Survival and contaminant analysis of juvenile spring Chinook salmon during herbicide treatment with 2,4-D and Diquat for control of noxious weeds in Drano Lake, 2006 Skamania County, Washington

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Background

In January 2006 the State of Washington Department of Ecology and Skamania County signed an agreement for "Drano Lake Milfoil Control". Skamania County subsequently contracted with a private source applicator to chemically treat Drano Lake to eradicate and/or control noxious weeds Eurasian watermilfoil *(Myriophyllum spicatum)* and curly leaf pondweed (*Potamogeton crispus*). Herbicide treatment occurred on July 31 and August 7, 2006.

The purpose for the chemical treatment was to eradicate and/or control the growth and proliferation of invasive noxious water plants that interfered with recreational and commercial uses in Drano Lake. The herbicides 2,4-D and Diquat were used in the treatment area. Information profiles for the herbicides, including ecological effects and environmental fate, are described in Appendix A.

Drano Lake is a backwater area at river kilometer 261 of the Columbia River at 45° 42' 30" N. Latitude and 121° 37' 30" N. Longitude, within Skamania County, Washington. This backwater is the result of flooding the lower Little White Salmon River, caused by the construction of Bonneville Dam in 1938. Bonneville Dam is located 16 miles downstream from the mouth of the Little White Salmon River.

Various fish species are found in Drano Lake throughout the year including juvenile and adult salmon, steelhead, and bull trout. In addition, the U.S. Fish and Wildlife Service (Service) operates Little White Salmon National Fish Hatchery on the Little White Salmon River just upstream of Drano Lake. The hatchery raises spring and fall Chinook salmon (*Oncorhynchus tshawytscha)* for release directly into the river during spring. Adult fish return to the hatchery in spring and fall. Drano Lake is also considered a cool water refuge during the upstream migration of adult salmon and steelhead in the Columbia River and has a popular fishery.

The Service's Columbia River Fisheries Program Office at the request of Little White Salmon National Fish Hatchery investigated the potential effect of herbicide application on juvenile fish raised at the hatchery. The Service and Skamania County cooperatively agreed to monitor the chemical treatment of Drano Lake, by exposing live fish to the treatment area then scientifically analyzing contaminant levels in these fish after 24 and 48 hours.

Study Objective

Our study objective was to determine the mortality and contaminant level in fish during herbicide application in Drano Lake. Basic water quality parameters such as dissolved oxygen, temperature and pH were also monitored. The Service also agreed to collect water samples for Skamania County for analysis.

Methods

Test periods. There were two, one-day herbicide application periods. The first period occurred on July 31, 2006 utilizing 2,4-D and the second application period occurred on August 7, 2006 utilizing Diquat.

Study site (Figure 1). The Drano Lake herbicide application area was divided into five areas including A) near outlet, B) west inlet, C) east inlet, D) mid-south shore, and E) far-east shore. The north shore (F) was an untreated area within Drano Lake. In addition, the hatchery raceways were used as a non-treated control area (G).

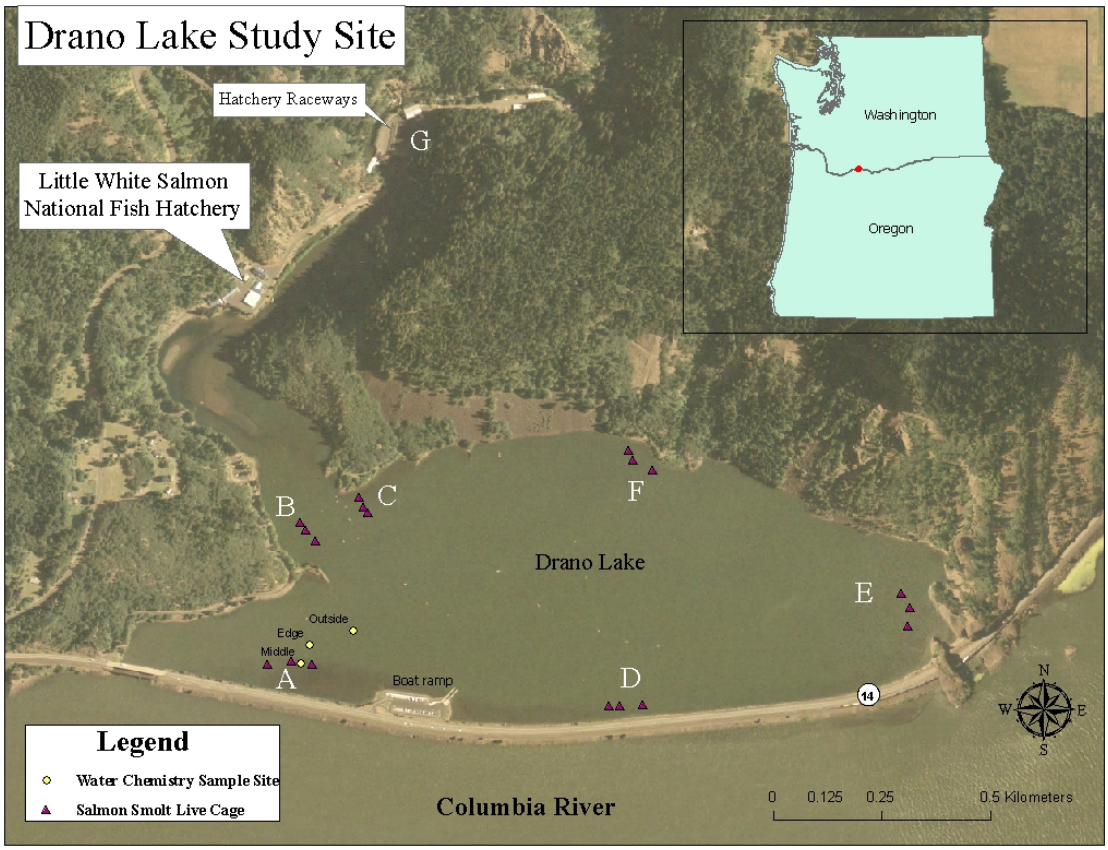

Figure 1. Drano Lake study site.

Test animals. The test animals were juvenile spring Chinook salmon (*Oncorhynchus tshawytscha)* from Little White Salmon National Fish Hatchery placed in live-cages. The live-cages were cylindrical (18 cm x 46 cm) shaped minnow traps, with the openings plugged so fish could neither enter nor escape. The live-cages were suspended in water (two to four feet suspended off the bottom) by a float and weighted line. Each live-cage contained 5 juvenile spring Chinook salmon. Average size of the test fish were 110 mm

and 13.4 gms on July 31, 2006 and 112 mm and 14.0 gms on August 7, 2006 (Speros Doulos, Little White Salmon National Fish Hatchery, pers. comm.).

One day prior to herbicide application, spring Chinook salmon from the hatchery were used to fill 22 live-cages, containing five fish each, 110 fish total. Live-cages were set in six sampling areas within Drano Lake (five areas of direct treatment, sites A, B, C, D, E and one area untreated, site F). Three live-cages were set in each area. In addition, four live-cages were placed in the hatchery raceway (site G) as our non-treated control group. Live-cage locations were marked by a Global Positioning System (GPS).

Live-cages were checked once per day for fish mortality over a three day period during each herbicide treatment as follows: 1) prior to herbicide application (24 hour pre-treatment), 2) one day after herbicide application (24 hour post-treatment), and 3) two days after herbicide application (48 hour post-treatment).

At the end of each study period, all fish not retained for contaminant analysis were released live into Drano Lake (sites A, B, C, D, E and F). Fish at the hatchery site G were released live into the raceway.

Contaminant Analysis in Fish Tissue. Ten groups of five fish each were selected for analysis (six 2,4-D groups, three diquat groups, and one control group; Table 1). The 2,4-D group was sampled twice, after 24 and 48 hrs and the diquat group was only sampled once after 48 hrs. The hatchery control group was only sampled once, at the same time as the diquat group. Fish from the randomly selected live-cage were placed in a uniquely labeled and sealed jar and put in a cooler with ice. The jars from the cooler were then placed in a freezer within eight hours of sample collection. Frozen samples were shipped with dry ice to the Mississippi State Chemical Laboratory for contaminant analysis. The laboratory at Mississippi State composited the livers from the five fish samples from each live-cage. The livers of the fish collected after the first herbicide treatment were analyzed for Chlorophenoxy acid herbicides, including 2,4-D. An analysis specific for Diquat and Paraquat was done on the fish livers collected after the second herbicide treatment.

Water Chemistry. Water chemistry sampling occurred each morning during each study period, between 07:15 and 08:40 am, including dissolved oxygen, pH, and temperature (°C). Data was collected using an InSitu Inc., Troll 9000 Multi-Parameter Water Quality Probe unit. All sampling occurred at site A, within the middle and edge of the herbicide application area (and approximately 107 meters outside from the edge of the treatment area during the Diquat treatment period). The middle, edge, and outside sampling locations were marked by GPS (Figure 1). Sampling occurred at the water surface and near the lake bottom.

Water temperature monitors were also attached to four live-cages (# 1005, 1009, 1012, and 1019, see Table 1). Temperature was automatically recorded hourly throughout the treatment periods.

