 160 L lactate solution was required to fill each tank to 10 cm above soil surface. The level of water was maintained through addition of water every two weeks. Levels of lactic acid were similarly maintained through addition of 10 mM lactate every two weeks.

The redox potentials in all tanks stabilized by February 21, 2006, which was considered the starting point for analyses. Anoxic incubation continued until June 8, 2006, at which time the water was removed from the tanks to begin the aerobic phase. Water was removed by siphon from the top of the tanks, and allowed to drain from the bottom through the drain.

Sample collection:

When redox potentials in the mesocosms stabilized, ten samples were taken every two weeks from each mesocosm to a depth of approximately 5 cm using a 50 ml disposable syringe with the end cut off. The use of a syringe was necessary because the soils in all mesocosms exhibited a slurry-like consistency that precluded the use of more traditional core samplers. The ten samples from each mesocosm were mixed in the laboratory to form composite samples. Gravimetric water contents were determined on a subsample of the composite, and the remaining sample was frozen at -80°C until analysis as described below.

Redox potential measurements:

Three redox probes were permanently installed in each tank (one at each end, approximately 30 cm from the wall, one in the center) at a soil depth of 5 cm. Redox potential (Eh) was determined at weekly intervals during the anoxic treatment. Eh was measured with permanently installed platinum electrodes; platinum wire (2 mm diameter, 15 mm length) was welded on a copper wire protected with insulation material. Eh was read using ORION SA 230, (Orion Research Inc., Boston). A photograph of permanently installed redox probes is presented in Fig. 4.

Water content:

Volumetric water content was measured for all samples collected. Samples were weighed, dried at 105°C overnight, and reweighed to calculate water contents.

Temperature measurement:

Temperature in each mesocosm was determined every two weeks at the time of sampling at a depth of 5 cm with a glass thermometer.

Organic acid analysis:

Acetate is an important metabolite of cellulose fermentation; thus, acetate concentrations provide information on the functioning of the system. Acetate and lactate concentrations in pore water were determined by high pressure liquid chromatography equipped with a UV detector (Waters, Co), with Aminex HP 87H column as the separating column (300 X 7.5 mm) and sulfuric acid (5mM) as eluent.

OCP analysis:

Toxaphene, DDT, DDE and DDD concentrations were determined by extraction with accelerated solvent extraction (ASE) and gas chromatography. Preliminary tests conducted to investigate the influence of soil moisture content on OCP extraction by ASE indicated that 50% water content yielded the greatest efficiencies of recovery. Moisture content had been previously shown to significantly affect OCP recovery by extraction (S. Richter, personal communication), such that moisture contents between 35% and 50% was found to be the optimum for OCP analysis by sonication.

Samples were air dried, ground, and sieved prior to analysis. Water contents of sieved soils were adjusted to 50% water content and allowed to equilibrate for three days in a closed dessicator.

Surrogates: Two surrogates (TCMX and decachlorobiphenyl) were added to all samples prior to extraction by ASE, including the calibration standards, the QC samples, and the field samples.

Accelerated Solvent Extraction:

Four grams of each treatment were mixed with 2 g hydromatrix to dry the samples, and extracted in a Dionex ASE 100 Accelerated Solvent Extractor. Extractant was an 80:20 mixture of methylene chloride:acetone. The extracted compounds were collected in the eluate fraction of between 30 and 40 mL of methylene chloride:acetone, and concentrated by an N₂ stream to remove acetone. The solvent was exchanged to hexane. If residual deposit was noted on the sides of the container, fresh hexane was added to the container in an effort to redissolve this precipitate, and the fresh mixture analyzed for OCP. A one mL aliquot was further purified by passage through Florisil for analysis of DDT, DDE and DDD, and another one mL aliquot purified by sulfuric acid treatment for further analysis of toxaphene (see below).

Florisil Cleanup:

DDT, DDD, and DDE were prepared by passage of the pesticide mixture in a 9:1 hexane:acetone mixture through a disposable prepacked Florisil column (Varian, Palo Alto, CA). Eluates were analyzed by gas chromatography.

Sulfuric Acid Cleanup:

Equal amounts of concentrated sulfuric acid and the sample were mixed in a 10 mL vial. The vial was capped and vortexed for 1 min. Phases were allowed to separate, and the top (organic) layer transferred to a clean vial for analysis by gas chromatography.

Gas Chromatography

OCP concentrations were evaluated by gas chromatography following EPA SW-846 Methods 8000B and 8081B.

Calibration Curve: A set of six different standards were run in triplicate during each set of analyses, such that each set of standards was run consecutively. The calibration curve was verified by analyzing one set of standards after every ten samples. The acceptance limits for the verification standards compared to the initial calibration are +/- 15%. If the verification standard recovery was not within these limits, all samples since the last verified control standard were evaluated for possible reanalysis. Concentrations of unknowns were determined via a weighted linear least squares regression approach.

Laboratory Control Sample (LCS): A clean sample (sand) spiked with a known amount of target analytes was included in each run. This LCS should yield a GC output of approximately half scale for as many analytes as possible. A matrix spike and duplicate were also included in each set of analyses.

Data Analysis

All results are reported on a dry weight basis. Data were analyzed using Statistical Analysis System by a blocking replicated factorial design. Pairwise comparisons for differences between means were performed by Tukey's Studentized test. The null hypothesis was rejected at $p < 0.10$, thus the probability of observing no difference among the pair wise is less than 10%.

Results:

Effects of water contents on OCP extraction efficiencies

The effects of soil moisture content on efficiency of extraction of OCPs via both ASE and Soxhlet extraction were investigated for the NSRA soil at the suggestion of District personnel. Three soil moisture contents (17%, 50%, and 70%) were tested (Table 2), and moisture content proved to be significant in determining extraction efficiency for toxaphene and DDT via ASE, and for DDD and DDT for Soxhlet. DDD and DDE extraction efficiencies tended to be higher at 50% moisture content for ASE, although not significantly so. On the basis of these data, samples were equilibrated to 50% moisture contents prior to extraction and analysis.

Mixing of soil

Homogeneous mixing of soil is of concern when working with volumes of soil as large as was required for this study. Mixing should be sufficiently complete prior to adding soil to the mesocosms. Initial OCP concentrations provided by Pacelabs are presented in Table 3. As can be seen from the coefficients of variation for these data (Table 4), starting concentrations were similar between the mesocosms (coefficients of variation typically ranging from 0.02 to 0.1), with the exception of the Control mesocosms. The relatively high coefficients of variation for this set of mesocosms (ranging from 0.28 to 0.37) are due to the low values measured for Control 1. The

exceptionally high variability observed for DDT in the lactate mesocosms (0.45) might be most easily explained by analytical problems rather than mixing problems; variabilities within the other analytes for the lactate mesocosms was approximately 0.02.

