U.S. Fish and Wildlife Service
Region 3
Contaminants Program

Upper Mississippi River Floodplain Water Quality for Keithsburg Division, Illinois, Mark Twain National Wildlife Refuge

U.S. Fish and Wildlife Service
4469 - 48th Avenue Court
Rock Island, Illinois 61201
1998

Upper Mississippi River Floodplain Water Quality Keithsburg Division, Illinois, Mark Twain National Wildlife Refuge

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ABSTRACT

Biologists from the U.S. Fish and Wildlife Service surveyed environmental quality at a backwater system of the Upper Mississippi River between 1994 and 1996. The backwater system is the Keithsburg Division management unit of the Mark Twain National Wildlife Refuge located in the floodplain of Navigation Pool 18. The surface water and groundwater sources for this backwater are agricultural drainage water and streams impacted by rural non-point source pollution. The environmental quality parameters that were studied included water quality, sediment quality and toxicity testing, organism health and ecological diversity. The chemistry data indicated nutrient rich surface water, groundwater and aquatic sediment resources in the refuge unit. The concentrations of toxic contaminants detected in the water and aquatic sediment resources were below lethal benchmark values for aquatic life. The concentrations of herbicide chemicals detected in surface water and aquatic sediments were above levels that may stress aquatic plants. Poor aquatic macroinvertebrate and plant diversity was observed in many areas around the refuge unit. We suggest that the loading of nutrients in these areas was sufficient to cause changes to the structure and composition of refuge plant communities and related shifts in invertebrate populations. The altered ecological communities in the polluted areas function to treat nitrogen inputs, providing less diverse wildlife habitats. Several lake and watershed management strategies are discussed to help increase habitat diversity and benefits to migratory birds. It is important to note that an increment of improvement in the watershed will not necessarily result in an increment of improvement in habitat quality in the backwater system. It is possible that little or no ecological change may occur or be observed at Keithsburg Division until a critical level is reached in the watershed resulting in a noticeable shift in refuge habitat quality.

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INTRODUCTION

This document reports and interprets water quality and ecological data for an Upper Mississippi River backwater system in Mark Twain National Wildlife Refuge known as Keithsburg Division (Figure 1). The data include results from biodiversity studies, toxicity testing, and chemical analyses of water and sediments.

The project was conducted between 1994 and 1996 by biologists from the U.S. Fish and Wildlife Service's Rock Island Field Office and Mark Twain National Wildlife Refuge as part of the agency's refuge contaminants program.

Background

In 1986, the U.S. Fish and Wildlife Service started a program to survey and catalog pollution problems on national wildlife refuges. As part of this program, in 1989 and 1992, contaminant studies were completed at the various management units that make up Mark Twain National Wildlife Refuge to determine if priority pollutants were present in aquatic sediments (Young 1991 and Coffey 1995).

No organic pollution from chemicals such as DDT, chlordane or PCB was detected in the sediments for refuge units along the Upper Mississippi River (Young 1991). Sediment heavy metal concentrations for the refuge study units were between normal and slightly elevated for some metals (Table 1) (Young 1991 and Coffey 1995).

However, poor water quality conditions as indicated by low dissolved oxygen concentrations and elevated ammonia concentrations were found at the Keithsburg Division unit (Coffey 1995). These findings prompted this study to characterize water quality for the Keithsburg Division basin.

The first phase for the study was to gather relevant contaminants information for the watershed. This included an inventory of agricultural chemical use and water quality data for the bordering rivers.

Local farmers and commercial applicators were periodically interviewed during the study period. The interview information indicated that eleven pesticide brands were used for the production of corn and soybean in the refuge unit's drainage area (Table 2).

We compiled and reviewed water quality data for the creeks and rivers bordering the refuge unit that included Pope Creek, Edwards River and the Mississippi River (Figure 2) (ILEPA 1993, ILEPA 1994 and USGS 1995).

The water quality reports for the creeks and rivers mentioned above indicated that a variety of chemical pollutants such as heavy metals, pesticide chemicals, phosphates and nitrogen compounds could be transported into the Keithsburg Division backwater system during flood stages.

Study Objectives

The four objectives for the study are outlined below.

- Monitor nutrient and pesticide chemicals in refuge surface water.
- Characterize aquatic sediment quality.
- Survey floodplain ecological communities for evidence of chemical stress.
- Develop water quality management alternatives for the refuge unit.

Table 1. Mean and maximum arsenic and heavy metal concentrations detected in sediments for backwater complexes in Mark Twain National Wildlife Refuge for 1992 and other background data for comparison (micrograms per gram, dry weight) (from Coffey 1995).

Analyte	1992 Mean (n=83)	1992 Maximum	U.S. Soil Background Average[1]	Illinois Stream Average[2]
Arsenic	5.32	8.18	-	<7.2
Cadmium	0.45	0.77	-	<2.0
Chromium	18.53	50.52	53	<37
Copper	21.05	34.65	25	<37
Nickel	21.36	44.30	20	<26
Lead	21.19	51.92	20	<60
Zinc	83.62	145.6	54	<170

[1] Average elemental concentrations in surficial materials (Schacklette *et al* 1971)

[2] Non-elevated stream sediments in Illinois (n=79, IEPA 1997).

STUDY AREA

Keithsburg Division is one of the 15 refuge management units that make up Mark Twain National Wildlife Refuge. The management units are scattered along a 358 mile reach of the Upper Mississippi River in Iowa, Illinois and Missouri, 45 miles in the middle and lower Iowa River in Iowa and 7 miles in the lower Illinois River in Illinois.

The specific management objectives for the Keithsburg Division management unit are listed below.

- Provide waterfowl with food, water and protection during migration.
- Improve and maintain existing habitat to perpetuate an optimum annual production of wood ducks (*Aix sponsa*).

Keithsburg Division is an 1800 acre backwater system in the floodplain of Navigation Pool 18 of the Mississippi River, Mercer County, Illinois. The backwater contains a mosaic of wetlands, shallow sloughs and bottomland hardwood forest. The backwater is bordered by the Edwards River to the north, Pope Creek to the south and the Mississippi River to the west (Figure 2). Surface water from these streams and rivers flows into the backwater only during flood stages. There are four un-named tributary ditches that intermittently flow into the refuge along the northeast edge.

Subsurface water and tile effluent regularly flow into the un-named tributary ditches mentioned above. Ground water intermittently discharges into Spring Slough from springs in the sandy bluff along the east side of the refuge unit (Figure 3).

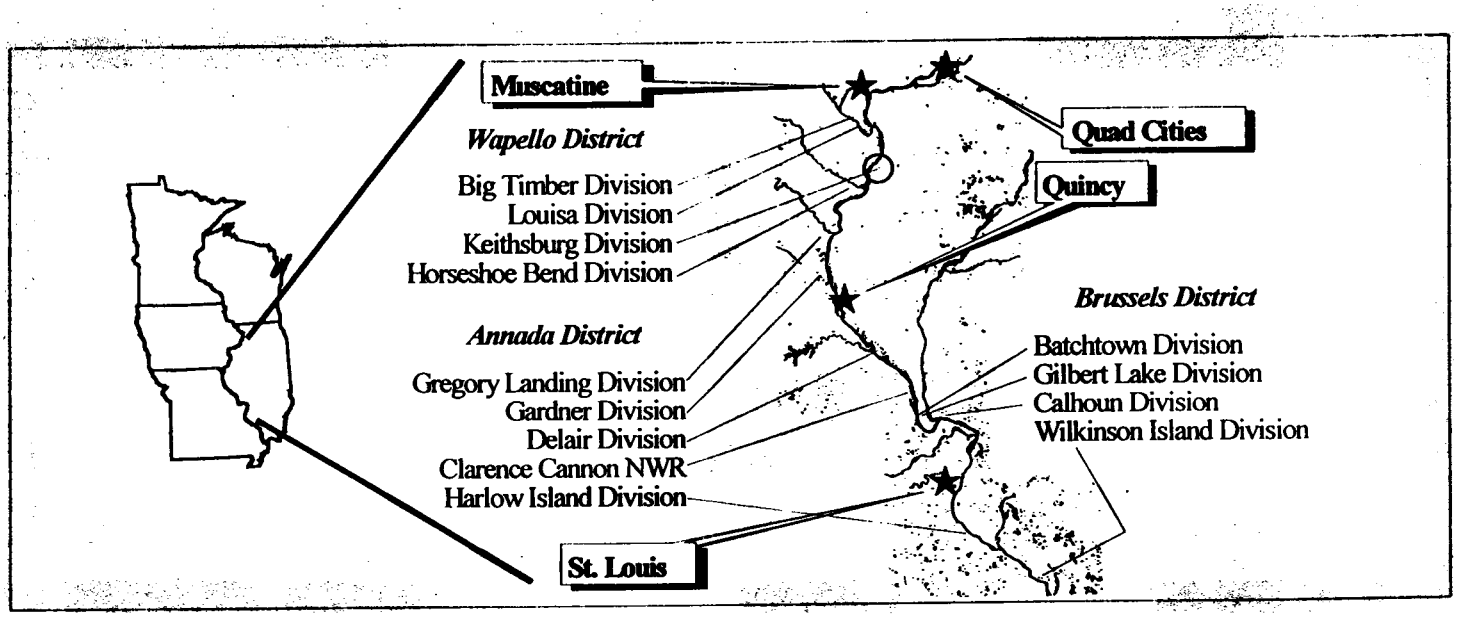

Figure 1. Map of Mark Twain National Wildlife Refuge with the location of Keithsburg Division.

Table 2. Pesticide use information for agricultural fields adjacent to Keithsburg Division refuge, 1992 through 1996.

Chemical	Trade Name	Use
Terbufos	Counter®	Systemic insecticide
Chlorpyrifos	Lorsban®	Systemic insecticide
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Atrazine	Atrazine®	Corn preemergent herbicide
Atrazine+ Alachlor	Bullet®	Corn preemergent herbicide
Acetochlor+ Atrazine	Harness Xtra®	Corn preemergent herbicide
Imidazolinone	Pursuit®	Soybean preemergent herbicide
Sulfonylurea	Pinnacle® & Synchrony®	Soybean postemergent herbicide

Systemic insecticides were generally used at corn planting time (early April through mid May) for rootworm control on fields that were not rotated to soybean, and again on young plants (June through July) for corn borer and/or cut worm control.

Seed corn was coated with systemic fungicide chemicals and insecticide chemicals for some users.

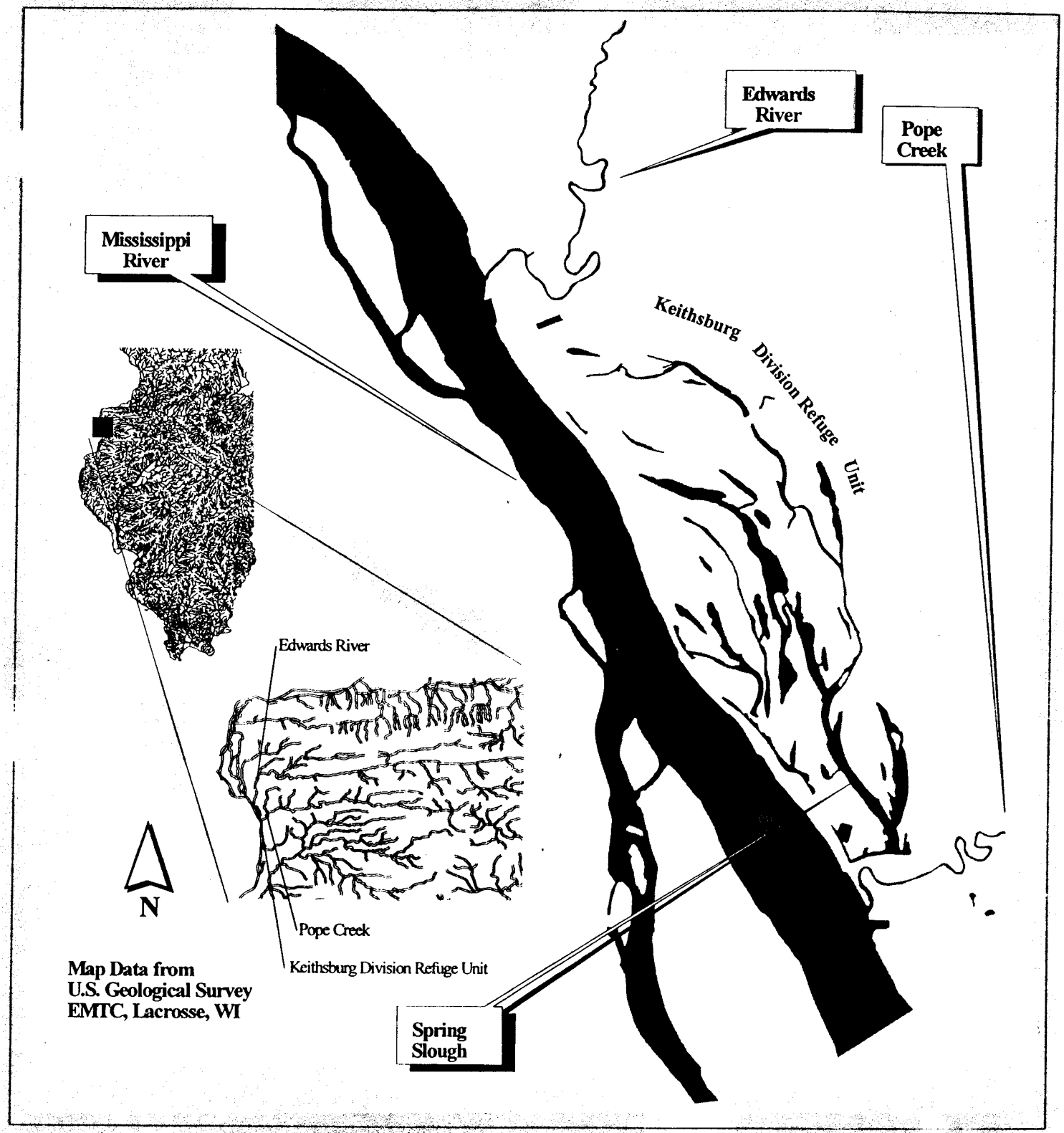

Figure 2. Surface water resources around Keithsburg Division refuge.

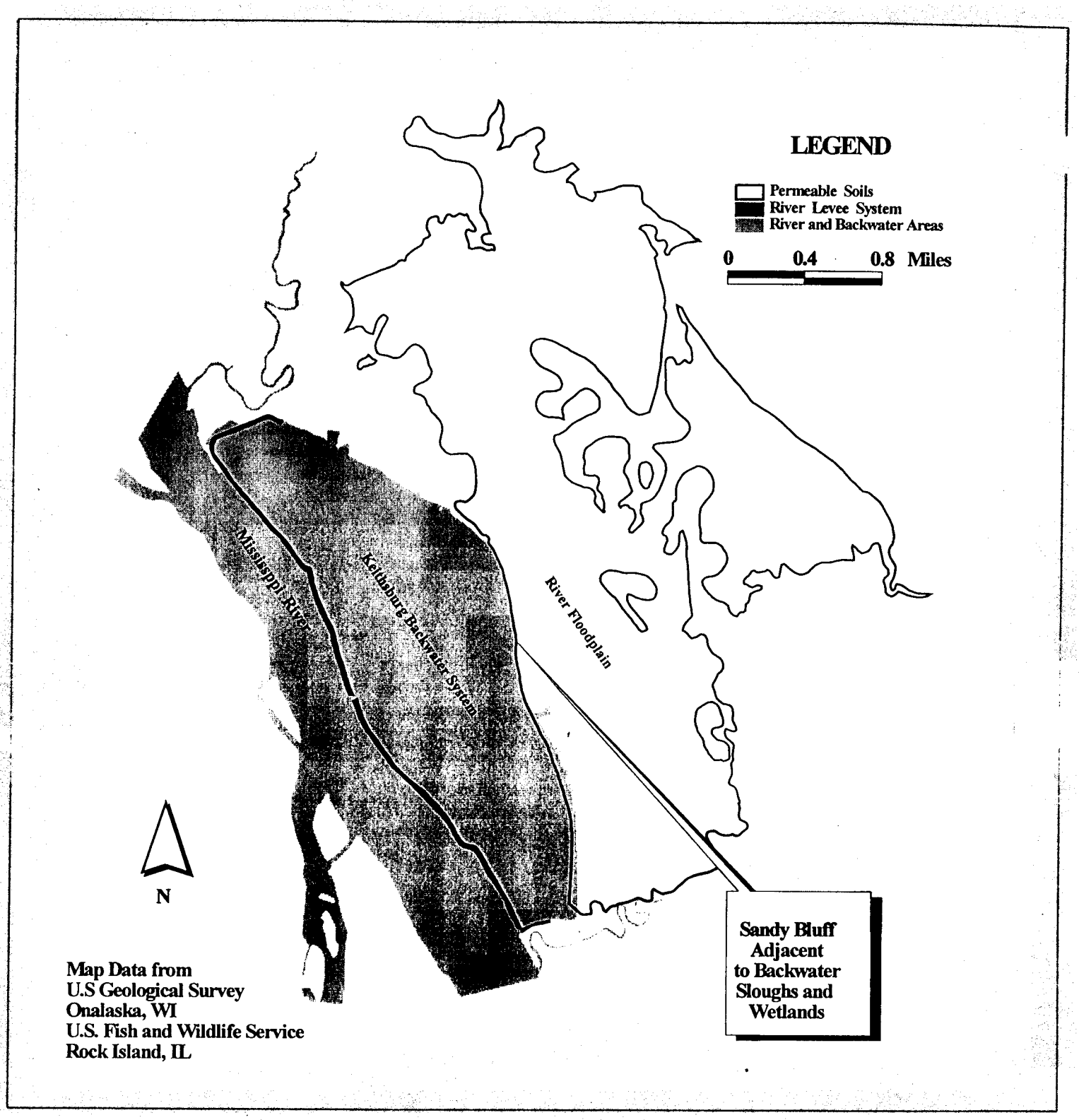

Figure 3. Location of sandy bluff and permeable soils that contain springs along the east side of the Keithsburg Division refuge.

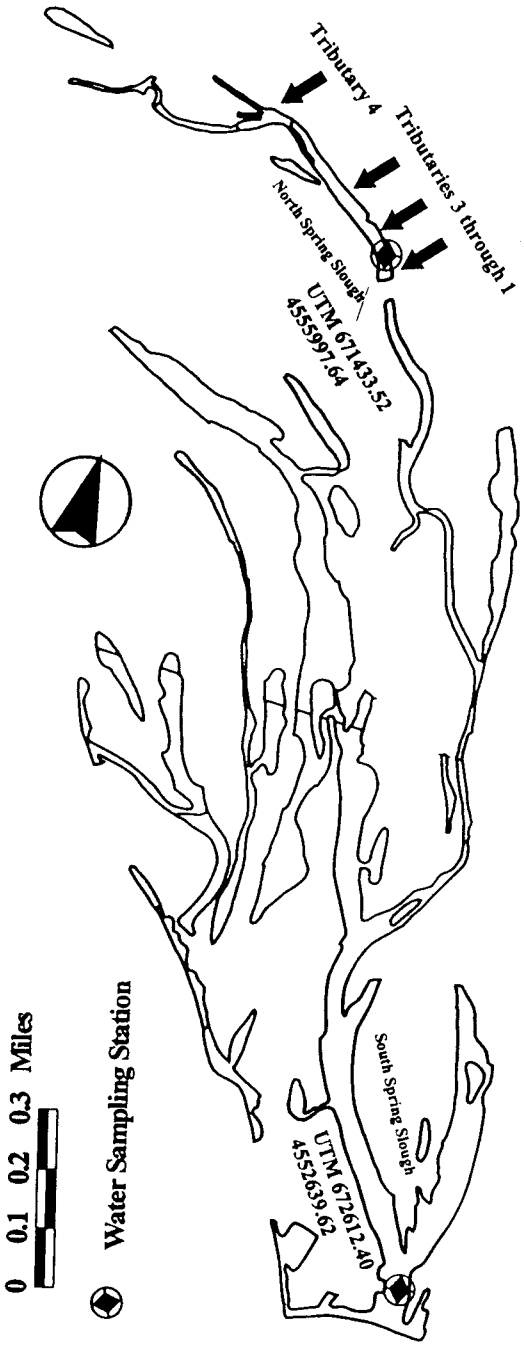

Figure 4. Water quality sampling locations for Keithsburg Division refuge.

Sampling Locations

Fixed sampling locations were established for the study around the backwater to regularly collect samples and field data (Figures 4 and 5).

We established the monitoring points for surface water sampling at the inlets of the four un-named tributary ditches along the northeast edge and two stations in the main slough that runs the length of the backwater known as Spring Slough.

In 1994, we examined sediment quality at several deep water areas within the backwater unit to document the status since the 1993 flood. In 1995, we re-examined sediment quality along a line transect down the center of the lake at the north end of Spring Slough to assess ecotoxicological impacts from elevated contaminant concentrations in those sediments.

Ecological surveys were completed around the water and sediment sampling stations discussed above and at two wetland sites. One of the wetland sites represented a polluted condition because it was connected to contaminant sources (Site A). The other wetland site represented an unpolluted condition (Site B). Wetland site B was not regularly connected to contaminant sources.

The selection of the deep water areas and study wetland sites was based on an independent desk top exercise and model process developed by the U.S. Fish and Wildlife Service known as the Contaminant Assessment Process (CAP). CAP is part of the Service's Biomonitoring of Environmental Status and Trends (BEST) program. The results of the desktop exercise and more information on BEST are available to view on the Internet at URL address:

http://orion.cr.usgs.gov/

Select CAP and refuge name Mark Twain NWR.

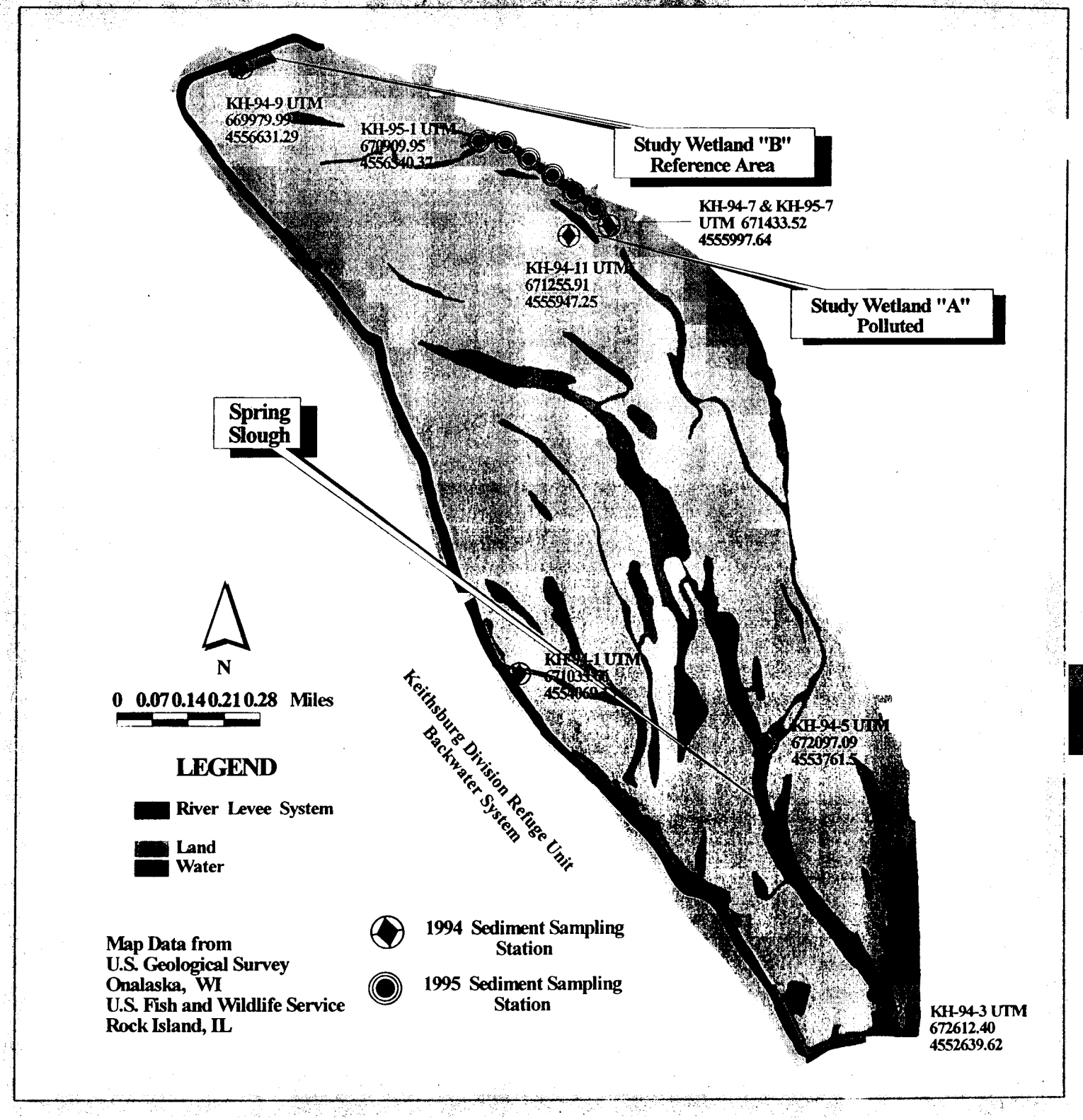

Figure 5. Locations of the sediment quality and ecological sampling sites for the contaminants investigation at the Keithsburg Division refuge.

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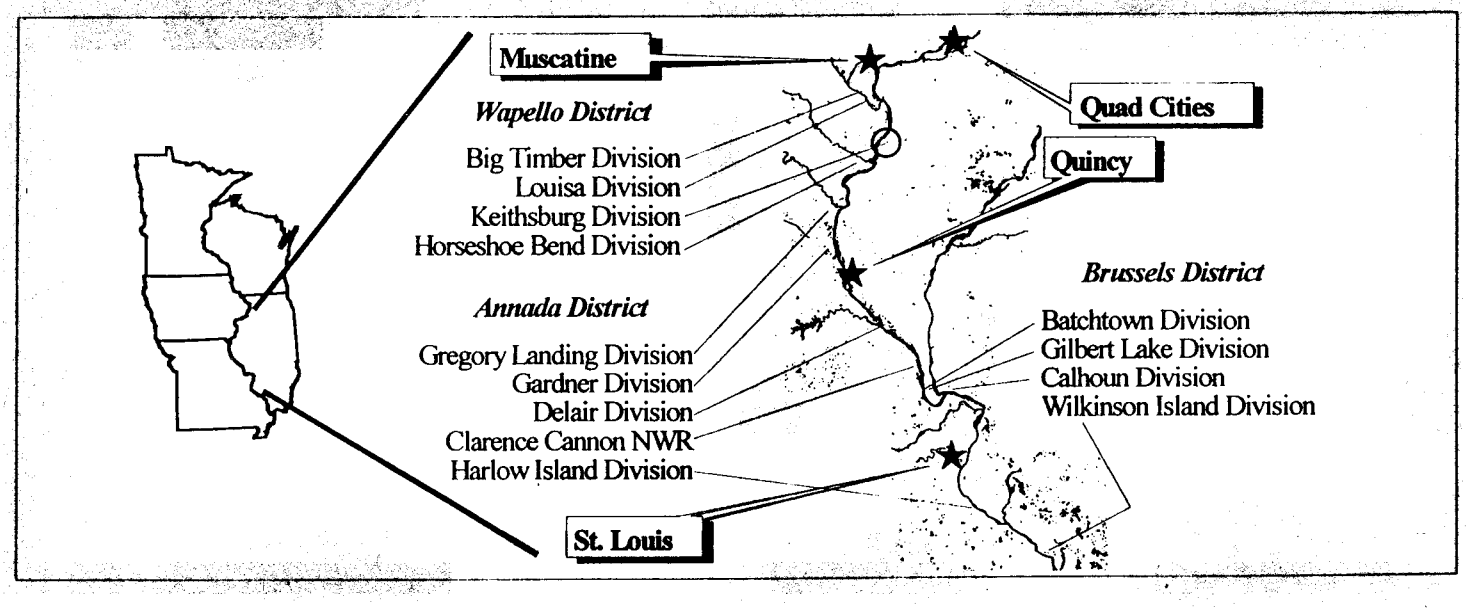

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Systemic insecticides were generally used at corn planting time (early April through mid May) for rootworm control on fields that were not rotated to soybean, and again on young plants (June through July) for corn borer and/or cut worm control.

Seed corn was coated with systemic fungicide chemicals and insecticide chemicals for some users.

METHODS

Water Quality

Water samples were collected at monthly intervals from the spring of 1994 to fall of 1995. Water samples were also collected following a couple of flood and storm events in 1995 and 1996.

Surface water was collected directly into bottles at one foot below the surface. The samples were maintained chilled in a cooler with blue ice and transported to the office for storage in a standard refrigerator. The samples were forwarded to a contract laboratory (University of Iowa Hygienic Laboratory, Iowa City and Des Moines, Iowa) for analysis within recommended holding times. The water samples were analyzed for a variety of pesticide and nutrient chemicals (Table 3). The water samples were not tested for insecticide, fungicide and some herbicide chemicals. The fungicide and some of the herbicide chemical tests are not readily available or are too costly. Insecticide chemicals are very short-lived and may be missed by routine monitoring.

A Solomat model 520c (Neotronics Company, Norwalk, CT) water quality meter was used to measure surface water temperature (°C), pH, dissolved oxygen (milligrams per liter - mg/L), conductivity (microSiemens per centimeter - μS/cm) and turbidity (NTU). Measurements were taken each time chemistry samples were collected. Readings were taken at one foot below the surface. Water depth was measured using a Hummingbird model LCR400 ID depth sounder (Eufaula, AL). Water transparency was measured using a standard Secchi disk (Wildco Company, Saginaw, MI). Observations on aquatic plant cover were noted for the sampling area.

Sediment Quality

Sediment samples were collected with a standard Ekman dredge (Wildco Company, Saginaw, MI). The dredge was dropped into the substrate, closed and raised for inspection. The contents were emptied into a stainless steel bowl if the dredge was at least three quarters full. If the grab was not complete, another grab was attempted approximately two meters in any direction from the last attempt.

The material in the bowl was gently mixed with a stainless steel spoon and portions were scooped into chemically clean jars for analyses.

The samples were maintained chilled in a cooler with blue ice and transported to the office for storage in a refrigerator. The samples were forwarded to the contract laboratory for analyses within recommended holding times. The sediment samples were analyzed for a variety of analytes (Table 4).

Separate samples were also collected as described above for pore water analysis. These sediments were scooped directly into large centrifuge vials and handled as discussed above. The samples were centrifuged at the contract laboratory within 48 hours after collection to separate the interstial pore water from the sediment grains and tested the same as for the water samples (Table 3).

Sediment sample temperature and pH were measured immediately in the field with a standard glass thermometer and a soil pH probe (OSK, Tokyo, Japan). The amount of coarse particulant matter such as leaf particles and color was noted on the sediment collection - field data sheet.

Table 3. Analytical methods, detection limits and preservative types for water quality samples.

Analyte	Method	Detection Limit	Preservative
Ammonia-nitrogen	Automated phenate	0.1 mg/L[1]	Sulfuric Acid
Nitrate-nitrogen	Automated cadmium reduction	0.1 mg/L	Sulfuric Acid
Phosphate-phosphorus	Automated ascorbic acid	0.1 mg/L	Sulfuric Acid
Alachlor	ELISA[2]	0.1 μg/L[3]	Refrigeration
Triazines[4]	ELISA	0.1 μg/L	Refrigeration
Metolachlor	ELISA	0.25 μg/L	Refrigeration
Imazethapyr	Gas chromatography	1.0 μg/L	Refrigeration

[1] Milligrams per liter or parts per million
[2] Enzyme-Linked Immunosorbent Assay
[3] Micrograms per liter or parts per billion
[4] Triazines include atrazine and cyanazine compounds

Table 4. Analytical methods and detection limits for the sediment quality samples.

Analyte	Method	Detection Limit
Ammonia-nitrogen	Automated phenate	1 mg/kg[1]
Phosphate-phosphorus	Automated ascorbic acid	1 mg/kg
Grain size distribution	Dry weight and sieve	
Total organic carbon content	Colormetric	
Herbicide scan[2]	HPLC[3]	0.1-0.2 mg/kg
Chlorinated hydrocarbon insecticide scan	Gas chromatography	0.05 mg/Kg
Polychlorinated biphenyl scan	Gas chromatography	0.5 mg/Kg
Total volatile hydrocarbon scan	Gas chromatography	100 μg/Kg[4]
Total extractable hydrocarbon scan	Gas chromatography	3 mg/Kg
Heavy metal scan	Various see Table 5	Various see Table 5

[1] Milligrams per kilogram or parts per million
[2] Atrazine, cyanazine, metolachlor, alachlor, metribuzin, butylate, trifluralin and acetochlor
[3] High Pressure Liquid Chromatography
[4] Micrograms per kilogram or parts per billion

Toxicity Testing

Sediment samples were collected in late summer of 1995 as described in the sediment quality section for use in a toxicity test. This time of year was selected because ammonia concentrations and potential toxicity may peak (Fraizer *et al* 1996). The test was designed to evaluate the acute toxicity of sediment ammonia to burrowing mayfly nymphs (*Hexagenia* species) (Ciborowshi *et al* 1992). *Hexagenia* are commonly found in Mississippi River backwater sediments. Limited toxicity data are available for those species of mayfly that actually dwell within the substrate like *Hexagenia* versus other types of mayfly (USEPA 1985).

The mayflies were hatched prior to the test from purchased wild caught eggs and raised in a sediment mixture prepared in the laboratory. The laboratory sediment mixture was used as un-contaminated reference sediment for the bioassay.

Four serial dilutions using the reference sediment were set up for the test. The test series was replicated five times and lasted 96 hours. A 200 milliliter volume of sediments was thoroughly mixed with prepared laboratory water at a ratio of 1:4. The solids were allowed to settle for about 60 hours before the introduction of the nymphs. The bioassay water was aerated.

After 96 hours, the number of dead versus live nymphs were counted. The bioassay was monitored for temperature, pH, dissolved oxygen and ammonia. Un-ionized ammonia concentrations were calculated using pH and temperature.

Aquatic Macrophytes

Aquatic plant diversity was surveyed at the two study wetland sites A and B during 1995. The methods followed those developed for the Upper Mississippi River Long Term Monitoring Program (USFWS 1995).

Briefly, the plant survey method included setting up a series of line transects at 100 meters apart and perpendicular the northeast shoreline. This shoreline was selected because it was accessible at both of the study wetlands. Three grab samples were collected with a rake every five meters along each transect. Relative abundance ratings were assigned for each plant species present.

In addition, the cover of aquatic macrophytes were described for the water and sediment quality sampling locations each time these sites were visited. This procedure provided a qualitative assessment of diversity and seasonal succession of macrophytes.

Phytoplankton

Phytoplankton samples were collected along with the water chemistry samples in 1995. Six replicate samples were collected at Secchi depth in a circular pattern around each of the two Spring Slough locations.

Surface water was pumped through an 80 micron plankton net for the amount of time necessary to filter 50 liters of water. Pump speed was timed before and after collections at each sampling location. The trapped matter from each replicate was rinsed with deionized water into sample bottles. Each sample bottle was topped with rinse water to 100 milliliters and preserved with Lugol's solution.

The sample was processed in the laboratory by mixing with a magnetic stirrer and extracting a 0.1 milliliter aliquot with a pipet pump. The pipet sample was placed in a Palmer counting cell for enumeration to genus level by student interns.

Benthic Macroinvertebrates

Benthic macroinvertebrate diversity was assessed in 1995 along with the sediment sampling activities and at the two study wetland sites A and B.

Triplicate sediment samples were collected with an Ekman dredge as described above for sediment quality. The contents of the dredge were emptied into a standard pan sieve with 0.595 millimeter mesh if the dredge jaws were closed and it contained at least a three fourths full compartment of sediment. If the dredge was not near full another grab was taken about two meters away in any direction from the first attempt.

The sediment was washed from the organisms and debris with surface water pumped through a hose at the boat. The material that was trapped by the sieve was gently back flushed into ziplock bags. This material was then preserved in 80 percent ethyl alcohol and three milliliters of a five percent solution of rose bengal dye for laboratory processing.

A different collection method was used at the two wetland study sites because of the presence of aquatic macrophytes and firm substrate. Three one meter grids were surveyed at each wetland. A standard kick net was used to sweep the grid area. The contents caught in the net were emptied into a white enamel pan. Coarse particulant matter was removed with caution as not to lose any invertebrates. The remaining material was rinsed in a container and preserved with 80 percent ethyl alcohol and three milliliters of a five percent solution of rose bengal dye for laboratory processing.