Water samples for contaminant analysis were collected just below the surface in standard glass containers (approximately 1000 ml) at the middle and outside areas of study Site A (Figure 1). Water samples were kept cool and dropped off at the Court House in Stevenson, Washington, for later analysis by Skamania County.

Table 1. Ten Tissue Sample Collection Sites indicated by Jar Labels (1-A, 1-B, 2-A, 2-B, 2-C, 2-D, 3-A, 3-B, 3-C, and 4-A).

Sample Area (see Figure 1)	Live-Cage Number	2,4-D Application Period			Diquat Application Period		
		Date			Date		
		24 Hour Pre-Application 7/31/2006	24 Hour Post-Application 8/1/2006	48 Hour Post - Application 8/2/2006	24 Hour Pre-Application 8/7/2006	24 Hour Post-Application 8/8/2006	48 Hour Post - Application 8/9/2006
Site A (Near Outlet)	1001						
	1002		1-A				3-A
	1003			2-A			
Site B (Inlet West)	1004						
	1005[a]						
	1006						
Site C (Inlet East)	1007						
	1008						
	1009[a]						
Site F (Mid North Shore-Not Treated, Control)	1010						
	1011						
	1012[a]			2-B			
Site E (Far East)	1014			2-C			
	1015		1-B				
	1016						3-B
Site D (Mid South Shore)	1017						
	1018						
	1019[a]			2-D			3-C
Site G (Hatchery Control)	Control						4-A

[a] Hobo temperature datalogger attached

Results

Herbicide application with 2,4-D on July 31, 2007. During the live-cage test period, July 30 through August 2, three fish mortalities occurred (Table 2).

The following is a breakdown for each day sampled: Day 1, July 31, 24 hour pre-treatment, 0% mortality (all 110 fish survived).

Day 2, August 1, 24 hour post-treatment, two fish died at Site E. Combining all five treatment areas, 2/75 fish died = 2.7% mortality, where Sites F (non-treated area) and G (hatchery control) had 0% mortality (0/35 fish died). Four live-cages within the treatment area were randomly selected (Sites A, C, D, and E) and fish removed for fish tissue contaminant analysis, 20 fish total (note: only fish from Site A (labeled jar 1A) and Site E (labeled jar 1B) were sent to the lab; Table 1).

Day 3, August 2, 48 hour post-treatment, one additional fish died from Site E. Combining all 5 treatment sites, 1/55 = 1.8% mortality, where areas F (non-treated area) and G (hatchery control) again had 0% mortality. 48 hour post-treatment cumulative mortality was 4.5% (2.7% from Day 2 + 1.8% from Day 3). 48 hour cumulative survival of the fish was 95.5% for treated areas and 100% for non-treated and hatchery control sites (Figure 2). Three live-cages within the treatment area were randomly selected for contaminant analysis (Sites A, D, and E; labeled as jars 2A, 2D, 2C respectively), as well as the non-treated Site F (labeled jar 2B), 20 fish total; Table 1.

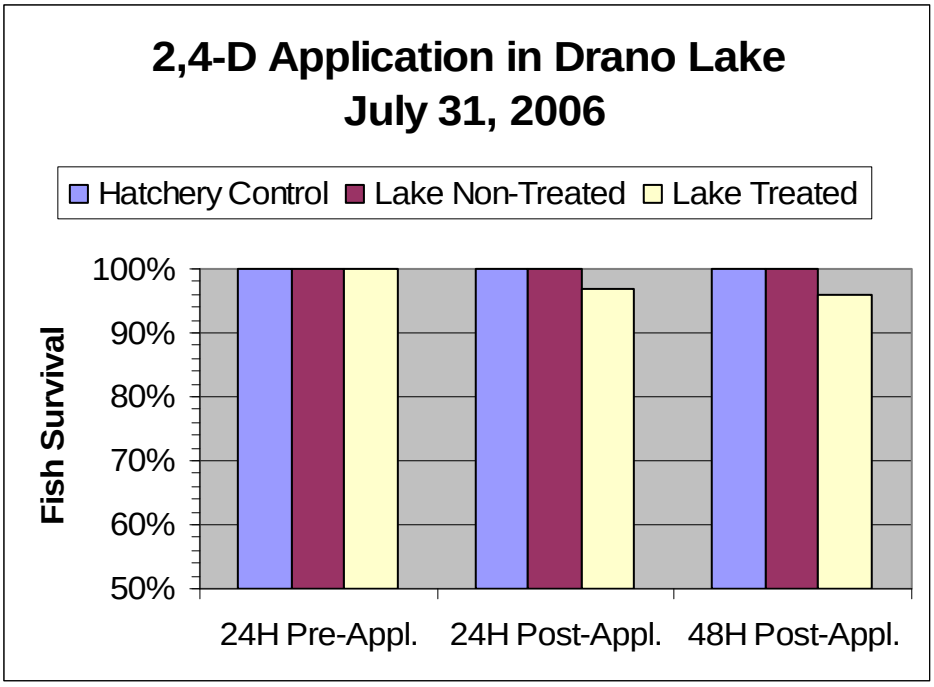

Figure 2. Survival of juvenile spring Chinook salmon before and after treatment with 2,4-D to control noxious weeds in Drano Lake, Skamania County, Washington.

Table 2. Live-Cage Daily Mortality Counts[e]

Sample Area (see Figure 1)	Water Depth (meters)	Live-Cage Number[f]	2,4-D Application Period				Diquat Application Period			
			Date			Total	Date			Total
			24 Hour Pre-Application 7/31/2006	24 Hour Post-Application 8/1/2006	48 Hour Post - Application 8/2/2006		24 Hour Pre-Application 8/7/2006	24 Hour Post-Application 8/8/2006	48 Hour Post - Application 8/9/2006	
Site A (Near Outlet)	2.4	1001								
	2.7	1002		[a]	[c]				1 [a]	1
	3.7	1003			[a]					
Site B (Inlet West)	4.6	1004								
	4.3	1005								
	4.3	1006								
Site C (Inlet East)	3.7	1007		[b]	[c]					
	4.6	1008								
	5.5	1009								
Site F (Mid North Shore-Not Treated, Control)	8.2	1010								
	8.8	1011								
	8.5	1012			[a]					
Site E (Far East)	5.8	1014			[a]			[d]	1	1
	5.2	1015		1 [a]	[c]	1		[d]		
	4.3	1016		1	1	2		[d]	1 [a]	1
Site D (Mid South Shore)	5.8	1017								
	4.9	1018		[b]	[c]					
	6.1	1019			[a]				2 [a]	2
Site G (Hatchery Control)	1	Control [g]							[a]	
		Total	0	2	1	3	0	0	5	5

[a] Live-cage pulled for tissue sample & analyzed; [b] Tissue samples collected but not sent to lab for analysis; [c] No Live-Cage Present; [d] Too windy, not sampled; [e] Zero values not shown; [f] Live-Cage 1013 not used; [g] Four live-cages.

Herbicide application with Diquat on August 7, 2007. During the live-cage test period, August 6 through August 9, five fish mortalities occurred (Table 2).

The following is a breakdown for each day sampled: Day 1, August 6, 24 hour pre-treatment, 0% mortality (all 110 fish survived).

Day 2, August 7, 24 hour post-treatment, no mortalities were found in the traps checked (0/95 fish died). Site E was not sampled because of windy, rough conditions on the lake. No live-cages were selected nor sampled for contaminants.

Day 3, August 8, 48 hour post-treatment, one fish died from Site A, two died from Site D, and two fish were found dead from Site E. Note that site E was not sampled on Day 2, so those mortalities could have previously occurred. Combining all five treatment sites, 5/75 = 6.7% cumulative mortality, where areas F (non-treated area) and G (hatchery control) again had 0% mortality. 48 hour cumulative survival of the fish was 93.3% for treated areas and 100% for non-treated and hatchery control sites (Figure 3). Live-cages were removed from Sites A, D, E, as well as hatchery control site G for contaminant analysis, 20 fish total (labeled jars 3A, 3C, 3B, and 4A respectively; Table 1).

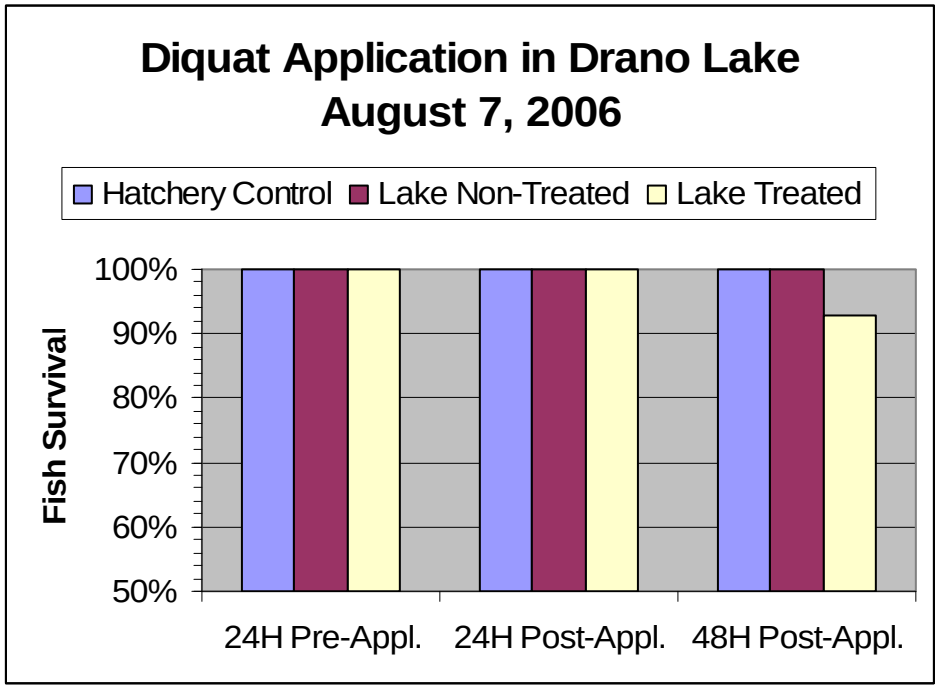

Figure 3. Survival of juvenile spring Chinook salmon before and after treatment with Diquat to control noxious weeds in Drano Lake, Skamania County, Washington.