Water Relations and Temperature within Mesocosms

Water relations: Redox potentials were monitored in all tanks following flooding and throughout the anoxic phase (Fig. 5). Redox potentials in all tanks with added carbon amendments reached a minimum of ca. -250 mV approximately one month after flooding. The control mesocosms continued to fall throughout the flooded phase, as might be expected in a soil that is limited in carbon. The amendments in the experimental mesocosms provided sufficient energy source to drive the redox potentials down rapidly.

Volumetric water content was monitored in all mesocosms following draining on June 8, 2006 (Fig. 6). Drainage was slow in some tanks, such that water above soil was siphoned off. The rate of drainage was not associated with a particular treatment; it is not clear why drainage was faster in some tanks than in others. This phenomenon may have been due to some soil clogging drains. Redox potentials rose rapidly following removal of water (Fig. 7), and were in the aerobic range within two weeks of draining. Maintaining constant water contents in the tanks proved to be difficult because of the difficulty in equally distributing water across the surface of the soils with a hose. There was no ponding of water on the surfaces, and the surface of the soil was level in all tanks.

Temperature: Temperature (Fig. 8) was monitored throughout anoxic and oxic phases of the experiment. Temperature in all mesocosms increased throughout the incubation, as would be expected from an incubation begun in winter and terminated in summer. Mesocosms were housed in a greenhouse with cooling, but the cooling was not efficient during the hotter days of July and August, when the mesocosm temperatures approached 35°C. It is unlikely that these temperatures inhibited decomposition of the OCPs. The locations of the mesocosms within the greenhouse led to additional variability in temperature due to the proximity of tanks to the cooling unit. The cooling unit for the greenhouse was closest to the Control-1 mesocosm, which maintained the lowest temperature for most time points.

OCP Degradation

Toxaphene: Toxaphene concentrations at the beginning of the flooded phase were similar in all tanks (Table 5), and steadily decreased in the mesocosms with added cattail and lactate throughout the anoxic incubation period (Fig. 9). No significant differences were observed for most of the timepoints between the cattail and lactate treatments, and both were significantly lower than the control mesocosms for most timepoints. Toxaphene concentrations in control mesocosms did not decrease significantly until May 3. This decrease in the control corresponds with a decrease observed in the lactate mesocosms, but does not correlate with changes in temperature, water content or redox potential. Toxaphene concentrations in control mesocosms did not significantly decrease after this timepoint. The concentrations within the lactate and cattail mesocosms continued to decrease until May 31, the end of the anoxic phase.

At the end of the anoxic phase, toxaphene concentrations in the treatment mesocosms were significantly lower than those observed in the control mesocosms

(Table 5). No significant difference was observed between the cattail and lactate treatments, and no significant loss of toxaphene was observed during the oxic phase.

DDT, DDD, and DDE: Concentrations of DDT and its major degradation products (DDD and DDE) were monitored with time (Table 5; Figs. 10, 11, 12). No differences were noted in the starting concentrations of DDT and its major degradation products (DDD and DDE) between the tanks at the beginning of the flooded phase (Table 5). Concentrations of DDT declined constantly in the cattail mesocosms throughout the anoxic phase. A similar decrease was not noted in the lactate mesocosms until May 3 (Fig. 10). Concentrations of DDT at the end of the anaerobic phase were significantly lower in the lactate and cattail mesocosms than in the control mesocosms (Table 5). No significant difference was observed for DDT at the end of the aerobic phase, likely due to high variability in the controls.

The primary decomposition products of DDT under anaerobic conditions are DDD and DDE (Fig. 13). DDD further degrades to a variety of compounds under both aerobic and anaerobic conditions (Figs. 13, 14); however, DDE degradation under anaerobic conditions is very slow. An increase in DDD concentrations in the cattail mesocosms was noted soon after the beginning of the flooded phase (Fig. 11), such that a corresponding decrease in DDT concentrations might be expected. Such a corresponding decrease was not observed (Fig. 10), although it may be that the higher variability observed in the DDT analyses than in the DDD (as indicated by the size of the error bars in Figs. 10 and 11) may have masked a decrease in DDT concentrations. An increase in DDD concentrations was observed in the lactate and cattail mesocosms toward the end of the flooded phase, corresponding with the relatively large decrease observed in those tanks for DDT and adding confidence that the observed degradation of DDT was real. DDD concentrations were significantly higher in the lactate mesocosms than in the control and cattail mesocosms at the end of the flooded phase, and DDD was higher in both lactate and cattail mesocosms than in the controls at the end of the aerobic phase, suggesting aerobic conversion of DDT to DDD. Levels of DDT were somewhat lower in the lactate and cattail mesocosms than in the controls at the end of the aerobic incubation, although not significantly so. This lack of significance is again likely due to the relatively high variabilities in the DDT measurements.

DDE degrades very slowly, if at all, under anaerobic conditions. No loss of DDE was observed in any treatment during either the anoxic or oxic phase (Fig. 12, Table 5).

Discussion

These mesocosm studies supported and extended findings observed during a previous laboratory scale microcosm on degradation of OCPs in NSRA soils. In general, added electron donors in the form of ground cattail and lactate increased degradation of toxaphene relative to control mesocosms, and stimulated production of DDD, the primary degradation product of DDT, under anoxic conditions.

Our results indicate that toxaphene and DDT degradation can be increased in these soils through incorporation of an appropriate electron donor. Both ground cattail and lactate stimulated degradation of toxaphene and DDT under anaerobic conditions, indicating that degradation in these soils was limited by electron donors. Soon after flooding, gas production was observed in all cattail mesocosms, which persisted for

several weeks. Gas production, coupled with the rapid decrease in redox potentials observed in all lactate and cattail mesocosms indicates a general response of the microbial community to the addition of carbon, and a limitation of available carbon in the soils. Potential nitrogen and phosphorus limitations at later stages of the incubation are possible although the degradation of DDD throughout the oxic phase argues against a general limitation of N and P.