Laboratory processing included counting and identifying the preserved organisms to recognizable taxonomic units (mostly family level) by student interns. Common chironomid subfamilies were identified by using eyespot morphology (Figure 6). Although identification of chironmids by eye spot morphology may include rare members from other subfamilies, it is a reliable and rapid method when the habitat is suspected of containing mostly common taxa.

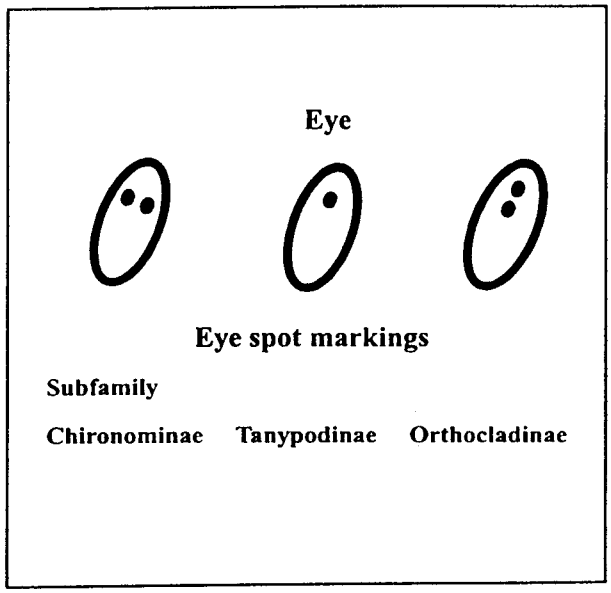


Figure 6. Eye spot morphology used for preliminary identification of members of the family Chironomidae (Delucchi pers. comm.).

Fish

Fish health growth potential and diversity were assessed during four studies completed during the study period for the Keithsburg refuge investigation.

One study included fish growth and diversity inventory surveys conducted by the U.S. Fish and Wildlife Service, Columbia Fisheries Resource Office (Columbia, MO). The second study was a cooperative winter creel census by Illinois Department of Natural Resources (Aledo, IL), Illinois Natural History Survey (Champaign, IL) and

this office. The third study was a fish tissue contaminant survey conducted by the author. The fourth study was biomonitoring of bluegill cholinesterase enzyme activity as an assessment of insecticide exposure. The biomonitoring was completed by the Department of Animal Ecology, Iowa State University (Ames, Iowa). Cholinesterase activity is altered by exposure to organophosphate and carbamate insecticides. Severe inhibition of cholinesterase activity is diagnostic of insecticide poisoning.

The field and analytical methods for fish studies are provided in technical reports in Appendix A. Method highlights for the fish studies are summarized below (taken from the reports in Appendix A).

The fish specimens that were saved for the growth survey and chemical analysis were collected from throughout both ends of Spring Slough.

The fish specimens that were saved for biomonitoring enzyme activity were collected from the north end of Spring Slough which was believed to be closer to agrichemical sources.

Fish were also collected from Wildcat Den State Park, Henry County, IL and Cooligar Slough, Louisa County, IA for use as reference specimens for the fish studies.

Abnormal fin conditions and external lesions were noted on all fish collected for the fish studies. Scales and the sagittal otolith were removed from 40 bluegill to evaluate growth rates using methods according to Schramm (1989).

Selected whole fish were saved in acid cleaned jars or plastic bags for analyses. The specimens were maintained in a cooler with blue ice for chemical analysis or frozen with dry ice for enzyme analysis. The specimens were transported to the office for storage in a standard freezer. The specimens were forwarded under dry ice to contract laboratories for analyses within recommended holding times.

Whole body common carp, large-mouth bass and bluegill specimens were analyzed for a variety of heavy metals (Table 5).

Table 5. Methods and detection limits for whole fish tissue chemical analysis.

Analyte	Analytical Method	Quantitation Limit
Arsenic	Graphite furnace AA[1]	0.5 $\mu g/g^2$
Cadmium	Graphite furnace AA	0.2 $\mu g/g$
Chromium	ICP[3]	1.0 $\mu g/g$
Copper	ICP	1.0 $\mu g/g$
Lead	Graphite furnace AA	5.0 $\mu g/g$
Mercury	Cold vapor AA	0.1 $\mu g/g$
Nickel	ICP	5.0 $\mu g/g$
Selenium	Graphite furnace AA	1.0 $\mu g/g$
Zinc	ICP	5.0 $\mu g/g$

[1] Atomic Absorption Spectroscopy
[2] Micrograms per gram or parts per million
[3] Inductively Coupled Plasma Emission Spectroscopy

Birds

Aquatic bird use at the two study wetlands sites A and B was monitored by members of a local ornithological club (Quad City Audubon Society, Davenport, Iowa). Volunteers visited the two study wetlands once a week according to personal schedules. The study wetlands were visited five times between March 8, 1995 and April 11, 1995. Estimated numbers of each species of aquatic birds flushed or observed on the wetland were tallied.

An experiment was designed to assess exposure to insecticides in birds living along the edge of cropfields. Cholinesterase activity was used as an indicator of exposure to insecticide chemicals used in the watershed for rootworm control. The potential exposure pathways included drinking pooled surface water that forms in cropfields after a rainstorm and ingestion of contaminated invertebrates.

The house wren (*Tachycineat bicolor*) was the bird species selected for study because it is common, attracted to artificial nest boxes and the wren's nesting season may overlap the rootworm control period.

Cardboard bluebird nest boxes (Forestry Suppliers, Inc., Jackson, MS) were set up along the riparian strip between Spring Slough and cropfields at the north end of Keithsburg Division. Another string of nest boxes were set up to collect reference data along the riparian strip that separates Cooligar Slough and a grassland in Flaming Prairie County Park (Louisa County, Iowa).

The boxes were set up in late April by volunteers from the Quad City Audubon Society (Davenport, Iowa). One box was wired to a tree at chest height every 50 paces along the riparian strip for a total of 19 boxes at Keithsburg Division and 17 boxes at the Cooligar Slough site. The volunteers checked the boxes biweekly.

In late June, six nestlings, two per nest, from each of the two study sites, were removed from the nest boxes for analysis.

The chick was weighed to the nearest tenth of a gram and decapitated. The brain was excised immediately and placed in a Whirl-pac™ plastic bag (Nasco, Fort Atkinson, WI). The samples were maintained frozen in a cooler with dry ice and transported to the office for storage in a standard freezer. The samples were forwarded to a contract laboratory (National Wildlife Health Laboratory, Madison, Wisconsin) for analysis within recommended holding times. Brain tissue was analyzed for cholinesterase activity according to methods by Ellman 1961.

Quality Control Plan

The quality control plan included decontamination procedures, calibration and quality assurance tests.

Decontamination Procedures

Instrument probes and sensors were rinsed with de-ionized water between uses. The sediment sampling gear was cleaned with surface water, decontaminated with acetone and rinsed with deionized water between uses. The de-ionized rinse water used on the sediment sampling gear was occasionally collected for analysis to test for cross contamination.

Instrument Calibration Checks

The water quality meter was calibrated and checked periodically throughout the year according to manufacturer procedures and specifications. The

calibration standards for the water quality meter were obtained from the manufacturer.

Quality Assurance

We randomly collected duplicate field samples of water and aquatic sediments and submitted them along with the original samples for chemical analysis along with the original samples. The results of the duplicate and original samples were compared as a test of laboratory performance.

The contract laboratories also analyzed split samples, blank samples and spiked samples according to their quality control program (Appendix B). The results from the ELISA tests were cross checked with standard analysis because ELISA is an inexpensive screening tool and may be less accurate.

Student identification work for the phytoplankton, aquatic plants and benthic macroinvertebrate organisms was monitored by their Biology Department university professors.

Data Management and Analysis

Field data, observations and instrument readings were recorded in the field in a bound write-in-the-rain book. Special field sheets generated for this study and the laboratory test results were archived in project binders maintained at the Rock Island Field Office, Rock Island, Illinois.

The data from the field sheets and laboratory reports were entered into spreadsheets (Microsoft Excel). The data were graphically and statistically analyzed using Microsoft Excel and SigmaStat (Jandel Corporation, San Rafael, CA).

Figure 7. Photograph of lake monitoring at Keithsburg Division refuge, 1995.

RESULTS AND DISCUSSION

Water Quality

Herbicides

The popular herbicides used for corn production were detected throughout the year in the water samples collected from the refuge unit and un-named tributary ditches (Appendix C). The concentrations were only slightly greater than analytical detection limits during most of the year. All of the observed herbicide chemicals had peak concentrations in May.

The peak concentration of atrazine measured for the routine monitoring program was 4.7 micrograms per liter (μg/L) or parts per billion (ppb). The peak concentration of cyanazine was 2.8 μg/L. The peak concentration of metolachlor was 3.9 μg/L. The peak concentration of alachlor was 3.34 μg/L. The peak concentration of acetochlor was 3.1 μg/L.

Herbicides used for corn production were detected at higher concentrations in surface run-off samples collected just outside the refuge unit near tributary #2 following a spring 1995 rain storm. The maximum concentration of atrazine was 19 μg/L, alachlor was 8.9 μg/L and metolachlor was 9.0 μg/L. A soybean herbicide, imidazolinone, was detected at 12 μg/L in the south end of Spring Slough during 1996 Pope Creek flood conditions

The concentrations of herbicides observed at Keithsburg Division were within the range of concentrations that have been detected in the rivers and streams for this watershed and throughout Illinois (Ciba Giegy 1992 and IEPA 1990).

The maximum concentrations of the various herbicides detected for this study were below lethal values for aquatic life (Fairchild *et al* 1993). Aquatic plants tend to be more sensitive to herbicides than aquatic invertebrates or vertebrates (Hartman and Martin 1985, Solomon *et al* 1996, Streit and Peter 1978 and Howe *et al* 1998).

Lethal concentrations for an algae species (*Selenastrum capricornutum*) to atrazine is 59 μg/L, metolachlor is 55 μg/L and alachlor is 36 μg/L (Solomon *et al* 1996). Lethal concentrations for a vascular plant species (*Potamogeton perfoliatus*) to atrazine is 53 μg/L (Forney and Davis 1981).

The concentrations of the herbicide chemicals detected at Keithsburg Division refuge may cause adverse effects in aquatic plants (Abou-Waly *et al* 1991, Brown and Lean 1995, Detenbeck *et al* 1996, Hersh and Crumpton 1987, Fleming *et al* 1995, Jones *et al* 1986 and Stay *et al* 1989).

Atrazine concentrations between 1 and 35 μg/L were shown to adversely affect a variety of phytoplankton species (Solomon *et al* 1996).

Atrazine concentrations of 20 μg/L in test ponds caused changes in phytoplankton community structure leading to the establishment of more resistant species over time (DE Noyelles *et al* 1982).

Huber (1993) reported that the community changes documented for phytoplankton at concentrations of 20 μg/L may be reversible and not permanent.

Photosynthesis and respiration rates for the aquatic macrophyte, sago pondweed (*Potamogeton pectinatus*), were affected at concentrations as low as 0.029 μg/L of atrazine and between 1 and 10 μg/L of alachlor (Fleming *et al* 1995).

The herbicides detected in refuge surface waters match those used on adjacent fields. The potential transportation pathways included surface run-off and subsurface water discharge. Herbicide chemicals may also be precipitated during rain storms following application times (Nations and Hallberg 1992). For example up to 40 µg/L of atrazine was detected in rain samples from central Iowa (Nations and Hallberg 1992).

Nutrients

The concentrations of phosphate and nitrogen compounds varied throughout the sampling times from very low to very high for all of the sampling stations (Table 6). The water nutrient chemistry data are in Appendix C.

The concentrations of total phosphate-phosphorus at the tributaries that fed into Spring Slough regularly exceeded the criteria to control biological nuisances. To prevent the development of biological nuisances, total phosphate-phosphorus should not exceed 50 µg/L for any tributary where it enters a lake (USEPA 1986).

Phosphates are not typically toxic to aquatic animals (USEPA 1986). Phosphate loading can cause plant blooms, eutrophication and poor water quality conditions (Wetzel 1983).

The maximum concentration of nitrate detected for this study was 40 mg/L. Levels of nitrate-nitrogen below 90 mg/L should have no adverse health affects on warmwater fish (Knepp and Arkin 1973). There is no restrictive criteria for nitrate-nitrogen for the protection of aquatic life because it is ubiquitous in the environment (USEPA 1986).

The nitrate drinking water criteria for human health is 10 mg/L (USEPA 1986). Nitrate is converted in the intestinal tract to nitrite which is toxic at very low concentrations.

Nitrate pollution is a concern for livestock producers for the same health reasons as for human health problems. There may also be health risks to wildlife production from drinking nitrate polluted water, but there is no information in the literature on nitrate toxicity to furbearers or other small mammals.

Like phosphorus, excess nitrogen in aquatic systems can cause nuisance aquatic plant blooms and eutrophication because it is a primary nutrient and fuels plant production.

There are likely several sources for nutrients. It is not know if there are inputs from the cottage septic systems located along the east side of the refuge. Fertilizers that contain anhydrous ammonia and phosphorus were applied yearly to the adjacent cropfields. Ammonia products are converted first to nitrite and then to nitrate by bacteria in the soil.

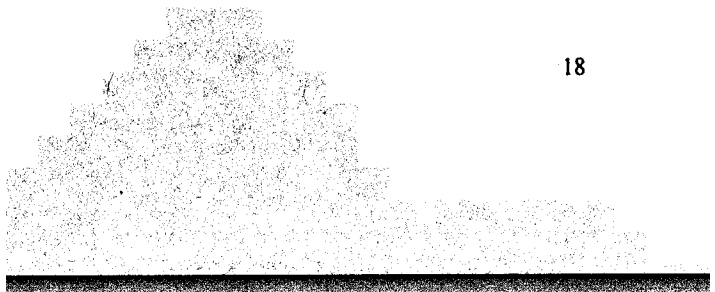
Fertilizer chemicals and nitrate may be transported into refuge wetlands and slough tributaries by surface run-off. Dissolved nitrate may accumulate in subsurface water below cropfields and contaminate groundwater resources (Stevenson 1982 and Kalkoff *et al* 1992). Cropfield tile drainage water and shallow groundwater was transported into refuge wetlands along the slough tributaries and along the east sandy bluff at intermittent springs. The groundwater in this region is contaminated as noted by the nitrate drinking water advisory for campgrounds in a State Park located 3 miles south.

Floodplain habitats especially bottomland forests can be naturally rich in nitrogen because of high production of organic matter and subsequent rapid decay during periods of seasonal flooding (Magee 1993).

Table 6. Range of phosphate and nitrogen chemicals and percent of samples that the phosphate and nitrogen chemicals were detected above the quantitation limit (in parenthesis) for the water quality sampling locations at Keithsburg Division refuge for 1994 and 1995.

Analyte	South End (n=23)	North End (n=20)	Trib 1 (n=18)	Trib 2 (n=16)	Trib 3 (n=16)	Trib 4 (n=11)
Phosphate-phosphorus (μg/L)[1]	100-400 (72)	100-200 (76)	100-500 (78)	100-1800 (94)	100-600 (87)	100-600 (90)
Ammonia-nitrogen (mg/L)[2]	0.1-0.3 (39)	0.1-0.3 (30)	0.1-0.5 (72)	0.1-1.9 (69)	0.1-0.4 (69)	0.1-0.7 (70)
Nitrate-nitrogen (mg/L)	0.1-8.5 (52)	0.7-8.5 (90)	0.5-40 (100)	0.1-7.6 (94)	0.1-4.1 (100)	0.1-2.3 (64)

[1] Micrograms per liter or parts per billion
[2] Milligrams per liter or parts per million



Aquatic bird feces may contribute as much as 70 percent of the phosphorus in an aquatic system (Manny *et al* 1994). High waterfowl use may stimulate the production of algae from the nutrient enrichment of bird feces (Skoruppa and Woodrin 1994). Large flocks of coot, waterfowl, double crested cormorant and white pelican use the refuge unit during migration periods and may contribute significantly to the internal nutrient load at the refuge unit.

There were periods when nutrient concentrations were low or not detected. This is likely related to two processes. The processes include plant processes and dilution.

Nitrate is assimilated by plants and converted to nitrogen by bacterial denitrification in wetland systems (Crumpton *et al* 1993). Phosphates are used by plants and sorbed to organic matter and sediments (Cooke and Kennedy 1977).

The refuge unit was flooded each spring during the study and less contaminated river flood water may have diluted nutrient concentrations in refuge surface waters at times. Likewise, the concentration of nutrients may be diluted if different, deeper and cleaner groundwater is occasionally discharged into refuge surface water. Surface water discharge and recharge of groundwater may occur in areas near alluvial stream valleys such as for the Edwards River and Pope Creek (Squillace *et al* 1996).

General Parameters

General water quality parameters were within normal ranges except for dissolved oxygen and pH (Appendix C). Parameters such as temperature and depth changed over the year as expected (Figure 8). Temperature ranged from 2.5 degrees Celsius in the winter to 30.2 degrees Celsius in the summer.

Water depth was usually between 1.2 to 1.8 meters except in May of 1995 it reached 2.5 meters deep related to Pope Creek flood conditions. Conductivity ranged from 300.1 to 498.3 μS/cm. Secchi depth ranged from 17 centimeters in late summer down to 67 centimeters during the winter. The low Secchi depth was usually due to phytoplankton blooms.

Short periods of up to a few weeks of very turbid conditions from suspended sediments were observed related to two processes. One process was during times when turbid flood water from Pope Creek flowed into the refuge. The other process was believed to be due to bioturbation from fish being concentrated into the south end Spring Slough as surrounding wetlands dried up during periods of very low water stage.

Dissolved oxygen and pH were extreme at times but within the criteria for the protection of aquatic life (USEPA 1986). Dissolved oxygen and pH varied daily depending on the time of day and seasonal status of primary production. Supersaturated dissolved oxygen concentrations and high pH (up to 9.2) were observed on some mid day readings. This was not unexpected because dissolved oxygen and pH may vary greatly daily in productive lakes related to photosynthesis and respiration cycles of phytoplankton.

High dissolved oxygen concentrations can occur on sunny days during an algae bloom from increased photosynthesis. This may be followed by low dissolved oxygen concentrations during the night time from algae respiration. A net low oxygen condition may occur after a few overcast days following an algae bloom. This is caused by low day time photosynthesis rates and continued high night time respiration rates. This cycle can cause a summer fish kill if the dissolved oxygen concentration drops below 5 mg/L.

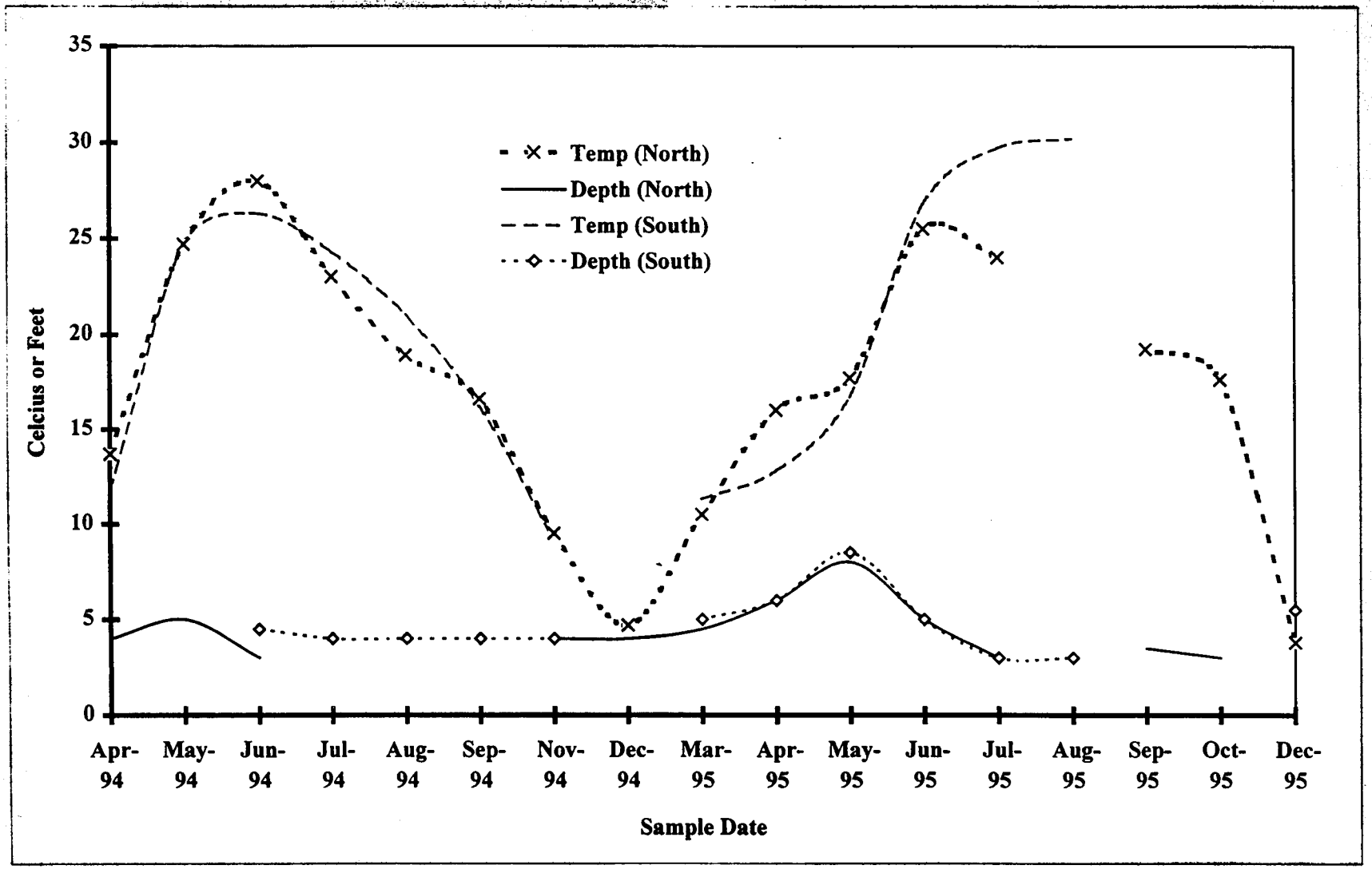


Figure 8. Seasonal changes in temperature (Celsius) and water depth (feet) in Spring Slough, Keithsburg Division refuge.

Sediment Quality - 1994 Post Flood Status

General

Surficial sediment texture was different between the 1994 sampling stations. The sand grain size distribution ranged from 3.8 percent to 32.1 percent in the backwater complex. The sampling stations with high sand content were close to potential sources such as the sandy upland bluff or overflow areas for the three adjacent rivers. Total organic carbon content ranged from 2.2 to 6.4. The sediment texture and chemistry data are in Appendix D.

Organic Contaminants

Gasoline, PCB, DDT, chlordane and similar organic contaminants were not detected in any of the 1994 sediment samples.

Petroleum hydrocarbon chemicals were detected at all of the sampling stations at concentrations between 43 and 130 mg/Kg. Total petroleum hydrocarbon concentration of 100 mg/Kg or less may be considered natural background levels caused by aquatic plant decay. Only one station had concentrations greater than 100 mg/Kg and that was in the middle section of Spring Slough.

Hydrocarbon chemicals were detected in the sediments at Keithsburg Division in 1989 (Young 1991). The source of the chemicals were believed to be from the natural production of blue-green algae (Coffey 1994). Blue-green algae can produce polycyclic aromatic hydrocarbon (PAH) chemicals (USEPA 1982). PAH chemicals would be detected by the total petroleum hydrocarbon test. The chemists that performed the total petroleum hydrocarbon test in 1994 indicated that the chromatographic profiles did not match their motor oil standards. These chemists suggested to us that the results may indicate natural hydrocarbon type chemicals found in aquatic substrates (Appendix D).

Metals

The average concentrations of arsenic and the heavy metals measured in 1994 were not above levels of concern and were at background concentrations for soil and aquatic sediments (see Table 2 for background concentrations) (Schacklette *et al* 1971, USEPA 1977 and ILEPA 1984). With the exception of chromium, the average metal concentrations in 1994 were lower compared to the pre-flood average concentrations measured in 1992 (Table 7) (Coffey 1995).

Note that cadmium was not detected above analytical detection limits in 1994 and cadmium had a mean concentration of 0.57 mg/Kg dry weight in 1992. This difference is because the cadmium test in 1994 had a detection limit of 2 mg/Kg which is much higher than the detection limit in the 1992 test.

Table 7. Average concentrations for sediment metals at Keithsburg Division for 1992 and 1994 (milligrams per kilogram, dry weight).

Metal	1994 (n =6)	1992 (n = 83)
Arsenic	4.8	6.1
Cadmium	0	0.57
Chromium	24.5	20.66
Copper	14.3	27.52
Nickel	19.0	21.1
Lead	14.6	21.6
Zinc	77.8	89.53

Nutrients

The concentrations of the two nutrients (ammonia and phosphates) that were examined in the 1994 sediment samples were slightly to moderately elevated (USEPA 1977). The 1994 sediment nutrient concentrations were less than 1992 values (Coffey 1995). Ammonia and phosphate concentrations had strong correlation with total organic carbon content (r=0.80 and 0.90 respectively) as was the case for ammonia in 1992 (r=0.75) (Coffey 1995).

The differences between 1992 and 1994 may or may not be related to the 1993 flood along the Mississippi River, but this is not significant because nutrient dynamics may vary greatly between years in natural systems.

The 1994 data were collected in the fall and the 1992 data were collected in the summer. Sediment ammonia production is expected to be lower in the fall versus the summer because it is temperature regulated (Rand and Petrolelli 1985).

We observed very little phytoplankton and macrophyte production during the 1993 flood. The limited primary production in 1993 could relate to lower organic loading to the substrate versus 1992 conditions which may result in less ammonia production (Rand and Petrocelli 1985).

Sediment Quality - 1995 Spring Slough

General

Surficial sediment texture was similar throughout the north Spring Slough sampling areas for 1995. The only difference was that the far north sampling point was very sandy. Otherwise the dominant grain sizes were silt and clay. Total organic carbon content ranged from 3.4 to 6.6 percent for all of the samples. The sediment texture and chemistry data are in Appendix D.

Sediment temperature and pH was the same between the sampling locations. Temperature was 29 degrees Celsius and pH was 7.

Herbicides

Alachlor was the only herbicide that was detected in the 1995 sediment samples. Alachlor was detected in two of the seven bulk sediment samples at 0.12 and 0.24 mg/kg dry weight. Alachlor was detected in four of the four pore water samples. The average concentration of alachlor in the pore water samples was 1.29 μg/L with a standard deviation of 0.18.

The concentration of the herbicide alachlor in solution within in the sediments was just above known sensitivity levels (1 - 10 μg/L) for a rooted aquatic plant species (Fleming *et al* 1995).

Nutrients

The concentrations of the two nutrients (phosphates and ammonia) that were examined in the 1995 sediment samples were moderately elevated and fairly uniform throughout the sampling area (Table 8) (USEPA 1977).

The average concentration of phosphate-phosphorus in the north Spring Slough sampling area (1720 mg/kg, n=5) was above the average background concentration of 703 mg/kg for sediments collected from 63 Illinois lakes (Sefton *et al* 1979).

Ammonia that is found in the substrate is mostly bound to surrounding fine grain particles in the form of ammonium (NH_4^+). The portion of ammonia is in solution is un-ionized ammonia (NH_3).

Table 8. Average concentrations and related information for sediment nutrients for Spring Slough, Keithsburg Division refuge in 1995.

Nutrient	Mean (n=5)	Standard Deviation
Bulk Sediments (milligrams per kilogram dry weight or parts per million)		
Ammonia-nitrogen	195.43	18.62
Phosphate-phosphorus	1720	193
Sediment Pore Water (milligrams per liter or parts per million)		
Ammonia-nitrogen	10.67	0.91

Sediment temperature and pH data may be used to calculate the un-ionized ammonia portion. Un-ionized ammonia remains in solution and may be used as a measure of ammonia toxicity (USEPA 1976).

The average un-ionized ammonia-nitrogen concentration for the sediment pore water within the north Spring Slough sampling area was 79.9 μg/L.

This concentration is between the mean and maximum values for another Upper Mississippi River study that detected an averaged concentration of un-ionized ammonia at 55 μg/L, and a maximum concentration of un-ionized ammonia at 175 μg/L in late summer (Frazer *et al* 1996).

The average concentration of un-ionized ammonia at Spring Slough was below lethal values for many species of pollution tolerant organisms.

Examples of acute toxicity values (un-ionized ammonia-nitrogen) for tolerant organisms include the oligochaete worm between 1200 to 3000 μg/L and the chironomid bloodworm between 1100 to 3200 μg/L (Williams *et al* 1986).

The average concentration of sediment un-ionized ammonia in Spring Slough was above concentrations that may cause harm but not kill pollution intolerant organisms.

An example of an organism for the Upper Mississippi River that is sensitive to poor water quality and pollution may be the fingernail clam (family Sphaeriidae). The growth of fingernail clams was inhibited by exposure to 30 μg/L un-ionized ammonia (Sparks and Sandusky 1981). The lowest concentration of un-ionized ammonia that affected survival of fingernail clams was between 90 and 160 μg/L (Zischke and Arthur 1987).

Lethal values for un-ionized ammonia to a bottom dwelling fish species, channel catfish (*Ictalurus punctaus*), is 500 μg/L which is well above the concentration that we observed (USEPA 1985).

Ammonia is produced in lake sediments from deamination of organic matter. Organic matter that is deposited in the substrate of aquatic and wetland systems is decomposed by bacteria which produce ammonia as a by-product. The ammonia is converted first to nitrites then to nitrates by bacteria. Ammonia production depends temperature, volume and quality of the organic matter (Rand and Petrocelli 1985).

The conversion of ammonia to nitrate can consume significant amounts of dissolved oxygen (Knowles and Lean 1987). This is often the cause of winter fish kills in isolated wetlands and ponds from chemical oxygen demand under an ice cap.

Toxicity Tests

The sediment toxicity tests did not cause mortality in the burrowing mayfly nymphs. Water quality parameters measured in the test vessels were unremarkable except for pH and ammonia. The pH values in the test vessels increased from 7.2 to 8.4 over the test period. This shift in pH caused un-ionized ammonia concentrations in the water for some test vessels to reach up to 700 μg/L. Similar results were observed for a toxicity test study on the Illinois River (USACOE 1992). The test illustrates the changes that may occur in potential toxicity of sediments from laboratory manipulation. Ammonia concentrations in the test vessels were much higher than that measured in the field and was related to an increase of 1.2 pH units.

Aquatic Macrophytes

Submerged or floating leaf aquatic macrophytes did not develop at the sampling locations during the study period. Beds of coontail (*Ceratophyllum demersum*) and American lotus (*Nelumbo lutea*) did develop in some locations in the backwater system. There were periods throughout the study when floating aquatic macrophyte production was high and dense mats developed, especially in late May and early June. The floating plant species included lesser duckweed (*Lemna minor*), greater duckweed (*Spirodela polyrhiza*) and waterfern *(Wolffia columbiana)*. There was a period only during the summer of 1994 that an extensive bed of waterfern (*Azolla mexicana*) covered most of the backwater. Waterfern species contain bacteria that can fix atmospheric nitrogen for survival in nitrogen deficient waters.

Study wetland A contained a thick and complete cover of floating plants throughout the growing season in 1994 and 1995. Wetland A also contained scattered stems of coontail and curly leaf pondweed (*Potamogeton crispus*) These plants were covered with algae or filamentous bacteria and seem to be in poor condition because the leaves sloughed off easily. Study wetland B contained scattered open water, duckweed areas, extensive and healthy looking beds of *Elodea* and coontail.

Phytoplankton

Maximum densities of algae cells in Spring Slough occurred during early summer (Figure 9). The maximum concentration measured was 404,000 cells per liter which was extremely high compared to historic information for other lakes through Illinois (Sefton *et al* 1979). High density of phytoplankton cells is a measurement and an indication of a highly productive and eutrophic lake.

Species richness for each phytoplankton phyla observed in the backwater system are listed in Table 9. The seasonal succession of phytoplankton included the presence of diatoms (phylum Chrysophyta), euglenophytes (phylum Euglenophyta) and large numbers of green algae (phylum Chorophyta) during the early and late parts of the growing season. A blue-green algae bloom (phylum Cyanophyta) occurred in mid growing season (Figure 10).

The density and sequence of the phytoplankton community succession were typical of fertilized lake patterns (Meyer 1994). Diatoms and euglenophytes are more tolerant of cold temperatures and appear in the fall, winter and early spring. Blue-green algae populations may develop if green algae production is eventually limited by nitrogen. Blue-green algae contain bacteria that make nitrogen available to the plant for survival in nitrogen deficient waters. Blue-green algae growth usually rises and falls with concentrations of phosphates.

Table 9. Phytoplankton taxa (phyla and genera) collected at Keithsburg Division in 1995.

Phylum Cyanophyta (blue-green algae)

Agmenellum
Anabaena
Anacystis
Aphanizomenon
Botryococcus
Chroococcus
Gomphosphaeria
Merismopedia
Microcystis
Oscillatoria
Tetrapedia

Phylum Chlorophyta (green alage)

Actinastrum
Ankistrodesmus
Chaetophora
Chlamydomonas
Chlorella
Clapdophora
Closteriopsis
Closterium
Coelastrum
Crucigenia
Cylindrocystis
Desmids
Dictyosphaerium
Elakatothrix
Eudorina
Kirchneriella
Mesotaenium
Microspora
Nannochloris
Pandorina
Pediastrum

Phylum Chlorophyta (continued)

Phytoconis
Platydorina
Pleodorina
Polytoma
Scenedesmus
Sphaeroplea
Spirogyra
Staurastrum
Tetraedron
Treubaria
Ulothrix
Volvox
Zygnema

Phylum Euglenophyta

Euglena
Phacus

Phylum Chrysophyta (diatoms)

Achnanthes
Asterionella
Attheya
Cyclotella
Dinobryon
Fragilaria
Melosira
Navicula
Nitzschia
Pseudostaurastrum
Stauroneis
Stephanodiscus
Synedra
Synura

Algal blooms have adverse ecological consequences. Extreme diurnal dissolved oxygen cycles may develop from algae photosynthesis and respiration setting up a fish kill. Dense blooms of algae may reduce water transparency and limit sunlight to submerged plants (Davis and Brinson 1976).

Large volumes of dead algal cells decay on the lake bottom which consumes dissolved oxygen and produces an anaerobic layer near the substrate. Natural hydrocarbon chemicals may accumulate in the substrate from decaying plant material (Rand and Petrocelli 1985).

Some blue-green algae collected at Spring Slough (*Anabena* and *Oscillatoria*) can produce biological toxins (Anatoxin-a) that can be lethal to wildlife (USEPA 1992).

Environmental Relationships

The crash in green algae numbers after the May 1995 bloom followed maximum concentrations of nitrate (Figure 11). Nitrate had a positive correlation with green algae density (r=0.91) and with total algae cell count (r=0.89).

Blue-green algae abundance had an inverse relationship with nitrogen and a more direct relationship with phosphate during this time frame (Figure 11). These trends are expected because algae production is nutrient dependent.

There may be other environmental stressors in addition to nutrient limitation present in the system that could have affected the crash in green algae numbers in May of 1995. These stressors did not stop the production of blue-green alage that increased during the period that green algae numbers decreased. The other environmental stressors included herbicide exposure, changes in temperature, discharge or flushing rates, planktivory and shading from duckweed cover (Figure 11).

The relationship between herbicide concentration and algae density was not as strong as for nitrate concentration and algae density. The correlation between herbicide concentration and green algae density was r=0.57, blue-green algae density was r=0.62 and total algae cell count was r=0.63.

Lange and Rada (1993) found that temperature and water discharge affected seasonal succession and standing crop of phytoplankton in a Mississippi River navigation pool. Perry *et al* (1990) found that reservoir flushing rates affected plankton densities.

The seasonal succession and diversity of the phytoplankton community we observed at Keithsburg Division in 1995 may have been influenced by basin flushing rate or discharge rate as with these other studies.

There was a relatively sudden discharge of surface water out of the backwater complex in late spring through the Pope Creek levee break and refuge water control structures as the Mississippi River stage receded from a spring high due to seasonal rains and northern ice melt. The sudden discharge was apparent by the decrease in water depth over a short period.