Table 3. Water chemistry at Site A (see figure 1) associated with herbicide application in Drano Lake.

Herbicide Application	Site	Water Depth (meter)	Sample Date	D.O. (mg/L)		pH		Temperature (°C)	
				Surface	Bottom	Surface	Bottom	Surface	Bottom
2,4-D	Middle	2.9	7/31/2006	8.96	8.7	8.97	8.87	20.04	20.03
			8/1/2006	9.44	9.79	9.06	8.85	19.89	19.74
			8/2/2006	9.23	9.16	9.02	8.76	19.94	19.93
	Edge	8.0	7/31/2006	9.18	9.01	8.99	8.72	20.17	19.74
			8/1/2006	9.42	9.47	9.07	8.8	20.18	18.71
			8/2/2006	9.26	9.84	9	8.41	19.95	13.4
	Outside	7.6	7/31/2006	not measured					
			8/1/2006						
			8/2/2006						
Diquat	Middle	3.1	8/7/2006	9.26	9.22	8.86	8.81	20.6	20.57
			8/8/206	8.99	8.98	8.9	8.9	20.19	20.5
			8/9/2006	9.09	9.11	8.93	8.97	20.08	20.08
	Edge	7.3	8/7/2006	9.49	9.75	8.87	8.29	20.7	14.21
			8/8/206	9.1	9.18	8.89	8.74	19.98	18.87
			8/9/2006	9.51	9.45	8.95	8.56	20.13	15.03
	Outside	7.5	8/7/2006	9.33	9.83	9.06	8.55	20.51	13.37
			8/8/206	9.18	9.8	8.85	8.48	19.95	14.18
			8/9/2006	9.3	9.59	8.98	8.57	20	14.71

Water sampling conducted between 07:15 and 08:40 each morning.

Contaminant Analysis in Fish Tissue. All fish (liver) samples analyzed for contaminants were below the detectable limit (DL) of 0.0100 ppm wet weight (Appendix B).

Water Chemistry. During the two herbicide application periods, dissolved oxygen, pH, and temperature were within the range expected for the time of year sampled (Table 3). Dissolved oxygen ranged from 8.7 to 9.8, pH ranged from 8.3 to 9.1, and temperature ranged from 13.4 to 20.7 °C, with shallow areas warmer than the bottom of the lake.

Water temperature in Drano Lake at the four live-cages ranged from 13.1 to 20.5 °C July 30 to August 2, 2006 and ranged from 13.4 to 21.4 °C August 6 to August 9, 2006 (Appendix C).

Water samples collected to test for contaminant concentrations will be analyzed and reported separately by Skamania County.

Discussion

There were very few fish mortalities in our live-cage study. When mortalities were observed, it was within the herbicide application area, however mortality cannot be explained by level of contaminant in the fish.

Contaminant analyses of fish indicated very low concentrations and were below detectable limits (<0.0100 ppm wet weight). The herbicide concentrations in the water samples will be reported separately by Skamania County. Based on the very low concentrations in the fish, we expect the herbicide concentrations in the water samples to be within acceptable limits.

Of the water chemistry parameters monitored, high water temperature in the lake may have affected survival of the juvenile spring Chinook salmon used in this study. The water temperature guidance[1] to protect salmon and trout in the Pacific Northwest has been recommended not to exceed 20 °C as the summer maximum. Not all sites in Drano Lake were monitored for water temperature; however, of the sites sampled, many approached the 20 °C maximum guideline for a short period of time during the study period. Water temperature at the deeper un-treated control site in the lake was cooler and averaged 14°C. The hatchery was cooler still and averaged approximately 8 °C.

Acknowledgements

Speros Doulos and staff from Little White Salmon National Fish Hatchery provided support, field assistance and fish for this study. Ann Gannam, Abernathy Fish Technology Center provided help in arranging lab work for contaminant analysis and provided helpful suggestions for the study and report. JR (Robert) Horel, Bill Brignon and Jen Poirier, Columbia River Fisheries Program Office and Charly Boyd, Advanced Planning Solutions, Inc., assisted with sampling and equipment.

[1] U.S. EPA, Region 10, 1200 Sixth Avenue, Seattle, WA 98101, April 2003. www.epa.gov/r10earth/temperature.htm

List of Appendices

Appendix A. Information Profiles for Herbicides 2,4-D and Diquat.

E X T O X N E T

Extension Toxicology Network

A Pesticide Information Project of Cooperative Extension Offices of Cornell University, Michigan State University, Oregon State University, and University of California at Davis. Major support and funding was provided by the USDA/Extension Service/National Agricultural Pesticide Impact Assessment Program.

Pesticide
Information
Profile

2,4-D

Publication Date: 9/93

TRADE OR OTHER NAMES

2,4-D is used in many commercial products. A few commercial names for products containing 2,4-D include Weedtrine-II, Aqua-Kleen, Barrage, Plantgard, Lawn-Keep, Planotox and Malerbane.

INTRODUCTION

2,4-D, a chlorinated phenoxy compound, functions as a systemic herbicide and is used to control many types of broadleaf weeds. There are many forms or derivatives (esters, amines, salts) of 2,4-D and these vary in solubility and volatility. Unless otherwise specified, this document will refer to the acid form of 2,4-D. This compound is used in cultivated agriculture and in pasture and rangeland applications, forest management, home and garden situations and for the control of aquatic vegetation. 2,4-D was a major component (about 50%) of the product Agent Orange used extensively throughout Vietnam. However most of the problems associated with the use of Agent Orange were associated with a contaminant (dioxin) in the 2,4,5-T component of the defoliant. The association of 2,4-D with Agent Orange has prompted a vast amount of study on the herbicide.

TOXICOLOGICAL EFFECTS

ACUTE TOXICITY

While the LD50 of 2,4-D suggests that it is only moderately toxic, the product carries the DANGER signal word on the label indicating that it is highly toxic. This is because 2,4-D has produced serious eye and skin irritation among agricultural workers (15,16).

The oral LD50 of 2,4-D in the rat ranges from 375 to 666 mg/kg; 370 mg/kg in the mouse; and less than 320 to 1,000 mg/kg in the guinea pig. The rat and rabbit have dermal LD50 values of 1,500 mg/kg and 1,400 mg/kg, respectively. In humans, prolonged breathing of 2,4-D causes coughing, burning, dizziness, and temporary loss of muscle coordination.

Symptoms of poisoning can be fatigue and weakness with perhaps nausea. On rare occasions there can be inflammation of the nerve endings with muscular effects following high levels of exposure (6). Symptoms vary with the different commercial products because of the specific amounts and types of

additives such as surfactants and solvents.

CHRONIC TOXICITY

Rats given moderate amounts (50 mg/kg) of 2,4-D in the diet for two years had no adverse effects. Some dogs fed lower amounts of the compound in their food for two years died, probably because dogs do not excrete organic acids efficiently. A human given a total of 16.3 grams in 32 days as "desperation therapy" lapsed into a stupor, showed signs of incoordination, weak reflexes, and urinary incontinence.

Reproductive Effects

Administration of drinking water dosed with moderate levels of 2,4- D (about 50 mg/kg) to pregnant rats did not result in any adverse effects on birth weights, or litter size. Rats fed higher levels (188 mg/kg) had fetuses with abdominal cavity bleeding and increased mortality. DNA synthesis in the testes was significantly inhibited when mice were fed large amounts (200 mg/kg) of 2,4-D (8). While there is some conflicting evidence about the reproductive effects of the compound in animals, most of the evidence suggests that 2,4-D causes reproductive effects at moderate doses in animals. This indicates that humans may be at risk with 2,4-D exposure though no direct evidence of reproductive problems associated with 2,4-D exposure exists.

Teratogenic Effects

2,4-D has a very limited ability to cause birth defects. However, rats fed 150 mg/kg on days 6-15 of pregnancy had an increase in skeletal abnormalities such as delayed bone development and wavy ribs (10) which are a function of general toxicity. The same conclusions may be drawn for 2,4-D's potential to cause teratogenic effects in humans as was noted above.

Mutagenic Effects

2,4-D has been very extensively tested for mutagenicity and found to be non-mutagenic in most systems. However, significant increases of damage occurred in chromosomes in cultured human cells at low exposure levels (17). 2,4-D did not damage DNA in human lung cells. The evidence is too equivocal to draw any conclusions.