Much of the observed degradation of OCPs occurred prior to draining, with significant degradation observed for toxaphene and DDT in the cattail and lactate mesocosms throughout the anoxic phase. Continued degradation was not observed during the oxic phase for these OCPs, suggesting that the primary route for toxaphene and DDT degradation in these soils is anaerobic. The lack of degradation of any OCP, with the exception of DDD, during the oxic phase was somewhat surprising. It was expected that toxaphene in particular would be susceptible to degradation following the anoxic phase. Reductive dehalogenation under anoxic conditions typically prepares halogenated compounds such as toxaphene for degradation under oxic conditions. The results suggest that the most efficient OCP degrading microorganisms are anaerobic, and that future strategies for OCP remediation at the NSRA should focus on anaerobic processes.

The observation that mesocosms with lactate and ground cattail were approximately equivalent in stimulating degradation of OCPs is worthy of note. Lactate was added every two weeks to the appropriate mesocosms, and was rapidly metabolized. No lactate or acetate was detected in porewaters of any of the mesocosms (data not shown). Relatively few microorganisms are capable of utilizing lactate as an electron donor under methanogenic conditions, and are largely restricted to secondary fermenters (syntrophs) in consortia with methanogens (e.g., Sekiguchi et al., 2006). Sulfate reducing bacteria are also capable of utilizing lactate as an electron donor either syntrophically or using their respective electron acceptors (Castro et al., 2000), and some have been implicated in halorespiration. Sulfate reduction rates in these mesocosms appeared to be quite low (i.e. no noticeable generation of sulfide), and it may be that a related organism may have utilized lactate or a fermentation product of lactate as an electron donor and the OCP as an electron acceptor (van Pee and Unversucht, 2003). Members of one group of related organisms, *Dehalococcoides* sp., have been shown to reductively dechlorinate certain compounds (Krajmalnik-Brown et al., 2004) using lactate as electron donor in consortia, and H₂ as an electron donor in pure culture (He et al., 2003). Lactate was thought to be fermented to H₂ in consortia in that study, which then served as the primary electron donor for reduction of trichloroethylene by *Dehalococcoides*. Significantly, both lactate and H₂ were identified as efficient electron donors for OCP degradation in the previous microcosm studies. *Dehalococcoides* were detected in NSRA soils (M. Coveney, personal communication); however, laboratory studies intended to enrich microorganisms capable of utilizing toxaphene as a terminal electron acceptor in the soils used in this study did not yield *Dehalococcoides* sp. In addition, polymerase chain reaction primers specific for *Dehalococcoides* sp. (Krajmalnik-Brown et al., 2004) did not amplify *Dehalococcoides* DNA sequences from soils collected from the nine mesocosms at the beginning of the mesocosm experiment (data not shown). This does not mean that *Dehalococcoides* were not involved in the dechlorination of OCPs in the mesocosms, only that they were not detected using the approaches we employed. Regardless, it is likely that an organism capable of using lactate, either directly or

indirectly, as an electron donor and OCP as an electron acceptor was responsible for degradation of OCPs in these soils. Fermentation of plant residues from the cattail treatments would also be expected to produce H₂ and lactate, thereby driving OCP degradation.

Summary and future directions

These experiments clearly demonstrated the potential for *in situ* remediation of toxaphene, DDT, and DDD in NSRA soils. Incorporation of electron donors in the form of plant residue provides a relatively inexpensive and technically simple means of stimulating biodegradation of these compounds. Anoxic conditions facilitated degradation of toxaphene more than oxic conditions, and the traditional anoxic/oxic cycle appeared to promote degradation of DDT and DDD. The capability of hydrology control at the NSRA may make such cycling feasible.

Additional strategies may provide greater degradation of both groups of compounds. The organisms responsible for metabolism of the compounds are present and can be stimulated, but they are likely limited by access to the target chemicals. It is likely that bioavailability of these hydrophobic chemicals is limited in the high organic content soils of the NSRA. Increasing the availability of OCPs to microbial attack would likely accelerate their degradation (Walters and Aitken, 2001). Possible strategies for increasing availability include use of environmentally friendly, biodegradable surfactants (Karanth et al., 1999; Kommalapati et al., 1997). Additional strategies, perhaps combined with surfactant production, might include phytoremediation (Gao et al., 2000; Lunney et al., 2004). Application of low levels of surfactants with incorporation of plant residue prior to flooding might result in increased degradation during a flooded phase, which could be coupled with an aerobic, phytoremediation strategy. Ideally, a plant may be found which would be both a source of surfactant and whose rhizosphere stimulates biodegradation of the OCPs. Most previously described phytoremediation strategies for DDT involve accumulation of the compound by the plant rather than transformation in the rhizospheres, although it may be that a thorough screening of selected plants would identify suitable candidates for transformation. Strategies such as these should be tested in laboratory studies prior to field studies.

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Table 1. Experimental design of mesocosm studies.

<u>Treatment/Control</u>	<u>Description</u>	<u>No. of Tanks</u>
Treatment – Lactate	20 mM lactate	3
Treatment – Cattail	0.04 g ground cattail per gram soil	3
Control- No electron donor	soil and water only	3
Total Mesocosms		9

Table 2: Effect of moisture content (17, 50, and 70 percent) on extraction of toxaphene, DDE, DDD, and DDT by using ASE and Soxhlet extraction. OCP concentrations are expressed as $\mu\text{g/g}$ dry weight. Asterisks (*) represent significantly different values ($p < 0.05$) within the soil and extraction method.

	ASE				Soxhlet			
	mean (17%)	mean (50%)	mean (70%)	p- value	mean (17%)	mean (50%)	mean (70%)	p- value
Toxaphene	12.637*	15.177	16.247	.0331	17.921	16.170	15.064	.3465
DDE	8.412	10.300	7.779	.2010	6.1111	7.6738	7.2111	.2271
DDD	2.9477	4.4051	2.7424	.2029	2.3340	4.1502*	2.8587	.0028
DDT	2.5978*	3.7073	4.5124	.0119	2.3437*	3.3876	4.1434	.0366

Table 3. Soil OCP concentrations in mesocosms prior to flooding. Concentrations expressed in µg/kg dry weight. Analyses provided by Pace Labs.

<u>OCP</u>	<u>Mesocosm</u>								
	<u>Control-1</u>	<u>Control-2</u>	<u>Control-3</u>	<u>Cattail-1</u>	<u>Cattail-2</u>	<u>Cattail-3</u>	<u>Lactate-1</u>	<u>Lactate-2</u>	<u>Lactate-3</u>
Toxaphene	15000	26000	25000	22000	24000	26000	23000	23000	22000
4,4'-DDT	100	220	170	160	180	150	190	170	160
4,4'-DDD	250	430	420	390	420	400	390	390	380
4,4'-DDE	1500	2800	2600	2700	2800	2200	2500	2500	2400

Table 4. Coefficients of variation of OCP concentrations for mesocosms within the three treatments prior to flooding.