Very dense blooms of daphnids and other zooplankton were observed by the author on several occasions while sampling for phytoplankton. Fish and zooplankton grazers can have significant effects on algae numbers (Carpenter *et al* 1990). Grazers affect phytoplankton communities by removing edible species and nutrient excretion (Carpenter *et al* 1990). Fish and plankton interactions are complex and can affect whole lake ecosystem chemical processes and nutrient cycles (Carpenter *et al* 1990).

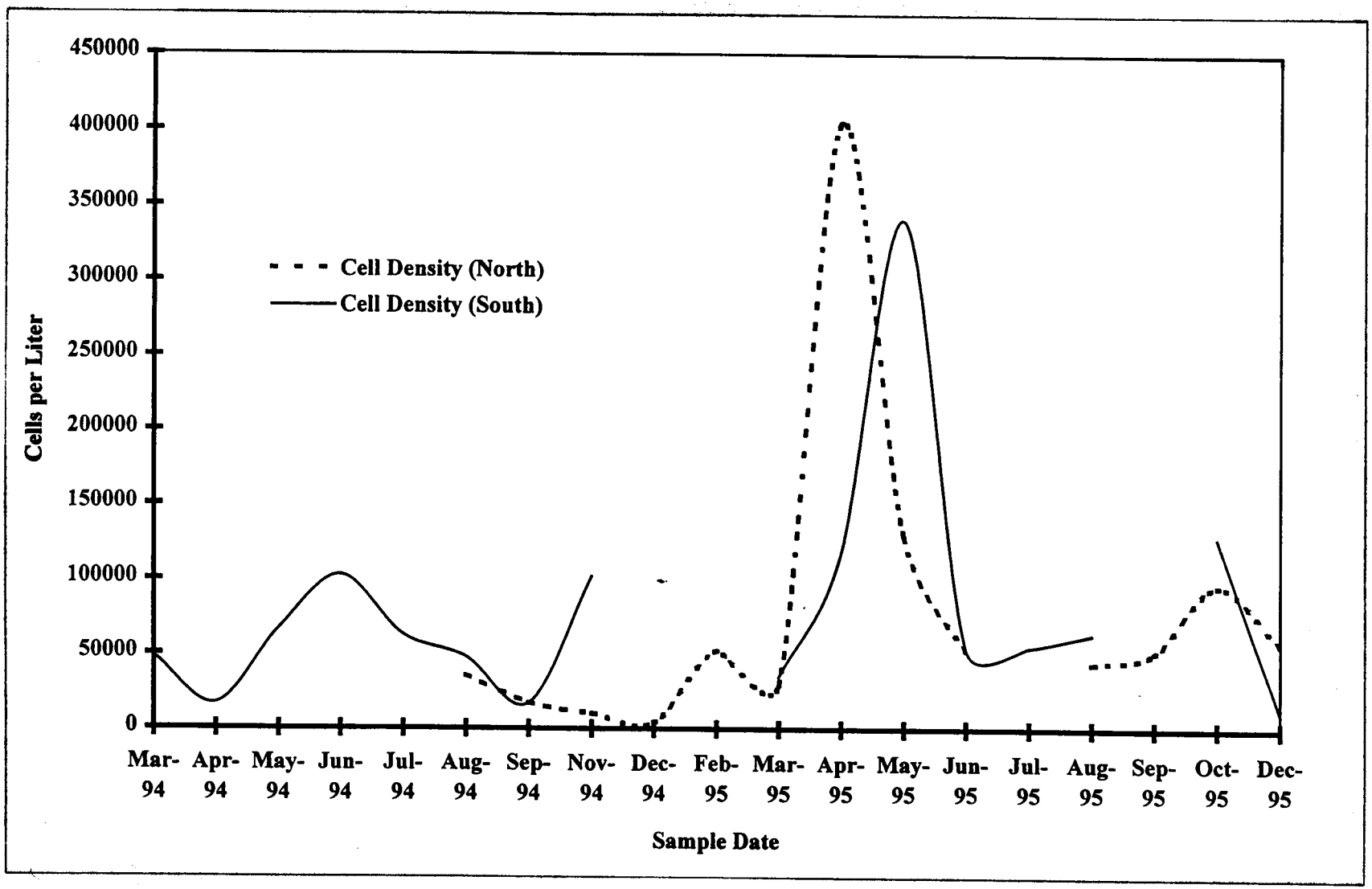


Figure 9. Seasonal changes in total phytoplankton cell density at Spring Slough, Keithsburg Division refuge.

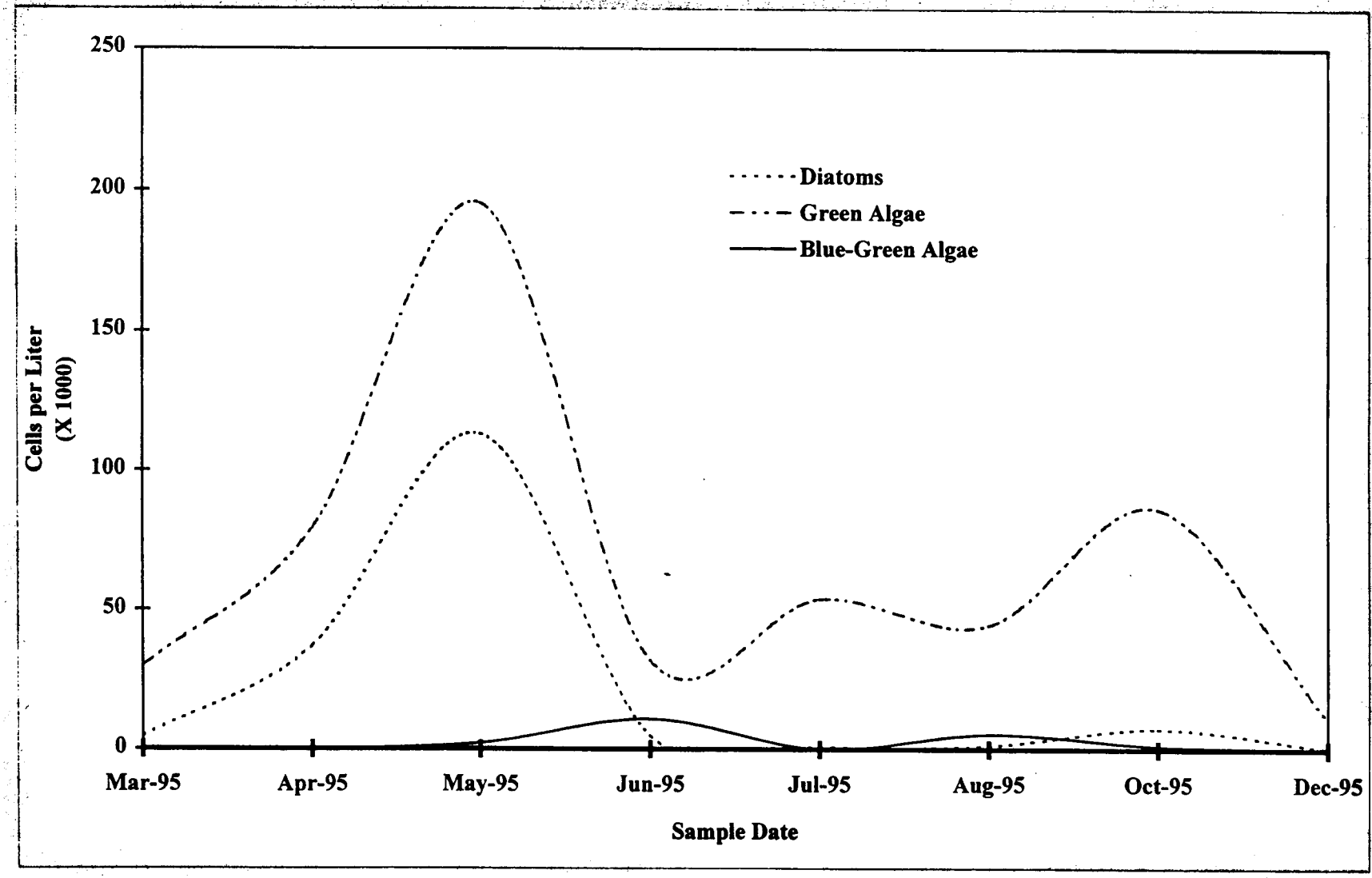


Figure 10. Seasonal succession of phytoplankton phyla at the south end of Spring Slough for 1995 , Keithsburg Division refuge.

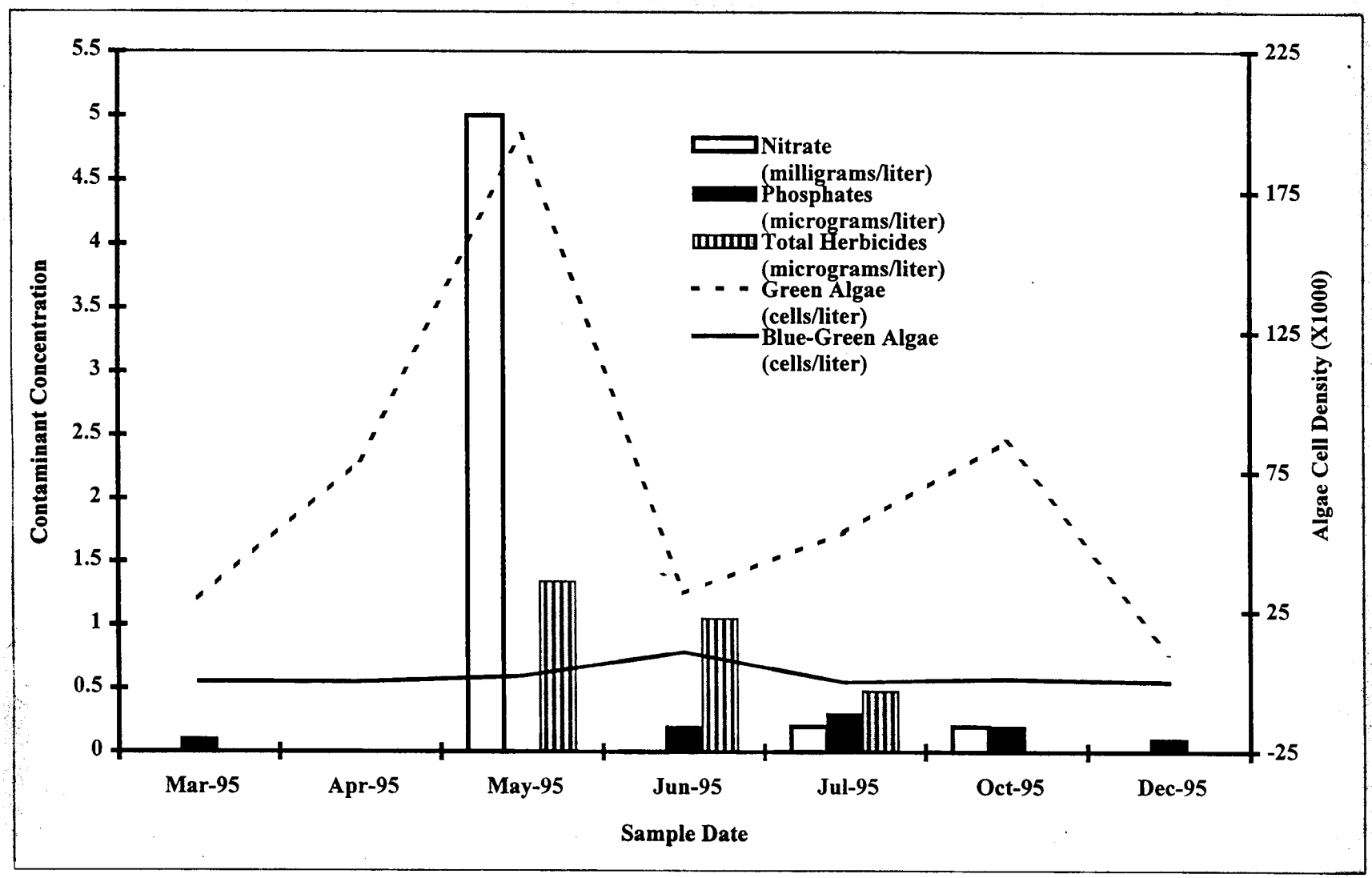


Figure 11. Trends in phytoplankton community structure and relationship to other environmental stressors in 1995 at Spring Slough, Keithsburg Division refuge.

Benthic Macroinvertebrates

Spring Slough Sediment Grab Samples

There were a few types of macroinvertebrates collected at the 1995 sediment sampling locations in Spring Slough (Table 10). The dominant infaunal (substrate dwelling) organism was aquatic worms (class Oligochaeta) with an average number per grab of 252.9. The other principal infaunal organism was the midge larvae (family Chironomidae) with an average number per grab of 4.74. The midge component of the assemblage was mostly members of the subfamily Chironominae (average number per grab 3.9) with few Orthocladine (average number per grab 0.8).

Oligochaetes and the midge subfamily Chironominae are indicators of eutrophic habitats (Bryce and Hobart 1972). Aquatic worms and chironomid midge larvae can tolerant poor sediment quality conditions (USFWS 1988).

The average number of fingernail clams per grab was 0.3. No burrowing mayfly nymphs were collected. Fingernail clams and burrowing mayflies may be common organisms in Upper Mississippi River backwaters (Eckblad 1990). Members of the midge subfamily Orthocladine mentioned above, fingernail clams and burrowing mayfly nymphs are indicators of good sediment quality (Byrce and Hobart 1972 and USFWS 1988).

The lack of a burrowing mayfly population in Spring Slough may be related to habitat suitability characteristics versus sediment toxicity as supported by the bioassays discussed earlier. These organisms prefer silty grain size and non-organic sediment texture. Spring Slough contains fine grain and organic sediments which tend to produce anaerobic conditions along the substrate water interface.

Study Wetland Kick Net Samples

The average total number of organisms per kick net sample was similar for study wetland A - polluted (245) and study wetland B - reference (197.2).

The Shannon Diversity Index values were similar between the two sites. The Shannon index for wetland A was 0.7448 and for B was 0.7572.

The Shannon Diversity Index is a measure of species richness and evenness between species. Shannon diversity is commonly reported as $\log_{10}$ (bits per individual). The maximum diversity possible occurs when each individual belongs to a separate species.
The Shannon index is defined as (Shannon 1949):

$$\text{Shannon Index} = -\Sigma(n_i/N)\log(n_i/N),$$

where n_i = number of individuals of species $_i$ and N = total number of individuals for all species.

The Jaccard Coefficient of Community Similarity Index value was moderate between the two wetland study sites. The Jaccard index was 0.56 on a scale from 0 to 1. The dissimilarity was related to relative abundances between taxa common to both sites and numbers of true bug, beetle and aquatic worm taxa.

The Jaccard index is a measure of the degree of similarity in taxonomic composition between sites in terms of taxa absence and presence.
The Jaccard index is defined as (Jaccard 1912):

$$\text{Jaccard Index} = a / (a + b + c),$$

where a = number of taxa common to both sites, b = number of taxa present in site B but not A and c = number of taxa present in site A but not B.

The differences between the two study wetlands in invertebrate relative abundances were likely related to the observed differences in habitat type related to plant community structure (WIDNR 1990).

Table 10. Aquatic invertebrate taxa and frequency for sediment grab samples from Spring Slough, Keithsburg Division refuge.

Class	Order	Family	Common Name	Average Number per Grab (n=21)	Standard Deviation
Nematoda			Round worms	1.43	1.47
Annelida			Segmented worms		
	Oligochaeta		Aquatic worms	252.90	109.25
	Hirudinea		Leeches	0.048	0.22
Crustacea			Crayfish, *etc.*		
	Cladocera		Water fleas	4.57	5.13
	Copepoda		Copepods	22.81	28.66
Insecta			Insects		
	Odonta		Dragonflies		
		Coenagrionidae	Damelsfly	0.048	0.22
	Diptera		Flies		
		Heleidae	Biting Midges	22.09	12.90
		Chironomidae	Midges		
		Subfamily Chironominae		3.90	5.45
		Subfamily Tanypodinae		0.81	0.87
		Subfamily Orthocladinae		0.05	0.22
Mollusca					
	Pelecypoda		Clams and mussels		
		Sphaeriidae	Fingernail clam	0.33	0.73

Table 11. Aquatic invertebrate taxa and average number per kick net grab sample for study wetland A (polluted, n=5) and wetland B (reference site, n=5), Keithsburg Division refuge.

Taxa	Average per sample	
	Wetland A (Polluted)	Wetland B (Reference)
Insecta		
Odonata		
Coenagrionidae	71.8	87.6
Libellulidae/Corduliidae	4.6	19.4
Ephemeroptera		
Baetidae	0.6	5
Caenidae	14	2.2
Hemiptera		
Belastomatidae	0	0.8
Corixidae	0.2	7.4
Coleoptera		
Hydrophilidae (adult)	0	2.8
Hydrophilidae (larvae)	0.6	14.6
Haliplidae (adult)	1	6.8
Scritidae	1.4	0
Diptera		
Dipteran pupae	1.6	4
Heleidae	3	8.6
Chironomidae		
Chironominae	43.4	57.2
Tanyopdinae	13.8	3.8
Stratiomyidae	0.8	0
Chaoboridae	0.6	0
Tipulidae	0	0.2
Crustacea		
Isopoda		
Asellus	0	0.2
Amphipoda		
Hyalella	29.6	23.6
Decapoda		
Palaemonetes	0	0.6
Annelida		
Oligochaeta	9.6	0.2
Hirudinidea		
Glossiphoniidae	0.6	0

Fish

Diversity Inventory

A total of 21 species of fish were collected during the resource management and ice creel surveys (Appendix A and reproduced in Table 12).

Bluegill, common carp, black crappie and largemouth bass were the most numerically abundant species. Common carp and bigmouth buffalo represented the majority of the total weight of fish collected. The scientific names for the fish species are listed in Table 12.

Bluegill production was high in 1995 with a catch of 26.7 kilograms of fish per hectare. Other species of winter sport fishes had catch ratios of less than 1 kilogram per hectare.

Two additional species were collected by the author during the invertebrate surveys that were not caught during the fish surveys: slough darter (*Etheostoma flabellare*) and grass pickerel (*Esox americanus*).

The author also observed very dense populations of gizzard shad (*Dorosoma cepedianum*) later in the summer near road culverts and throughout the backwater complex evident in boat propeller wash.

Health and Growth Rates

There were no signs of abnormal health in the fish collected in 1994 and 1995, except for a bass with eye lesions and several specimens with trauma lesions.

The mean length of fish collected in 1995 were within normal ranges for the respective age classes. The bluegill produced in 1992 were at the low end of the size range for this age class.

There were no differences in mean length for the most abundant fish, the bluegill, collected in 1995 between the north end and the south end of Spring Slough except for the 1992 year class.

The bluegill produced in 1992 and collected in 1995 at the north end of Spring Slough were smaller (161.59 mm) compared to the fish produced at the south end of the refuge (176.38 mm) (t test, $p=0.008$).

There was also a difference in back-calculated lengths of the bluegill produced in 1992 for year 1992 and for year 1993 (Appendix A). Fish produced in 1992 from the north end were smaller for years 1992 and 1993 compared to the south end (t test, 1992: $p=0.047$ and 1993: $p=0.061$).

An explanation for the differences in average fish length for the 1992 year class may be related to the development of a nuisance aquatic plant bloom during the year they were produced.

Keithsburg Division was visited several times in 1992 by the author to finish a previous study. Water levels were low in 1992 and extensive and dense beds of coontail developed throughout the refuge (Coffey 1994).

Fish can become crowded in dense plant cover causing overpredation of food items and lower fish growth (Engel 1990). Heavy vegetation prevents thinning of bluegill numbers by a primary predator the largemouth bass (Trebitz *et al* 1994).

Individual sunfish growth rates may be greatest in lakes with intermediate plant cover and for lakes with abundant open water areas or open water lanes in the vegetation (Crowder and Cooper 1982, Wiley *et al* 1984, Crowder and Cooper 1982 and Johnson and Jenning 1998).

Table 12. Fish taxa and percent of total number collected during the fisheries management studies conducted at Keithsburg Division refuge in 1994 and 1995.

Common Name	Scientific Name	Percent Total Number
Bigmouth Buffalo	*Ictiobus cyprinellus*	4.3
Black Bullhead	*Ictalurus melas*	3.1
Black Crappie	*Pomoxis nigromaculatus*	9.8
Bluegill	*Lepomis macrochirus*	38.0
Bowfin	*Amia calva*	3.3
Brown Bullhead	*Ictalurus nebulosus*	3.1
Common Carp	*Cyprinus carpio*	11.4
Channel Catfish	*Ictalurus punctatus*	0.5
Emerald shiner	*Notropis atherinoides*	1.3
Freshwater Drum	*Aplodinotus grunniens*	2.2
Gizzard shad	*Dorosoma cepedianum*	12.0
Golden shiner	*Notemigonus crysoleucas*	2.7
Largemouth Bass	*Micropterus salmoides*	9.2
River Carpsucker	*Carpiodes carpio*	0.5
Sauger	*Stizostedion canadense*	0.5
Shortnose Gar	*Lepisosteus platostomus*	1.6
Smallmouth Buffalo	*Ictiobus bubalus*	2.7
Walleye	*Stizostedion vitreum*	1.6
Warmouth	*Lepomis gulosus*	1.1
White Bass	*Morone chrysops*	1.1
Yellow Bullhead	*Ictalurus natalis*	0.5

Insecticide Exposure

A total of 221 adult bluegill were collected and analyzed in 1995 and 1996. A combined summary of mean body weights, brain weights and cholinesterase activity is reported in Table 13.

There was no evidence of brain cholinesterase (ChE) inhibition in the sampling date group means for the Keithsburg Division refuge fish compared to reference site samples or literature values (Appendix A). None of the samples had greater than a twenty percent cholinesterase inhibition. Twenty percent inhibition is commonly accepted as evidence of exposure to a ChE inhibitor (Ludke *et al* 1975).

Cholinesterase activity was significantly different between collection dates (MANOVA, $p=0.0001$). Mean cholinesterase activities were higher on later collection dates. Increases in mean ChE activities were probably due to differences in fish size (Beauvais 1997; Zinkl *et al* 1987).

Lake water samples were never tested for insecticide chemicals. The potential for exposure to insecticide chemicals was inferred based on the proximity and lack of buffer zone to cropfields being treated for insect pests. It is possible that no system wide insecticide contamination by ChE inhibitors occurred during the study time frame because chemicals were not widely used that year or the lack of off site migration.

Heavy Metal Concentrations

There were differences in the concentrations of the heavy metals copper and mercury detected in the whole fish from Keithsburg Division compared to the reference site at Big Timber Division. The other metals (arsenic, cadmium, chromium, lead, nickel and selenium) were not detected in any of the fish specimens and zinc was detected in all fish. The fish tissue chemistry data are in Appendix E.

Seven out of the ten common carp collected at Keithsburg Division had trace concentrations of copper between 1.1 to 1.5 mg/Kg wet weight. Copper was not detected in the common carp collected at Big Timber Division.

Five out of the five large-mouth bass collected at Keithsburg Division had trace concentrations of mercury between 0.11 to 0.17 mg/Kg wet weight. Mercury was detected at 0.20 mg/Kg wet weight in one out of the five bass collected at Cooligar Slough in Big Timber Division.

There were no statistically significant difference in zinc concentrations between the common carp collected at Keithsburg Division compared to Big Timber Division (Mann-Whitney Rank Sum Test, $p=0.358$). There was no statistically significant difference in zinc concentrations between the large-mouth bass collected at Keithsburg Division compared to Big Timber Division (t test, $p=0.887$).

The mean concentrations (mg/Kg, wet weight) of the heavy metals copper (1.24), mercury (0.23) and zinc (38.3) measured in carp and bass collected at Keithsburg Division were above the geometric means developed for the national contaminant biomonitoring program (Schmitt and Brumbaugh 1990). The national contaminant biomonitoring program mean for copper was 0.65 mg/Kg wet weight, mercury was 0.10 mg/Kg wet weight and zinc was 21.7 mg/Kg wet weight (Schmitt and Brumbaugh 1990). The national biomonitoring program analyzed bottom dwelling species like the common carp and a predator like the bass.

Trace concentrations of mercury were also measured in eight of the ten bluegill collected from Keithsburg

Division. No bluegill were tested for mercury from the reference sites. The bluegill mercury data were used to estimate food chain pathway risks to fish eating birds and mammals.

The mean concentration for these eight bluegill was 0.063 and the maximum concentration was 0.125.

The mean concentration of mercury in the bluegill was below the risk level for the protection of fish eating birds. For the protection of sensitive birds, total mercury concentrations in prey items should probably not exceed 0.057 mg/Kg wet weight (USEPA 1997).

Five out of the ten fish tested for mercury at Keithsburg Division were at or just above the criteria to protect wildlife and only one fish in the test group was above 0.06 mg/Kg wet weight.

The fish chemistry data indicated that the heavy metals copper and mercury were bioavailable to biota at Keithsburg Division as compared to Big Timber Division. The source for the heavy metals, copper and mercury, may be from historic use of agricultural seed fungicide chemicals that contained the contaminants.

Prior to the 1970's, copper and mercurial compounds were used in corn seed fungicides and likely applied to cropfields adjacent to the refuge unit.

The absence of copper and mercury in the fish from Big Timber Division may be explained by the fact that this backwater has been isolated by a levee from local surface run-off of agricultural chemicals used on adjacent floodplain cropfields.

A similar explanation is given for decreased copper and mercury levels in fish collected over years from agricultural lands in California San Joaquin Valley (Saiki and May 1988).

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The sources for zinc are not known. Generally, zinc is toxic at higher concentrations than copper and mercury (Rand and Petrocelli 1985). Previous studies indicated that zinc concentrations in aquatic sediments at Keithsburg Division and Big Timber Division were elevated above geological background levels (see Table 1).

Birds

Waterfowl Use Inventory

There were differences in total numbers and types of waterfowl that were observed using study wetland A versus study wetland B (χ^2 test, $p < 0.001$).

A total of 138 dabbling ducks were observed using Wetland B and 59 on Wetland A during the 1995 spring migration survey time frame. A total of 325 diving ducks were observed using wetland B during the 1995 spring migration survey time frame and none were observed on wetland A (Table 14).

The differences in waterfowl use was likely related to the differences that were observed and described in this report in the plant and invertebrate communities between the two study wetland sites.

Wetland B had a relatively balanced aquatic macrophyte community, a diverse aquatic invertebrate assemblage and had greater waterfowl use compared to study wetland A.

Aquatic macrophytes and invertebrates are important food resources for waterfowl (ILNHS 1959). Wetlands with high production of aquatic invertebrates tend to attract waterfowl (CWS 1987; ILNHS 1966 and Payne 1992) .

Table 13. Combined summary statistics for bluegill cholinesterase tests, Keithsburg Division refuge and Wildcat Den Hollow State Park, Henry County, Illinois, 1995 and 1996 (Appendix A).

Mean Value	1995 Data (n=131)	1996 Data (n=90)
Length (millimeters)	161	147
Weight (grams)	97.06	70.2
Brain Weight (grams)	0.1017	0.0925
Cholinesterase Activity[1]	9.73	11.38

[1] Reported as micromoles acetylthiocholine hydrolyzed per minute per gram of brain tissue.

Table 14. Species and total numbers of aquatic birds observed during weekly visits using study wetland sites A and B at Keithsburg Division during the spring of 1995.

Study Wetland	Mallard (*Anas platyhynchos*)	Northern Shoveler (*A. clypeata*)	Blue-winged Teal (*A. biscors*)	American Wigeon (*A. americana*)	Gadwall (*A. strepera*)	Wood Duck (*Aix sponsa*)
A "Polluted"	19	15	2	0	0	10
B "Reference"	85	13	13	4	2	10

	Lesser Scaup (*Aythya affubus*)	Ring-necked Duck (*A. colaris*)	Redhead (*A. americana*)
A "Polluted"	0	0	0
B "Reference"	86	94	2

	Pied-billed Grebe (*Podilymbus podiceps*)	American Coot (*Fulica americana*)	Great-blue Heron (*Ardea herodias*)
A "Polluted"	1	34	2
B "Reference"		17	0

Wren Biomonitoring

All of the bird nest boxes were set up by early April. By June 13, none of the house wren eggs at Keithsburg Division had hatched and half of the active nest boxes at Big Timber Division had a full clutch of freshly hatched nestlings. This was a relatively late start at Keithsburg Division for the house wrens and thus missed insecticide use periods.

Six nestlings of various ages from each of the two sites were tested for brain ChE activity. The ChE activity for all of the nestlings collected were within normal ranges for immature birds (Custer and Ohlendorf 1989; Grue and Hunter 1984). Standard morphometric measurements and ChE activity for each nestling tested are outlined in Table 15.

Brain ChE activity generally increased with nestling size for the nestlings collected from the reference site at Big Timber Division (Figure 12). Wingcord had good correlation with brain ChE activity (r=0.97). Tarsus length and bill length had weak correlation to brain ChE activity (r=0.37 and r=0.59, respectively).

There was poor correlation between the bird size data and brain ChE activity in the nestlings from Keithsburg Division; r values were from 0.31 to -0.21.

Brain ChE activity data from other studies indicated an increase with age in other altricial bird species (Custer and Ohlendorf 1989; Grue and Hunter 1984). It is possible that the Keithsburg Division nestlings collected were selectively exposed to other ChE inhibitor chemicals other than farm insecticides or more variation is present in younger birds.

The period that the wren nestlings were present in the nest boxes did not overlap the general use times for rootworm insecticide chemicals (see Table 2).

Ecological risk from exposure to rootworm insecticide chemicals was therefore believed to be low for the 1995 season. Birds may be at different levels of insecticide exposure risk from year to year depending on insect pest cycles, method of pest control and post control weather conditions.

Table 15. Morphometric measurement and brain cholinesterase activity data for nestling house wrens collected at Keithsburg Division refuge and Big Timber Division refuge, 1996.

Nest Number and Chick Reference Number	Body Weight (grams)	Wing Cord (millimeter)	Tarsus Length (millimeter)	Bill Length (millimeter)	Cholinesterase Activity[1]
Keithsburg - 1a	4.0	13.1	13.3	4.97	8.3
Keithsburg - 1b	4.5	14.07	14.35	6.06	15.9
Keithsburg - 6a	6.0	18.39	15.75	5.88	10.4
Keithsburg - 6b	6.0	17.01	15.63	6.16	10.1
Keithsburg - 12a	7.75	23.43	17.83	6.93	10.4
Keithsburg - 12b	6.5	20.86	16.33	5.87	10.3
Big Timber - 2a	-	39.01	20.29	7.75	18.9
Big Timber - 2b	-	37.24	19.63	8.23	18.9
Big Timber - 11a	-	27.02	18.22	6.61	9.5
Big Timber - 11b	-	28.77	19.95	8.02	10.2
Big Timber - 15a	-	27.84	19.8	7.26	11.6
Big Timber - 15b	-	28.44	18.77	6.24	9.8

[1] Reported as micromoles acetylthiocholine hydrolyzed per minute per gram of brain tissue.

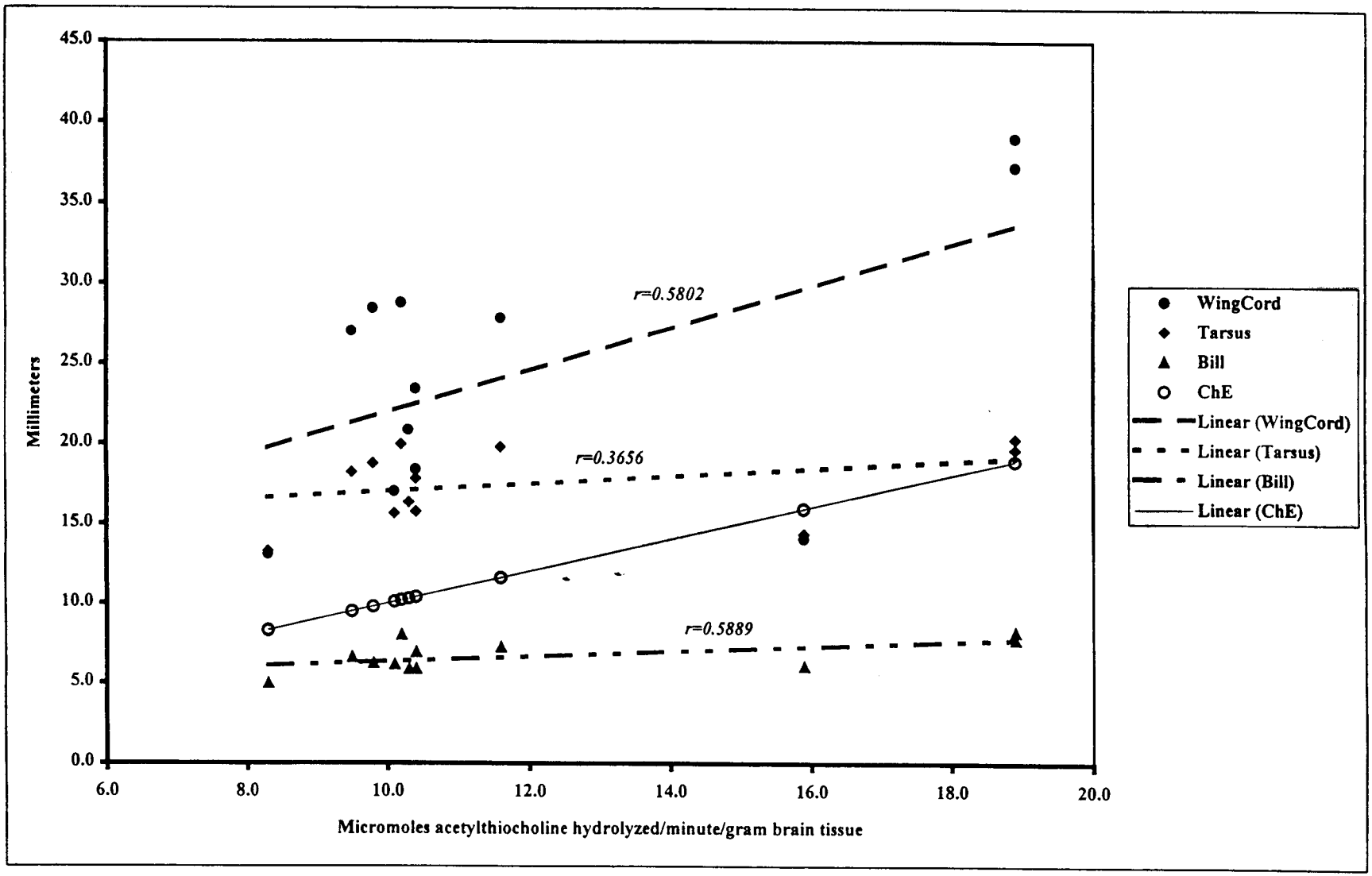

Figure 12. Relationships between brain cholinesterase activity and nestling house wren size parameters for nestling house wrens collected at Keithsburg Division refuge and Big Timber Division refuge.

Quality Control

The water quality instruments used for this study were purchased new each year and no anomalies were observed during field use and testing with standards. We replaced the pH probe and dissolved oxygen sensor each field season.

Information on quality assurance performance tests for the ChE methods are contained in the respective technical reports from the National Wildlife Health Center and Iowa State University Department of Animal Ecology (Appendix A). No anomalies were reported by the ChE method contract laboratories.

There were a few instances where the contract laboratory's water chemistry data differed between field split samples (Table 16). The differences in the duplicate water chemistry data are likely related to inherent differences in accuracy and precision for the ELISA analytical method.

The differences between the field spilt samples analyzed by two different methods (ELISA and GC) may be explained by differences in accuracy of the GC method compared to the ELISA method.

No other anomalies were reported by the water chemistry contract laboratory.

The sediment contaminant chemistry agreed between the field spilt samples (Table 17). The slight differences observed between duplicate sample metal concentrations are likely related to the heterogenous distribution of contaminants in grab type samples.

No other anomalies were reported by the sediment chemistry contract laboratory.

Several trip blank and equipment rinse blank samples were analyzed during the sediment quality surveys. Contaminants were not detected in the trip blank and rinse blank samples except for copper in two of the three rinse blank samples. Copper was detected at 0.03 and 0.01 mg/L. The source of the copper is believed to be the sediment sampling device (Ekman dredge). The Ekman dredge contains copper alloys.

No anomalies were reported by the fish tissue chemistry contract laboratories.

Table 16. Field duplicate water chemistry data for samples submitted to the contract laboratory for the Keithsburg Division contaminants study. All concentrations are in milligrams per liter.