Carcinogenic Effects

Low doses fed to rats for two years caused an increase in malignant tumors (10). There was some question about whether the tumors were associated with specific organs or were non-specific. Female mice given a single injection of 2,4-D developed cancer (reticulum-cell sarcomas) (10).

The studies of 2,4-D carcinogenicity mentioned above are considered to be inadequate by IARC (International Agency for Research on Cancer). New studies, completed in 1986, show a low incidence of brain tumors at moderate exposure levels (45 mg/kg/day) over a lifetime.

In humans, a variety of studies give conflicting results. Several studies in Sweden and the United States (Kansas (1) and Nebraska (21)), suggest an association of 2,4-D exposure with cancer. An increased occurrence of non-Hodgkin's lymphoma was found among a Kansas and Nebraska farm population associated with the spraying of 2,4-D. Other studies done in New Zealand, Washington, New York, Australia, and on Vietnam veterans from the United States were all negative. There remains

considerable controversy about the methods used in the various studies and thus with the results of the various studies (20). Investigations are continuing.

Organ Toxicity

Most symptoms disappear within a few days but there is a report of liver dysfunction from long term exposure (6).

Fate in Humans and Animals

The absorption of the herbicide is almost complete in mammals after ingestion and nearly all of a dose is excreted in the urine. The compound is readily absorbed through the skin and lungs also. When five men were given 5 mg/kg, they excreted most of the dose (about 82%) as unchanged 2,4-D.

Only traces of the compound have been found in the milk of lactating animals for six days following exposure. The half-life is between 10 and 20 hours in living organisms. There is little evidence to suggest that the compound accumulates to any significant level in mammals or in other organisms (18). Peak concentrations of 2,4-D were found in the blood, liver, kidney, lungs and spleen with lower levels in muscle and brain between six and eight hours after small doses (1 mg/kg) were given to rats. After 24 hours there were no detectable tissue residues. 2,4-D passes through the placenta in pigs and rats. In rats, about 20% was detected in the uterus, placenta, fetus, and amniotic fluid (12). Chickens given moderate amounts of 2,4-D in drinking water from birth to maturity had very low levels of the compound in egg yolks and only a trace in the egg whites.

ECOLOGICAL EFFECTS

2,4-D is slightly toxic to wildfowl. Mallards, pheasants, quail, and pigeons had LD50 levels of >1000, 472, 668, and 668 mg/kg, respectively.

Some formulations of 2,4-D are highly toxic to fish while others are less so. For example the LC50 ranges between 1.0 mg/l to 100 mg/l in cutthroat trout, depending on the formulation used. Channel catfish had less than 10% mortality at 10 mg/l in 48 hours. Green sunfish when exposed to 110 mg/l for 41 hours showed no effect on swimming response. Limited studies indicate a half-life of less than 2 days in fish and oysters when exposure is discontinued (11).

Brood production was severely impaired when honeybees were fed moderate doses, but, at lower levels of exposure they lived significantly longer than the controls. The honeybee LD50 is 11.5 micrograms/bee.

Concentrations of 10 mg/l for 85 days did not adversely affect the survival of adult dungeness crabs. The early immature stages had an LC50 of greater than 10 mg/l in 96 hours indicating that the compound is only slightly toxic to these organisms. Brown shrimp had a small increase in mortality at 2 mg/l over a 48 hour exposure period.

ENVIRONMENTAL FATE

2,4-D applied at 1.16 lb/acre to bluegrass turf in a laboratory experiment had a half-life of ten days. Other half-life figures for the herbicide in soil are seven days (15-25 degree C with 65% moisture) and ten days in non-sterile soil and 1.5 to 16 days in other studies. Soil microbes are primarily responsible

for its disappearance in soil. Studies in Alaska and Canada failed to detect leaching in 22 weeks or from spring to fall (10), but 2,4-D has been included on the EPA list of compounds that are likely to leach from soil.

In aquatic environments microorganisms readily degrade 2,4-D and breakdown by sunlight is not a major reason for loss. Rates of breakdown increase with increased nutrients, sediment load and dissolved organic carbon. Under oxygenated conditions the half-life can be short, in the order of one week to several weeks. 2,4-D interferes with normal plant growth processes. Uptake of the compound is through leaves, stems and roots; however, it is generally nonpersistent. In one study when 2,4-D was applied to grass, there were 80 ppm at day zero, 45 ppm at 14 days, and 6 ppm at 56 days. Breakdown in plants is by a variety of biological and chemical pathways (11).

Despite its short half-life in soil and in aquatic environments, the compound has been detected in groundwater supplies in at least five States and in Canada (18). It has also been detected in surface waters throughout the United States at very low concentrations.

DIOXIN CONTAMINATION

Although recently manufactured 2,4-D technical acids have consistently been free of dioxin contamination, the amine and ester products may have measurable levels of some forms of dioxin. According to a study of 2,4-D manufactured in Canada (9), of 26 amine samples tested, 8 were positive. The levels ranged from 5 ppb to nearly 500 ppb. Several different forms of dioxin were present in the different products. All but one of 21 ester samples were positive.

Since an earlier study had reported finding hexachlorodioxin in 2,4-D the samples were analyzed for mono- to octachlorodioxin but no other isomers were found above the detection limit of 10 ppb. A subsequent study of 2,4-D manufactured in the United States found very little dioxin contamination. Measurable amounts of one form of the compound (2,7 DCDD) were found in 3 of 30 samples, with traces of other isomers. The amounts found do not have biological significance.

Exposure Guidelines:

NOEL (rats):	1 mg/kg
ADI:	0.3 mg/kg (WHO)
MCL:	0.07 mg/l
HA:	70 ug/l (lifetime)
TLV-TWA:	10 mg/m3
TLV STEL:	20 mg/m3
Dangerous Exposure:	500 mg/m3 (OSHA/NIOSH)
RfD:	0.01 mg/kg/day
LEL:	5 mg/kg/day (rat)

Physical Properties:

CAS #:	94-75-7
Chemical Name:	(2,4-dichlorophenoxy) acetic acid
Chemical class/use:	phenoxy herbicide

Solubility in water:	890 mg/l
Solubility in other solvents:	ethanol and acetone, 9.5 g/100 g; benzene, 1.07 g/100g
Melting Point:	138 degrees C
Vapor Pressure:	8 x 10-6 mm Hg
Partition Coefficient:	2.81 (octanol/water)

BASIC MANUFACTURER

Rhone-Poulenc, Inc.
PO Box 12014
2 T.W. Alexander Dr.
Research Triangle Park, NC 27709
Telephone: 919-549-2000
Emergency: 800-334-7577

Review by Basic Manufacturer:

Comments solicited: October, 1992
Comments received:

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For more information relative to pesticides and their use, please contact the PMEP staff at:

5123 Comstock Hall
Cornell University
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Cornell University

Questions regarding the development of this web site should be directed to the PMEP Webmaster

EXTOXNET

Extension Toxicology Network

Pesticide Information Profiles

A Pesticide Information Project of Cooperative Extension Offices of Cornell University, Oregon State University, the University of Idaho, and the University of California at Davis and the Institute for Environmental Toxicology, Michigan State University. Major support and funding was provided by the USDA/Extension Service/National Agricultural Pesticide Impact Assessment Program.

EXTOXNET primary files maintained and archived at Oregon State University

Revised June 1996

Diquat dibromide

Trade and Other Names: Trade names include Aquacide, Aquakill, Dextrone, Diquat, Reglone, Reglox, Reward, Tag, Torpedo, Vegetrole, and Weedtrine-D.

Regulatory Status: Diquat dibromide is a moderately toxic compound in EPA toxicity class II [1,2]. It is a General Use Pesticide (GUP). Labels for products containing diquat dibromide must bear the Signal Word WARNING.

Chemical Class: dessicant

Introduction: Diquat dibromide is a nonselective, quick-acting herbicide and plant growth regulator, causing injury only to the parts of the plant to which it is applied. Diquat dibromide is referred to as a desiccant because it causes a leaf or an entire plant to dry out quickly. It is used to desiccate potato vines and seed crops, to control flowering of sugarcane, and for industrial and aquatic weed control. It is not residual; that is, it does not leave any trace of herbicide on or in plants, soil, or water.