<u>OCP</u>	<u>Control</u>	<u>Cattail</u>	<u>Lactate</u>
Toxaphene	0.28	0.08	0.03
4,4'-DDT	0.37	0.09	0.45
4,4'-DDD	0.28	0.04	0.02
4,4'-DDE	0.3	0.13	0.02

Table 5. Pair-wise comparison of the treatments on the degradation of pesticides. Concentrations are presented in $\mu\text{g/g}$ dry. Concentrations within a given time point sharing the same letter did not show significant difference at the $P < 0.05$ level.

	Toxaphene	DDT	DDD	DDE
Starting concentrations (Feb. 21, 2006)	Control	3.17(A)	1.54(A)	3.16(A)
	Cattail	3.34(A)	1.83(A)	3.53(A)
	Lactate	3.14(A)	1.47(A)	3.44(A)
Anaerobic period (Ending May 31, 2006)	Control	3.16(A)	1.42(B)	3.92(A)
	Cattail	2.08(B)	1.90(B)	4.18(A)
	Lactate	2.55(AB)	4.10(A)	4.11(A)
Aerobic period (June 8-Aug. 21, 2006)	Control	3.07(A)	1.87(B)	3.93(A)
	Cattail	2.74(A)	4.02(A)	4.24(A)
	Lactate	2.23(A)	4.34(A)	3.98(A)

Figure 1. Soil collection site ZSS0750 in the North Shore Restoration Area at Lake Apopka, Orange County, Florida.



Figure 2. Soil mixed in a 13 ft x 13 ft plot at site ZSS0750 by a trackhoe prior to collection and transport to the greenhouses at the Department of Soil and Water Science, University of Florida, Gainesville.



Figure 3. Mesocosm tanks in the greenhouse at the Department of Soil and Water Science, University of Florida, Gainesville.



Figure 4. Permanently installed redox electrodes in mesocosm tank. Three redox electrodes were installed in each tank.

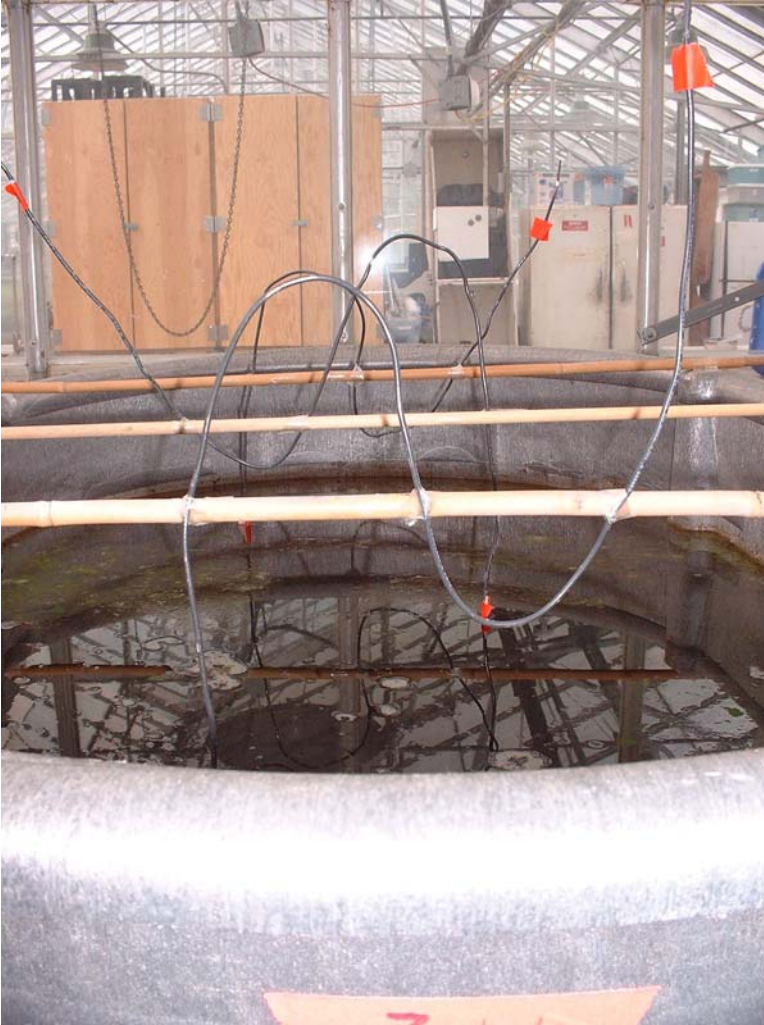


Figure 5. Redox potentials with mesocosm tanks during flooded phase.

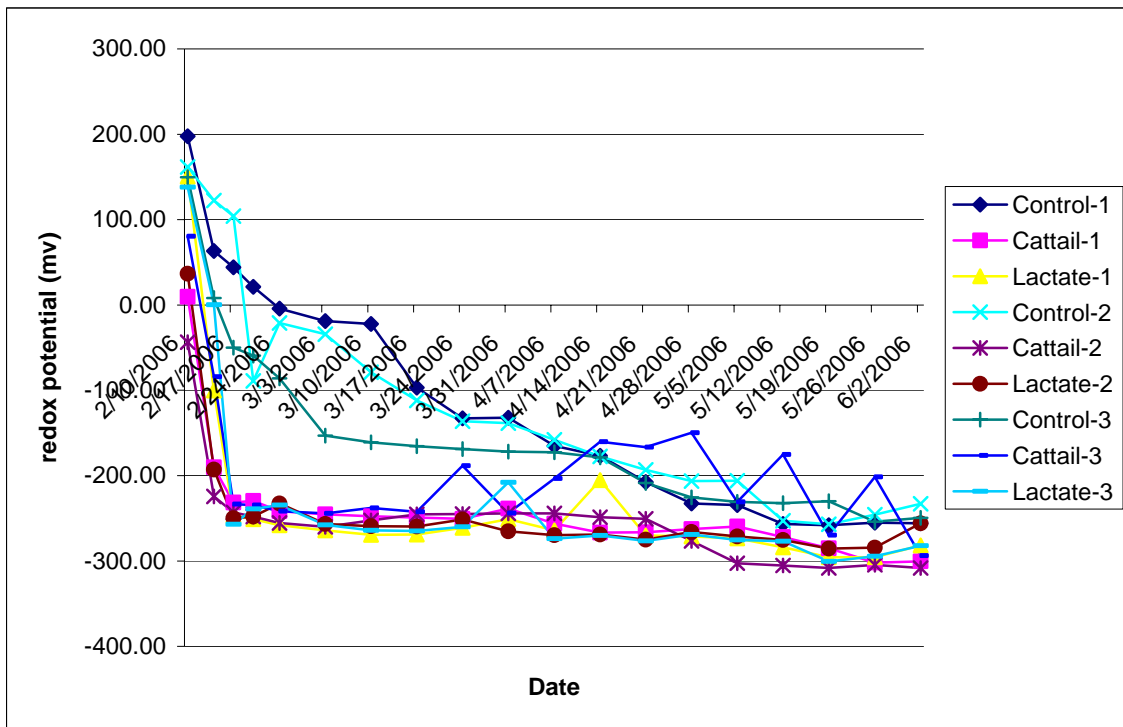


Figure 6. Volumetric water contents following draining.