	Duplicates	
Analyte	Split 1	Split 2
Ammonia	6.2	6.2
Atrazine	0.32	0.30
Alachlor	1.09	0.89

	Methods	
	ELISA[1]	GC[2]
Atrazine	0.12	ND
Atrazine	0.19	0.16
Alachlor	0.29	ND
Metolachlor	0.37	0.1

[1] Enzyme-Linked Immunosorbent Assay

[2] Gas Chromatography

Table 17. Field duplicate sediment chemistry data for samples submitted to the contract laboratory for the Keithsburg Division contaminants study. All concentrations are in milligrams per kilogram, dry weight, unless otherwise noted.

Analyte	Split 1	Split 2
Total ammonia	160	160
Total phosphate	1700	1600
Total organic carbon	6.4%	7.1%
Clay content	25.5%	21.6%
Arsenic	7.2	5.6
Chromium	18	17
Copper	14	14
Lead	14	17
Nickel	17	17
Zinc	67	67
Total Petroleum Hydrocarbons	130	130

CONCLUSIONS

Between 1994 and 1996, biologists from the U.S. Fish and Wildlife Service monitored water quality and surveyed biological diversity at a backwater system of the Upper Mississippi River. The backwater system that was studied is a management unit of the Mark Twain National Wildlife Refuge known as Keithsburg Division part of the Wapello District.

Water quality conditions at Keithsburg Division do not limit the site specific management objectives for the refuge unit. The specific management objectives for the refuge unit are to: 1) provide waterfowl with food, water and protection during migration; and 2) improve and maintain existing habitat to perpetuate an optimum annual production of wood ducks.

Water quality problems at Keithsburg Division limit the production of food for waterfowl. Many refuge wetlands now function to treat pollution versus the functions of providing wildlife habitat and food resources. The shift in wetland functions from "wildlife habitat" to "chemical treatment" appears to be the result of nutrient enrichment.

The nitrate and phosphate loading from upland run-off and groundwater coupled with natural floodplain nutrient cycling are sufficient to cause major changes to the structure and composition of refuge plant communities.

The high nutrient loads cause nuisance aquatic plant blooms. The nuisance plant populations do not produce seeds preferred by waterfowl and do not provide substrate for invertebrate production. These altered plant communities covered a large extent of the backwater system at certain times of the year.

The production of desirable aquatic invertebrate species in an estimated area of over half of the refuge unit does not achieve its potential every year. We believe that the standing crop of preferred aquatic invertebrate food resources would be greater with better water quality conditions.

Improved water quality conditions may promote the development of permanent and balanced aquatic plant communities throughout the refuge. These plant communities could generate high quality seeds and substantial invertebrate biomass on an annual basis to the benefit of waterfowl and migratory bird productivity.

The primary water sources for Keithsburg backwater, upland run-off and groundwater, are polluted with nutrients and agricultural herbicide chemicals. Water quality in the backwater system can be improved by targeting these primary water sources. The rivers bordering the refuge unit were not typically connected and do not exchange water except at Pope Creek through the railroad grade levee break.

Agricultural herbicide chemicals were detected above levels of concern. Although, herbicide chemical concentrations measured in refuge

surface waters did not reach levels that would kill aquatic plants and animals, they did reach levels that could harm aquatic plants.

The aquatic plant community at Keithsburg Division is dominated by monotypic blooms of pest species during the study years. The pest species included blue-green algae, duckweeds and coontail. The plant populations were apparently not affected by exposure to the herbicides in the water and sediment. This may be because these aquatic plants quickly recolonize after short term and critical herbicide exposure periods in high nutrient waters (Lozano and Pratt 1993).

However, our concern is that repeated annual herbicide exposure that may have the effect of culling sensitive species from plant communities over time and reducing refuge biodiversity.

The concentrations of herbicides in refuge surface waters may have been influenced in some undetermined manner by flood events. The years that the backwater was studied (1994 to 1996) included significant floods and the river floodplain was completely underwater during the spring and early summer each year.

The upland water sources and groundwater that discharged into refuge wetlands were contaminated with nitrate and phosphates. Nutrient enrichment in refuge sloughs and wetlands from these sources caused hypereutrophic conditions at times.

The hypereutrophic conditions were noted by very dense algae and duckweed blooms, low to supersaturated dissolved oxygen concentrations and elevated sediment ammonia concentrations. Dense growth of phytoplankton and duckweed limited water transparency and appeared to inhibit the production of aquatic macrophytes in many sloughs. Aquatic macrophytes are needed to provide substrate for a variety of invertebrate species (Miller *et al* 1989).

The benthic macroinvertebrate community was poorly represented in the sloughs. The benthic macroinvertebrate community was dominated by high numbers of a few pollution tolerant species.

Sediment toxicity test results indicated that the ammonia contamination was not severe enough to kill a common backwater invertebrate, the burrowing mayfly nymph. Therefore, we suspect that poor oxygen conditions and lack of plant stems coupled with chemical stress limited high quality benthic macroinvertebrate production.

It is interesting to note that pelagic fish and sunfish species were very abundant in the sloughs at times. Immature pelagic fish and sunfish species likely benefited from the high plankton production which is an important food source for these immature fishes.

Large numbers of fish eating birds such as the double-crested cormorant (*Phalacrocorax auritus*) and white pelican (*Pelecanus erythrorhynchos*) were attracted to the site especially during migration times probably related to the abundant food source.

Backwater wetlands that were isolated from nitrate sources contained balanced plant communities and produced a diverse invertebrate community. Large numbers of diving ducks were attracted to these wetlands.

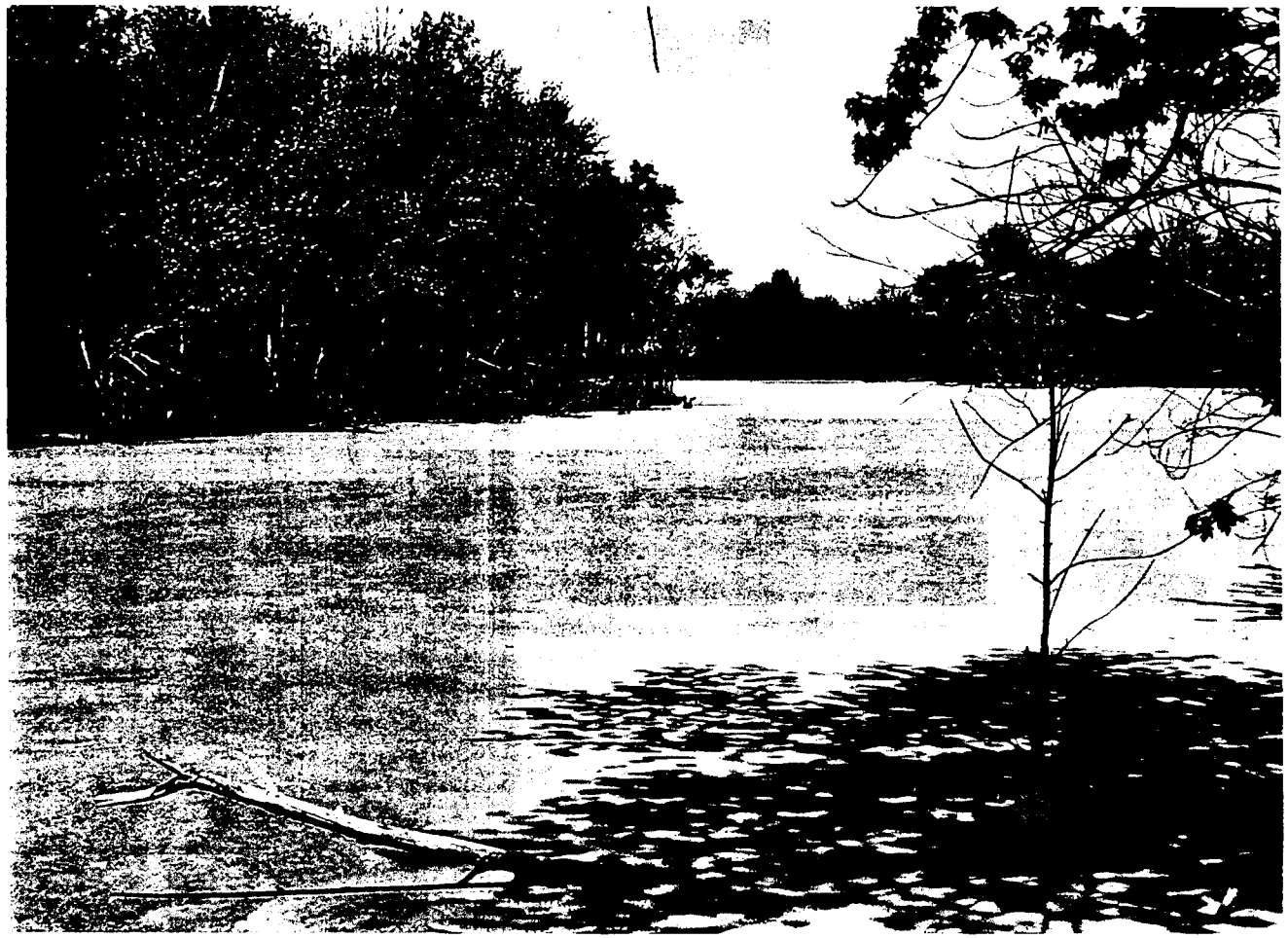

Figure 13. Photograph of nuisance aquatic plant bloom (Duckweed species) at the north end of Keithsburg Division refuge, 1995.

RECOMMENDATIONS

Eutrophication from nutrient enrichment is the leading problem facing Illinois water resources (IEPA 1998). The U.S. Environmental Protection Agency is developing enforceable water quality standards for nutrients under the Clean Water Act. Refer to Internet address "http://www.epa.gov:80/ostwater/Rules/nutstra3.pdf" for more information on the proposed national criteria.

The water quality at the Keithsburg Division refuge may recover over time by implementing standard lake management strategies to improve conditions and watershed management strategies to affect the nutrient sources.

Lake Management Strategies

There are three lake management strategies that may be used to guide hypereutrophic shallow water bodies toward desired ecological states (WIDNR 1995).

1. **Drawdowns** kill undesirable fish, stimulate wetland forb production and solidify loose sediments. A drawdown of surface water will transport dissolved nutrients out of the backwater system.

2. **Biomanipulation** can be used to control nuisance plant populations with cutting and herbicide application operations. Stocking of predator fish can regulate nuisance fish species.

3. **Water level control** to match natural fluctuations may be used to regulate inputs of surface water from less desirable sources.

Watershed Management Strategies

There are three general watershed management strategies that may be used to control nutrient sources. The three strategies are outlined below and described for consideration in future management plans.

1. **Reduce** the nutrient source by improving land management practices.

2. **Dilute** the nutrient source by increasing the flushing rate of the system with clean water.

3. **Treat** the nutrient polluted water sources before this water enters the system.

Reduce the Source

Nutrient loads to the backwater system may be decreased by reducing the amount of fertilizer chemicals applied to and transported from the cropfields adjacent to the Keithsburg Division refuge. This could reduce the amount of phosphates and nitrogen compounds available to plant resources in refuge surface waters.

There are four approaches to reduce the source of nutrients:

1. Acquiring the lands adjacent to the refuge and restore prior converted wetlands.

2. Institute conservation easements for lands adjacent to the refuge and regulate the use of fertilizer chemicals.

3. Stimulate local environmental quality incentive programs (EQIP) and other nutrient management programs administered by the National Resource Conservation Service (NRCS). For more information on EQIP and other related programs refer to the Internet URL address "http://www.nrcs.usda.gov/NRCSProg.html".

4. Develop conservation buffers between refuge wetlands and adjacent cropfields. Buffer strips may be designed to mitigate the transportation of nutrients from cropfields to surface water and groundwater resources.

Internal phosphorus cycling between the water column and sediments may or may not sustain highly productive conditions for some time in the hypereutrophic sloughs. Internal phosphorus loads may be stored in the sediments and rendered unavailable for recycling in mesotrophic waters (McCabe *et al* 1982). After several years of reduced phosphate inputs the system may be guided to a less productive condition.

Dilute the Source

A strategy to dilute nutrient loads in the surface water resources at Keithsburg Division is to mix in additional sources of clean water from adjacent rivers or groundwater.

There is not an accessible source of clean water for the use of increasing flushing rates in the Keithsburg Division backwater system. The Mississippi River, Edwards River and Pope Creek are polluted with nitrogen and phosphate compounds and have high sediment loads. The water sources of drainage ditches, upland run-off and shallow groundwater are moderately to highly polluted with nitrate.

Treat the Source

A strategy to treat nutrient rich water is by routing the polluted water through artificial chemical treatment wetlands. Several artificial chemical treatment wetlands placed higher in the watershed and closer to the specific nutrient sources may be more effective than one larger treatment wetland lower in the watershed as is the case for the Keithsburg Division watershed at this time (De Laney 1995).

Wetlands can be very efficient in treating or assimilating and converting nitrate to less ecologically harmful chemicals such as atmospheric nitrogen gas (Figure 14). The dominant mechanism for nitrate conversion in a wetland is denitrification (Figure 14). Bacterial denitrification accounted for about 80 percent and plant assimilation accounted for about 14 percent of the nitrate removed from a test treatment wetland (Crumptom *et al* 1993).

Chemical treatment wetlands can trap sediments and filter phosphate compounds. Sediment loading may fill an artificial wetland over time and thus require maintenance. An artificial wetland may become saturated with phosphate compounds and the efficiency to filter phosphate may decrease over time.

The function of chemical treatment often reduces the value of a artificial wetland for fish and wildlife habitat which is a critical trade off. A wetland may respond to nutrient loading by adverse changes in water quality and development of nuisance plants.

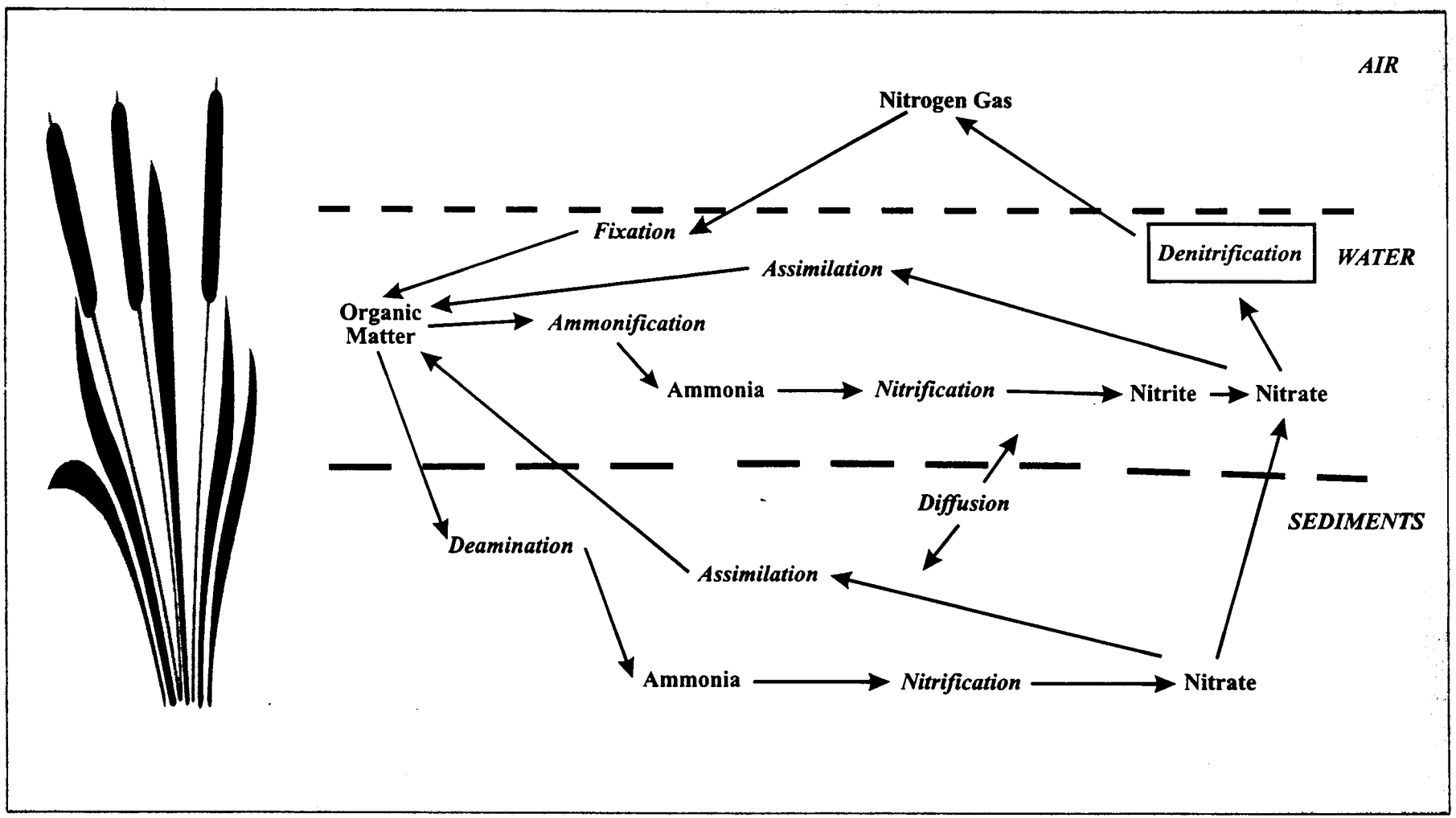

Figure 14. Generalized nitrogen cycle in wetlands.

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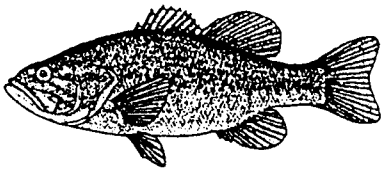

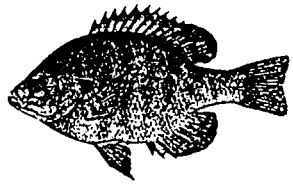

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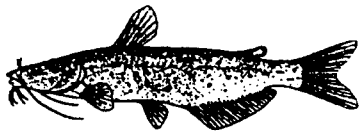

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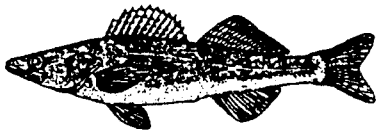

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U.S. Fish and Wildlife Service
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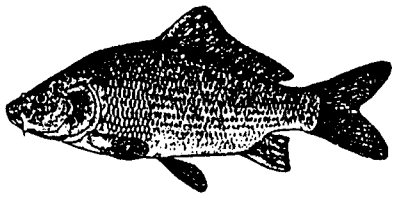
March, 1996



INTRODUCTION

Fishery resource surveys are conducted annually at the Keithsburg Division, Mark Twain National Wildlife Refuge by the U.S. Fish and Wildlife Service's Fisheries Resources Office, Columbia, Missouri and Illinois Department of Natural Resources fisheries personnel. The following report describes the results of the 1995 survey, compares the results to those of previous years, and discusses potential fishery management alternatives. In addition to the annual survey, conducted in June 1995, additional fish were collected in August 1995 as part of a contaminants survey being conducted by the Rock Island, Illinois Ecological Services office. Fish collected in August were sampled in two distinct locations, South End and Spring Slough (Appendix 1).

METHODS

The inshore fish community of several areas in Keithsburg Lake was sampled using boom mounted electrofishing boats. Pulsed DC current (707 Volts, 6.5 amps, 60 pulses per second) was used by USFWS staff on June 26-27, 1995 and August 29, 1995. Three-phase AC current was used by IDNR staff on August 29, 1995. All fish species were weighed (g), and measured (mm). Scale samples were removed from some bluegill collected in the South End in June to assist in year-class determination. Saggital otoliths were removed from approximately forty bluegill in each location in August to aid in determining if there was a difference in growth rates between the two sites.

Two gill nets and one trammel net were set on June 27, 1995. The nets fished throughout the night and were pulled the following morning. The gill nets were 100 feet long with alternating 25 foot panels of 1 and 2 inch mesh. The trammel net was 300 feet long and 6 feet deep with a 16 inch outer mesh and a 3 inch inner mesh.

RESULTS

A total of 208 fish of eighteen species weighing 71.3 kg (157.3 lbs) were collected by electrofishing in Keithsburg Lake in June 1995 (Table 1). Bluegill, common carp, black crappie, and largemouth bass were the most numerically abundant species making up respectively 38.0, 11.4, 9.8, and 9.2 percent of the population. Common carp and bigmouth buffalo represented 50.9 percent of the total weight of fish collected.

Sixty-four fish of ten species weighing 81.6 kg (179.9 lbs) were collected by gill nets and trammel nets (Table 2). Common carp, bigmouth buffalo, and bowfin were both the most numerically abundant species (respectively 46.9, 18.8, and 10.9 percent of the population) and represented the majority of the total weight of fish collected (42.9, 34.0, and 13.8 percent of the total weight, respectively).

It should be noted in the following discussion that data previous to 1995 includes fish collected by Illinois Department of Natural Resources staff with three-phase AC electrofishing. Data from June 1995 does not include their effort as personnel were unavailable at that time. Their effort is included in the August sample discussion in Appendix A.

Largemouth Bass

Fewer largemouth bass were present in Keithsburg Lake in 1995 than in 1994 as evidenced by both the decreased relative abundance (percent of sample) and catch per unit effort (number of fish per hour) (Figures 1 and 2). The catch rate (CPUE) of largemouth bass totaled 11.3 fish per hour of electrofishing. This is the lowest CPUE for largemouth bass since 1991.

Relative Weight (Wr) is a measure of body condition. The measured weight of a fish is compared to an established standard weight of a fish the same length. Wr values greater than 100 indicate the individual fish weighs more than the standard weight. The ideal range of Wr values is 90-110. Fish populations with Wr values close to 100 are in balance with their food supply. Fish with Wr values less than 90 are underweight, while fish with Wr values greater than 110 are overweight. Either of these extremes indicate predator:prey ratios are not balanced. The bass sampled were in good condition, with an average Wr of 95.1 (Figure 3).

Proportional Stock Density (PSD) is an index of the size structure of a population. It also represents the percentage of fish that are attractive to an angler. The larger the PSD percentage, the greater the number of large fish. The desirable range for bass is 40-60%. PSD values larger than 60% would indicate a larger proportion of largemouth bass are 12 inches (quality size) or larger than would be desirable for maintaining balance. PSD of this sample was 57.1% (Figure 2). This is within the desirable range. Relative Stock Density (RSD) is a measure of the size structure of fish > 15 inches. Desirable ranges are 20-30%. The RSD of this sample is 14.3% This is below the desirable range and indicates a deficiency of fish larger than 15 inches (380 mm) (Figure 3). No largemouth bass greater than 400 mm (15.7 inches) were collected. This may indicate either limited recruitment of overharvest of large fish.

A low PSD in 1994 (15.5%) indicated large numbers of fish 8-12 inches. The higher PSD, accompanied by a low RSD in the 1995 sample, indicates largemouth bass have grown into the 12-15 inch range.

Bluegill

The relative abundance of bluegill increased from 27.5% in 1994 to 38% in 1994 (Figure 1). Bluegill have been increasing in relative abundance, from a low of 1% in 1990 to 38% in 1995.

CPUE increased between 1990 and 1992, but declined from 72.0 fish/hour in 1992 to 47.3 fish/hour in 1995 (Figure 4). PSDs for bluegill should range between 20% - 40%, with stock size = 76.2 mm (3 in.) and quality or harvestable size = 152 mm (6 in.). PSD for bluegill in 1995 was very high at 65.7 (Figure 4). This indicates a large proportion of fish in the 6-8 inch (152-203 mm) size range. A large year class in 1990 has grown beyond minimum harvestable size to greatly increase the number of fish > 152 mm (Figures 4 & 5). This large year class should increase angler catch rates. Forty-six percent of the bluegill sampled were of harvestable size (greater than 6 inches). Harvestable size is the length at which anglers will generally decide a fish is worth keeping. There is no RSD value for this population as no bluegill of preferred (8 inches), memorable (10 inches), or trophy size (12 inches) were collected (Figure 5). The condition of individual bluegill is good with an average Wr of 107.6, well within the 90-110% range (Figure 5). The small numbers of bluegill between 50 mm - 100 mm may be cause for concern. These fish represent the future catchable population. The smaller bluegills may be under represented in our sample. Although another year's data may be needed to clarify any potential problems, this was also seen in the 1994 data and may indicate a reproduction problem.

Average relative weight (Wr) for bluegill in 1995 was good at 107.6% indicating good growth. Stunting in bluegills is a common problem in Midwestern impoundments. Stunting generally results from reduced predator communities and overcrowding which leads to slow growth. Keithsburg has a good predator community and bluegills exhibit good growth at all sizes indicating stunting is not a problem at this time (Figure 5).

Black Crappie

Although relative abundance of black crappie increased from 1.5% in 1994 to 9.8% in 1995, CPUE dropped from 67 fish/hour to 12 fish/hour (Figures 1 and 6). The majority of crappie collected in 1994 were from the 1990 year-class. These fish were greater than 203 mm in length in 1994. Most black crappie collected in 1995 were less than 210 mm, indicating the majority of the 1990 year-class have left the system, probably due to angling and natural mortality (Figure 7). PSD for crappie should range between 20-40% with stock size = 127 mm (5 inches) and quality or harvestable size = 203 mm (8 inches). The PSD of this sample was 37.5%, well within the desirable range. The crappie sampled were in good condition with an average Wr of 101.1. This is significantly higher than the Wr value of 86 found in the 1994 sample. The large 1990 year-class had been competing for food, thus exhibiting reduced growth rates. Crappie fishing will decline for a few years until a new strong year-class cycles through.

Common Carp and Bigmouth Buffalo

Commercially harvestable populations of carp and buffalo are present. Encouraging commercial harvest would capitalize on this underutilized resource.

Relative abundance of carp has decreased from the levels seen in the 1994 and 1992 samples (Figure 1). Although most carp sampled were of harvestable size (greater than 12 inches), they were in less than ideal condition with an average Wr of 85.3% in electrofishing samples and 87.0% in gill net and trammel net samples (Figure 8).

The relative abundance (4.3%) of bigmouth buffalo caught by electrofishing was the same as that of 1994 but dropped from 17.6% in 1992 (Figure 1). Bigmouth buffalo were in better condition than carp with an average Wr of 105.1 in electrofishing samples and 109.4 in gill net and trammel net samples (Figure 9).

Other Fish Species

Although electrofishing is not very selective for catfishes, one channel catfish was collected. It exceeded the harvestable size of 10 inches (250 mm). The catfish was in less than ideal condition with a Wr of 88.3.

Only two walleye were collected by electrofishing. Both were of harvestable size (15.5 inches), but in less than ideal condition with an average Wr of 83.4. One walleye collected in a gill net was also in poor condition with a Wr of 84.5.

Two species which have previously been present at Keithsburg, but were not well represented in the 1994 and 1995 samples are northern pike and golden shiner. Gill nets and trammel nets caught 5 pike in 1991 and 3 in 1992. Neither the 1994 or 1995 surveys captured northern pike. Northern pike are present in low densities and may have simply avoided capture. Golden shiner at one time was very abundant comprising 28% of all fish collected in 1991. The 1994 sample did not collect any golden shiner. Four golden shiner were collected by electrofishing in 1995. Post levee repair conditions at Keithsburg must have favored golden shiner and as the conditions changed and predator numbers increased, the population declined.

Trends

The levee breach in the flood of 1986 and subsequent post flood conditions have had a large impact on fish populations at Keithsburg. Following the levee break in 1986, a drought in 1987 lowered Mississippi river levels allowing most of the water to be drawn off at Keithsburg. Water levels remained low until levee repairs were completed in 1989. Gamefish did not survive this period of low water, however, roughfish did. A 1990 fish survey found high populations of goldfish, carp, buffalo, and golden shiner. These species comprised 87% of all fish collected.

When the water level returned to normal in 1990 several species produced large year classes. These species included; carp, buffalo, bluegill, and black crappie. These

young of year fish had little competition for resources and grew well. In 1994 these large cohorts made up most of the harvestable size fish for these species as can be seen with the dominant black crappie year class in Figure 7. This "boom" fishery will be short lived as natural mortality and angling pressure reduce this year class of fish.

Conclusions

Gamefish populations are doing well. Largemouth bass appear to be increasing, and a few bass > 15 inches were captured in our sample. A protected slot limit should be considered to increase numbers of larger bass. An improved largemouth bass fishery may affect bluegill and crappie fisheries. Bluegill and crappie populations have been doing well. The large 1990 year class has increased harvestable populations of bluegill and crappie. Fishing for bluegill should be excellent for the next year or two. As this large year class is removed by angling and natural mortality, fishing success will decline. Black crappie fishing may already be declining. The break in the railroad levee at the south end has allowed walleye, sauger, and catfish to utilize the Keithsburg area. As long as Keithsburg is allowed to remain connected to the Mississippi River, fishing for these species will improve.

The flood of 1993 has had a positive impact on the fishery at Keithsburg. Bass and panfish populations appear to be increasing. Riverine species such as walleye, sauger, white bass, flathead and channel catfish have been added to the gamefish community. The connection to the Mississippi River will provide access to spawning and nursery habitat for riverine fish during high flow events and contributes to the process of reestablishing vital links between the river and its floodplain.

Recommendations

1) Consider a 12 - 15 inch protected slot limit on largemouth bass to increase the number of bass >15 inches.

2) Solicit and encourage commercial fishing for carp and buffalo. Allow use of 3 inch or larger mesh trammel nets only. Close netting during waterfowl migration.

3) If the damaged railroad levee on the south end of Keithsburg is to be repaired, consider constructing a high flow spillway or notch(es) to retain connectivity to the Mississippi River at elevations consistent with the existing conditions.

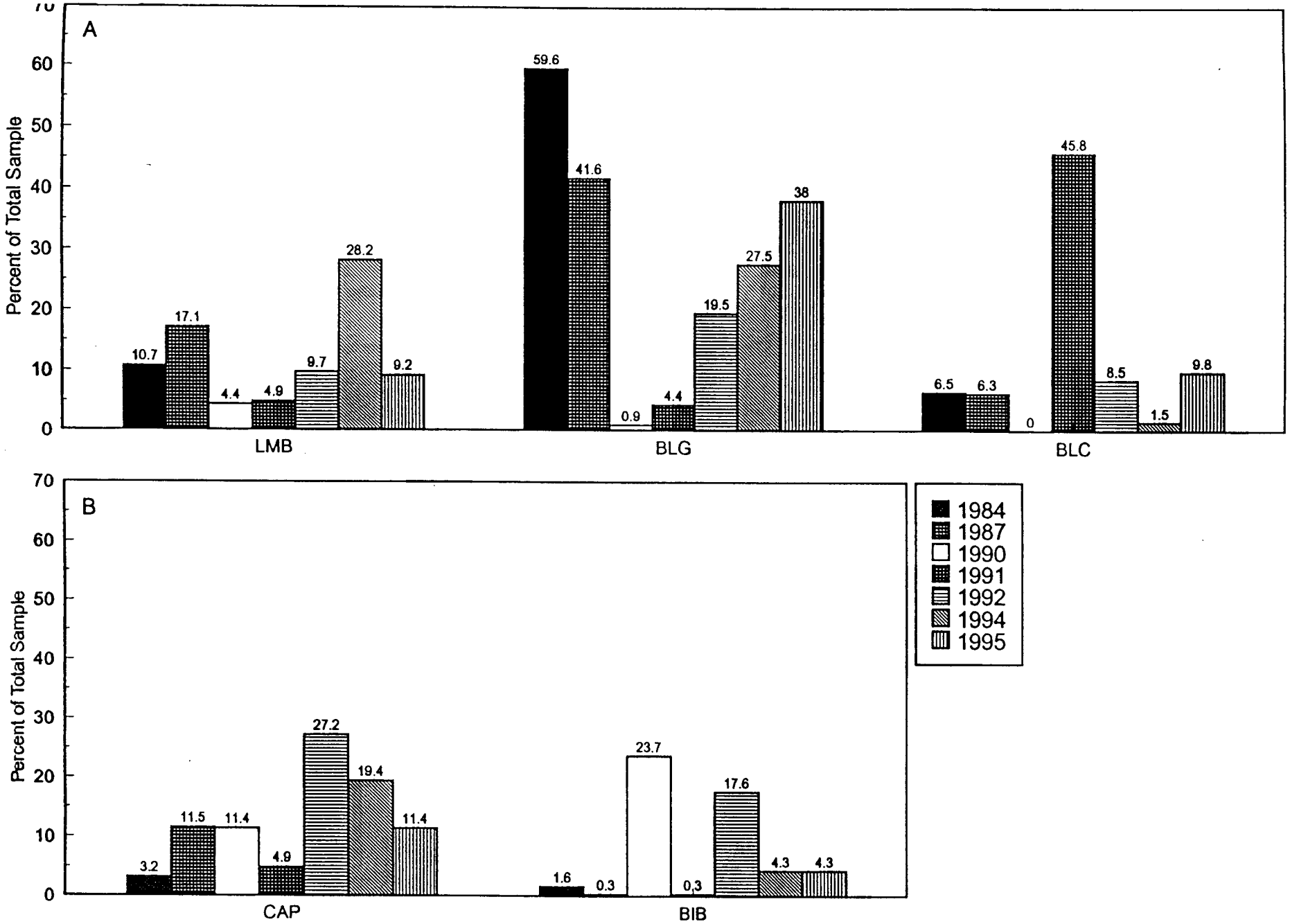


Figure 1. Relative abundance as a percentage of total fish sampled for (a) selected sportfish species and (b) selected rough fish species collected in Keithsburg District, 1984-1995.

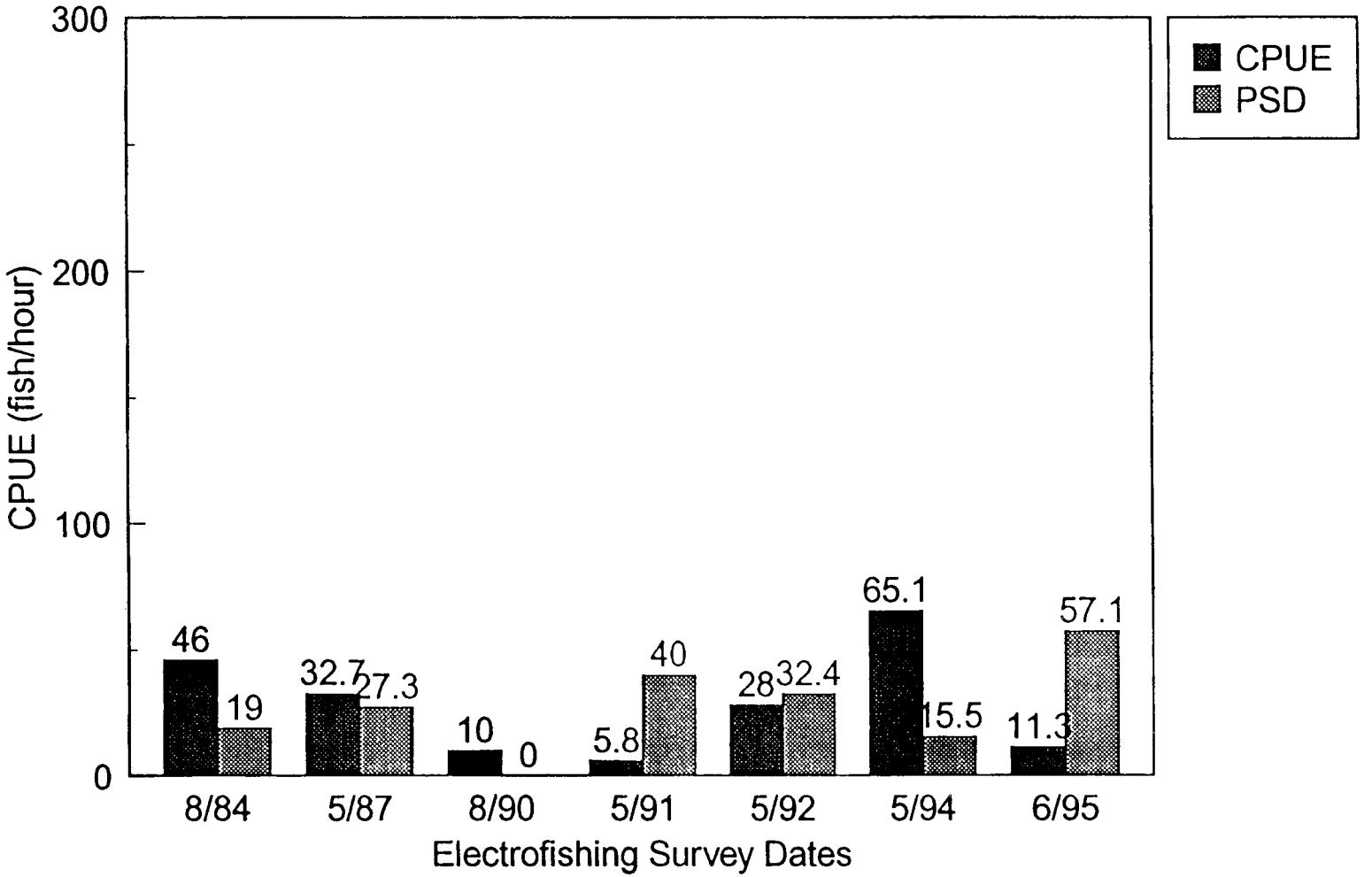


Figure 2. Catch per unit effort and PSDs of largemouth bass collected by electrofishing at Keithsburg District, Mark Twain National Wildlife Refuge, 1984-1995.