Formulation: Not Available

Toxicological Effects:

- **Acute toxicity:** Diquat dibromide is moderately toxic via ingestion, with reported oral LD50 values of 120 mg/kg in rats, 233 mg/kg in mice, 188 mg/kg in rabbits, and 187 mg/kg in guinea pigs and dogs [1,87]. Cows appear to be particularly sensitive to this herbicide, with an oral LD50 of 30 to 56 mg/kg [17]. The acute dermal LD50 for diquat dibromide is approximately 400 to 500 mg/kg in rabbits, indicating moderate toxicity by this route as well [58,87]. A single dose of diquat dibromide was not irritating to the skin of rabbits, but repeated dermal dosing did cause mild redness, thickening, and scabbing [58]. Moderate to severe eye membrane irritation occurred when diquat dibromide was administered to rabbits [88]. Ingestion of sufficient doses may cause

severe irritation of the mouth, throat, esophagus, and stomach, followed by nausea, vomiting, diarrhea, severe dehydration, and alterations in body fluid balances, gastrointestinal discomfort, chest pain, diarrhea, kidney failure, and toxic liver damage [87]. Skin absorption of high doses may cause symptoms similar to those that occur following ingestion [89]. Very large doses of the herbicide can result in convulsions and tremors [88]. Test animals (rats, mice, guinea pigs, rabbits, dogs, cows, and hens) given lethal doses of diquat dibromide showed a delayed pattern of illness, with onset approximately 24 hours following dosing, subsequent lethargy, pupil dilation, respiratory distress, weight loss, weakness and finally death over the course of 2 to 14 days after dosing [58,87,89]. There have been reports of workers who have had softening and color changes in one or more fingernails after contact with concentrated diquat dibromide solutions [87]. In some instances, the nail was shed, and did not grow in again [87]. Several cases of severe eye injury in humans have occurred after accidental splashings [87]. In each case, initial irritation was mild, but after several days, serious burns and sometimes scarring of the cornea developed. Direct or excessive inhalation of diquat dibromide spray mist or dust may result in oral or nasal irritation, nosebleeds, headache, sore throat, coughing, and symptoms similar to those from ingestion of diquat [87].

- **Chronic toxicity:** Chronic effects of diquat dibromide are similar to those of paraquat [87]. Cataracts, a clouding of the eyes which interferes with light entering the eye, occurred in rats and dogs given 2.5 mg/kg/day and 5 mg/kg/day of diquat dibromide, respectively [87]. Cataracts increased in proportion to the dose given in test animals (cats and dogs) [17,88]. Chronic exposure is necessary to produce these effects [87]. Other effects on the eye (hemorrhage, retinal detachment) may occur at higher dosages [87]. Rats fed dietary doses of 2.5 mg/kg/day over 2 years did not exhibit signs of toxicity other than reduced food intake and decreased growth [17]. In another study using rats, oral doses of 4 mg/kg/day over 2 years produced no behavioral or other changes in general condition [87]. At this dose level no evidence of change in the kidneys, liver, or mycocardium (heart muscle) were seen. This dosage (but not 2 mg/kg/day) caused changes in lung tissues [87]. Repeated or prolonged dermal contact may cause inflammation of the skin, and, at high doses, systemic effects in other parts of the body. These may include damage to the the kidneys [58]. Chronic exposure may damage skin, which may increase the permability of the skin to foreign compounds [88].
- **Reproductive effects:** Diquat dibromide generally did not reduce fertility when tested in experimental animals [89]. Rats receiving 1.25 mg/kg/day decreased their food intake and showed slowed growth, but had unchanged reproduction [89]. Fertility was reduced in male mice given diquat dibromide during different stages of sperm formation [87]. Neither fertility nor reproduction was affected in a three-generation study in rats given dietary doses of 12.5 or 25 mg/kg/day of diquat dibromide, although some growth retardation was seen at the 25 mg/kg/day dose [87]. Based on this evidence it is unlikely that diquat dibromide will cause reproductive effects in humans under normal circumstances.
- **Teratogenic effects:** Offspring of pregnant rats given a fatal injected dose of 14 mg/kg of diquat dibromide showed evidence of skeletal defects of the collar bone, as well as little or no ear bone formation upon examination [58,87]. No deformities were found in the unborn offspring of pregnant rats that were injected intraperitoneally with 0.5 mg/kg/day of diquat daily during organogenesis, the stage of fetal development in which organs are formed [26]. Growth retardation was seen in test animals given extremely high doses of diquat. While no actual teratogenesis occurred in rats given single abdominal injections during days 7 to 14 of pregnancy, many rats did not have normal weight gain and bone formation in the unborn was decreased [23]. It is unlikely that diquat dibromide will cause teratogenic effects in humans under normal circumstances.
- **Mutagenic effects:** There is no evidence that diquat dibromide causes permanent changes in genetic material [87]. For example, no mutagenic effects were seen in mice given oral doses of 10 mg/kg/day for 5 days [23].

- **Carcinogenic effects:** An 80-week feeding study showed that dietary doses of 15 mg/kg/day of diquat did not cause tumors in rats [90]. Likewise, dietary levels of 36 mg/kg/day for 2 years did not induce tumors in rats [87]. Based on the evidence, it appears that diquat dibromide is not carcinogenic.
- **Organ toxicity:** In animals, diquat dibromide may affect the gastrointestinal tract, eyes, kidneys or liver, and the lungs.

- **Fate in humans and animals:** Absorption of diquat dibromide from the gut into the bloodstream is low [87]. Oral doses are mainly metabolized within the intestines, with metabolites being excreted in the feces [87,30]. Rat studies showed only a small percentage of the applied oral dose (6%) was absorbed into the bloodstream and then excreted in the urine [87]. Dermal, inhalation, or intravenous exposure results in little processing and rapid elimination in the urine [87]. Following subcutaneous injection in rats, excretion of about 90% of the dose occurred in the urine on the first day and almost all of the remainder on the next day [87]. Complete elimination of the herbicide was seen in urine and feces of rats within 4 days of administration of single oral doses of 5 to 10 mg/kg of diquat dibromide [87].

Ecological Effects:

- **Effects on birds:** Diquat dibromide ranges from slightly to moderately toxic to birds [91]. The reported acute oral LD50 in young male mallards is 564 mg/kg [8]. The oral LD50 for diquat dibromide is 200 to 400 mg/kg in hens [8]. The 5-day dietary LC50 is about 1300 ppm in Japanese quail [36].
- **Effects on aquatic organisms:** Diquat dibromide is moderately to practically nontoxic to fish and aquatic invertebrates. The 8-hour LC50 for diquat dibromide is 12.3 mg/L in rainbow trout and 28.5 mg/L in Chinook salmon [28]. The 96-hour LC50 is 16 mg/L in northern pike, 20.4 mg/L in fingerling trout, 245 mg/L in bluegill, 60 mg/L in yellow perch, and 170 mg/L in black bullhead [37,92]. Research indicates that yellow perch suffer significant respiratory stress when herbicide concentrations in the water are similar to those normally present during aquatic vegetation control programs [93]. There is little or no bioconcentration of diquat dibromide in fish [8].
- **Effects on other organisms:** Diquat dibromide is not toxic to honey bees [1]. Since diquat dibromide is a nonselective herbicide, it may present a danger to non-target plant species [91]. Cows are particularly sensitive to the toxic effects of this material [17].

Environmental Fate:

- **Breakdown in soil and groundwater:** Diquat dibromide is highly persistent, with reported field half-lives of greater than 1000 days [11]. It is very well sorbed by soil organic matter and clay [11]. Although it is water soluble [11], its capacity for strong adsorption to soil particles suggest that it will not easily leach through the soil, be taken up by plants or soil microbes, or broken down by sunlight (photochemical degradation). Field and laboratory tests show that diquat usually remains in the top inch of soil for long periods of time after it is applied [94].
- **Breakdown in water:** Studies on the erosion of diquat-treated soils near bodies of water indicate that diquat dibromide stays bound to soil particles, remaining biologically inactive in surface waters, such as lakes, rivers, and ponds [95]. When diquat dibromide is applied to open water, it disappears rapidly because it binds to suspended particles in the water [95]. Diquat dibromide's half-life is less than 48 hours in the water column, and may be on the order of 160 days in sediments due to its low bioavailability [94,95]. Microbial degradation and sunlight play roles in the breakdown of the compound [95]. At 22 days after a weed infested artificial lake was treated, only 1% of the applied diquat dibromide remained in the water and 19% was adsorbed to

sediments [9].

- **Breakdown in vegetation:** Diquat dibromide is rapidly absorbed into the leaves of plants, but usually kills the plant tissues necessary for translocation too quickly to allow movement to other parts of the plant. The herbicide interferes with cell respiration, the process by which plants produce energy. Diquat dibromide is broken down on the plant surface by photochemical degradation [58]. It is rapidly absorbed by aquatic weeds from the surrounding water and concentrated in the plant tissue [8]. Thus, even low concentrations of the herbicide can control aquatic weeds [8].

Physical Properties:

- **Appearance:** Technical diquat dibromide, which is greater than 95% pure, forms white to yellow crystals [1].
- **Chemical Name:** 1,1'-ethylene-2,2'-bipyridyldiylium dibromide salt [1]
- **CAS Number:** 85-00-7
- **Molecular Weight:** 344.06
- **Water Solubility:** 700,000 mg/L @ 20 C; v.s. [1]
- **Solubility in Other Solvents:** i.s. in nonpolar solvents such as chloroform, diethyl ether, and petroleum ether [1]; s.s in alcohol and hydroxylic solvents [1]
- **Melting Point:** Decomposes above 300 C [1]
- **Vapor Pressure:** Negligible @ 20 C [1]
- **Partition Coefficient:** -4.6021 [1]
- **Adsorption Coefficient:** 1,000,000 (estimated) [11]

Exposure Guidelines:

- **ADI:** 0.002 mg/kg/day [12]
- **MCL**: 0.02 mg/L [65]
- **RfD:** 0.0022 mg/kg/day [13]
- **PEL:** Not Available
- **HA**: Not Available
- **TLV**: 0.1 mg/m3 (8-hour) (respirable fraction) [17]

Basic Manufacturer:

Zeneca Ag Products
1800 Concord Pike
Wilmington, DE 19897

- **Phone:** 800-759-4500
- **Emergency:** 800-759-2500

References:

References for the information in this PIP can be found in Reference List Number 10

DISCLAIMER: The information in this profile does not in any way replace or supersede the information on the pesticide product labeling or other regulatory requirements. Please refer to the pesticide product labeling.