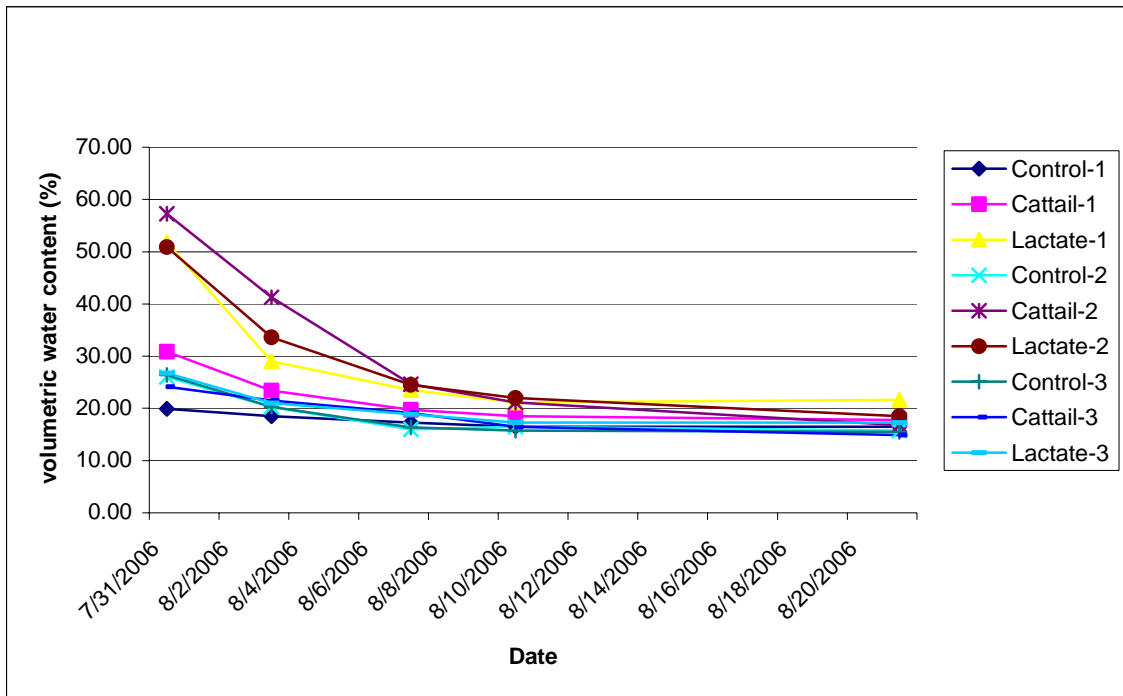


Figure 7. Redox potentials following draining.

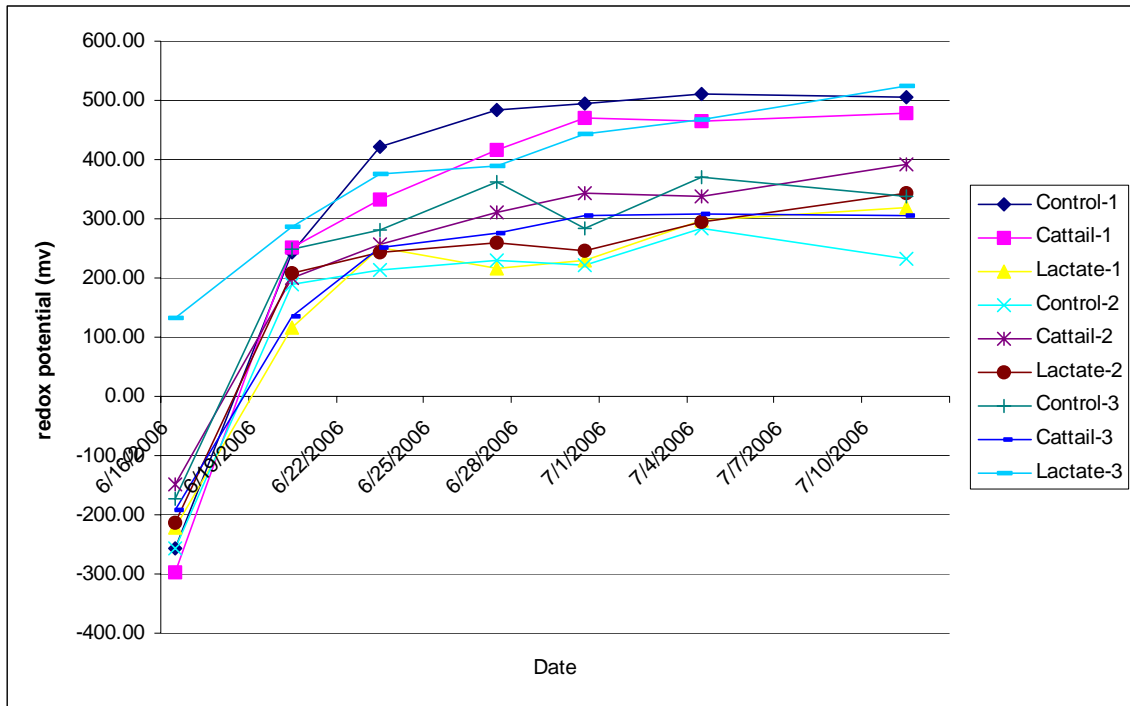


Figure 8. Temperatures within selected mesocosms from three locations at a depth of 5 cm. Vertical black arrow indicates date of draining (June 8). Mesocosms for temperature monitoring were selected on the basis of location within the greenhouse (northern, central, and southern areas).