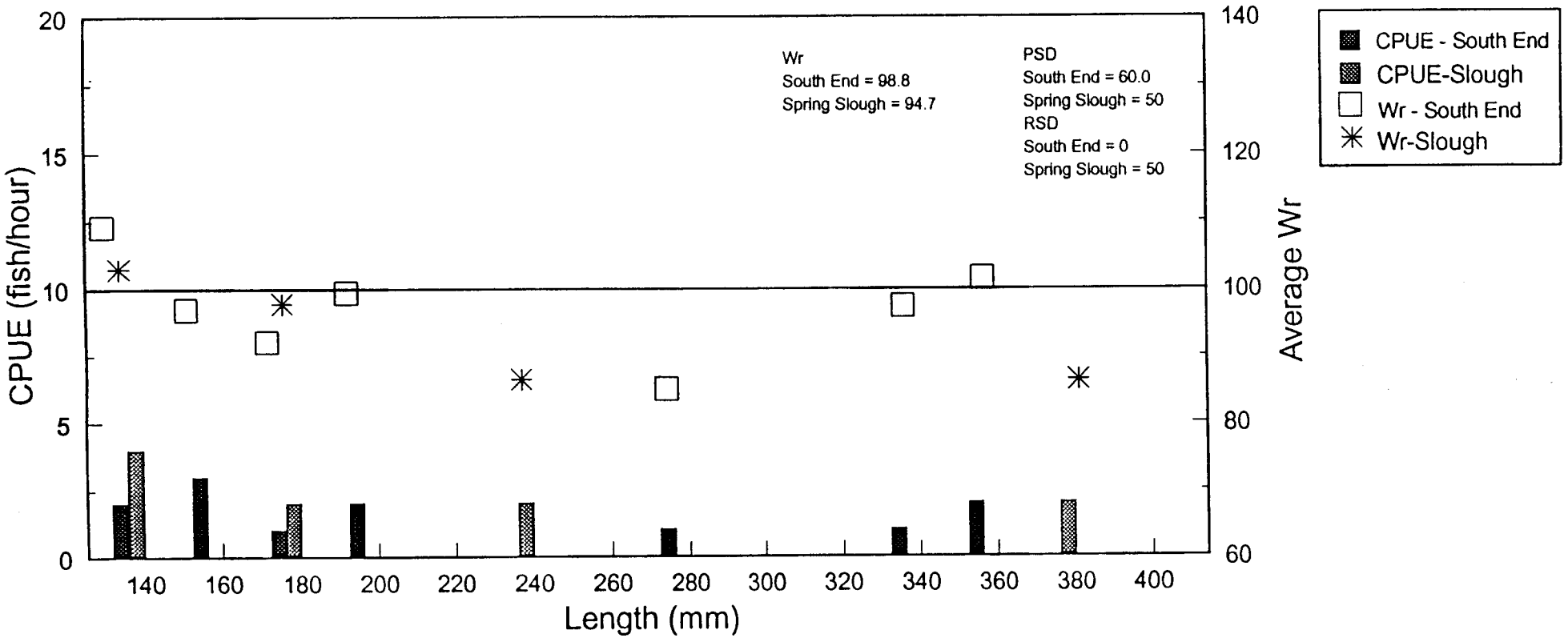


Figure 3. Largemouth bass collected by electrofishing at Keithsburg District, Mark Twain National Wildlife Refuge, on June 26-27, 1995.

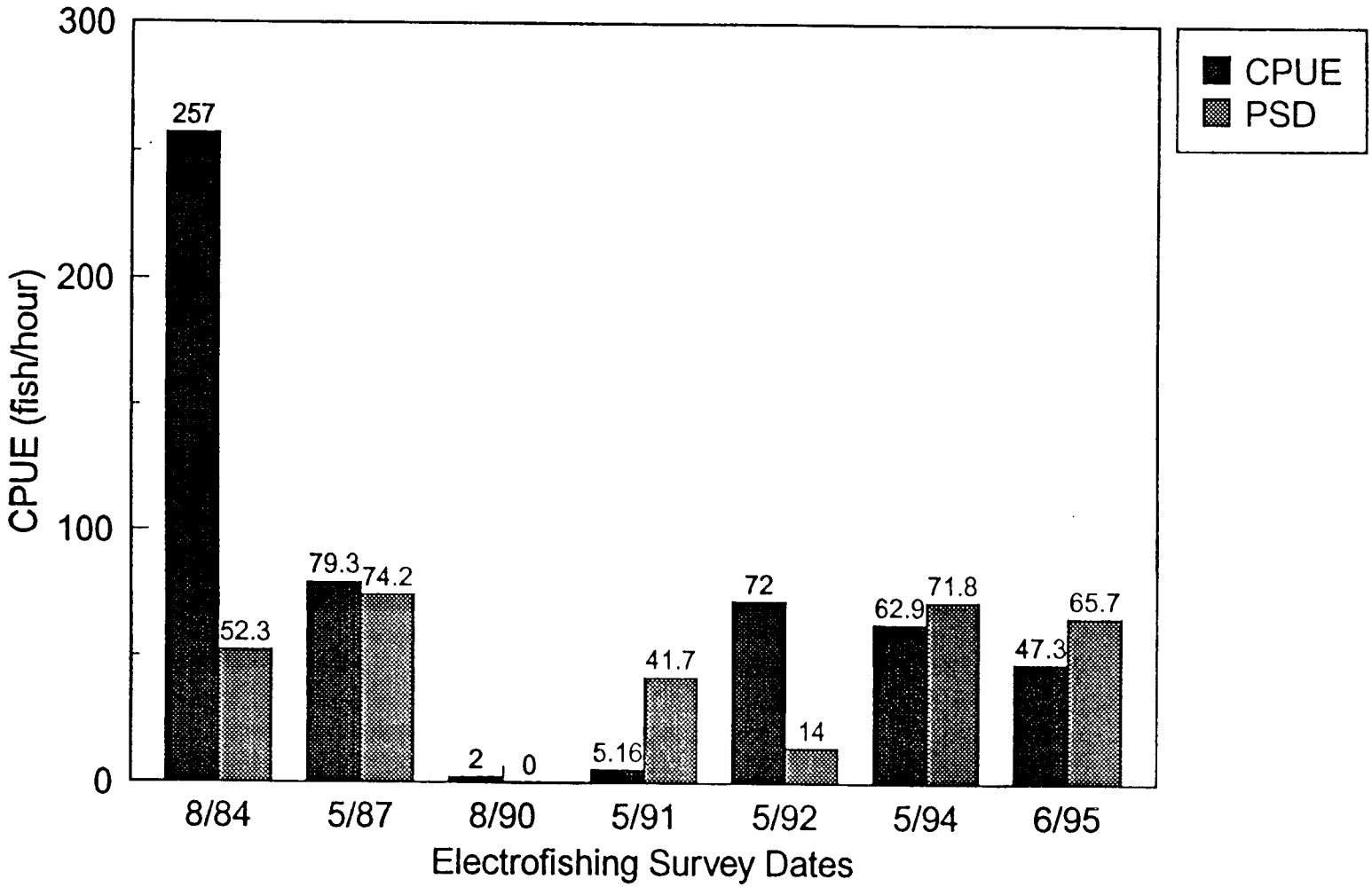


Figure 4. Catch per unit effort and PSDs of bluegill collected by electrofishing at Keithsburg District, Mark Twain National Wildlife Refuge, 1984-1995.

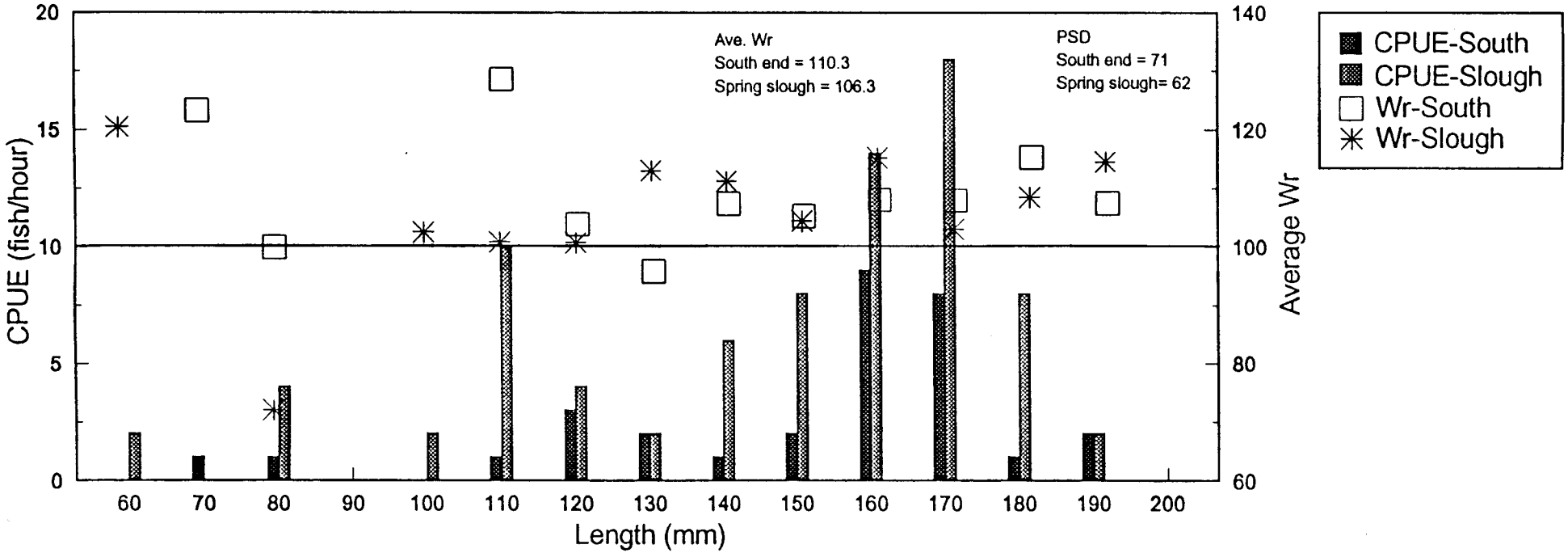


Figure 5. Bluegill collected by electrofishing at Keithsburg District, Mark Twain National Wildlife Refuge, on June 26-27, 1995.

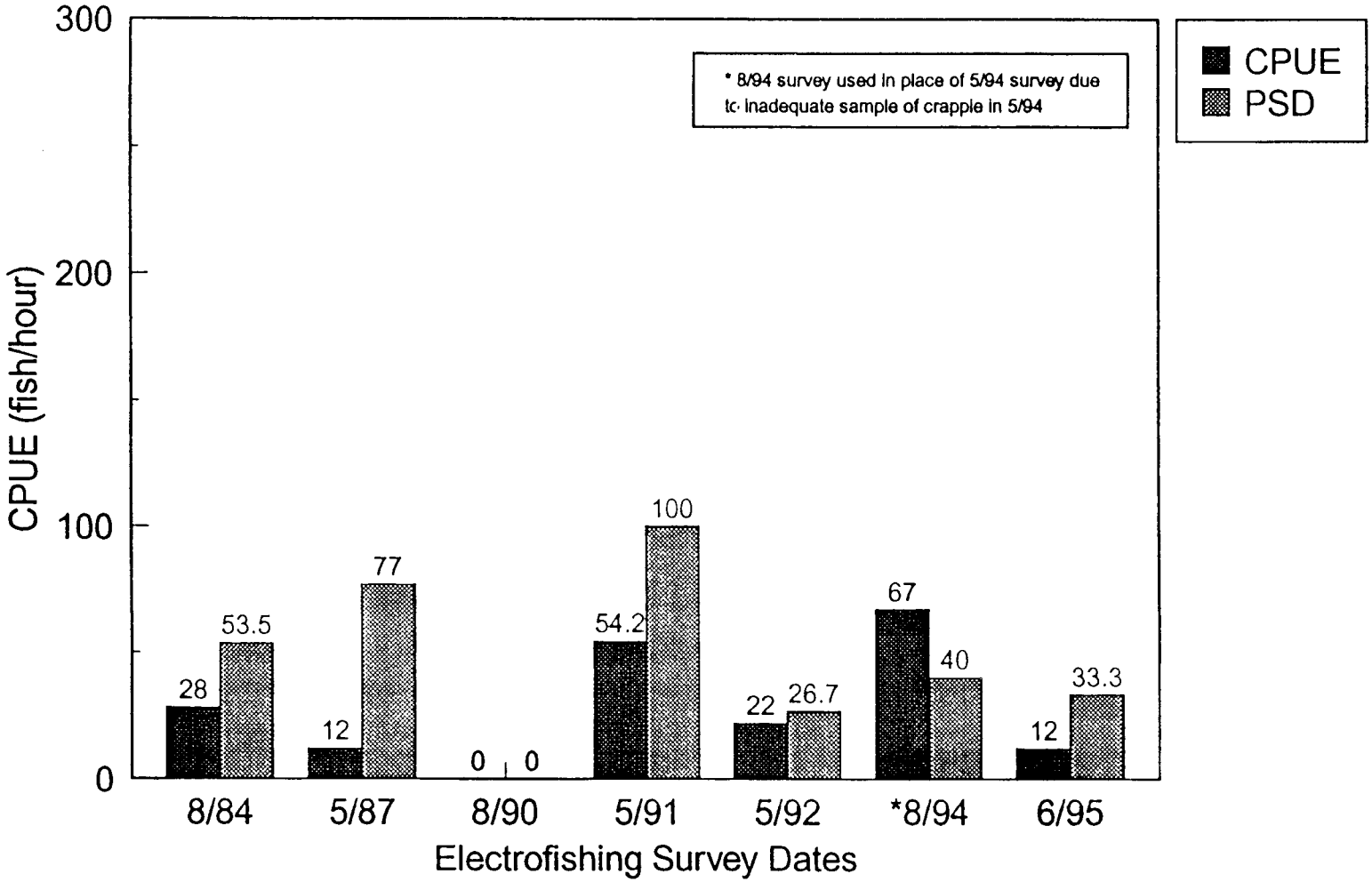


Figure 6. Catch per unit effort and PSDs of black crappie collected by electrofishing at Keithsburg District, Mark Twain National Wildlife Refuge, 1984-1995.

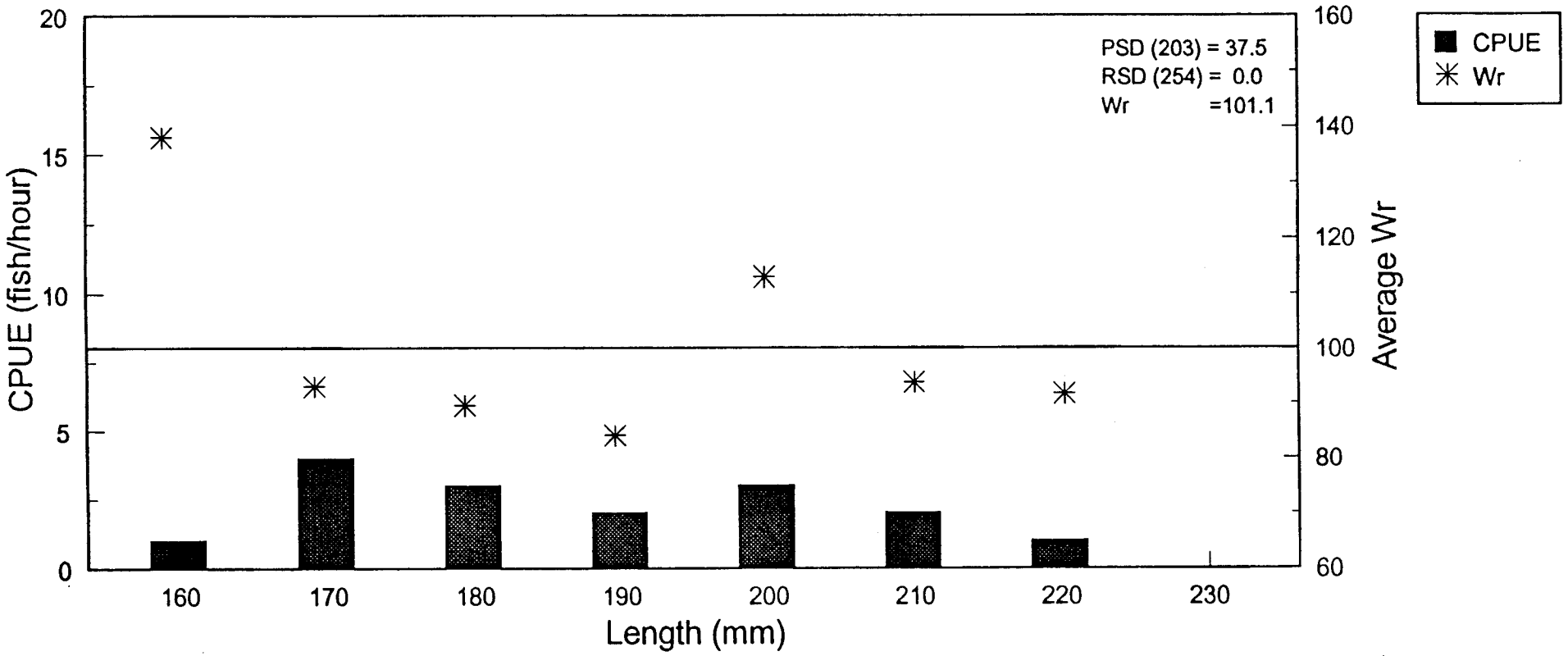


Figure 7. Black crappie collected at Keithsburg District, Mark Twain National Wildlife Refuge, on June 26-27, 1995.

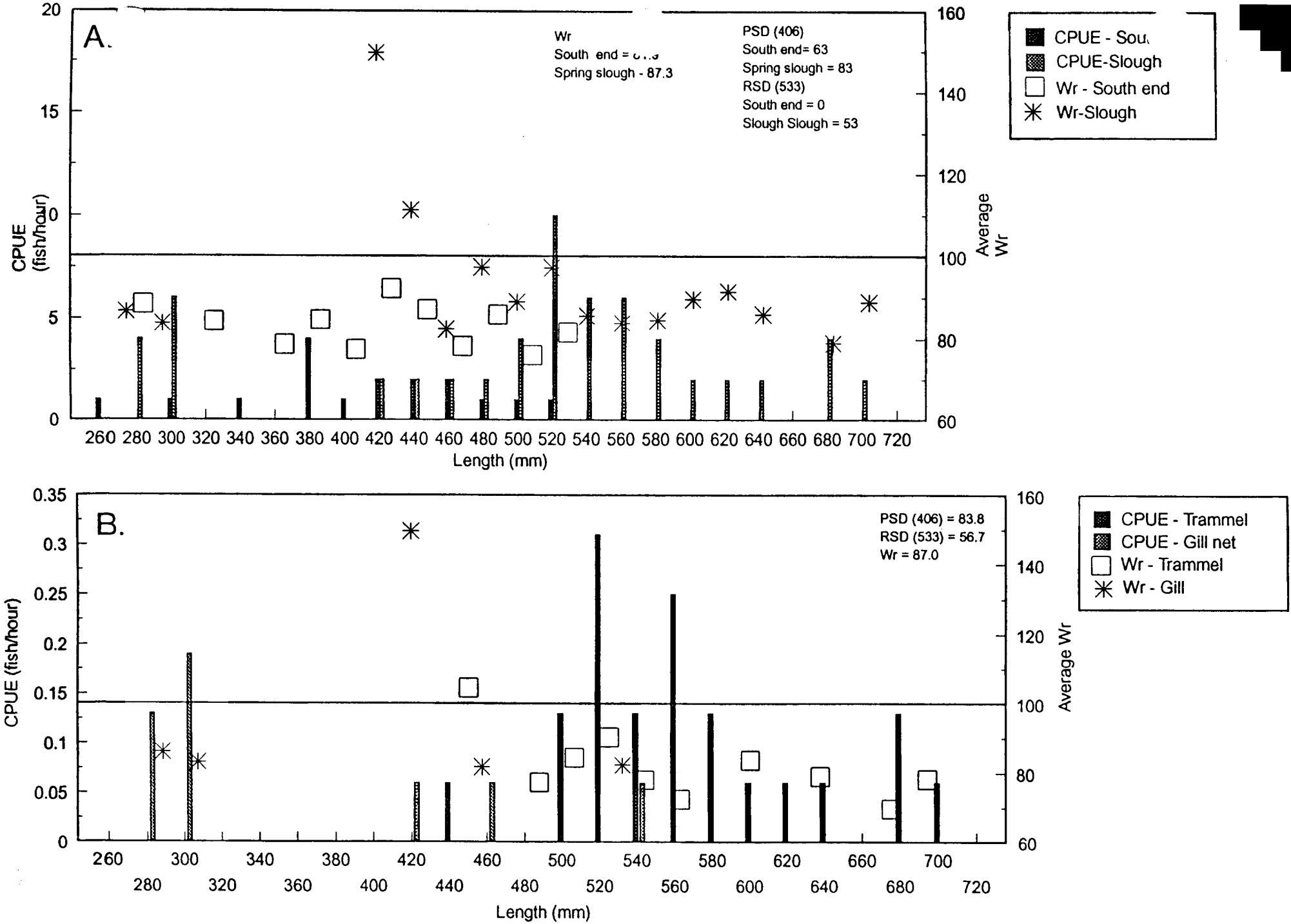


Figure 8. Catch per unit effort and relative weights of common carp collected by A.) electrofishing and B.) gill nets and trammel nets in Keithsburg District, Mark Twain National Wildlife Refuge, on June 26-27, 1995.

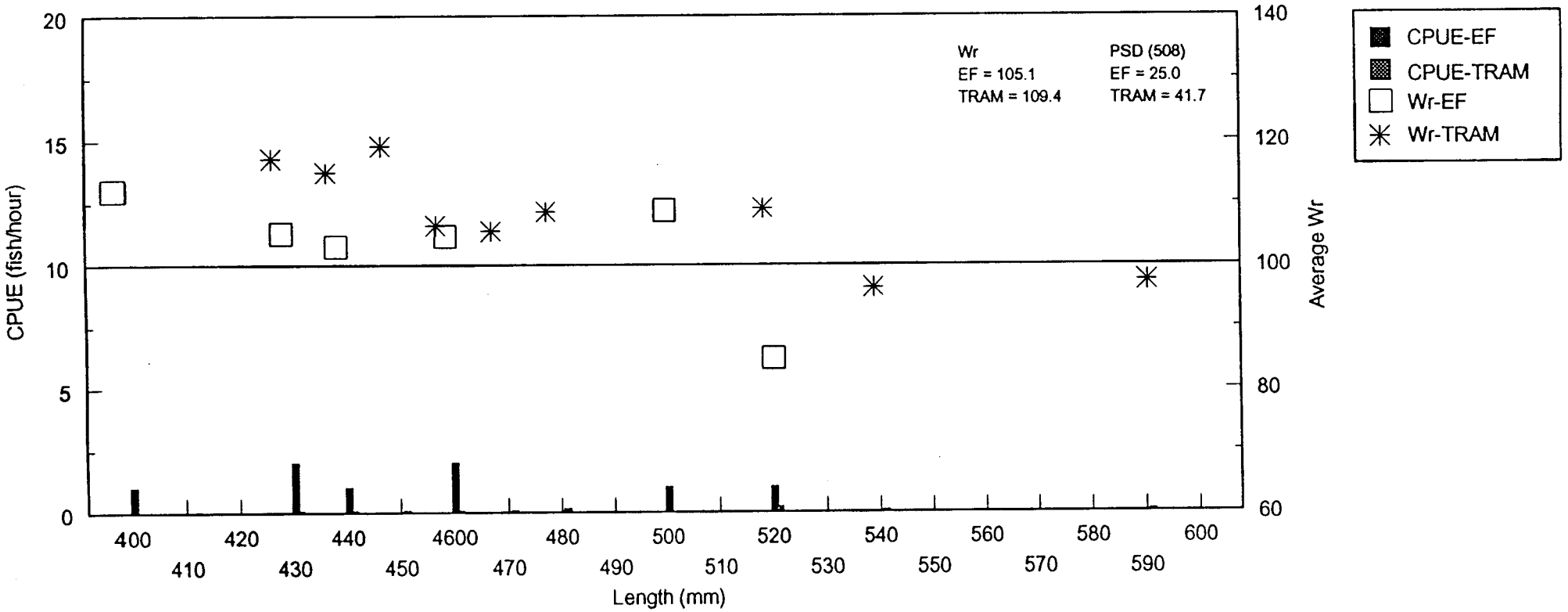


Figure 9. Bigmouth buffalo collected by electrofishing (EF) and trammel nets (TRAM)at Keithsburg District, Mark Twain National Wildlife Refuge, on June 26-27, 1995.

Table 1. sburg electrofishing data summary, 6/27/95 (Total effort; 90 es (1.5 hour))

Species	Number	Total Weight Kg.	Average Weight Grams	Average Total Length (mm)	Total Length Range (mm)	Percent Total Weight	Percent Total Number	C.P.U.E. No./Hr.	Number* Harvestable and (%)
BLG	70	6.5	86.4	144.6	64-196	9.4	38.0	46.7	46, (66)
LMB	17	4.2	243.8	228.6	141-383	6.0	9.2	11.3	3, (18)
BLC	18	2.1	117.5	192.4	164-220	3.0	9.8	12	8, (44)
CAP	21	21.5	1025.5	409.7	263-522	30.9	11.4	14	19, (91)
BIB	8	13.9	1732.5	459.0	401-520	20.0	4.3	5.3	8, (100)
SAB	5	2.6	510.2	313.2	295-328	3.7	2.7	3.3	4, (80)
CCF	1	0.3	280	326	326	0.4	0.5	0.7	1, (100)
BON	6	10.6	1772.5	589.7	495-663	15.3	3.3	4	1,(100)
WHB	2	0.3	135	220	215-225	0.4	1.1	1.3	1, (100)
WAE	3	1.5	481.7	377.7	335-426	2.2	1.6	2	3, (100)
FRD	4	1.8	437.5	320.5	248-386	2.6	2.2	2.7	
SAR	1	0.3	305	336	336	0.4	0.5	0.7	
RCS	1	1.1	1120	419	419	1.6	0.5	0.7	
YEB	1	0.2	190	234	234	0.3	0.5	0.7	1, (100)
WAR	2	0.03	25	99	77-121	0.04	1.1	1.3	0
TOTAL	184	69.48							
GZS	18	1.8	149.7	219.9	182-306	2.5	12.0	7	N/A
GOS	4	0.04	10.3	107.5	102-111	0.06	2.7	2	N/A
ELS	2	0.01	4	74.5	71-78	0.01	1.3	2	N/A

* Bluegill and other sunfish - 15cm & Greater
Crappie - 20cm & Greater
Largemouth bass - 35cm & Greater
Catfish - 25cm & Greater
Bullhead - 20cm & Greater
Carp & Buffalo - 30cm & Greater
Walleye - 30cm & Greater
Flathead catfish - 40cm & Greater

Metric Equivalents
1 inch = 25.4 mm
2.21 lbs = 1 Kg.
1 lb = 453.6 g

Table 2. Keithsburg gill netting and trammel netting data summary, 6/27/95 (Total effort; 3 net-nights)

Species	Number	Total Weight Kg.	Average Weight Grams	Average Total Length (mm)	Total Length Range (mm)	Percent Total Weight	Percent Total Number	C.P.U.E. No./net-night	Number* Harvestable and (%)
BIB	12	27.7	2307.1	496.9	438-593	34.0	18.8	4	12, (100)
BLB	2	0.4	194.0	109.5	91-128	0.5	3.1	0.67	
BLC	2	0.2	92.0	178.0	173-183	0.2	3.1	0.67	0
BRB	2	0.6	301.0	257.5	197-318	0.7	3.1	0.67	1, (50)
BON	7	11.3	1600.7	1600.7	488-686	13.8	10.9	2.3	
CAP	30	35.0	2110.3	496.5	288-694	42.9	46.9	10.0	28, (93)
CCF	6	4.8	803.2	387.2	250-626	5.9	9.4	2.0	6, (100)
LMB	1	0.6	642	353	353	0.7	1.6	0.33	1, (100)
SNG	1	0.3	280	473	473	0.4	1.6	0.33	
WAE	1	0.7	664	422	422	0.9	1.6	0.33	1, (100)
TOTAL	64	81.6							

* Bluegill and other sunfish - 15cm & Greater
Crappie - 20cm & Greater
Largemouth bass - 35cm & Greater
Catfish - 25cm & Greater
Bullhead - 20cm & Greater
Carp & Buffalo - 30cm & Greater
Walleye - 30cm & Greater
Flathead catfish - 40cm & Greater

Metric Equivalents
1 inch = 25.4 mm
2.21 lbs = 1 Kg.
1 lb = 453.6 g

Common Fish Names and Abbreviations

American Eel (AEL)
Bigmouth Buffalo (BIB)
Bigmouth Shiner (BMS)
Black Buffalo (BKB)
Black Bullhead (BLB)
Black Crappie (BLC)
Blackchin Shiner (BCS)
Blacknose Shiner (BNS)
Blue Catfish (BCF)
Bluegill (BLG)
Bluntnose Minnow (BNM)
Bowfin (BOW)
Brook Silverside (BSS)
Brown Bullhead (BRB)
Bullhead Minnow (BHM)
Carp (CAP)
Channel Catfish (CCF)
Chestnut Lamprey (CHL)
Emerald Shiner (ELS)
Fathead Minnow (FHM)
Flathead Catfish (FCF)
Freshwater Burbot (FWB)
Freshwater Drum (FRD)
Gizzard Shad (GZS)
Glass Shiner (GLS)
Golden Redhorse (GRH)
Golden Shiner (GOS)
Goldeye (GNE)
Green Sunfish (GSF)
Highfin Carpsucker (HFC)
Johnny Darter (JND)
Lamprey (LAM)
Largemouth Bass (LMB)
Logperch (LOG)
Longear Sunfish (LSF)
Longnose Gar (LNG)
Mooneye (MOE)
Muskellunge (MUE)
Mud Darter (MDD)
Northern Common Shiner (NCS)
Northern Hogsucker (NHS)
Northern Pike (NOP)
Northern Redhorse (NOR)
Orange Spotted Sunfish (OSS)
Other Sunfish (OSF)
Paddlefish (PAH)
Pirate Perch (PRP)
Pugnose Minnow (PNM)
Pugnose Shiner (PNS)
Pumpkinseed Sunfish (PKS)
Quillback Carpsucker (QCS)
Rainbow Darter (RBD)
Redear Sunfish (RSF)
River Carpsucker (RCS)
River Redhorse (RRH)
River Shiner (RVS)
Rock Bass (ROB)
Sand Shiner (SDS)
Sauger (SAR)
Shorthead Redhorse (SHR)
Shortnose Gar (SNG)
Silver Chub (SLC)
Silver Redhorse (SRH)
Silvery Minnow (SVM)
Smallmouth Bass (SMB)
Smallmouth Buffalo (SAB)
Spotfin Shiner (SFS)
Spottail Shiner (STS)
Spotted Sucker (SPS)
Tadpole Madtom (TPM)
Walleye (WAE)
Warmouth (WAM)
Weed Shiner (WDS)
Western Sand Darter (WSD)
White Bass (WHB)
White Crappie (WHC)
White Sucker (WHS)
Yellow Bass (YLB)
Yellow Bullhead (YEB)
Yellow Perch (YEP)
Trout Perch (TRP)
Hybrid Sunfish (HSF)
Silverband Shiner (SBS)

Appendix A.

Contaminants Investigation - Keithsburg Division,Wapello District, Mark Twain National Wildlife Refuge - Fisheries Component

Bluegill were collected by USFWS staff using pulse DC electrofishing on June 26-27, 1995 and August 29, 1995 at the Keithsburg Division. Fish were also collected by IDNR staff using three-phase AC current on August 29, 1995. Fish were collected in two distinct sampling locations, South End and Spring Slough. All fish were weighed to the nearest gram and measured to the nearest mm. Scale samples were removed from some fish collected in the South End in June to assist in year-class determination. Saggital otoliths were removed from approximately forty bluegill in each location in August to aid in determining if there was a difference in growth rates between the two sites.

June Sample:
Bluegill were numerous in Keithsburg. Bluegill comprised 25.6 % of the total number (5.2% of the total weight) of fish collected in the South End and 59.7% of the total number (30.1% of the total weight) of fish collected in Spring Slough (Tables A1 and A2). Relative weights (Wr) of bluegill were very good, averaging 110.3 in the South End and 106.6 in Spring Slough (Figure A1). Catch Per Unit Effort (fish per hour) was greater in Spring Slough (80 fish/hour) than in the South End (30 fish/hour) (Tables A1 and A2, Figure A1).

Overall, CPUE of bluegill was larger in the August sample than in the June sample. Water levels had dropped between June and August, cutting the refuge's hydrologic connection to the river, therefore, the fish were more concentrated. Bluegill also reproduced during the summer, increasing their numbers.

August Sample:
Bluegill comprised 52.3% of the total number (18.9% of the total weight) of fish collected in the South End and 51.9% of the total number (14.5% of the total weight) of fish collected in Spring Slough (Tables A3 and A4). CPUE was still greater in Spring Slough (328 fish/hour) than in the South End (243 fish/hour) (Tables A1 and A2, Figure A1).

Sagittal otoliths were read and ages assigned to each of the bluegill. Fish were grouped by year-class. Mean lengths of each year-class were compared between the two areas using an independent t-test. There was no difference in mean lengths for fish 1 and 2 years of age (the 1993 and 1994 year-classes). The only difference in mean length discovered was for 3-year old fish (the 1992 year-class). Mean lengths could not be compared for fish older than age-3 due to a small sample size (Table A5). Mean lengths of all year-classes fell within the ranges historically found for bluegill in Iowa and Illinois (Table A5).

A modified Fraser-Lee formula was used to back-calculate the lengths of each fish at each annulus. Back- calculated lengths at each annulus were compared using an independent t-test. There was a difference in back-calculated lengths of 1992 year-class bluegill between the two sites in 1992 and 1993 (Table A6).

The differences in both measured lengths, and back-calculated lengths at ages 1 and 2, may be attributable to a water draw down in 1992. Dense coontail blooms appeared in the sloughs. These populations crashed in late summer leading to elevated sediment ammonia concentrations and subsequent mortality of selected slough benthos (Mike Coffey, personal communication). The 1992 year-class faced periods of limited food supply, thus decreasing their growth rate and reducing their fitness to compete the following spring.

Mean relative weights (Wr) of each bluegill year-class were compared between the two study areas using an independent t-test. Although mean Wr of all year-classes fell within the desired range of 80-100%, relative weights of bluegill sampled were significantly higher in the South End (Table A7, Figure A2).

The differences found in bluegill Wr between study areas may be due to the different densities of bluegill. Bluegill comprised a greater portion of the fish sample and were more abundant in Spring Slough than in the South End. This would increase intraspecific competition for food and reduce the condition of individual fish. Although it is also likely the differences in Wr may be attributed to differences in water quality and productivity, any potential differences in water quality did not have the same effect on other fish species in Spring Slough.

As water quality data are unavailable at this time, Wrs of other fish species collected in sufficient numbers in June and August were compared between the two study areas using independent t-tests. No differences were found between study areas for largemouth bass, black crappie, and common carp collected in June. Differences were found between study areas for black crappie, and common carp in August, but not for largemouth bass (Table A8). Unlike the results seen in the bluegill sample, Wrs of carp and black crappie were higher in Spring Slough than in the South End in August.

This information should be regarded with some caution. Without water quality and invertebrate data, no cause and effect relationship has been proven.