Appendix B. Analytical Results Report for Contaminant Concentrations in Juvenile Spring Chinook Salmon in Drano Lake.

Analytical Results Report TOC

1. ECDMS Analytical Results Report 1/4/2007

Catalog Number	Purchase Order Number	Lab ID	Catalog Submitter	ECDMS User ID
1200005	94420-06-Y731	MSCL	Gannam, Ann - Longview, WA	r1aftc

Catalog Title	DRANO LAKE INVASIVE PLANT REHAB
Lab Name:	Mississippi State Chemical Lab
Regional Study ID:	R1061932002
Regional Study Title:	Drano Lake near Cook, WA.

Notes, Symbols and Abbreviations Used
Based on the report options selected the report should be **printed in landscape mode**
Notes, Symbols and Abbreviations Used The following may appear before a reported result (e.g. < 1234). < - Less than symbol indicates that the actual result is less than the reported detection limit. > - Greater than symbol indicates that the actual result is greater than the reported result.
All results are reported as 3 significant digits.
All results are reported as parts per million (ppm), or percent, unless otherwise noted.

1. Integrity Report

Lab Receipt Date	09/28/2006	**Lab Approval Date**	09/28/2006

Catalog Problems
No problems reported
Problem Resolution

2. Bulk Data

Sample Number	Sample Matrix	Sample Weight (grams)	Percent Moisture
1-A	Liver	2.4	84.3
1-B	Liver	2.7	80.8
2-A	Liver	5	81.5
2-B	Liver	2.6	80.9
2-C	Liver	5.3	80.1
2-D	Liver	2.8	79.0
3-A	Liver	2.6	
3-B	Liver	2.9	
3-C	Liver	4.6	
4-A	Liver	12.4	78.0

4. Contaminant Concentrations

Analyte	Sample Number	Sample Matrix	Dry Weight (ppm)	DL Dry Weight (ppm)	Wet Weight (ppm)	DL Wet Weight (ppm)
2,4,5-T						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
2,4-D						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
2,4-DB						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
Acifluorfen						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100

Analyte	Sample Number	Sample Matrix	Dry Weight (ppm)	DL Dry Weight (ppm)	Wet Weight (ppm)	DL Wet Weight (ppm)
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
Clopyralid						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
Diquat						
	3-A	Liver			< 0.0100	0.0100
	3-B	Liver			< 0.0100	0.0100
	3-C	Liver			< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
Paraquat						
	3-A	Liver			< 0.0100	0.0100
	3-B	Liver			< 0.0100	0.0100
	3-C	Liver			< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
Picloram						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100

Analyte	Sample Number	Sample Matrix	Dry Weight (ppm)	DL Dry Weight (ppm)	Wet Weight (ppm)	DL Wet Weight (ppm)
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
Quinclorac						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
Triclopyr						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
dicamba						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
dichlorprop						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100

Analyte	Sample Number	Sample Matrix	Dry Weight (ppm)	DL Dry Weight (ppm)	Wet Weight (ppm)	DL Wet Weight (ppm)
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100
silvex						
	1-A	Liver	< 0.0637	0.0637	< 0.0100	0.0100
	1-B	Liver	< 0.0521	0.0521	< 0.0100	0.0100
	2-A	Liver	< 0.0541	0.0541	< 0.0100	0.0100
	2-B	Liver	< 0.0524	0.0524	< 0.0100	0.0100
	2-C	Liver	< 0.0503	0.0503	< 0.0100	0.0100
	2-D	Liver	< 0.0476	0.0476	< 0.0100	0.0100
	4-A	Liver	< 0.0455	0.0455	< 0.0100	0.0100

5. Procedural Blanks

Analyte	Lab Sample Number	Lab Sample Matrix	Result Total UG	** BEC (ppm/%)	Basis
2,4,5-T					
	74448	Animal Tissue	0.000	< 0.0100	Wet
2,4-D					
	74448	Animal Tissue	0.000	< 0.0100	Wet
2,4-DB					
	74448	Animal Tissue	0.000	< 0.0100	Wet
Acifluorfen					
	74448	Animal Tissue	0.000	< 0.0100	Wet
Clopyralid					
	74448	Animal Tissue	0.000	< 0.0100	Wet
Diquat					
	74450	Animal Tissue	0.000	< 0.0100	Wet
Paraquat					
	74450	Animal Tissue	0.000	< 0.0100	Wet
Picloram					
	74448	Animal Tissue	0.000	< 0.0100	Wet
Quinclorac					
	74448	Animal Tissue	0.000	< 0.0100	Wet
Triclopyr					
	74448	Animal Tissue	0.000	< 0.0100	Wet
dicamba					
	74448	Animal Tissue	0.000	< 0.0100	Wet
dichlorprop					
	74448	Animal Tissue	0.000	< 0.0100	Wet
silvex					
	74448	Animal Tissue	0.000	< 0.0100	Wet

** Blank Equivalent Concentration

6. Duplicates

Analyte	Sample Number	Sample Matrix	Basis	Initial Result (ppm/%)	Duplicate Result (ppm/%)	Average	Relative Percent Diff.
% Moisture							
	4-A	Liver	Percent	78.0	77.2	77.6	1.03
2,4,5-T							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
2,4-D							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
2,4-DB							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
Acifluorfen							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
Clopyralid							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
Picloram							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
Quinclorac							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
Triclopyr							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
dicamba							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
dichlorprop							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000
silvex							
	4-A	Liver	Wet	< 0.0100	< 0.0100	0.00500	0.000

7. Spike Recoveries

Analyte	Sample Number	Sample Matrix	Basis	Spike Level (ppm/%)	Amount Recovered (ppm/%)	*** Spike Background	Percent Recovery
2,4,5-T							
	74449	Animal Tissue	Wet	1.00	1.00		100.
2,4-D							
	74449	Animal Tissue	Wet	2.00	2.00		100.
Acifluorfen							
	74449	Animal Tissue	Wet	1.00	0.700		70.0
Clopyralid							
	74449	Animal Tissue	Wet	1.00	1.10		110.
Diquat							
	74451A	Animal Tissue	Wet	1.00	0.850		85.0
	74451B	Animal Tissue	Wet	1.00	0.830		83.0
Paraquat							
	74451A	Animal Tissue	Wet	1.00	0.790		79.0
	74451B	Animal Tissue	Wet	1.00	0.810		81.0
Picloram							
	74449	Animal Tissue	Wet	1.00	0.860		86.0
Quinclorac							
	74449	Animal Tissue	Wet	1.00	0.820		82.0
Triclopyr							
	74449	Animal Tissue	Wet	1.00	1.00		100.
dicamba							
	74449	Animal Tissue	Wet	1.00	1.20		120.
dichlorprop							
	74449	Animal Tissue	Wet	1.00	1.00		100.
silvex							
	74449	Animal Tissue	Wet	1.00	0.970		97.0

*** For a spike to be a valid measure of method accuracy, this ratio must be higher than 1.0.

9. Laboratory Notes

Analyte	Sample Number	Result Modifier
% Moisture		
	74449	Lab Generated
2,4,5-T		
	74449	Lab Generated
2,4-D		
	74449	Lab Generated
Acifluorfen		
	74449	Lab Generated
Clopyralid		
	74449	Lab Generated
Diquat		
	74451A	Lab Generated
	74451B	Lab Generated
Paraquat		
	74451A	Lab Generated
	74451B	Lab Generated
Picloram		
	74449	Lab Generated
Quinclorac		
	74449	Lab Generated
Triclopyr		
	74449	Lab Generated
dicamba		
	74449	Lab Generated
dichlorprop		
	74449	Lab Generated
silvex		
	74449	Lab Generated

Code List

If appropriate, labs are instructed to use the following codes when entering laboratory notes. The labs may use one or more of the codes in each note displayed above.

Code	Comment
A	Values reported based on Aldrin response factor.
C	Sample possibly compromised due to improper handling / packaging.
D	Sample was deleted from the catalog by the submitter.
H	Due to sample characteristics it was difficult to obtain adequate sample homogeneity - precision was impacted.
I	Interferences occurred during analysis.
L	Sample compromised or destroyed during shipment - sample not analyzed.
M	Compound identity was confirmed by GC/MS.
N	Sample was not analyzed.
P	Sample destroyed during preparation at lab - sample not analyzed.
Q	Insufficient sample quantity to perform requested analysis.
R	Sample is highly decomposed - results may be impacted.
S	Sample was substituted by the submitter.
T	Retention time relative to Aldrin.
U	GC/MS identifies the unknown compound to be _____ (fill in analyte).
W	Insufficient sample quantity to perform duplicate / spike analyses.
Y	Sample was analyzed but results may be impacted (see 'C')

10. QAQC Summary

1. Procedural Blank Summary

Procedural Blank Summary of Blank Equivalent Concentration (BEC) Data

Within a lab sample matrix, there must be three or more Blank results for a given analyte in order to generate a report.