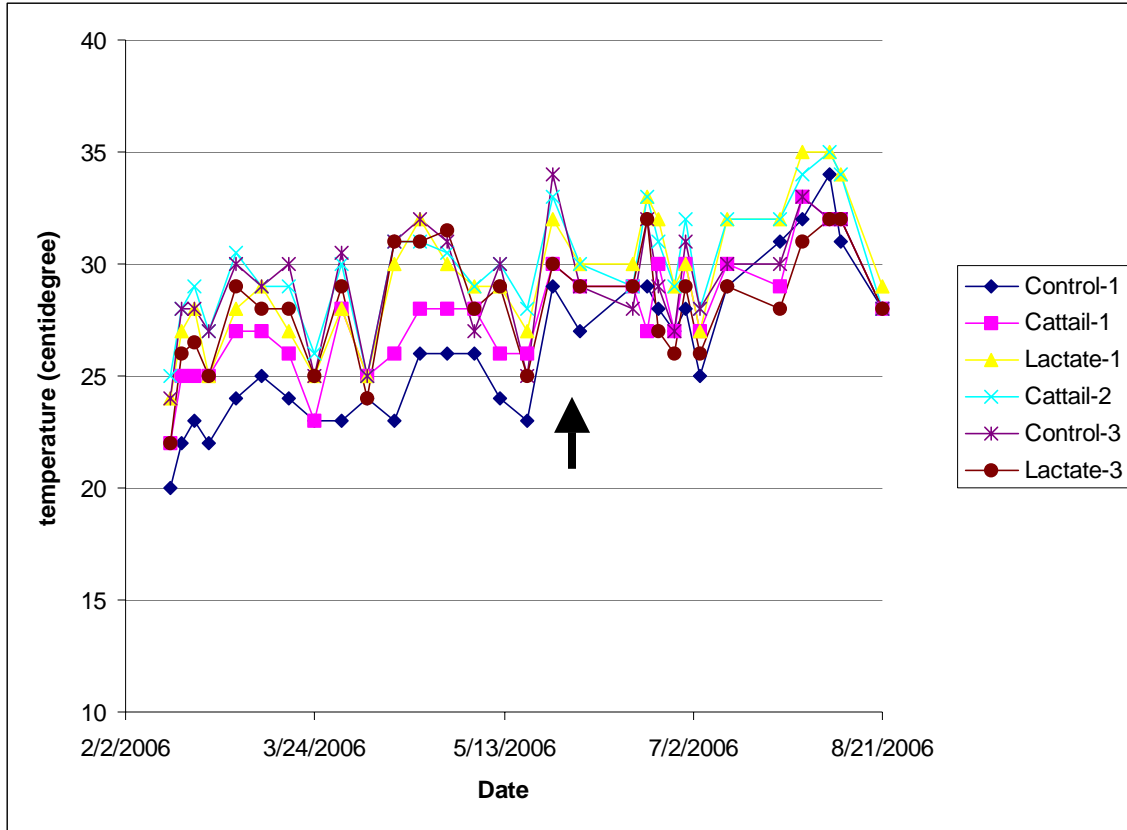


Figure 9. Concentrations of toxaphene during study. Error bars represent +/- 1 standard deviation. Vertical black arrow indicates date of draining (June 8).

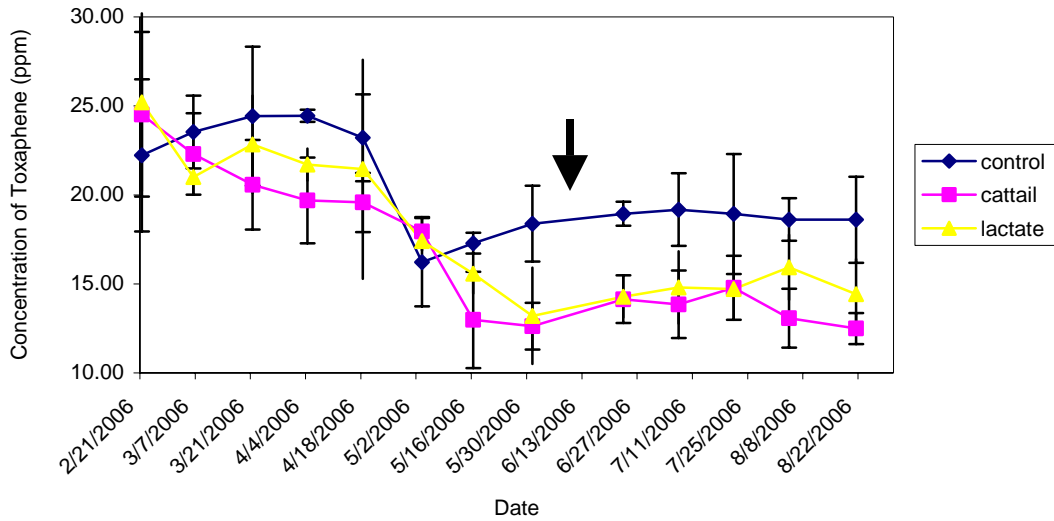


Figure 10. Concentrations of DDT during study. Error bars represent +/- 1 standard deviation. Vertical black arrow indicates date of draining (June 8).

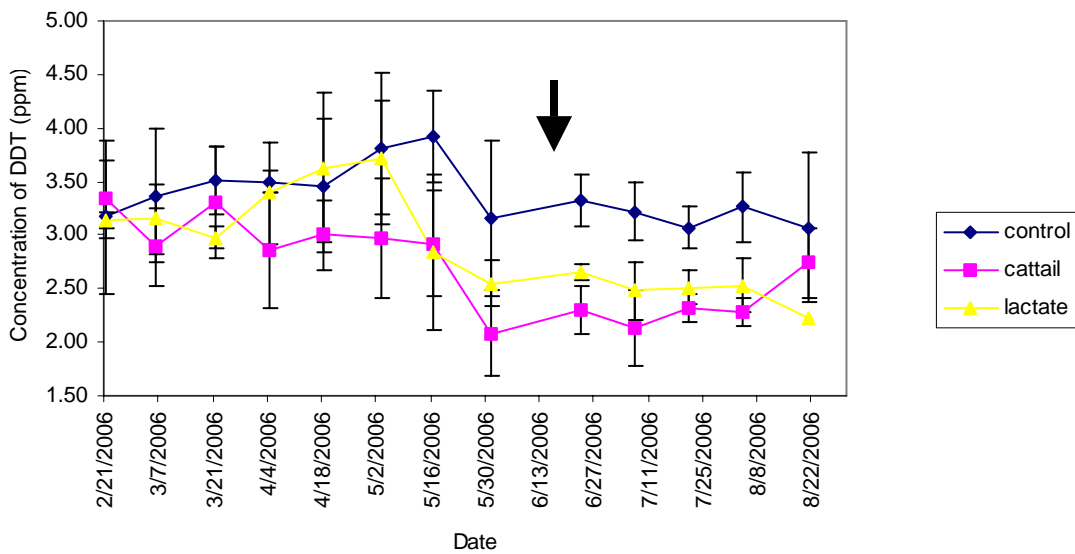


Figure 11. Concentrations of DDD during study. Error bars represent +/- 1 standard deviation. Vertical black arrow indicates date of draining (June 8).