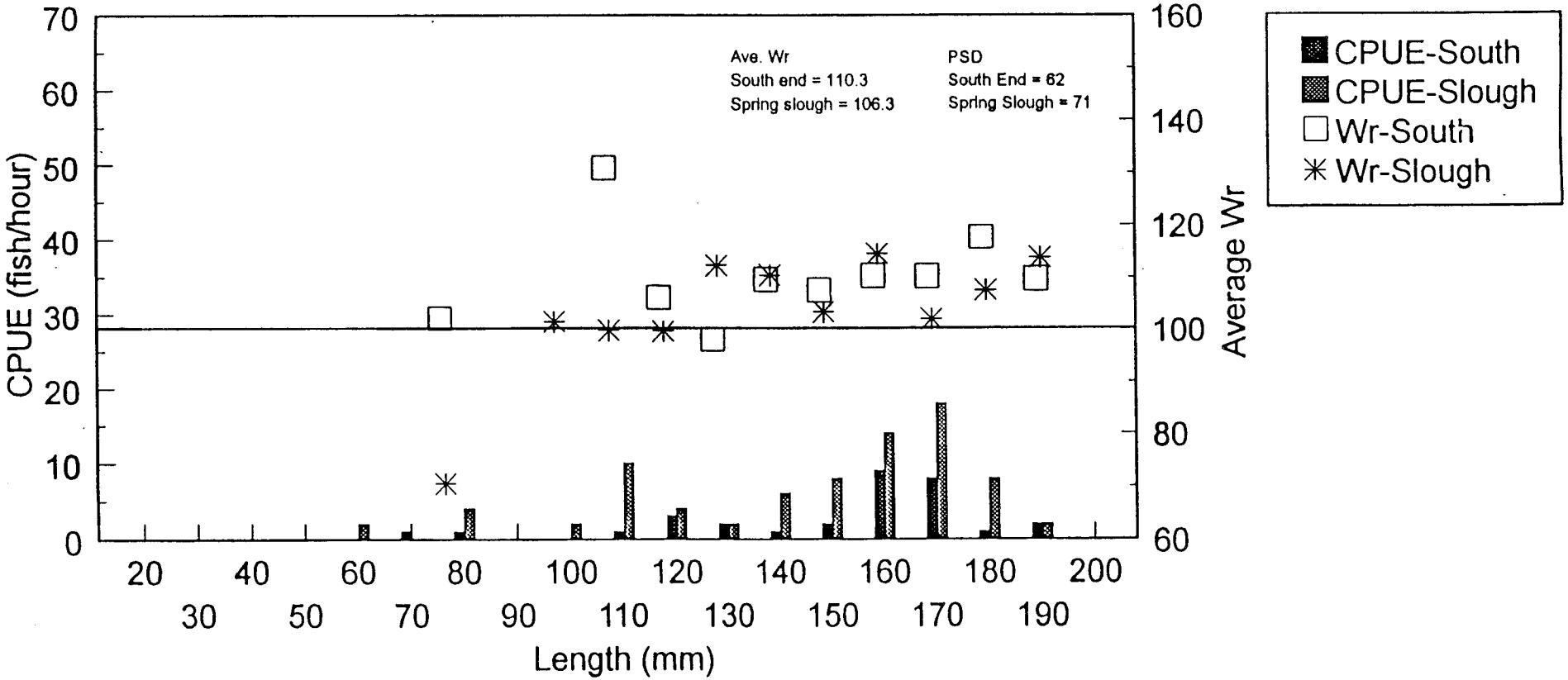


Figure A1. Bluegill collected by electrofishing at Keithsburg District, Mark Twain National Wildlife Refuge, on June 26-27, 1995.

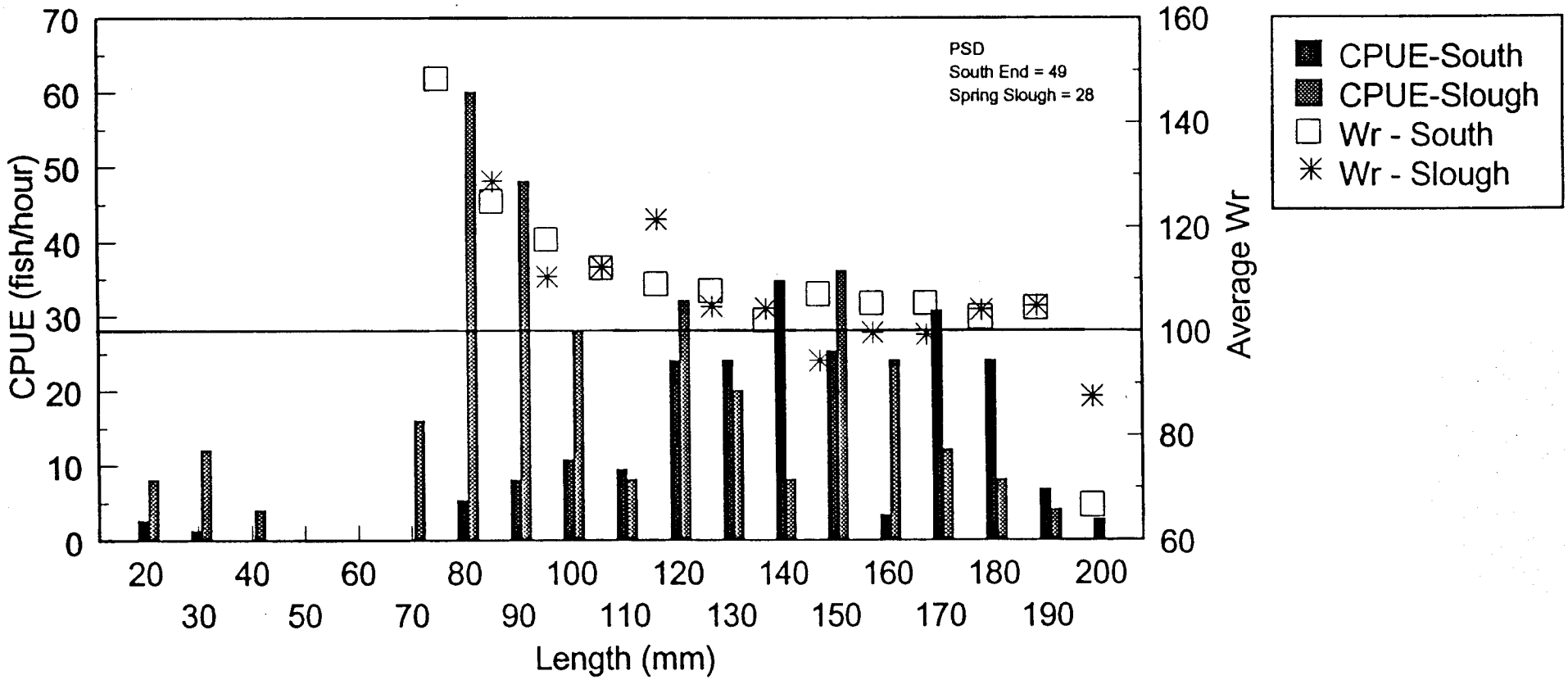


Figure A2. Bluegill collected by electrofishing at Keithsburg District, Mark Twain National Wildlife Refuge, on August 29, 1995.

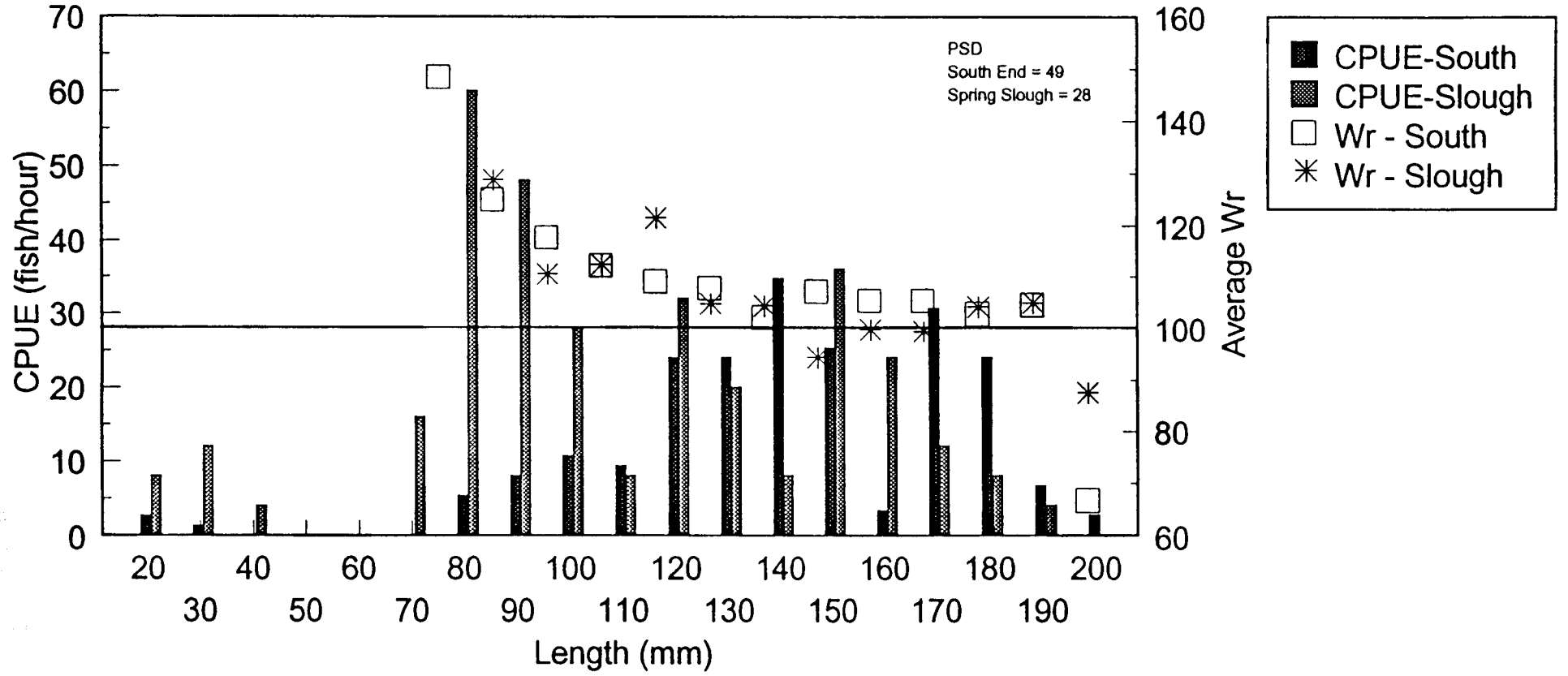


Figure A2. Bluegill collected by electrofishing at Keithsburg District, Mark Twain National Wildlife Refuge, on August 29, 1995.

Table A1. Keithsburg (South End) electrofishing data summary, 6/27/95 (Total effort; 60 minutes (1 hour))

Species	Number	Total Weight Kg.	Average Weight Grams	Average Total Length (mm)	Total Length Range (mm)	Percent Total Weight	Percent Total Number	C.P.U.E. No./Hr.	Number* Harvestable and (%)
BLG	30	3.0	95.7	155.2	83-196	5.2	25.6	30	19, (63)
LMB	12	3.1	257.9	232.6	145-379	5.4	10.3	12	2, (17)
BLC	16	1.9	117.3	191.4	164-220	3.3	13.7	16	6, (38)
CAP	17	17.5	1029.8	417.6	273-522	30.2	14.5	17	16, (94)
BIB	8	13.9	1732.5	459.0	401-520	24.0	6.8	8	8, (100)
SAB	5	2.6	510.2	313.2	295-328	4.5	4.3	5	4, (80)
CCF	1	0.3	280	326	326	0.5	0.9	1	1, (100)
BON	6	10.6	1772.5	589.7	495-663	18.3	5.1	6	1,(100)
WHB	2	0.3	135	220	215-225	0.5	1.7	2	1, (100)
WAE	3	1.5	481.7	377.7	335-426	2.6	2.6	3	3, (100)
FRD	1	0.7	700	386	386	1.2	0.9	1	
SAR	1	0.3	305	336	336	0.5	0.9	1	
RCS	1	1.1	1120	419	419	2.0	0.9	1	
YEB	1	0.2	190	234	234	0.3	0.9	1	1, (100)
WAR	2	0.03	25	99	77-121	0.04	1.7	2	0
TOTAL	117	57.86							
GZS	7	0.8	116.9	219.9	182-292	1.4	6.0	7	N/A
GOS	2	0.02	11	110	109-111	0.03	1.7	2	N/A
ELS	2	0.01	4	74.5	71-78	0.02	1.7	2	N/A

* Bluegill and other sunfish - 15cm & Greater
Crappie - 20cm & Greater
Largemouth bass - 35cm & Greater
Catfish - 25cm & Greater
Bullhead - 20cm & Greater
Carp & Buffalo - 30cm & Greater
Walleye - 30cm & Greater
Flathead catfish - 40cm & ⁽

Metric Equivalents
1 inch = 25.4 mm
2.21 lbs = 1 Kg.
1 lb = 453.6 g

Table A2. Keithsburg (Spring Slough) electrofishing data summary, 6/27/95 (total effort; 30 minutes (1/2 hour))

Species	Number	Total Weight Kg.	Average Weight Grams	Average Total Length (mm)	Total Length Range (mm)	Percent Total Weight	Percent Total Number	C.P.U.E. No./Hr.	Number* Harvestable and (%)
BLG	40	3.5	86.5	149.5	64-193	30.1	59.7	80	27, (68)
LMB	5	1.1	209.8	219.0	141-383	9.5	7.5	10	1, (20)
BLC	2	0.2	119.5	200.5	200-201	1.7	2.9	4	2, (100)
CAP	4	4.0	1007.0	371.3	263-435	34.4	6.0	8	3, (75)
FRD	3	1.1	350.0	298.7	248-367	9.5	4.5	6	8, (100)
TOTAL	67	11.62							
GZS	11	1.7	152.9	236.5	202-306	14.6	16.4	22	N/A
GOS	2	0.02	9.5	106.0	102-108	0.2	3.0	4	N/A

* Bluegill and other sunfish - 15cm & Greater
Crappie - 20cm & Greater
Largemouth bass - 35cm & Greater
Catfish - 25cm & Greater
Bullhead - 20cm & Greater
Carp & Buffalo - 30cm & Greater
Walleye - 30cm & Greater
Flathead catfish - 40cm & Greater

Metric Equivalents

1 inch = 25.4 mm
2.21 lbs. = 1 Kg
1 lb = 453.6 g

Table A3. Fish collected by electrofishing at Keithsburg District (South End) on 08/29/95 using pulse DC current (total effort 45 minutes)

Species	Number	Total Weight (kg)	Average Weight (g)	Average Total Length (mm)	Total Length Range (mm)	Percent Total Weight	Percent Total Number	C.P.U.E. No./Hr.	Number* Harvestable and (%)
BLG	162	14.27	88.1	147.1	27-204	18.9	52.3	243	92 (57)
BLC	49	5.59	114.1	196.1	142-262	7.4	15.8	73.5	1 (02)
CAP	33	39.16	1186.7	42.5	300-625	52.0	10.7	49.5	33 (100)
LMB	20	3.73	186.6	204.9	100-377	5.0	6.5	30	2 (10)
WAM	8	0.62	77.5	144.9	108-181	0.8	2.6	12	3 (38)
BON	6	6.38	106.3	495.0	437-582	8.5	1.9	9	
WHC	4	0.36	90.3	193.8	182-210	0.5	1.3	6	1 (25)
SAU	2	0.45	225.0	310.5	271-350	2.5	0.7	27	
FRD	2	0.55	276.5	272.5	222-323	0.7	0.7	3	
WHB	2	0.14	71.5	173.0	170-176	0.2	0.7	3	
BIB	1	0.89	890	375	375	1.2	0.3	1.5	1 (100)
SAB	1	0.17	170	223	223	0.2	0.3	1.5	0
OSF	1	.01	11	79	79	0.01	0.3	1.5	
RCS	1	1.10	1100	406	406	1.5	0.3	1.5	
TOTAL	310	75.31							
GZS	18	1.89	105.0	211.7	115-295	2.5	5.8	27	N/A

* Bluegill and other sunfish - 15 cm & Greater
Crappie - 20 cm & Greater
Largemouth Bass - 35 cm & Greater
Catfish - 25 cm & Greater
Bullhead - 20 cm & Greater
Carp & Buffalo - 30 cm & Greater
Walleye - 30 cm & Greater
Flathead Catfish - 40 cm & Greater

Table A4. collected by electrofishing at Keithsburg District (Spring Slo[illegible] n 08/29/95 using pulse DC current (total effort 15 minutes)

Species	Number	Total Weight (kg)	Average Weight (g)	Average Total Length (mm)	Total Length Range (mm)	Percent Total Weight	Percent Total Number	C.P.U.E. No./Hr.	Number* Harvestable and (%)
BLG	82	3.39	41.4	113.1	26-199	14.5	51.9	328	21 (26)
BLC	21	1.71	81.3	173.7	133-213	7.3	13.3	84	3 (14)
CAP	4	5.38	1346.3	460.0	430-520	23.0	2.5	16	4 (100)
LMB	6	1.61	268.2	214.4	165-350	6.9	3.8	24	1 (17)
NOP	1	1.38	1375.0	610	610	5.9	0.6	4	1 (100)
BON	1	1.51	1510.0	575	575	6.4	0.6	4	
WHC	2	0.04	20.5	109.5	85-134	0.2	1.3	8	0
WAE	2	1.27	634.5	412.0	385-439	23.0	1.3	16	4 (100)
WHB	1	0.19	185.0	245	245	0.8	0.6	4	
BIB	2	2.75	1375.0	444.0	404-484	11.7	1.3	8	2 (100)
SAB	1	0.45	445.0	286	286	1.9	0.6	4	0
TOTAL	158	23.44							
GZS	28	3.64	251.3	229.4	115-335	15.5	17.7	112	N/A
GOS	7	0.12	17.7	115.4	69-132	0.5	4.4	28	N/A

* Bluegill and other sunfish - 15 cm & Greater; Bullhead - 20 cm & Greater
Crappie - 20 cm & Greater; Carp & Buffalo - 30 cm & Greater
Largemouth Bass - 35 cm & Greater; Walleye - 30 cm & Greater
Catfish - 25 cm & Greater; Flathead Catfish - 40 cm & Greater

Table A5. Mean Lengths of Bluegill Collected in Keithsburg District, Mark Twain National Wildlife Refuge, on August 29, 1995.

Year-Class	Age	South End	Spring Slough	Statistical Difference in Mean Length?	IL bluegill* studies	IA bluegill* studies
1994	1	95.63	94.0	No (t=0.364, df=18, p=0.720)	102 (51-178)	94 (61-183)
1993	2	146.09	137.0	No (t=1.038, df=21, p=0.311)	134 (102-190)	126 (81-208)
1992	3	176.38	161.59	Yes (t=2.817, df=31, p=0.008)	158 (124-208)	149 (102-224)
1991	4	183.00	none	N/A	162 (119-203)	172 (107-228)
1990	5	191.50	none	N/A	164 (127-229)	184 (127-229)

* Mean of means (range). Excerpted from: Carlander, K.D. 1977. Handbook of Freshwater Fishery Biology Volume 2. The Iowa State University Press, Ames, Iowa, 431pp.

Table A6. Mean Back-Calculated Lengths of Age-3 Bluegill Collected in Keithsburg District, Mark Twain National Wildlife Refuge, on August 29, 1995.

Year	Annulus	South End	Spring Slough	Statistical Difference in Mean Length?
1994	3	157.78	148.83	No (t=1.594, df=31, p=0.121)
1993	2	123.00	112.32	Yes (t=2.537, df=31, p=0.61)
1992	1	59.06	49.92	Yes (t=2.068, df=31, p=0.47)

Table A7. Mean Relative Weights (Wr) of Bluegill Collected in Keithsburg District, Mark Twain National Wildlife Refuge, on August 29, 1995.

Year Class	Age	South End	Spring Slough	Statistical Difference in Mean Wr?
1994	1	139.45	102.30	Yes (t=3.714, df=18, p=0.002)
1993	2	111.97	100.90	Yes (t=3.638, df=21, p=0.002)
1992	3	107.71	98.17	Yes (t=3.281, df=29, p=0.003)
1991	4	94.5	None	N/A
1990	5	74.75	None	N/A

Table A8. Mean Relative Weights of Several Fish Species Collected in Keithsburg District, Mark Twain National Wildlife Refuge, on June 27, 1995 and August 29, 1995.

Species	Sample Date	South End	Spring Slough	Statistical Difference in Mean Wr?
Carp	June	81.89	106.82	No (t=-0.962, df=3.0, p=0.407)
Carp	August	78.65	84.57	Yes (t=-2.740, df=9.3, p=0.023)
Black Crappie	June	111.09	98.80	No (t=2.005, df=15.9, p=0.063)
Black Crappie	August	99.07	103.36	Yes (t=-2.058, df=28.5, p=0.049)
Largemouth Bass	June	100.62	100.04	No (t=0.087, df=5.0, p=0.934)
Largemouth Bass	August	111.97	102.67	No (t=1.214, df=8.0, p=0.264)

MEMO

To: Ed Walsh and Dan Sallee
From: Steve Sobaski, INHS-Center for Aquatic Ecology
Subject: Analysis of the 1995 Ice Creel Survey at Keithsburg Refuge
Date: Tuesday, May 2, 1995

Attached are **final** effort, harvest, catch, supplementary, completed trip and length frequency data from the **1995 DAY ICE CREEL** at **Keithsburg Refuge**. Results include data collected from 1/1/95 - 2/22/95. The format of these outputs is identical to that used for the report on the walleye creel from the Lock and Dam 17 region of the Mississippi River that I sent last year. Unlike that report, there aren't separate analyses for any specific subset of anglers, targetting one fish species or group of species, such as I ran just for walleye/sauger anglers in the Lock and Dam 17 report. These tables summarize fishing for all anglers, regardless of the fish species being targeted.

Rather than giving a long explanation of the creel analysis and interpretation of the attached outputs in this cover letter (I think I can hear the sighs of disappointment all the way in Champaign), I've included several write-ups separately, that explain the creel analysis methodology. The first, "What's Included in the INHS Interim and Final Creel Reports" discusses all of the outputs that we generate from the Apple *IIe*/General Manager creel databases, such as the one for Keithsburg. Three creel output programs, custom written for our creel databases, produce (1) the **EFFORT** Table, (2) the **HARVEST** and **CATCH** (*the total of harvest plus released fish*) Tables, and (3) the **Supplementary** outputs, covering information such as the trip length, degree of angler satisfaction, fish species sought, distance traveled to fish, the completed trip angler catch rates for specific species and the species length frequency histograms for Harvested and Released (rather than total Catch) fish.

Following the protocol that we've previously used for analyzing impoundment ice creel data, all anglers interviewed during the Keithsburg creel and counted in instantaneous counts have been recoded as Boat anglers. The rationale for this is that ice fishing generally occurs out on the water rather being than restricted to the shoreline area. Therefore, whenever we survey a lake for an entire year and need to run an analysis of the entire year of fishing, the ice creel data is more logically reported with the boat data from the ice-free portion of the year, on the basis of "habitats being sampled" by anglers, rather than analyzed with the ice-free shore angler catch. For this survey's results, though, because the analysis only pertains to ice fishing, angler coding really doesn't make any difference, as long as all anglers are given the same code . Coding all anglers as Shore anglers would have resulted in the same estimate of total hours of fishing, total harvest, total catch, etc. So, if you're wondering what happened to the angler interviews and instantaneous counts that were recorded as "SHORE", they've simply been entered as "Boat" in the General Manager database for Keithsburg and are reported in the Boat Angler totals (whenever a distinction is made between boat and shore angling in these outputs).

The acreage value of 380 reported at the top of these outputs is based on estimates calculated by our GIS software, ARC/INFO. We used the most detailed maps available to us here at the Survey, in this case USGS 7.5" topo maps, as the basis for the survey area estimates. To input water boundaries into the GIS we digitized the area defined by the shaded zone on the Mississippi River chart map provided by Ed (a copy is attached to this report). The area of that map colored fully in black, corresponds to the area actually sampled, as noted by Ed. This area was estimated to be **96.80 acres** of water. The total water acreage in the shaded zone came to **380.13 acres,** which is pretty close to the USFW estimate of 400 acres of open water. As for which acreage to report, I opted for a conservative approach, in terms of reporting # or biomass of fish/acre, and used the larger area value. The assumption inherent in doing this is that the creel clerks were missing just a very insignificant portion of all of the ice anglers in the refuge during their instantaneous counts as well as an insignificant amount of the total fish catch on the day periods sampled. If you feel more confident in using the smaller value (that of the actual area sampled as noted in black) simply multiply all ***per acre*** values reported in these outputs by **3.926963**.

As for the choice of the individual species listed in the harvest and catch reports, our creel analysis software, **STATCALC,** will only permit up to nine separate species to be reported with the calculation and report of the total fish harvest and catch. The estimated totals do account for all of the species recorded during the survey. To individually report each species taken by anglers during the ice creel, I've run two STATCALC analyses. Thus, you'll find two harvest tables and two catch tables included with this report. The first page of each covers the nine primary species taken by ice anglers, while the second page covers the remaining two species reported by anglers (white bass and bowfin). Note that the totals given between each Harvest or Catch page are identical. The first page includes separate listing for black crappie (BLC), white crappie (WHC), and unidentified crappie (CRP). Since the weight of fish harvested or caught is based on the length of the fish taken converted to weight using the standard length to weight conversion of LOG WT = a + b* LOG TL (where the a and b parameters are species specific), I used an average of the white crappie and black crappie a and b parameters to calculate an estimated weight of unidentified crappie (CRP) taken.

One other note regarding the CRP code to avoid some obvious potential confusion. Length Frequency histograms Figs. 10 and 11 report " Harvested 'CRP' " and " Released 'CRP' ". These histograms summarize length frequency for crappie as a **group,** rather than just representing unidentified crappie (as is the case in the Harvest and Catch tables). The frequencies shown in these two plots are the sum of all black crappie (BLC), white crappie (WHC), and unidentified crappie that were reported by anglers during the creel. Or, in other words, Figure 10 is the combination of Figures 6, 8, and 18, while Figure 11 is a combination of Figures 7, 9, and 19. Since the Supplementary output program only recognizes CRP as a group code (it automatically searches through the entire database and totals all harvest or released crappie records when analyzing for code CRP), I had to recode the unidentified crappie records as "UCR" to generate a separate **# FISH Harvested/Released by # Angler-Completed Trips** table for this set of fish (these are the table that breakdown all 40 interviewed anglers reporting a completed trip based on the number fish of a given species that they individually harvested or released). In hindsight "UCR" probably would have been a wiser choice for coding unidentified crappie for the harvest and catch analysis too. But, in lieu of rerunning the

analyses, I hope this explanation clears up any confusion over the use of the "CRP" code in these tables. When listed in the Harvest and Catch tables it refers to UNIDENTIFIED CRAPPIE. In all other tables and plots it refers to ALL CRAPPIE.

The last plot included in these outputs, attached just prior to the map, is the result of a regression analysis relating the number of ice anglers to the number of ice huts or shanties. This plot is based on 52 observations (or instantaneous counts) made during the creel. The relationship of **# Ice Anglers = (1.5598 * #Huts) - 0.03945** is, not surprisingly, pretty tight. Basically, on average, this works out to 1-2 anglers per hut. For future ice creels, if time is limited for taking a complete instantaneous count of fishermen, this relationship might make counting huts a useful substitute for actually visiting each hut to determine the total number of anglers. Also not surprising is that the South Access point had considerably more fishing pressure than the North Access area. The average number of ice fishermen ($\pm$ 1 standard error) from 52 instantaneous counts was 15.54 anglers ($\pm$ 1.27) for the South Access. By contrast, only 2.04 anglers ($\pm$ 0.56) were at the North Access.

If you have any questions about these analysis or on how to interpret these results, and the enclosed writeup on interpretting results doesn't help, I'll be glad to entertain your questions (honest). Feel free to call me at (217)-333-3312.

EFFORT TABLE FOR THE FULL DAY *** DAY ***

REGION :=1 LAKE :=KEITHSBURG REFUGE
DISTRICT :=04 YEAR :=95
ACREAGE :380 SAMPLING RATIO :=55/159 = 34.5%
NUMBER OF INTERVIEWS:351

YEAR PERIOD 01/01 TO 02/15 OF SECTION 1 COALESCED WITH
YEAR PERIOD 02/16 TO 02/22 OF SECTION 1

	ANGL HRS	95% CONF INTVL		HRS/ ACRE	95% CONF INTVL		% EFF INTVD
BOAT FISHING:							
WEEKDAY	4676	3088-6264	(34%)	12	8-16	(34%)	9.26
WKND/HOL	3069	2620-3518	(15%)	8	7-9	(15%)	18.71
STR TOTAL	7745	6099-9391	(21%)	20	16-25	(21%)	13.01
SHORE FISHING:							
WEEKDAY	0		(0%)	0		(0%)	
WKND/HOL	0		(0%)	0		(0%)	
STR TOTAL	0		(0%)	0		(0%)	0
BOAT/SHORE COALESCED:							
WEEKDAY	4676	3088-6264	(34%)	12	8-16	(34%)	9.26
WKND/HOL	3069	2620-3518	(15%)	8	7-9	(15%)	18.71
STR TOTAL	7745	6099-9391	(21%)	20	16-25	(21%)	13.01
BOAT/SHORE STRATIFIED:							
WEEKDAY	4676	3088-6264	(34%)	12	8-16	(34%)	9.26
WKND/HOL	3069	2620-3518	(15%)	8	7-9	(15%)	18.71
STR TOTAL	7745	6099-9391	(21%)	20	16-25	(21%)	13.01

HARVESTED AND CPUE TABLE BY SUBSTRATUM ACROSS STRATA *** DAY ***
REGION :=1 LAKE :=KEITHSBURG REFUGE
STRICT :=04 YEAR :=95
EAGE :380 SAMPLING RATIO :=55/159 = 34.5%
RTIO OF EFFORT HOURS INTERVIEWED := 1007.2/7748.9 = 12.99%
NUMBER OF INTERVIEWS: 351

COMBINED ACROSS STRATA:

YEAR PERIOD 01/01 TO 02/15 OF SECTION 1 COALESCED WITH
YEAR PERIOD 02/16 TO 02/22 OF SECTION 1

MSC SPECIES CAUGHT:
BOW WHB
SUBSTRATUM:
DAY PERIODS STRATIFIED
WEEKDAY/WEEKEND: WEEKDAY/WEEKEND STRATIFIED
FISHING TYPE: BOAT/SHORE COALESCED
FISH: HARVESTED

SPEC	#/HR	95% CI		# HARVST	95% CI		#/HA	#/ACRE
BLC	.076	.038-.114	(50 %)	787	424-1149	(46 %)	5.11	2.07
BLG	3.351	1.707-4.996	(49 %)	31163	14757-47570	(53 %)	202.64	82.01
CAP	.002	+-.006	(220 %)	12	+-31	(166 %)	.08	.03
CRP	.008	+-.025	(219 %)	69	+-219	(219 %)	.45	.18
GWH	.002	+-.007	(201 %)	27	+-71	(167 %)	.17	.07
LMB	.002	+-.005	(143 %)	21	+-48	(129 %)	.14	.06
WAE	*** NOT RECORDED ***			*** NOT RECORDED ***				
WAM	.000	+-.000	(213 %)	3	+-10	(213 %)	.02	.01
WHC	.005	+-.010	(103 %)	35	5-66	(86 %)	.23	.09
MSC	.000	+-.000	(213 %)	2	+-7	(213 %)	.01	.00
TOT	3.447	1.782-5.112	(48 %)	32119	15450-48788	(52 %)	208.85	84.52

SPEC	KG/HR	95% CI		KG HARVST	95% CI		KG/HA	AVG WT(G)
BLC	.010	.005-.015	(52 %)	96	52-140	(46 %)	.624	122.1
BLG	.361	.188-.533	(48 %)	3360	1617-5104	(52 %)	21.851	107.8
CAP	.001	+-.004	(186 %)	10	+-26	(154 %)	.066	862.8
CRP	.001	+-.004	(225 %)	10	+-34	(225 %)	.068	152.2
GWH	.000	+-.001	(211 %)	4	+-11	(175 %)	.025	146.3
LMB	.001	+-.002	(130 %)	11	+-25	(126 %)	.071	524.7
WAE	*** NOT RECORDED ***			*** NOT RECORDED ***				
WAM	.000	+-.000	(213 %)		+-1	(213 %)	.003	140.2
WHC	.001	+-.004	(169 %)	8	+-19	(127 %)	.054	233.5
MSC	.000	+-.000	(213 %)		+-.96103	(213 %)	.002	147.3
TOT	.376	.201-.551	(46 %)	3501	1727-5275	(51 %)	22.764	109.0

SPEC	LB/HR	95% CI		LB HARVST	95% CI		LB/ACRE	AVG WT(LB)
BLC	.021	.010-.032	(52 %)	212	115-308	(46 %)	.557	.2692
BLG	.796	.415-1.176	(48 %)	7408	3565-11252	(52 %)	19.495	.2377
CAP	.003	+-.008	(186 %)	22	+-57	(154 %)	.059	1.9020
CRP	.003	+-.009	(225 %)	23	+-75	(225 %)	.061	.3356
GWH	.000	+-.002	(211 %)	9	+-24	(175 %)	.023	.3225
LMB	.002	+-.005	(130 %)	24	+-55	(126 %)	.064	1.1567
WAE	*** NOT RECORDED ***			*** NOT RECORDED ***				
WAM	.000	+-.000	(213 %)		+-3	(213 %)	.002	.3091
WHC	.003	+-.008	(169 %)	18	+-41	(127 %)	.048	.5147
MSC	.000	+-.000	(213 %)		+-2	(213 %)	.002	.3247
OT	.829	.443-1.214	(46 %)	7718	3807-11629	(51 %)	20.310	.2403

HARVESTED AND CPUE TABLE BY SUBSTRATUM ACROSS STRATA *** DAY ***
REGION :=1 LAKE :=KEITHSBURG REFUGE
DISTRICT :=04 YEAR :=95
ACREAGE :380 SAMPLING RATIO :=55/159 = 34.5%
RATIO OF EFFORT HOURS INTERVIEWED := 1007.2/7748.9 = 12.99%
NUMBER OF INTERVIEWS: 351

COMBINED ACROSS STRATA:

YEAR PERIOD 01/01 TO 02/15 OF SECTION 1 COALESCED WITH
YEAR PERIOD 02/16 TO 02/22 OF SECTION 1

MSC SPECIES CAUGHT:
BLG LMB BLC CRP CAP WHC GWH WAM WAE
SUBSTRATUM:
DAY PERIODS STRATIFIED
WEEKDAY/WEEKEND: WEEKDAY/WEEKEND STRATIFIED
FISHING TYPE: BOAT/SHORE COALESCED
FISH: HARVESTED

SPEC	#/HR	95% CI		# HARVST	95% CI		#/HA	#/ACRE
BOW	.000	+-.000	(213 %)	2	+-7	(213 %)	.01	.00
WHB		*** NOT RECORDED ***			*** NOT RECORDED ***			
MSC	3.447	1.782-5.112	(48 %)	32117	15447-48786	(52 %)	208.84	84.52
TOT	3.447	1.782-5.112	(48 %)	32119	15450-48788	(52 %)	208.85	84.5[illegible]

SPEC	KG/HR	95% CI		KG HARVST	95% CI		KG/HA	AVG WT(G)
BOW	.000	+-.000	(213 %)		+-.96103	(213 %)	.002	147.3
WHB		*** NOT RECORDED ***			*** NOT RECORDED ***			
MSC	.376	.201-.551	(46 %)	3501	1726-5275	(51 %)	22.762	109.0
TOT	.376	.201-.551	(46 %)	3501	1727-5275	(51 %)	22.764	109.0

SPEC	LB/HR	95% CI		LB HARVST	95% CI		LB/ACRE	AVG WT(LB)
BOW	.000	+-.000	(213 %)		+-2	(213 %)	.002	.3247
WHB		*** NOT RECORDED ***			*** NOT RECORDED ***			
MSC	.829	.443-1.214	(46 %)	7717	3806-11628	(51 %)	20.309	.2403
TOT	.829	.443-1.214	(46 %)	7718	3807-11629	(51 %)	20.310	.2403

CAUGHT AND CPUE TABLE BY SUBSTRATUM ACROSS STRATA *** DAY ***
REGION :=1 LAKE :=KEITHSBURG REFUGE
DISTRICT :=04 YEAR :=95
AREAGE :380 SAMPLING RATIO :=55/159 = 34.5%
RATIO OF EFFORT HOURS INTERVIEWED := 1007.2/7748.9 = 12.99%
NUMBER OF INTERVIEWS: 351