10.2. Duplicate Summary

Duplicate Summary of Relative Percent Difference (RPD) Data

Within a lab sample matrix and concentration range, there must be three or more Duplicate results for a given analyte in order to generate a report.

10.3. Spike Summary

Spike Summary of Percent Recovery (PR) Data

Within a lab sample matrix, there must be three or more Spike results for a given analyte in order to generate a report.

10.4. SRM Summary

Standard Reference Material Summary of Percent Recovery (PR) Data

Within an SRM ID, there must be three or more Recoveries for a given analyte in order to generate a report.

11. QA/QC Anomalies

1. Blank Frequency Anomalies

The required number of blank analyses were performed.

11.2. Duplicate Frequency Anomalies

The required number of duplicate sample analyses were performed with the following exceptions.

Analyte	Lab Matrix	Number of Samples	Number of Duplicates	Frequency (%)	See QA/QC Note No.
Diquat	Animal Tissue	4	0	0	1
Paraquat	Animal Tissue	4	0	0	2

11.3. Spike Frequency Anomalies

The required number of spike sample analyses were performed with the following exceptions.

Analyte	Lab Matrix	Number of Samples	Number of Spikes	Frequency (%)	See QA/QC Note No.
2,4-DB	Animal Tissue	7	0	0	3

11.4. Reference Material Frequency Anomalies

No Standard Reference Material data exists in this set of results; therefore, the anomaly test was not performed.

11.5. Mass Spec Frequency Anomalies

No Carbamate, OC, or OP data exists in this set of results; therefore, the anomaly test was not performed.

11.6. Limit of Detection Anomalies

Limits of Detection were within the contract requirements.

11.7. Blank Anomalies

Procedural Blank analyses were acceptable.

11.8. Duplicate Anomalies

All duplicate results were within normal limits.

11.9. Spike Anomalies

All spike results were within normal limits.

11.10. S.R.M. Anomalies

No SRM data exists in this set of results; therefore, the anomaly test was not performed.

11.11. QA/QC Notes

QA/QC Note Number and Comments
1-2. Insufficient sample size for duplicate analysis for these analytes. 3. It is not practical to spike with every analyte. The procedure used is acceptable. The results include one new analyte, Acifluorfen (CAS 5094-66-6). This analyte has been added to the list of chlorophenoxy acid herbicides. Judy Bischoff, Ph.D. Branch Chief, ACF 12-28-06

12. Analytical Methods

Below are the analytical methods used by MSCL to produce the results included in this report.

Method Codes:	034

Lab Matrix	Analyte
Animal Tissue	% Moisture
	2,4,5-T
	2,4-D
	2,4-DB
	Acifluorfen
	Clopyralid
	Picloram
	Quinclorac
	Triclopyr
	dicamba
	dichlorprop
	silvex

Method Code: 034

LABORATORY: Mississippi State Chemical Laboratory

Analysis for Chlorophenoxy Acid Herbicides in Liver

Weigh 0.5 gm of liver into a size 22 Duall tissue grinder. Add 2 ml PRQ water and 6 drops 6N HCl. Grind the liver. Extract the liver with 4 ml diethyl ether by grinding. Centrifuge and remove ether to a screw top tube. Extract the liver 2X with 2 ml diethyl ether and once with 2 ml pet. ether. This combined extract contains chlorophenoxy acid herbicides.

Derivitization: Reduce sample volume to approximately 0.5ml and ethylate using diazoethane (15 min.). Exchange to hexane (N-EVAP) and reduce volume to 0.2ml.

Column Clean-Up:

Column clean-up: Place 2.0g of 1% deactivated silica gel in a 7mm id. chromatography column (#22 Kontes). Top with 1cm Na2SO4 and prewet column with 10ml hexane. Collect sample eluents in three fractions as follows:

Fraction A: add sample and rinse container with two 0.5ml washes of 20% benzene in hexane. Elute with 9ml of the same solution. (Contains PCP.)

Fraction B: add 10ml 40% benzene in hexane. Add 10ml 60% benzene in hexane. (Contains Dalapon, Silvex, Dinoseb, portion of Dicamba.)

Fraction C: add 10ml 80% benzene in hexane. Add 10ml 100% benzene. (Contains remaining Dicamba, Dichlorprop, 2,4-D, 2,4,5-T, 2,4-DB, Bentazon, Blazer.)

[Reference for column clean-up for acid herbicides: Shafik, T. A.,H. C. Sullivan, H. R. Enos, 1973." Multiresidue Procedure for Halo- and Nitrophenols. Measurement of Exposure to Biodegradable Pesticides Yielding These Compounds as Metabolites." J. Agr. Food Chem. 21:295-298.]

[Reference for extraction: A modification of Determination of Pentachlorphenol in Blood, Manual of Analytical Methods for the Analysis of Pesticides in Humans and Environmental Samples, Section 5,A,(3),(b).EPA June 1980.

Recoveries: N=3

Dicamba	93%
Dichloroprop	101%
24D	115%
PCP	96%
Silvex	105%
245T	114%
24DB	98%
Blazer	90%

Method Codes:	075

Lab Matrix	Analyte
Animal Tissue	Diquat
	Paraquat

Method Code: 075
LABORATORY: Mississippi State Chemical Laboratory

Analysis of Paraquat and Diquat in Vegetation, Soil and Tissue

Reagents and Materials:

Distilled Deionized Water (DIDW)
250mL round bottomed flask
250mL column
Saturated NaCl solution
Saturated NH4Cl solution
Sulfuric acid (conc't)
50mL calibrated polypropylene vial
Dowex 50W8-100 ion exchange resin
Ion pair concentrate solution
Ion pair concentrate preparation solution

Reagent Preparation:

Ion pair concentrate preparation solution:

Add 13.5mL of orthophosphoric acid and 10.3mL of diethylamine to 500mL of DIDW and dilute to 1000mL with DIDW/

Ion pair concentrate solution:

Dissolve 3.75g of 1-hexanesulfonic acid in 15mL of Ion pair concentrate preparation solution and dilute to 25mL in volumetric flask with the Ion pair concentrate preparation solution.

Extraction:

Water

- Measure out 250mL of sample into 400mL beaker. Slowly add 15mL of conc't sulfuric acid while stirring contents.

Tissue, Soil, and Vegetation

- Weight out 10g of sample into 250mL round bottomed flask. Add 100mL DIDW and slowly add 10mL of conc't sulfuric acid while stirring contents.

- Reflux for five hours and let cool. After this step, the samples can sit overnight.

Column preparation and Use:

- Set up 250mL column with stopcock and glass wool. Add approximately 15mL of DIDW.

- Measure out 3.5g of Dowex resin. Make a slurry with DIDW and pour into 250mL column with plenty of washing until the resin is sufficiently packed.

- Wash the column with first: 50mL DIDW, 25mL saturated NaCl solution, finally 25mL DIDW.

- Sample is added to the column while the flow rate is adjusted to approximately 1mL/min.

- After entire sample is percolated through the column, wash again with 50mL DIDW.

- All elutants at this point should be discarded and washed down the sink with plenty of water.

- Sample is slowly eluted off the column with 50mL of saturated NH4Cl solution. Collect this solution. Sample is collected in 50mL calibrated polypropylene vial, while sample is being eluted off, add 200uL of ion pair concentrate solution.

- Samples are brought up to volume with saturated NH4Cl solution. Samples are then filtered into HPLC vials through 0.45um Nylon filters.

- Samples are now ready to be shot on the HPLC.

HLPC Mobile Phase: 8.3g NaCl / 1L DIDW adjusted to pH 2 with 6N HCl
HPLC column: DuPont Zorbax SIL 4.6 x 250mm
Flow rate: 1.0 mL/min
UV wavelengths: 257nm for Paraquat, 308nm Diquat

Appendix C. Hourly Water Temperature Profile at Four Live Cage Sites in Drano Lake.