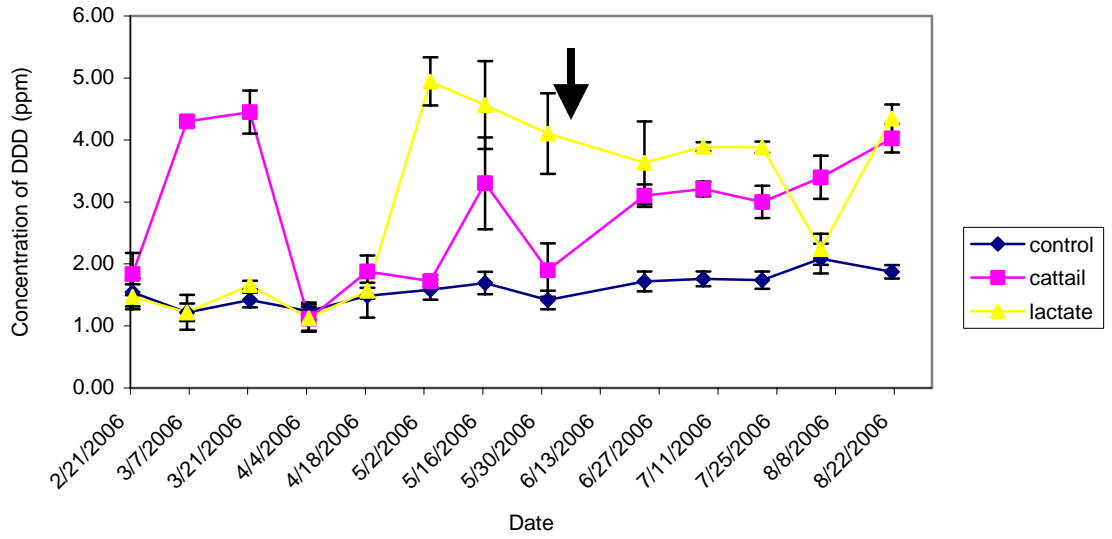
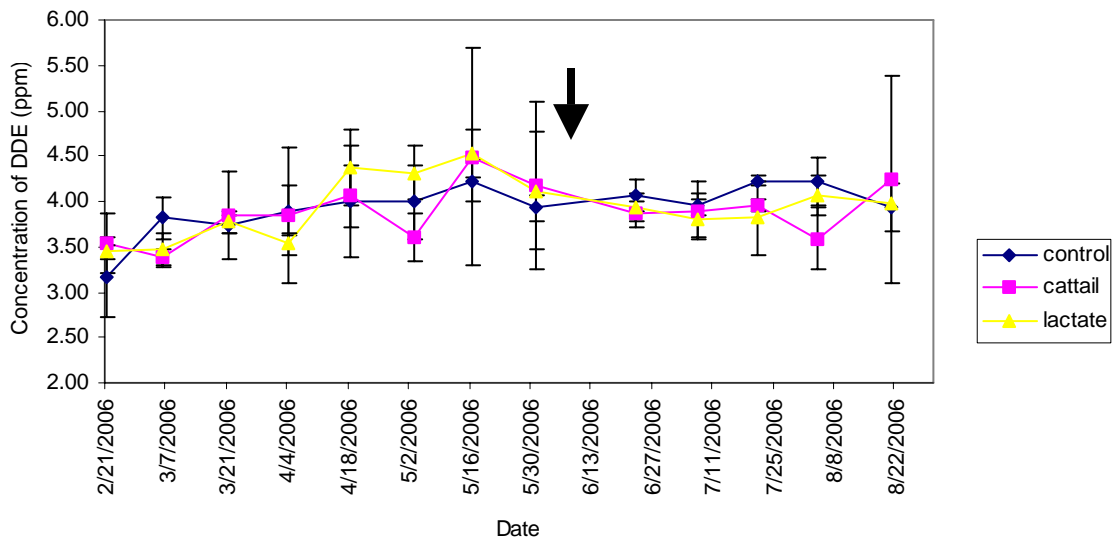


Figure 12. Concentrations of DDE during study. Error bars represent +/- 1 standard deviation. Vertical black arrow indicates date of draining (June 8).