COMBINED ACROSS STRATA:

YEAR PERIOD 01/01 TO 02/15 OF SECTION 1 COALESCED WITH
YEAR PERIOD 02/16 TO 02/22 OF SECTION 1

MSC SPECIES CAUGHT:
BOW WHB
SUBSTRATUM:
DAY PERIODS STRATIFIED
WEEKDAY/WEEKEND: WEEKDAY/WEEKEND STRATIFIED
FISHING TYPE: BOAT/SHORE COALESCED
FISH: CAUGHT

SPEC	#/HR	95% CI		# CAUGHT	95% CI		#/HA	#/ACRE
BLC	.087	.035-.140	(60 %)	864	459-1269	(47 %)	5.62	2.28
BLG	5.848	3.595-8.100	(39 %)	56460	30774-82146	(45 %)	367.14	148.58
CAP	.002	+-.006	(220 %)	12	+-31	(166 %)	.08	.03
CRP	.036	.000-.071	(98 %)	273	+-562	(106 %)	1.77	.72
GWH	.002	+-.007	(201 %)	27	+-71	(167 %)	.17	.07
LMB	.063	.017-.108	(72 %)	846	+-1723	(104 %)	5.50	2.23
WAE	.000	+-.000	(213 %)	3	+-10	(213 %)	.02	.01
WAM	.002	+-.004	(148 %)	27	+-66	(145 %)	.18	.07
WHC	.007	.000-.014	(92 %)	58	+-116	(101 %)	.38	.15
MSC	.001	+-.004	(212 %)	12	+-37	(206 %)	.08	.03
TOT	6.048	3.725-8.372	(38 %)	58582	31980-85183	(45 %)	380.93	154.16

SPEC	KG/HR	95% CI		KG CAUGHT	95% CI		KG/HA	AVG WT(G)
BLC	.010	.004-.016	(56 %)	99	53-144	(46 %)	.642	114.2
BLG	.437	.247-.627	(43 %)	4106	2122-6090	(48 %)	26.700	72.7
CAP	.001	+-.004	(186 %)	10	+-26	(154 %)	.066	862.8
CRP	.003	+-.005	(105 %)	20	+-43	(112 %)	.133	74.7
GWH	.000	+-.001	(211 %)	4	+-11	(175 %)	.025	146.3
LMB	.008	.003-.014	(67 %)	115	7-223	(94 %)	.747	135.9
WAE	.000	+-.000	(213 %)		+-2	(213 %)	.004	201.9
WAM	.000	+-.000	(141 %)	2	+-6	(163 %)	.014	82.3
WHC	.001	+-.004	(167 %)	8	+-19	(125 %)	.055	145.6
MSC	.000	+-.000	(201 %)	1	+-4	(194 %)	.008	106.5
TOT	.462	.266-.658	(42 %)	4367	2304-6430	(47 %)	28.395	74.5

SPEC	LB/HR	95% CI		LB CAUGHT	95% CI		LB/ACRE	AVG WT(LB)
BLC	.022	.010-.035	(56 %)	218	118-318	(46 %)	.573	.2518
BLG	.964	.546-1.383	(43 %)	9052	4678-13427	(48 %)	23.822	.1603
CAP	.003	+-.008	(186 %)	22	+-57	(154 %)	.059	1.9020
CRP	.006	+-.012	(105 %)	45	+-95	(112 %)	.118	.1646
GWH	.000	+-.002	(211 %)	9	+-24	(175 %)	.023	.3225
LMB	.018	.006-.030	(67 %)	253	16-491	(94 %)	.667	.2997
WAE	.000	+-.000	(213 %)	1	+-4	(213 %)	.004	.4451
WAM	.000	+-.000	(141 %)	5	+-13	(163 %)	.013	.1815
WHC	.003	+-.008	(167 %)	19	+-42	(125 %)	.049	.3210
MSC	.000	+-.000	(201 %)	3	+-8	(194 %)	.007	.2348
TOT	1.018	.586-1.450	(42 %)	9627	5079-14175	(47 %)	25.334	.1643

CAUGHT AND CPUE TABLE BY SUBSTRATUM ACROSS STRATA *** DAY ***
REGION :=1 LAKE :=KEITHSBURG REFUGE
DISTRICT :=04 YEAR :=95
ACREAGE :380 SAMPLING RATIO :=55/159 = 34.5%
RATIO OF EFFORT HOURS INTERVIEWED := 1007.2/7748.9 = 12.99%
NUMBER OF INTERVIEWS: 351

COMBINED ACROSS STRATA:

YEAR PERIOD 01/01 TO 02/15 OF SECTION 1 COALESCED WITH
YEAR PERIOD 02/16 TO 02/22 OF SECTION 1

MSC SPECIES CAUGHT:
BLG LMB BLC CRP CAP WHC GWH WAM WAE
SUBSTRATUM:
DAY PERIODS STRATIFIED
WEEKDAY/WEEKEND: WEEKDAY/WEEKEND STRATIFIED
FISHING TYPE: BOAT/SHORE COALESCED
FISH: CAUGHT

SPEC	#/HR	95% CI		# CAUGHT	95% CI		#/HA	#/ACRE
BOW	.000	+-.000	(213 %)	2	+-7	(213 %)	.01	.00
WHB	.001	+-.004	(245 %)	10	+-34	(245 %)	.06	.03
MSC	6.047	3.724-8.370	(38 %)	58569	31969-85170	(45 %)	380.85	154.13
TOT	6.048	3.725-8.372	(38 %)	58582	31980-85183	(45 %)	380.93	154.1

SPEC	KG/HR	95% CI		KG CAUGHT	95% CI		KG/HA	AVG WT(G)
BOW	.000	+-.000	(213 %)		+-.96103	(213 %)	.002	147.3
WHB	.000	+-.000	(245 %)		+-3	(245 %)	.006	98.0
MSC	.462	.266-.658	(42 %)	4365	2302-6428	(47 %)	28.386	74.5
TOT	.462	.266-.658	(42 %)	4367	2304-6430	(47 %)	28.395	74.5

SPEC	LB/HR	95% CI		LB CAUGHT	95% CI		LB/ACRE	AVG WT(LB)
BOW	.000	+-.000	(213 %)		+-2	(213 %)	.002	.3247
WHB	.000	+-.000	(245 %)	2	+-7	(245 %)	.006	.2159
MSC	1.018	.586-1.450	(42 %)	9624	5076-14172	(47 %)	25.326	.1643
TOT	1.018	.586-1.450	(42 %)	9627	5079-14175	(47 %)	25.334	.1643

TABLE KEITHSBURG REFUGE 1995 ICE CREEL DATA (1/1-2/22/95)
DAYTIME DATA FOR LAKE=KEITHSBURG REFUGE CREEL BEGUN IN YEAR=95

SECTION 1 FROM 01/01 TO 02/15
SECTION 1 FROM 02/16 TO 02/22

HOURS PER COMPLETED TRIP:

	MEAN	95% CONF.INTVL. OF MEAN			MIN.	MAX.	#SAMPLES
BOAT	4.1	3.3 -	5	(20%)	1.6	10.2	28
SHORE	***	NO DATA	***				
BOAT & SHORE	4.1	3.3 -	5	(20%)	1.6	10.2	28

11 SAMPLES WERE FROM SPLIT INTERVIEWS OF COMPLETED TRIPS
7.8% OF ALL 359 INTERVIEWS WERE COMPLETED TRIPS

SUPPLEMENTARY DATA:

QUESTION	MEAN	95% CONF.INTVL. OF MEAN			MIN.	MAX.	#SAMPLES
DISTANCE TRAVELLED IN MILES	49.3	44.1 -	54.5	(11%)	0	200	358
SUCCESS RATING 1-10?	5	4.7 -	5.2	(5%)	1	10	339

IS CATCH ILLEGAL?
CLERK NOTED 1 OUT OF 359 INTERVIEWS HAD ILLEGAL CATCHES

INTERVIEWS (AND %) PER SPECIES SOUGHT

ANY	119 (33.1%)	BLG	230 (64.1%)
CRP	9 (2.5%)	BLB	1 (.3%)

PARTY SIZE VS. # INTERVIEWS

	BOAT	SHORE
1	182	1
2	143	2
3	30	3
4		4
5	3	5
6	1	6
7		7
8		8
9		9
10+		10+

TABLE 1995 KEITHSBURG REFUGE ICE CREEL REPORT. CONTINUED
DAYTIME DATA FOR LAKE=KEITHSBURG REFUGE CREEL BEGUN IN YEAR=95

FISH HARVESTED/RELEASED BY # ANGLER-COMPLETED TRIPS FOR DIFFERENT TAXA

HARVESTED FISH:

ALL SIZES #LMB	#ANGLERS	ALL SIZES #BLG	#ANGLERS	ALL SIZES #BLG	#ANGLERS	ALL SIZES #GWH	#ANGLERS	ALL SIZES #GWH	#ANGLERS
0	38	0	4	16		0	40	16	
1	2	1		17	1	1		17	
2		2		18	1	2		18	
3		3	1	19		3		19	
4		4	2	20	1	4		20	
5		5	2	21		5		21	
6		6	3	22	1	6		22	
7		7	1	23		7		23	
8		8	3	24		8		24	
9		9		25	1	9		25	
10		10	2	26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30	1	14		30	
15+		15	2	31+	14	15		31+	

RELEASED FISH:

ALL SIZES #LMB	#ANGLERS	ALL SIZES #BLG	#ANGLERS	ALL SIZES #BLG	#ANGLERS	ALL SIZES #GWH	#ANGLERS	ALL SIZES #GWH	#ANGLERS
0	38	0	6	16		0	40	16	
1		1	1	17	3	1		17	
2		2		18		2		18	
3	1	3	2	19		3		19	
4	1	4	1	20	1	4		20	
5		5		21		5		21	
6		6	2	22		6		22	
7		7		23		7		23	
8		8	3	24		8		24	
9		9		25	2	9		25	
10		10	2	26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13	4	29		13		29	
14		14		30	1	14		30	
15+		15		31+	12	15		31+	

(TAXA FOR L.FREQS.= LMB BLG GWH)

TABLE 1995 KEITHSBURG REFUGE ICE CREEL REPORT. CONTINUED
DAYTIME DATA FOR LAKE=KEITHSBURG REFUGE CREEL BEGUN IN YEAR=95

SH HARVESTED/RELEASED BY # ANGLER-COMPLETED TRIPS FOR DIFFERENT TAXA

HARVESTED FISH:

ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES	
#BLC	#ANGLERS	#WHC	#ANGLERS	#WHC	#ANGLERS	#	#ANGLERS	#	#ANGLERS
0	29	0	39	16		0		16	
1	7	1	1	17		1		17	
2	3	2		18		2		18	
3		3		19		3		19	
4	1	4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

RELEASED FISH:

ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES	
#BLC	#ANGLERS	#WHC	#ANGLERS	#WHC	#ANGLERS	#	#ANGLERS	#	#ANGLERS
0	40	0	40	16		0		16	
1		1		17		1		17	
2		2		18		2		18	
3		3		19		3		19	
4		4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

(TAXA FOR L.FREQS.= BLC WHC CRP)

TABLE KEITHSBURG REFUGE 1995 ICE CREEL DATA (1/1-2/22/95). CONTINUED
DAYTIME DATA FOR LAKE=KEITHSBURG REFUGE CREEL BEGUN IN YEAR=95

FISH HARVESTED/RELEASED BY # ANGLER-COMPLETED TRIPS FOR DIFFERENT TAXA

HARVESTED FISH:
UNIDENTIFIED CRAPPIE

ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES	
#UCR	#ANGLERS	#	#ANGLERS	#	#ANGLERS	#	#ANGLERS	#	#ANGLERS
0	37	0	40	16		0	40	16	
1	3	1		17		1		17	
2		2		18		2		18	
3		3		19		3		19	
4		4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

RELEASED FISH:
UNIDENTIFIED CRAPPIE

ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES	
#UCR	#ANGLERS	#	#ANGLERS	#	#ANGLERS	#	#ANGLERS	#	#ANGLERS
0	36	0	40	16		0	40	16	
1	2	1		17		1		17	
2	1	2		18		2		18	
3	1	3		19		3		19	
4		4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

(TAXA FOR L.FREQS.= UCR)
↳ same as 'CRP' in HARVEST and CATCH tables)

TABLE KEITHSBURG REFUGE 1995 ICE CREEL DATA (1/1-2/22/95). CONTINUED
DAYTIME DATA FOR LAKE=KEITHSBURG REFUGE CREEL BEGUN IN YEAR=95

FISH HARVESTED/RELEASED BY # ANGLER-COMPLETED TRIPS FOR DIFFERENT TAXA

HARVESTED FISH:

ALL CRAPPIE (BLC+WHC+CRP-unknown)

ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES	
#CRP	#ANGLERS	#	#ANGLERS	#	#ANGLERS	#	#ANGLERS	#	#ANGLERS
0	26	0	40	16		0	40	16	
1	9	1		17		1		17	
2	4	2		18		2		18	
3		3		19		3		19	
4	1	4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

RELEASED FISH:

4. PPIE (BLC+WHC+CRP-unknown)

ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES	
#CRP	#ANGLERS	#	#ANGLERS	#	#ANGLERS	#	#ANGLERS	#	#ANGLERS
0	36	0	40	16		0	40	16	
1	2	1		17		1		17	
2	1	2		18		2		18	
3	1	3		19		3		19	
4		4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

(TAXA FOR L.FREQS.= CRP)

TABLE 1995 KEITHSBURG REFUGE ICE CREEL REPORT. CONTINUED
DAYTIME DATA FOR LAKE=KEITHSBURG REFUGE CREEL BEGUN IN YEAR=95

FISH HARVESTED/RELEASED BY # ANGLER-COMPLETED TRIPS FOR DIFFERENT TAXA

HARVESTED FISH:

ALL SIZES #WHB	#ANGLERS	ALL SIZES #BOW	#ANGLERS	ALL SIZES #BOW	#ANGLERS	ALL SIZES #WAM	#ANGLERS	ALL SIZES #WAM	#ANGLERS
0	40	0	40	16		0	40	16	
1		1		17		1		17	
2		2		18		2		18	
3		3		19		3		19	
4		4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

RELEASED FISH:

ALL SIZES #WHB	#ANGLERS	ALL SIZES #BOW	#ANGLERS	ALL SIZES #BOW	#ANGLERS	ALL SIZES #WAM	#ANGLERS	ALL SIZES #WAM	#ANGLERS
0	40	0	40	16		0	40	16	
1		1		17		1		17	
2		2		18		2		18	
3		3		19		3		19	
4		4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

(TAXA FOR L.FREQS.= WHB BOW WAM)

TABLE KEITHSBURG REFUGE 1995 ICE CREEL DATA (1/1-2/22/95) CONTINUED
DAYTIME DATA FOR LAKE=KEITHSBURG REFUGE CREEL BEGUN IN YEAR=95

JH HARVESTED/RELEASED BY # ANGLER-COMPLETED TRIPS FOR DIFFERENT TAXA

HARVESTED FISH:

ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES	
#WAE	#ANGLERS	#CAP	#ANGLERS	#CAP	#ANGLERS	#	#ANGLERS	#	#ANGLERS
0	40	0	37	16		0	40	16	
1		1	3	17		1		17	
2		2		18		2		18	
3		3		19		3		19	
4		4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

RELEASED FISH:

ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES		ALL SIZES	
#WAE	#ANGLERS	#CAP	#ANGLERS	#CAP	#ANGLERS	#	#ANGLERS	#	#ANGLERS
0	40	0	40	16		0	40	16	
1		1		17		1		17	
2		2		18		2		18	
3		3		19		3		19	
4		4		20		4		20	
5		5		21		5		21	
6		6		22		6		22	
7		7		23		7		23	
8		8		24		8		24	
9		9		25		9		25	
10		10		26		10		26	
11		11		27		11		27	
12		12		28		12		28	
13		13		29		13		29	
14		14		30		14		30	
15+		15		31+		15		31+	

TAXA FOR L.FREQS.= WAE CAP)

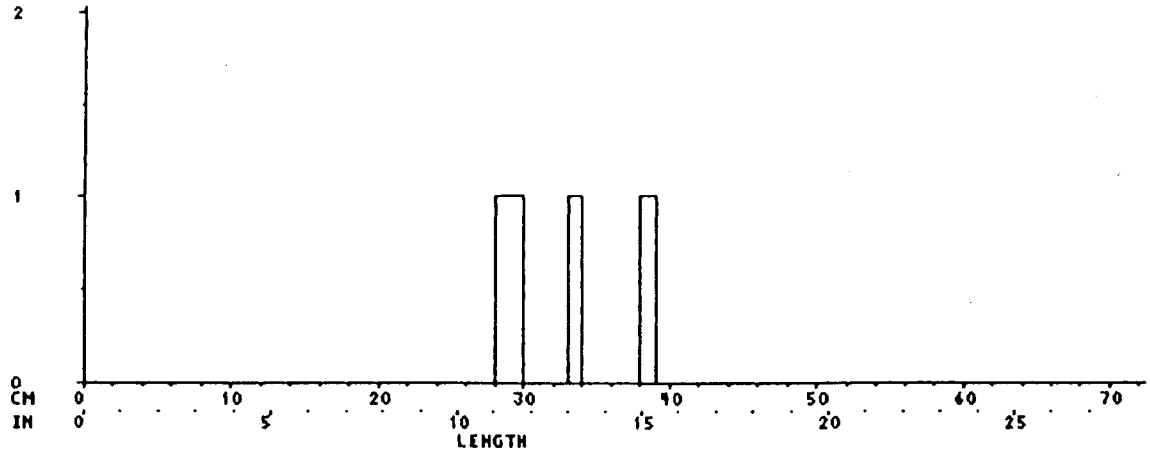


FIGURE 1. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. HARVESTED 'LMB' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 4 INDIVIDUALS

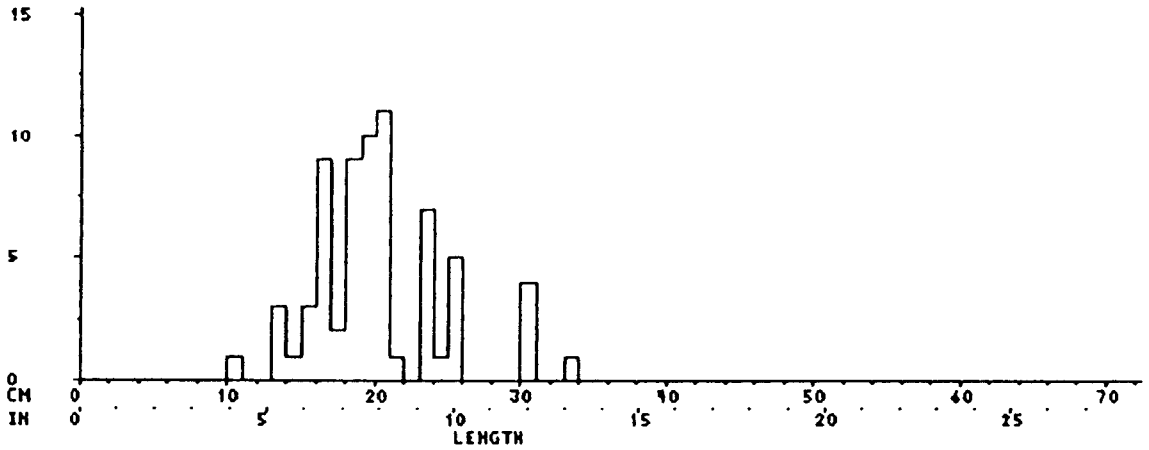


FIGURE 2. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. RELEASED 'LMB' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 68 INDIVIDUALS

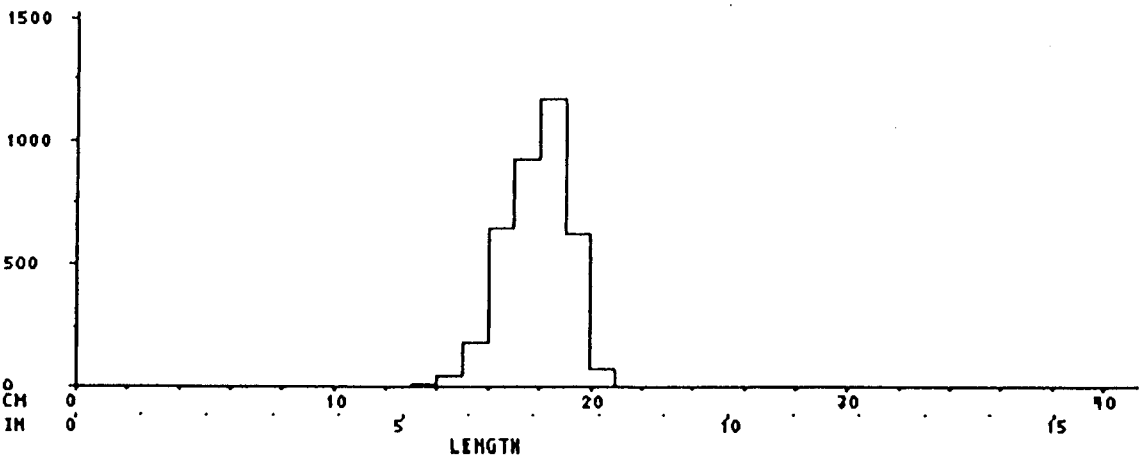


FIGURE 3. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. HARVESTED 'BLG' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 3659 INDIVIDUALS

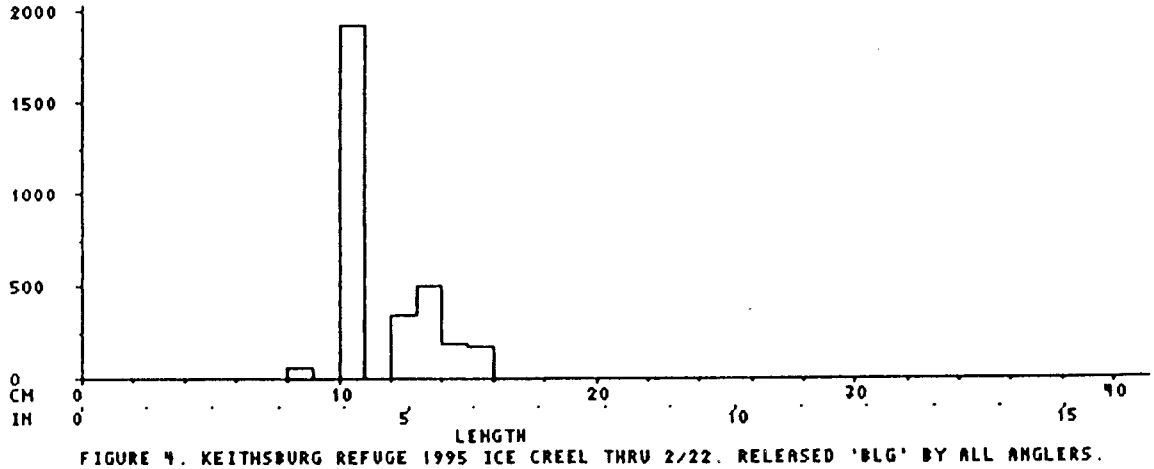


FIGURE 4. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. RELEASED 'BLG' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 3191 INDIVIDUALS

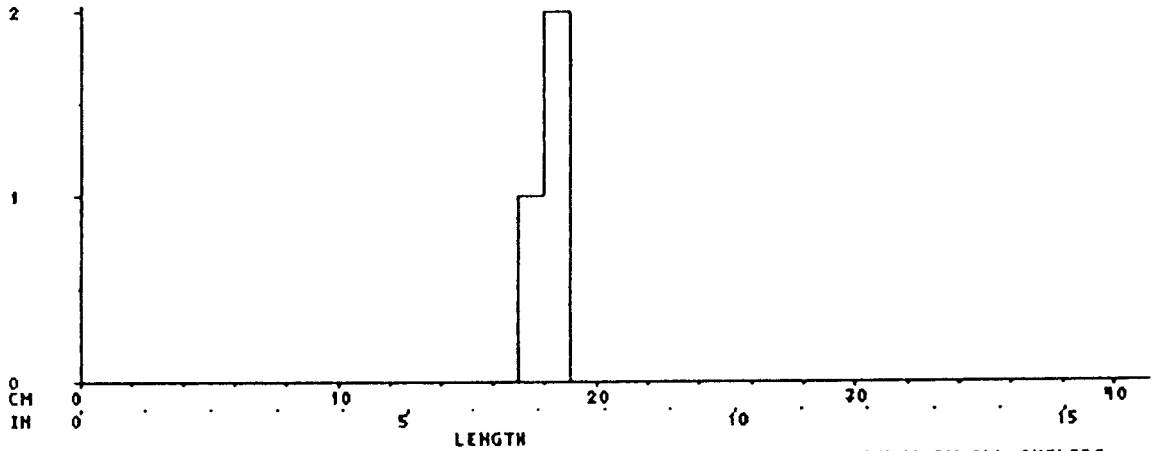


FIGURE 5. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. HARVESTED 'GWH' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 3 INDIVIDUALS

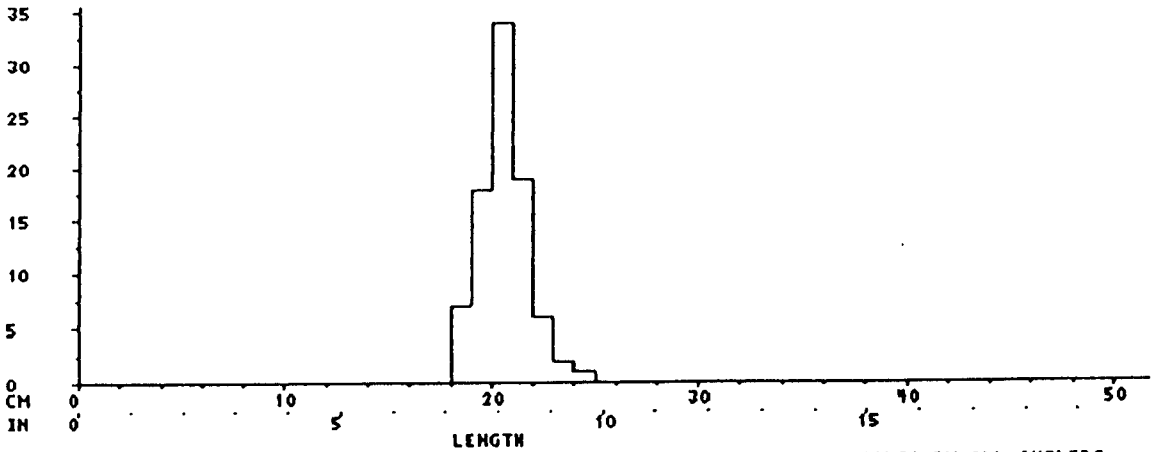


FIGURE 6. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. HARVESTED 'BLC' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 87 INDIVIDUALS

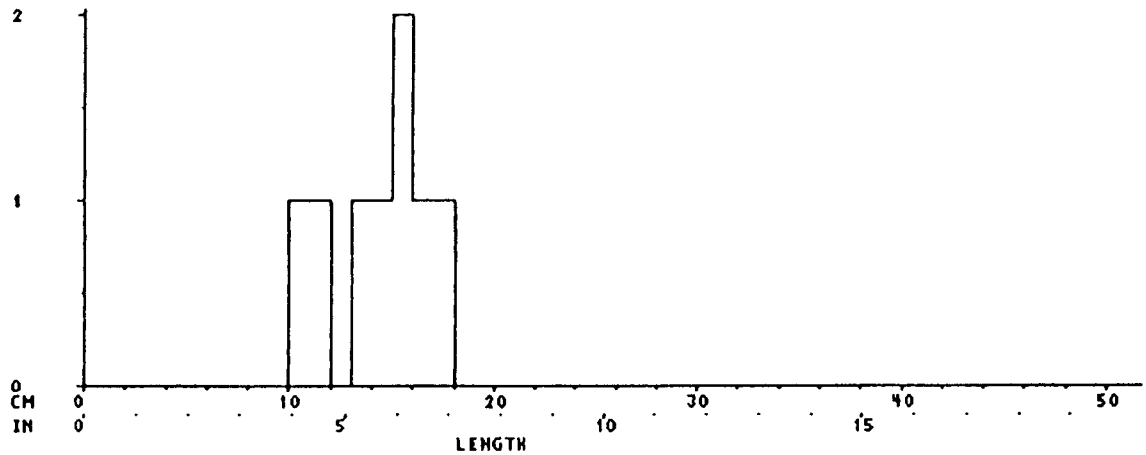


FIGURE 7. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. RELEASED 'BLC' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 8 INDIVIDUALS

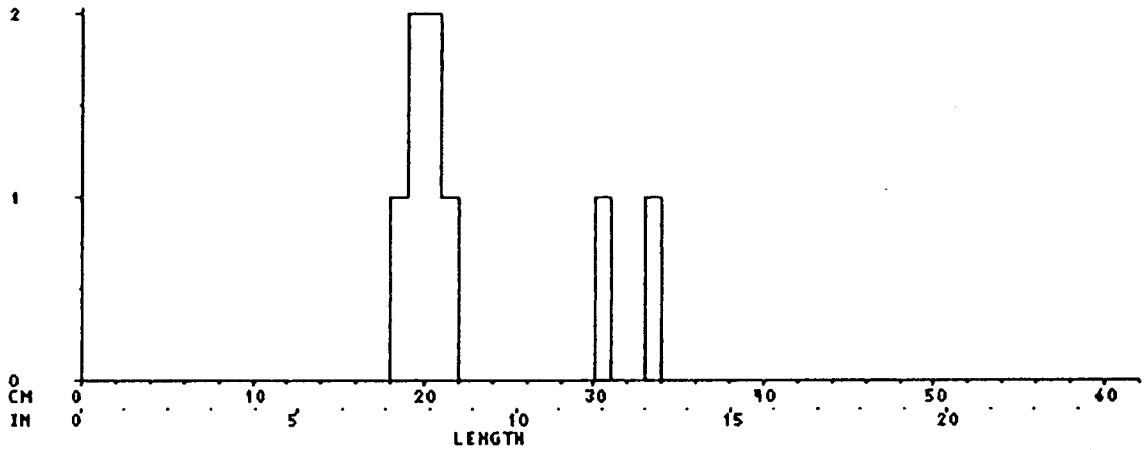


FIGURE 8. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. HARVESTED 'WHC' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 8 INDIVIDUALS

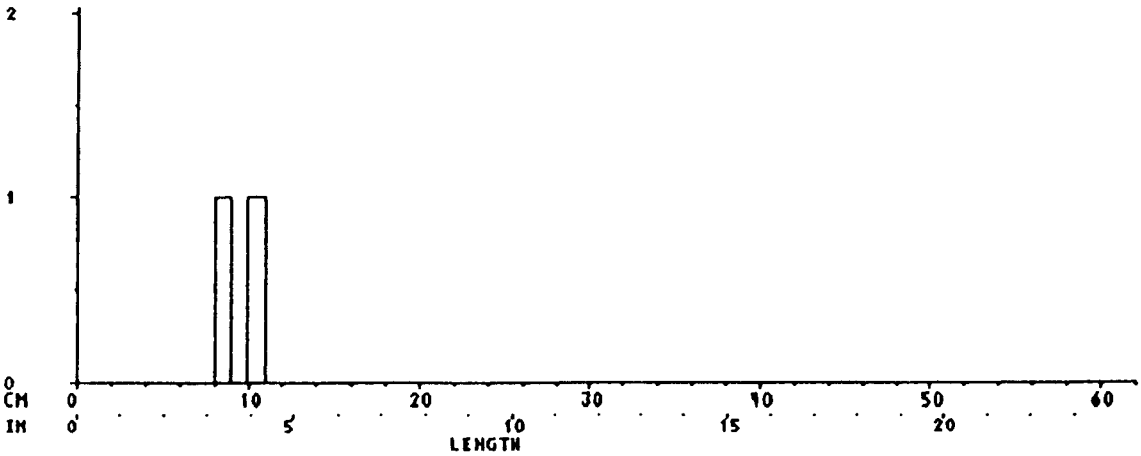


FIGURE 9. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. RELEASED 'WHC' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 2 INDIVIDUALS

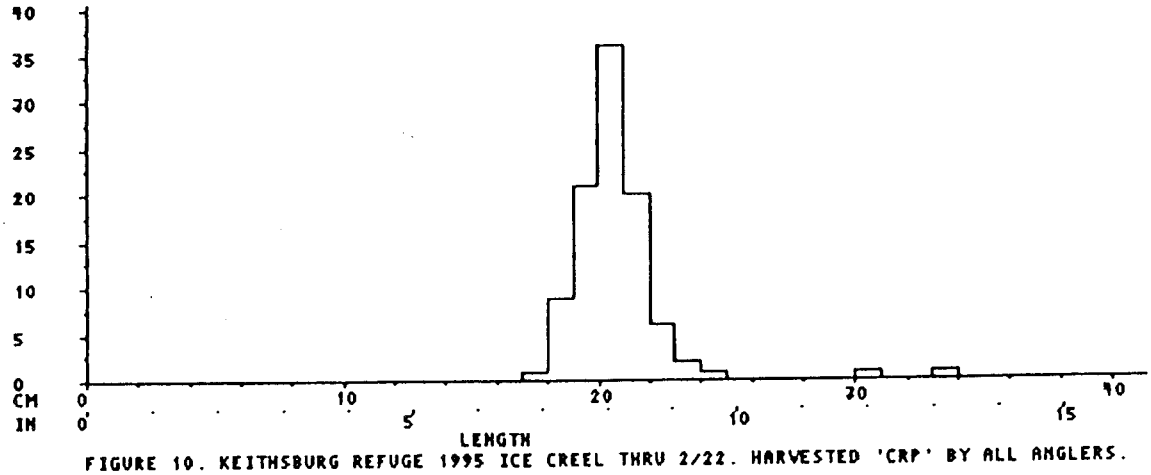


FIGURE 10. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. HARVESTED 'CRP' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 98 INDIVIDUALS

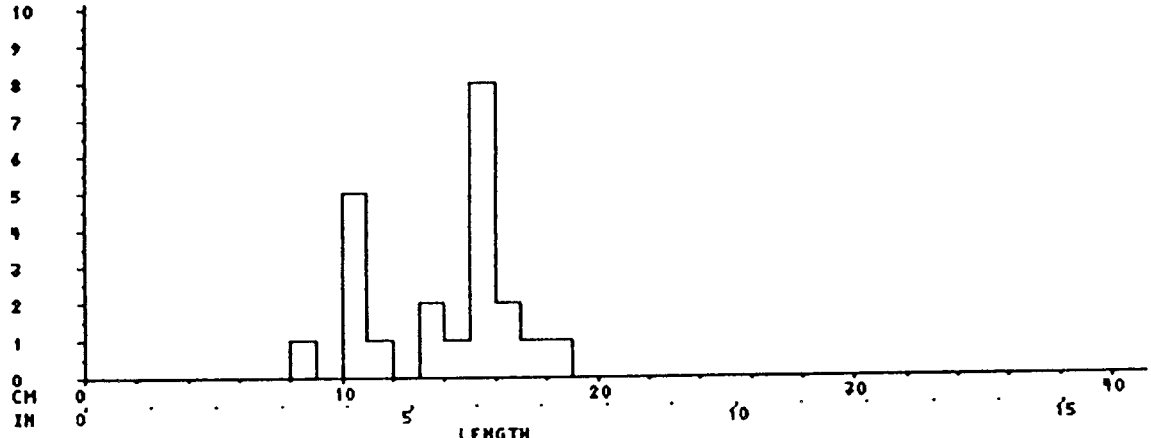


FIGURE 11. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. RELEASED 'CRP' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 22 INDIVIDUALS