Date	Live-Cage# Time	1005 Temperature (°C)	1012 Temperature (°C)	1019 Temperature (°C)	1009 Temperature (°C)
7/30/2006	13:00:00	18.13	14.01	20.5	18.81
7/30/2006	14:00:00	18.29	14.01	20.5	18.81
7/30/2006	15:00:00	18.45	14.01	20.5	18.81
7/30/2006	16:00:00	17.97	13.72	20.19	19.11
7/30/2006	17:00:00	18.13	13.72	20.19	19.41
7/30/2006	18:00:00	17.48	13.72	20.19	19.11
7/30/2006	19:00:00	18.29	13.43	19.89	19.11
7/30/2006	20:00:00	18.13	13.72	19.89	18.81
7/30/2006	21:00:00	19.27	14.31	19.89	19.71
7/30/2006	22:00:00	18.94	14.01	19.89	19.71
7/30/2006	23:00:00	17.48	14.01	19.89	19.11
7/31/2006	0:00:00	18.29	13.72	19.89	18.81
7/31/2006	1:00:00	17.81	13.72	19.59	19.41
7/31/2006	2:00:00	18.94	13.72	19.59	19.41
7/31/2006	3:00:00	18.78	13.72	19.59	19.41
7/31/2006	4:00:00	18.29	13.72	19.59	19.11
7/31/2006	5:00:00	18.45	14.01	19.59	19.11
7/31/2006	6:00:00	17.48	13.72	19.59	19.11
7/31/2006	7:00:00	17.81	13.72	19.59	19.41
7/31/2006	8:00:00	18.13	13.72	19.59	19.41
7/31/2006	9:00:00	15.42	13.72	19.59	19.41
7/31/2006	10:00:00	15.74	13.72	19.59	18.81
7/31/2006	11:00:00	18.62	13.43	19.89	18.21
7/31/2006	12:00:00	18.94	14.01	20.19	16.73
7/31/2006	13:00:00	18.78	13.72	20.19	19.11
7/31/2006	14:00:00	17.97	13.72	20.5	19.41
7/31/2006	15:00:00	18.13	13.43	20.5	18.51
7/31/2006	16:00:00	18.78	13.43	20.5	18.51
7/31/2006	17:00:00	18.62	13.72	20.5	18.21
7/31/2006	18:00:00	19.43	13.72	20.5	19.41
7/31/2006	19:00:00	19.27	13.72	20.5	18.21
7/31/2006	20:00:00	18.62	13.72	20.19	18.81
7/31/2006	21:00:00	18.29	13.72	20.19	19.41
7/31/2006	22:00:00	17.97	13.43	20.19	19.41
7/31/2006	23:00:00	15.89	13.43	20.19	19.11
8/1/2006	0:00:00	16.84	13.43	19.89	19.41
8/1/2006	1:00:00	15.1	13.43	19.89	18.51
8/1/2006	2:00:00	15.26	13.43	19.89	19.41
8/1/2006	3:00:00	15.89	13.43	19.59	19.41
8/1/2006	4:00:00	14.94	13.43	19.59	18.51
8/1/2006	5:00:00	14.78	13.43	19.59	17.62
8/1/2006	6:00:00	17.97	13.43	19.59	18.51
8/1/2006	7:00:00	15.42	13.72	19.59	18.21
8/1/2006	8:00:00	15.89	13.72	19.59	19.11
8/1/2006	9:00:00	15.42	13.72	19.59	18.81
8/1/2006	10:00:00	16.06	13.43	19.59	18.81

8/1/2006	11:00:00	17.97	13.43	19.89	19.11
8/1/2006	12:00:00	17.48	13.43	19.89	18.81
8/1/2006	13:00:00	17.97	13.72	20.19	19.11
8/1/2006	14:00:00	17.17	13.72	20.19	18.51
8/1/2006	15:00:00	17.48	13.72	20.19	18.81
8/1/2006	16:00:00	18.78	13.72	20.19	19.41
8/1/2006	17:00:00	17.97	13.43	20.19	19.41
8/1/2006	18:00:00	18.94	13.43	20.19	19.41
8/1/2006	19:00:00	19.11	14.31	20.19	19.41
8/1/2006	20:00:00	19.11	14.01	19.89	19.41
8/1/2006	21:00:00	18.13	13.43	19.89	19.41
8/1/2006	22:00:00	19.43	13.14	19.89	18.51
8/1/2006	23:00:00	19.27	13.72	19.59	18.81
8/2/2006	0:00:00	18.29	14.01	19.89	19.41
8/2/2006	1:00:00	15.42	13.72	19.89	19.41
8/2/2006	2:00:00	15.1	13.43	19.59	18.51
8/2/2006	3:00:00	15.58	13.43	19.59	18.21
8/2/2006	4:00:00	16.37	13.43	19.29	19.11
8/2/2006	5:00:00	15.89	14.01	19.29	19.11
8/2/2006	6:00:00	14.62	14.01	19.29	18.81
8/2/2006	7:00:00	15.74	13.72	19.29	16.73
8/2/2006	8:00:00	16.06	14.01	19.29	18.81
	Average	17.51	13.69	19.90	18.93
	Max	19.43	14.31	20.5	19.71
	Min	14.62	13.14	19.29	16.73

Appendix C. Hourly Water Temperature Profile at Four Live Cage Sites in Drano Lake.

Date	Live-Cage# Time	1005 Temperature (°C)	1012 Temperature (°C)	1019 Temperature (°C)	1009 Temperature (°C)
8/6/2006	9:00:00	19.11	13.72	19.29	20.61
8/6/2006	10:00:00	19.11	14.01	20.5	17.62
8/6/2006	11:00:00	19.59	14.31	20.5	17.02
8/6/2006	12:00:00	19.59	14.31	20.81	19.41
8/6/2006	13:00:00	19.43	14.01	20.81	19.41
8/6/2006	14:00:00	19.11	13.72	20.81	19.41
8/6/2006	15:00:00	19.27	13.72	20.81	19.71
8/6/2006	16:00:00	19.59	13.43	21.11	20.01
8/6/2006	17:00:00	19.76	14.31	21.11	20.31
8/6/2006	18:00:00	19.92	14.01	21.11	20.31
8/6/2006	19:00:00	20.08	14.01	21.11	20.31
8/6/2006	20:00:00	20.08	13.72	21.11	20.01
8/6/2006	21:00:00	19.76	13.72	21.11	18.21
8/6/2006	22:00:00	19.27	13.72	21.11	20.01
8/6/2006	23:00:00	18.94	14.01	21.11	20.61
8/7/2006	0:00:00	20.08	13.72	21.11	20.61
8/7/2006	1:00:00	18.45	14.01	20.81	17.92
8/7/2006	2:00:00	18.13	14.01	20.81	18.81
8/7/2006	3:00:00	19.11	14.59	20.81	20.31
8/7/2006	4:00:00	17.81	14.01	20.81	20.31
8/7/2006	5:00:00	19.59	14.01	20.5	20.31
8/7/2006	6:00:00	20.08	13.72	20.5	20.31
8/7/2006	7:00:00	19.92	13.72	20.5	19.71
8/7/2006	8:00:00	18.13	13.72	20.5	20.01
8/7/2006	9:00:00	19.11	13.72	20.81	18.81
8/7/2006	10:00:00	19.92	14.31	20.81	19.41
8/7/2006	11:00:00	19.92	14.59	20.81	20.01
8/7/2006	12:00:00	19.76	14.01	21.11	20.01
8/7/2006	13:00:00	19.76	14.31	21.11	20.01
8/7/2006	14:00:00	19.76	14.01	21.11	20.31
8/7/2006	15:00:00	19.59	14.01	21.11	20.01
8/7/2006	16:00:00	19.76	14.01	21.42	19.41
8/7/2006	17:00:00	18.29	14.59	21.11	18.21
8/7/2006	18:00:00	16.84	15.47	21.11	17.32
8/7/2006	19:00:00	19.43	14.01	21.11	19.11
8/7/2006	20:00:00	16.53	14.88	20.81	20.61
8/7/2006	21:00:00	19.27	13.72	20.81	20.01
8/7/2006	22:00:00	19.92	13.72	20.81	19.11
8/7/2006	23:00:00	19.11	15.47	20.5	18.81
8/8/2006	0:00:00	19.43	14.88	20.5	20.31
8/8/2006	1:00:00	19.59	14.59	20.5	20.31
8/8/2006	2:00:00	19.27	14.01	20.5	20.31
8/8/2006	3:00:00	18.62	14.31	20.19	16.43
8/8/2006	4:00:00	14.78	15.17	20.19	19.41
8/8/2006	5:00:00	15.26	14.31	19.89	20.01
8/8/2006	6:00:00	18.94	14.31	19.89	19.71

8/8/2006	7:00:00	15.74	14.59	19.59	19.41
8/8/2006	8:00:00	15.89	14.59	19.59	19.71
8/8/2006	9:00:00	15.26	14.88	19.59	19.11
8/8/2006	10:00:00	15.58	14.31	19.59	19.41
8/8/2006	11:00:00	19.27	14.01	19.59	18.51
8/8/2006	12:00:00	15.1	14.01	19.89	18.51
8/8/2006	13:00:00	15.58	16.64	20.19	19.11
8/8/2006	14:00:00	14.94	14.88	19.89	18.51
8/8/2006	15:00:00	16.69	14.88	20.19	19.11
8/8/2006	16:00:00	17.97	14.59	20.5	19.41
8/8/2006	17:00:00	18.13	14.01	20.19	19.41
8/8/2006	18:00:00	19.11	14.01	20.19	19.41
8/8/2006	19:00:00	19.27	15.17	20.19	19.41
8/8/2006	20:00:00	18.13	14.01	20.19	19.71
8/8/2006	21:00:00	19.27	14.01	20.19	19.71
8/8/2006	22:00:00	19.27	14.31	20.19	19.71
8/8/2006	23:00:00	19.11	14.01	20.19	19.71
8/9/2006	0:00:00	18.94	14.01	20.19	19.41
8/9/2006	1:00:00	18.13	14.01	20.19	19.71
8/9/2006	2:00:00	16.69	14.31	20.19	19.41
8/9/2006	3:00:00	16.84	14.31	19.89	19.71
8/9/2006	4:00:00	15.42	14.31	19.89	19.41
8/9/2006	5:00:00	14.94	14.01	19.89	19.11
8/9/2006	6:00:00	17.17	14.01	19.89	18.81
8/9/2006	7:00:00	16.21	14.31	19.59	18.81
8/9/2006	8:00:00	15.89	14.31	19.59	19.11
	Average	18.35	14.24	20.48	19.45
	Max	20.08	16.64	21.42	20.61
	Min	14.78	13.43	19.29	16.43