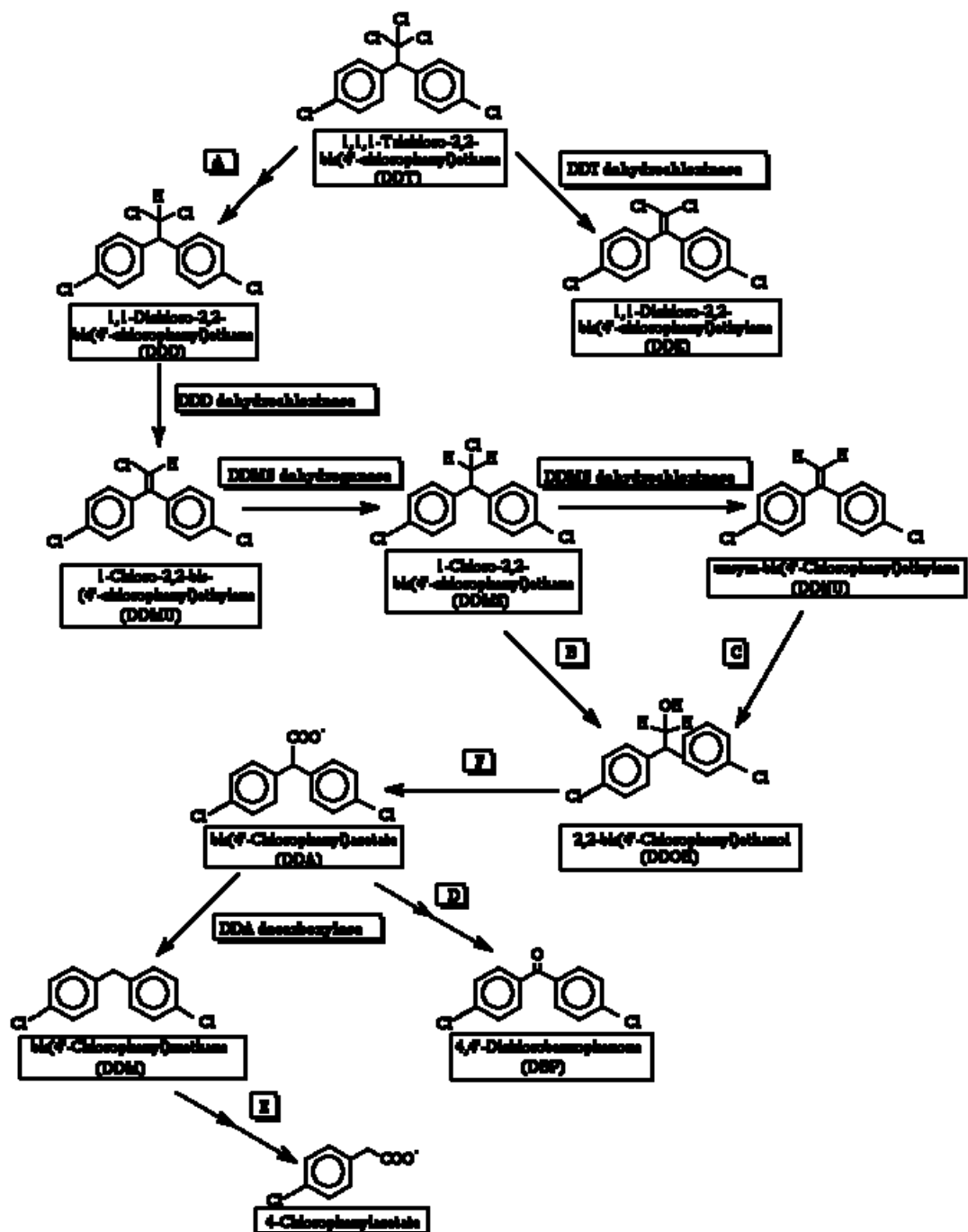


Fig. 13: Anaerobic degradation pathway of DDT (A, D, E and F are multiple steps whose intermediates are not identified yet.)
 (http://umbbd.ahc.umn.edu/ddt2/ddt2_map.html).

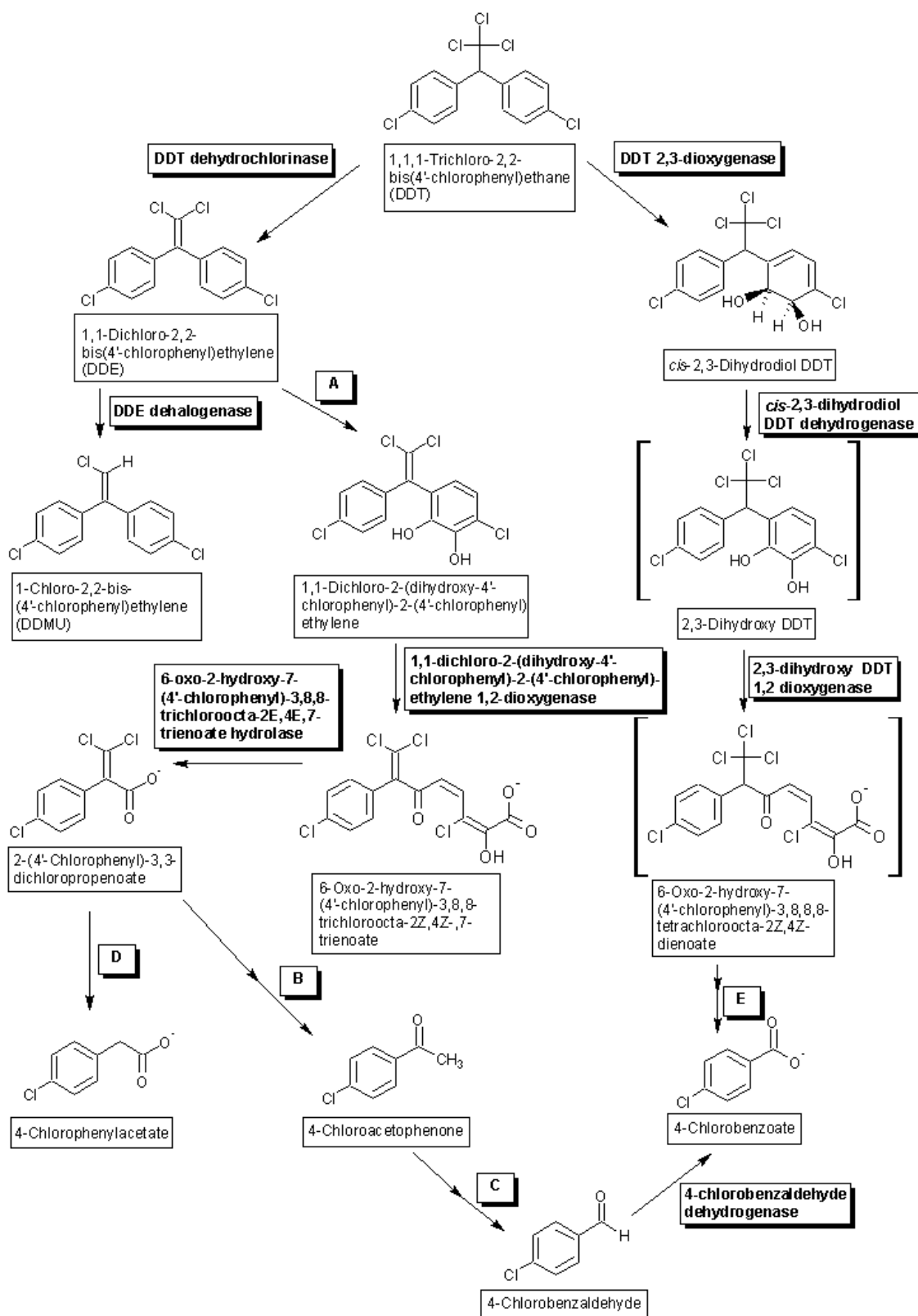


Fig. 14: Aerobic degradation pathway of DDT (A, B, C, D, and E are multiple steps whose intermediates are not identified yet.).
 (http://umbbd.ahc.umn.edu/ddt/ddt_map.html)