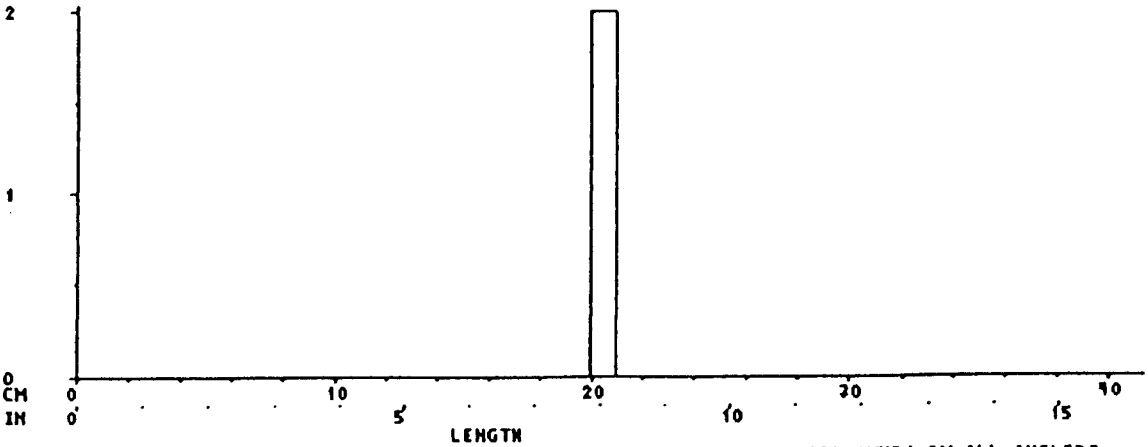


FIGURE 12. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. RELEASED 'WHB' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 2 INDIVIDUALS

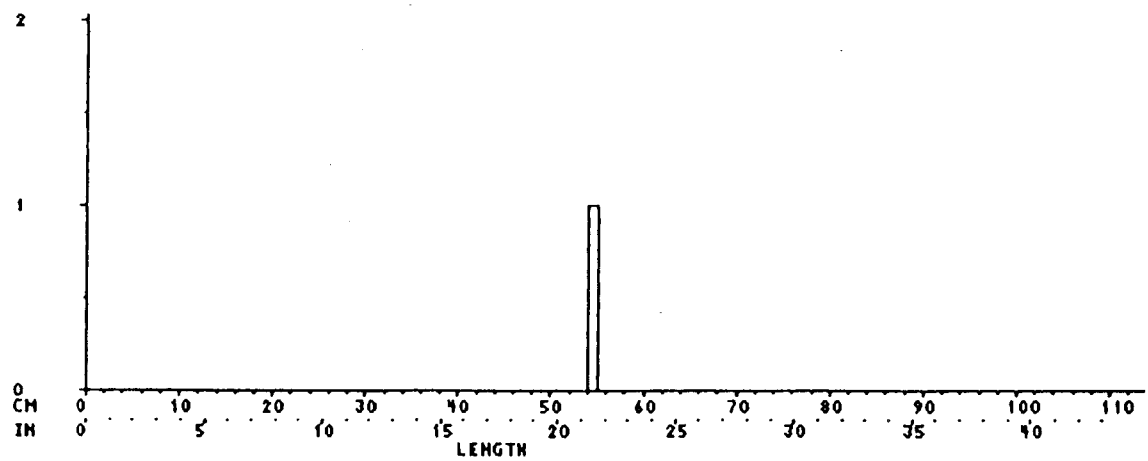


FIGURE 13. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. HARVESTED 'BOW' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 1 INDIVIDUALS

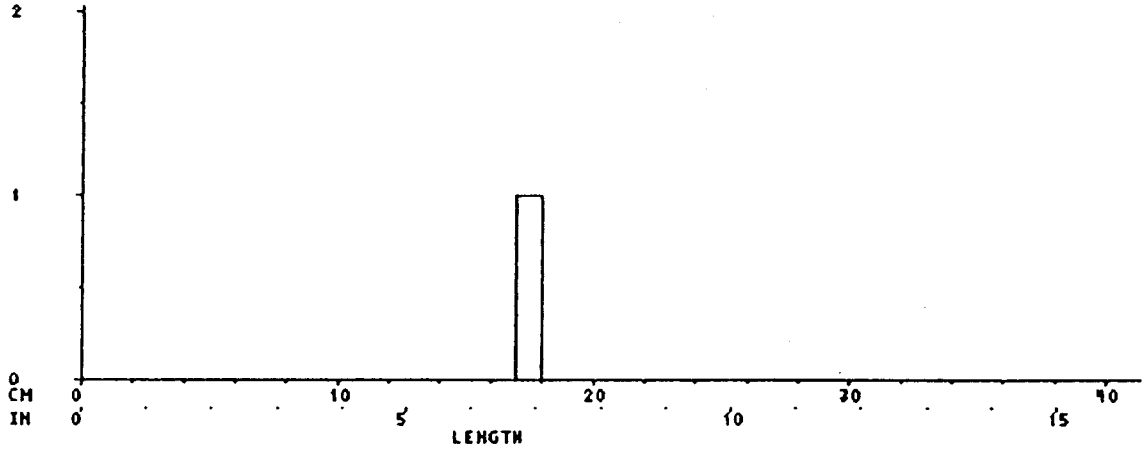


FIGURE 14. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. HARVESTED 'WAM' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 1 INDIVIDUALS

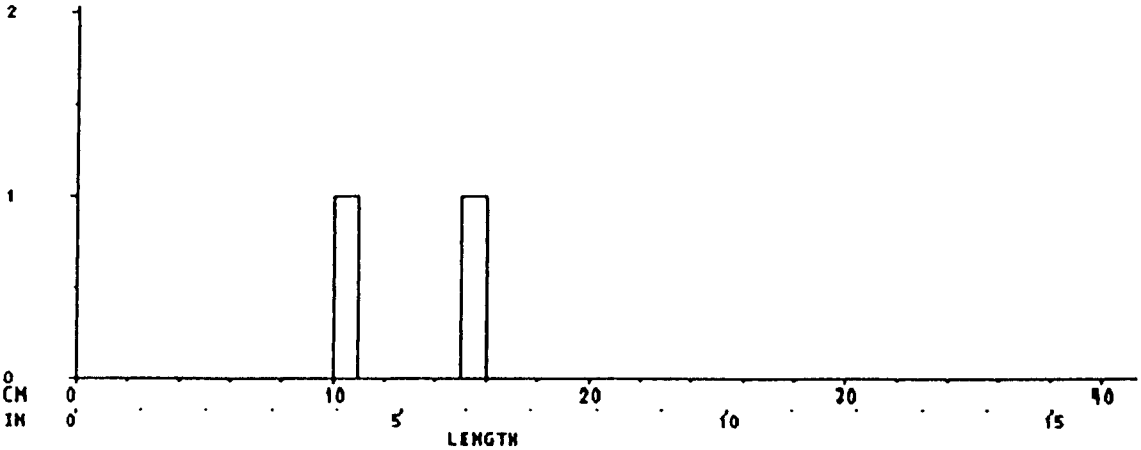


FIGURE 15. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. RELEASED 'WAM' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 2 INDIVIDUALS

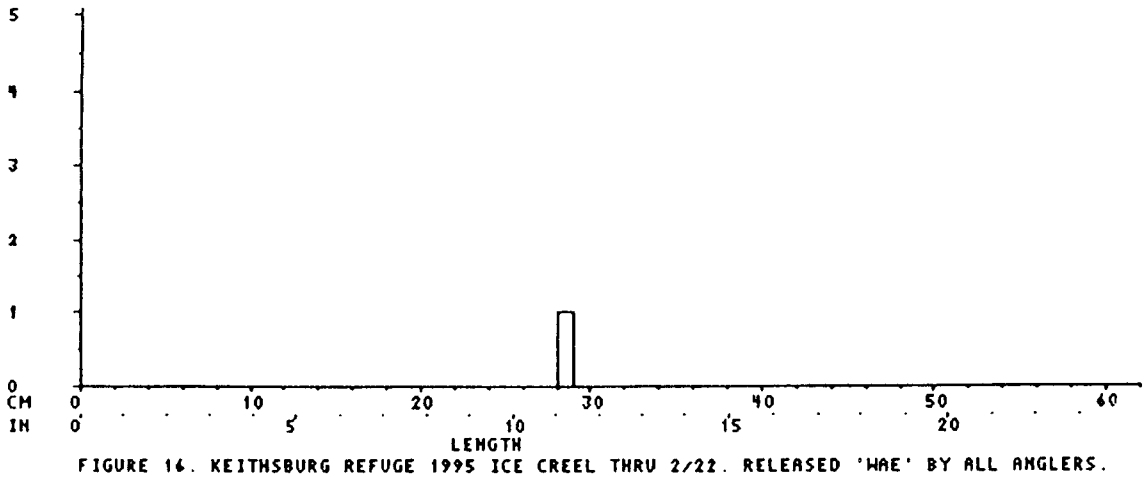


FIGURE 16. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. RELEASED 'WAE' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 1 INDIVIDUALS

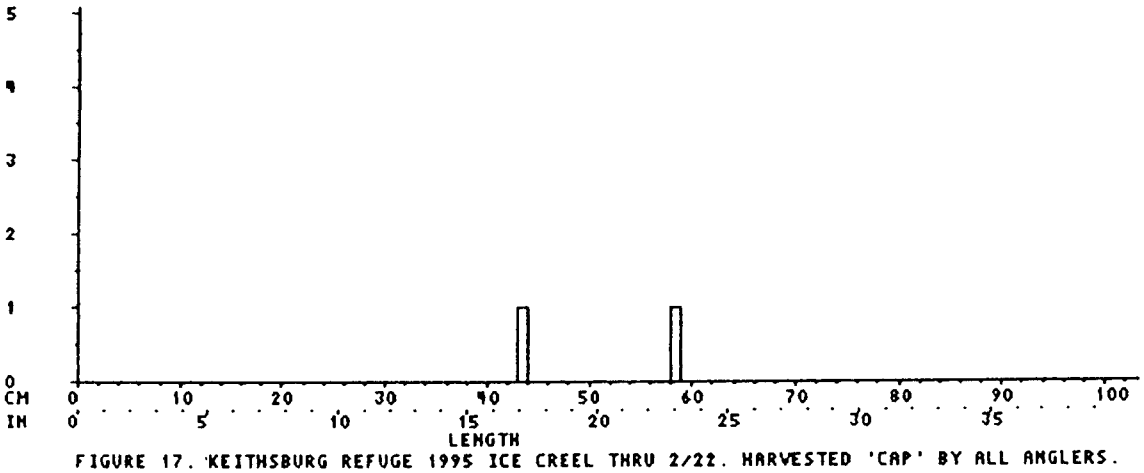


FIGURE 17. KEITHSBURG REFUGE 1995 ICE CREEL THRU 2/22. HARVESTED 'CAP' BY ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 2 INDIVIDUALS

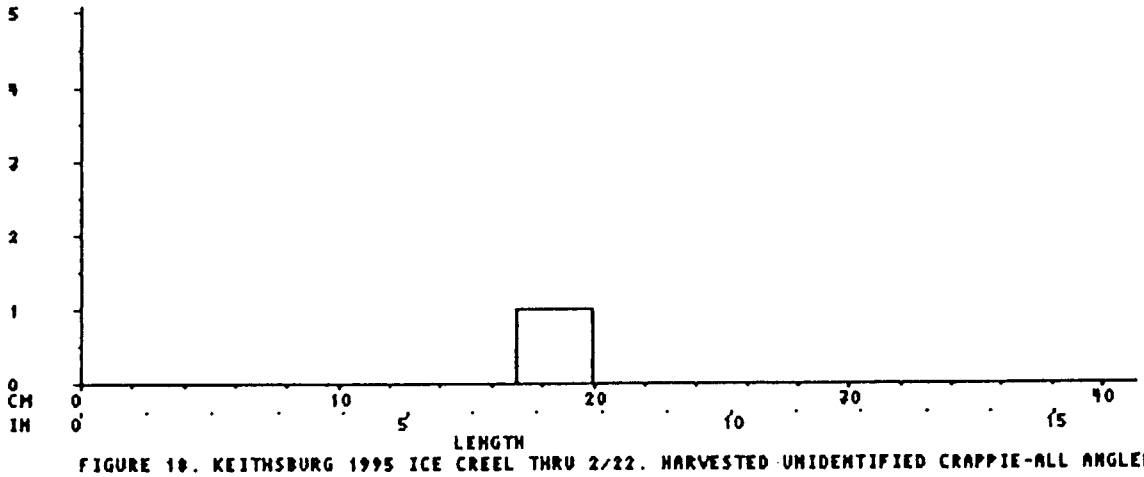


FIGURE 18. KEITHSBURG 1995 ICE CREEL THRU 2/22. HARVESTED UNIDENTIFIED CRAPPIE-ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 3 INDIVIDUALS

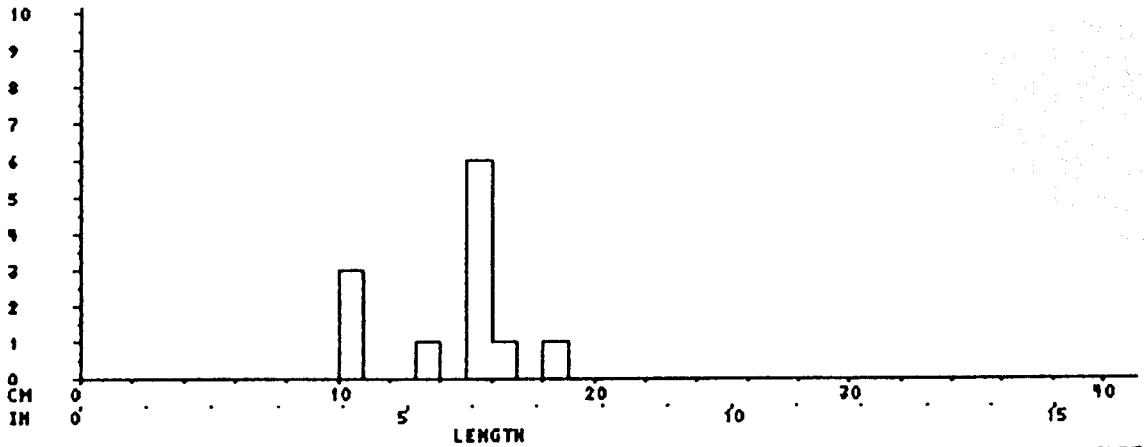
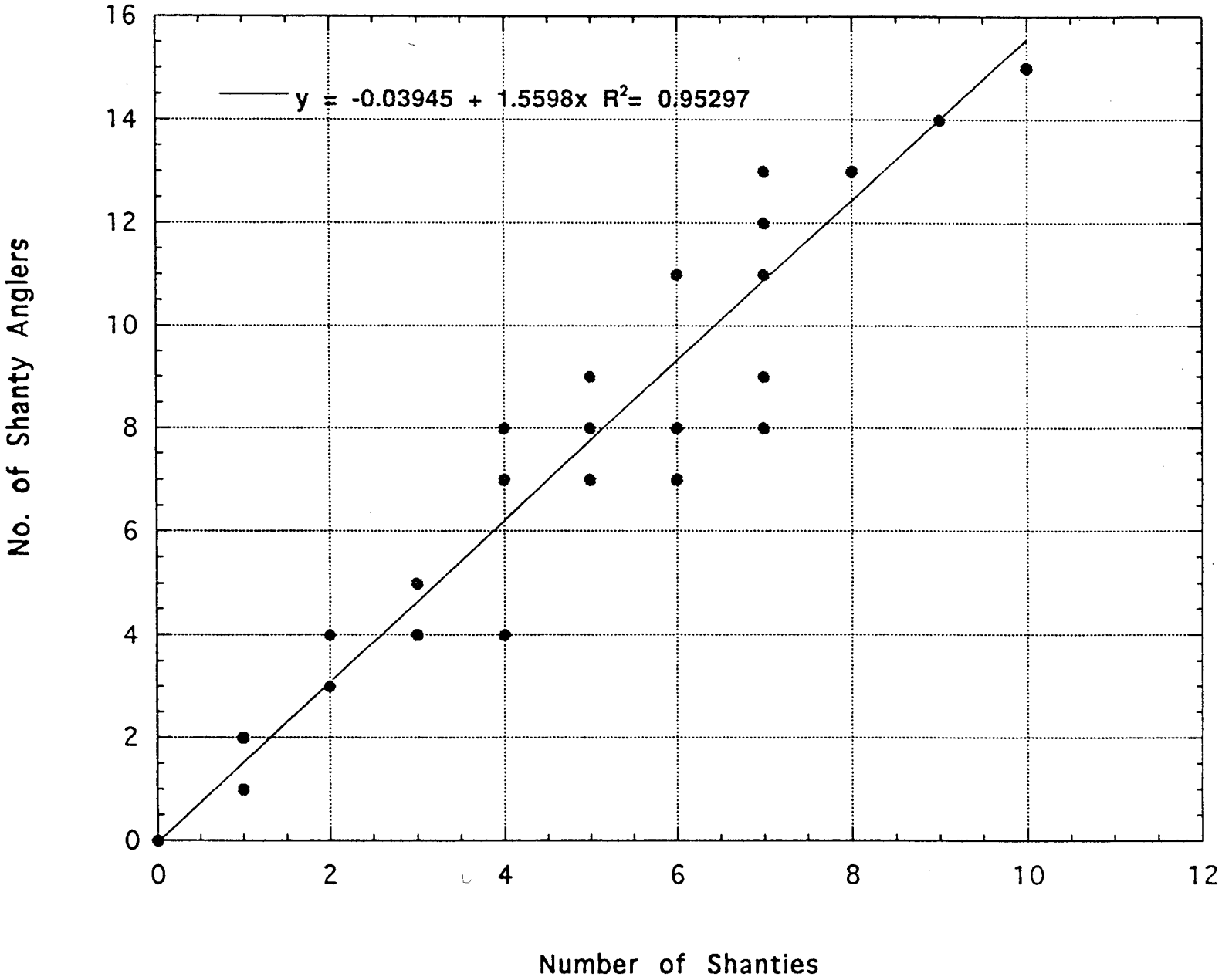
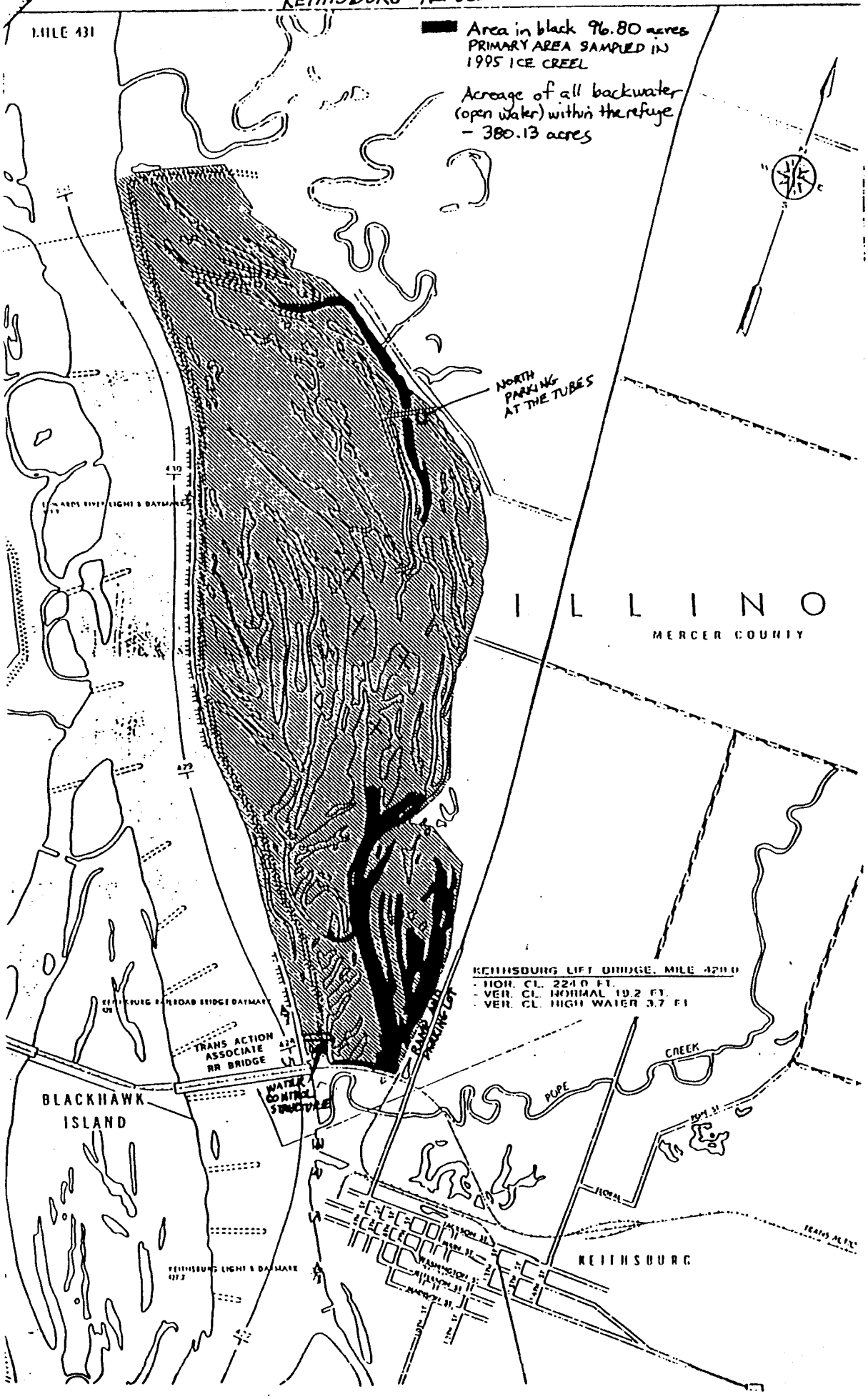


FIGURE 19. KEITHSBURG 1995 ICE CREEL THRU 2/22. RELEASED UNIDENTIFIED CRAPPIE-ALL ANGLERS.
FREQUENCY VS. FISH LENGTH 12 INDIVIDUALS

Keithsburg Refu 1995 Ice Creel Survey
Relationship between the No. of Ice Shanties
and the Total Number of Shanty Anglers
(4/17/95)



KEITHSBURG REFUGE



S.T. Sobaski - INHS Center for Aquatic Ecology 10/26/94

What's Included in the INHS Interim and Final Creel Reports

To help you interpret the Interim and Final Creel Reports from the Illinois Natural History Survey, we've included this guide to explain the contents of various pages. You will also find two companion documents enclosed: (1) the current list of three letter FAS Fish Species and Group codes and (2) a copy of the **Statistical Design and Calculation of Each Creel,** Appendix A. of the 1990 Illinois Natural History Survey report 90/10: **Creel Survey Manual for the District Fisheries Analysis System (FAS): A Package for Fisheries Management and Research.** This appendix describes how the creel data are collected, their subdivision for analysis by five different categories: specifically the Year Period, Lake Section, Time of Day (morning/midday/late afternoon), Day Type: (weekday/weekend and holidays), and Angler Type: (boat/shore) that the data were collected from (in other words, the stratification scheme applied to the creel data), and, for those of you so inclined, the statistical methodology used to calculate the estimated total hours of fishing, harvest, and catch.

Each creel report is composed of the following pages (in this chronological order):

I. EFFORT Table:

This first page, created by the District FAS-Apple *IIe* program **Final Effort**, reports the estimated total hours of fishing by all anglers for the Year Periods and Lake Sections listed near the top of the page. Unless otherwise noted, reports will always apply to all pole and line fishing activity on the entire lake. Lakes are split into several sections whenever (a) they're too large to allow for the completion of an Instantaneous Count of anglers within one hour or (b) if a lake area has some unique characteristic (e.g. the warm water arm of cooling lake).

As described in the **Statistical Design and Calculation of Each Creel,** the effort estimate, i.e. the estimated total hours of fishing, is calculated separately for boat and shore anglers as well as for all anglers for each day period sampled. These estimates are based on the instantaneous counts of anglers scaled up by the effective hours available for fishing for that time of day and year, rather than on the hours of fishing reported in angler interviews. An estimated total effort is then calculated for each combination (i.e. *stratum*) of Year Period x Lake Section x Day Period x Day Type x Angler Type by averaging the total hours of fishing from all days sampled within the stratum, then extrapolating that average by the total number of days within the stratum. Finally, each stratum total effort is added together to give the separate estimates of total hours of fishing for boat and shore anglers for the lake and time period of interest.

A weighted estimate of the total hours of fishing for all anglers is then calculated in two ways:

The first, **BOAT/SHORE COALESCED,** calculates an average total effort value for each stratum by (1) *combining the boat and shore instantaneous counts* of each sample,

then (2) multiplying this total count by the effective hours of fishing available for that day period to give the total hours of fishing in that day period (this is done for each day period-date sampled), (3) then averaging the total effort estimates of all samples of that day -period for each stratum (year period-weekday-lake section or year-period-weekend/holiday-lake section combination), and (4) extrapolating this average across all days sampled within that stratum. These stratum totals are then added together to give the total effort values for all anglers that you see reported in the table. The precision estimate of the total (the 95% confidence interval listed with the mean) is calculated directly from the combined variances of all strata. The implicit assumption in this approach is that there's no significant difference between the variability of boat versus shore fishing within the stratum, thus justifying combining the two counts together for each day period creeled. As you can see, this is sufficiently complex to require a computer program, STATCALC, to do these calculations. It also compels us to occasionally offer sacrifices to our Apple IIe's.

The second method, which we have found preferable for the better precision that it generally provides, is the **BOAT/SHORE STRATIFIED** approach to estimating total effort. Rather than combining the boat and shore I-counts of each sample and ignoring any potential difference in the day to day variability of boat vs. shore fishing, the stratified approach *first calculates separate estimates of total effort for boat and for shore anglers* for the entire time period being reported. These totals and their variances are then combined to give the overall total estimated hours of fishing.

As part of this project's research into obtaining more precise creel estimates we've found, with few exceptions, that the BOAT/SHORE STRATIFIED estimates are more precise than those provided by the COALESCED method. That is, the 95% confidence interval covers a smaller range of numbers around the estimated total. Consequently, you'll find the **BOAT/SHORE STRATIFIED:** totals circled on the Effort Table. We recommend that you use these values whenever reporting the total hours of fishing for a given lake and time period.

(a) Effort Table Header Information:

Most of the information given at the top of the page should be easily understood (e.g. lake name, region, district). The following are possible exceptions:

The information reported at the top of this page includes all of the individual combinations of "**YEAR PERIOD**" and lake "**SECTION**" included in the analysis. This same listing will also be found as part of the header information of the Harvest and Catch Tables as well as the first page of the Supplementary/Completed Trip/Length Frequency output. In cases where **COALESCED WITH** is printed after a YEAR PERIOD/SECTION line, data of this stratum were combined, during the analysis, with those of the next YEAR PERIOD/SECTION listed immediately below that line. Generally, this lumping of data into a single stratum is done only when a larger set of samples is required to calculate a variance term for the estimated mean effort, harvest, or catch of a single stratum. Data from different lake sections may also be coalesced for

a given year period if the lake has been sectioned solely to accommodate the angler instantaneous count time limit of one hour, rather than for biological or limnological reasons (such as retaining the warm water arm of a cooling lake as a separate stratum from other areas of the lake).

The **SAMPLING RATIO** value, listed directly below **YEAR**, is the ratio of the number of day periods worked vs. the total number of day-period samples covered by the creel survey analysis. In short, **the SAMPLING RATIO** gives an index of the intensity of the sampling schedule. For example, there may be 128 instantaneous counts taken between 3/15 - 6/15. Each count is specific to a third of a single date (i.e. a *day period*). To calculate the Sampling Ratio, the total number of day periods sampled is divided by the total number of day periods occurring during that span of dates. In this example, there are 93 days within the span of 3/15-6/15, thus 3 x 93 or 279 day periods. The Sampling Ratio = (128/279) X 100 or 45.8%.

(b) Effort Values:

Estimates of the total hours of fishing (the **ANGL HRS** column) by **Boat** anglers, **Shore** anglers, and **Total** anglers are reported in separate blocks in the table. The total estimates for each type of angler (the **STR TOTAL** line of each block) are further subdivided by *day type* (Weekday vs. Weekend/Holiday). The "**STR TOTAL**" is an abbreviation for Strata Total.

You'll notice **95% CONF INTVL** or **95% CI** columns following estimated totals such as **ANGL HRS**, on this and other report pages. These report the 95% confidence interval for the estimated totals. In other words, 95% percent of the time we'd expect the true total to fall somewhere within that given range. In cases where the lower limit of the confidence interval is a negative number, a "+" is printed.

The percentage listed in ( ) after the confidence interval is another indicator of the precision of the estimate. This percentage is calculated as the (Upper value of the 95% Confidence Interval - Estimated Total) / Estimated Total. The larger this percentage is, the more imprecise or less accurate the estimate. For example, if the Total Angler Hours Estimate is **30,293**, with an upper 95% confidence interval of **34,952**, the precision percentage is calculated as **(34,952 - 30,293)/30,293** or **15.38%**. The percentage is rounded to the nearest integer for the tabular output.

The **HRS/ACRE** column gives the Hours of Fishing per acre of lake. This is calculated by dividing the **ANGL HRS** value in each row by the **ACREAGE** value given at the top of the page.

The final column, **% EFF INTVD**, located on the right margin of the effort table, is the percentage of the estimated total effort actually accounted for by angler interviews. This number is calculated by summing the total hours of fishing reported by anglers from each stratum (i.e. day period - year period - weekday/weekend - boat/shore combination) and dividing it by the estimated total effort (calculated from the

instantaneous counts) for that period.

For instance, a total of 120 hours of weekday fishing might be reported by Boat anglers for Day Period 1 (sunrise-10:00 A.M.) between 6/1 - 6/15/94. The estimated total boat effort, however, based on the average boat angler instantaneous counts of Day Period 1 extrapolated by the 11 weekdays within 6/1-6/15/94, turns out to be 360 hours. The **% Effort Interview** value for this stratum would be: 120 hrs. of fishing (from interviews)/ 360 hrs. (based on I-counts)) x 100 = 33.33%. Like SAMPLING RATIO, this number gives an indication of the effectiveness of the sampling intensity. A higher % Effort Interview value indicates a more complete job of obtaining information on all of the angling activity for that type of angler. If you sampled every day within a stratum and interviewed every angler (in other words conducted a *census* rather than a survey), this percentage would approach or possibly exceed 100%.

II. HARVEST and CATCH Tables:

These tables make up the next two or more pages of the report, with the Harvest table(s) preceding the Catch table(s). Both tables are created by the District FAS-Apple *IIe* program **Final Catch**. The table's header information is identical to that of the effort page, except for the additional line of the total **RATIO OF EFFORT HOURS INTERVIEWED** (the total % EFF INTVD value from the bottom right of the Effort table) reported just below the **SAMPLING RATIO**.

These pages present estimates of fish harvest and catch (harvest + released fish combined). These estimates are given both as rates (**#/HR, KG/HR,** and **LB/HR**) and as totals (**# HARVST** or **CAUGHT, KG HARVST** or **CAUGHT,** and **LB HARVST** or **CAUGHT**) respectively. These estimates are computed for both individual species and for all fish creeled by all anglers.

The FAS creel analysis program, STATCALC, can produce only 9 separate species summaries per table (or per analysis) to accompany the Total (**TOT**) estimate of Harvest or Catch. This limitation stems from memory constraints with the of Apple *II*es, dating back to when 64KB was considered an abundant amount of memory in a computer. Consequently, to provide information on more individual species, you may find additional harvest and catch tables in your report. While each additional pair of harvest and catch tables has a different set of individual species reported, the **TOT** lines values will be identical. All fish species caught but not individually analyzed for a given harvest and catch analysis are lumped together by STATCALC as **MSC** (Miscellaneous species). A list of species making up this miscellaneous group is listed under "**MSC SPECIES CAUGHT:**". You'll find this just below the YEAR PERIOD ##/## - SECTION # listings near the top of each page.

One word of caution related to the miscellaneous species grouping. The **MSC SPECIES CAUGHT** line of both tables actually reports all other species caught (reported

as harvested and/or released by interviewed anglers) during the survey period being analyzed. The **MSC** line given above the totals **(TOT)** of the Harvest Table, therefore, may apply only to a subset of all of the species listed under **MSC SPECIES CAUGHT.**

- For example, the MSC SPECIES CAUGHT list seen on both the Harvest and Catch Tables might include BOW YLB WAM GSF CAP. However, of these 5 species, only Warmouth and Yellow Bass were harvested, while all BOW CAP and GSF were reported as released. Therefore, the **MSC** harvest summaries above the **TOT**(al) lines, would actually apply to only two rather than all five species listed in the MSC SPECIES CAUGHT. Since the Catch Table reports on harvest and released fish combined, the MSC line represents all species listed as **MSC SPECIES CAUGHT.**

As for the choice of species to be individually analyzed, we try to report on all games species in the lake plus any commonly caught species. Additional analyses for individual species can be run for any of our creel surveys. We encourage you to contact us if you would either like additional analyses of surveys that we've conducted in your district or if you're interested in attempting to use the FAS Creel software for the Apple *IIe* to run your own analyses. This software should be available at each district office and is also available directly from us.

Harvest/Catch Values:

The first block of values on the harvest and catch pages give:

SPEC - the FAS Fish Species code
#/HR - the NUMBER/HOUR estimate of fish harvested or caught.
95% CI - the 95% Confidence Interval around that HPUE or CPUE value.
 # HARVST or CAUGHT - the estimate of the Total Number of Fish Harvested or Caught.
95% CI - the 95% Confidence Range around the estimated Total Number of fish.
#/HA - the Number of Fish/Hectare harvested or caught.
#/ACRE - the Number/Acre harvested or caught.

In cases where an individual species is analyzed, but either (*a*) was not reported by anglers as harvested or (*b*) not reported at all in any interview, you'll see "*** **NOT RECORDED** ***" displayed. This is most commonly seen in the Harvest table, where all fish of a given species have been caught, but not kept. In these cases, all estimates in the Harvest Table for that species will appear as *** **NOT RECORDED** ***, while numbers will appear in the Catch Table for #/HR, #CAUGHT, KG CAUGHT, etc. for that species. In cases where a species was neither reported as harvested or released, then *** **NOT RECORDED** *** will appear in *both* the Harvest and Catch Tables for all estimates of that species .

Rate estimates (**#/HR, KG/HR, LB/HR**) with a value of .000, have a harvest or catch rate that is **less than 0.001** fish/hr or lbs/hr but **greater than zero.** A *zero* rate is

reported as *** NOT RECORDED ***, rather than as ".000". If a BLANK space appears for an estimate, this indicates that the estimate is so small than it can't be printed for the table. For instance, a total **KG CAUGHT** or **LB CAUGHT** estimate that is less than 1 will be reported as a blank. This is also true for any **#/HA** or **#/ACRE** estimate less than **0.01**, and **LB/HA, LB/ACRE, KG/HA,** or **KG/ACRE** estimates less than **0.001.**

Following the block of abundance (number of fish) estimates, each table presents two blocks which summarize the total weight of fish harvested or caught.

The first of these presents biomass estimates in metric units (**KG** = kilograms). The other block (the last part of the table) presents the same information in English units (**LB** = pounds). Rather than measuring fish weights directly during interviews, weights are estimated based on the standard length to weight relationships (Weight = a * Total Length $^{b)}$ developed for each species from IDOC population survey data stored in the Illinois STATE FAS database or from fisheries literature. Average fish weights reported in the far right column (**AVG WT (G)** and **AVG WT(LB)**) are calculated by dividing the estimated total biomass caught (e.g. **KG HARVST**) by the estimated total number caught (e.g. **# HARVST**) for that species.

III. SUPPLEMENTARY output pages:

The pages following the effort, harvest, and catch tables summarize various data collected during angler interviews such as: (a) the average length of fishing trips, (b) the average distance travelled to the lake by fisherman, (c) the average success rating that anglers gave to their fishing trip, (d) the number of illegal catches noted by the clerk, (e) a breakdown of the species being targeted by fishing parties, (f) a breakdown of the size of fishing parties interviewed, (g) a breakdown of the success of completed trip anglers in catching individual species or species groups, and finally (h) length frequencies histograms for individual species or species groups harvested and released. Numbers reported here differ from those of the previous tables since these numbers are unweighted averages based solely on interview data rather than estimated totals for an entire year. Rather than stratifying these data as is done for the effort, harvest, and catch estimates, these tables take all interview data, combine it regardless when it was collected during the survey, and report simple averages. These tables are all generated by the District FAS-Apple *IIe* program **Comp.Trip/Supp/L-Freq.**

(A.) Supplementary Interview Data Summaries:

The first page of supplementary output presents a list of SECTIONS and YEAR PERIODS included in the analysis followed by:

(1) Hours Per Completed Trip -

This is the average length of completed boat and shore trips. These averages are

based on the length of the fishing trip per angler party rather than per angler. By definition, this excludes all interviews were anglers intended to continue fishing (incomplete trips). With each mean, the 95% Confidence interval is present in the same format as the previous tables, followed by the minimum and maximum times of completed trips and the number of interviews included in the calculation.

The last two lines of this first section report the number of completed trips that spanned more than one day period and proportion of all interviews that were of completed trips.

(2) **Supplementary Data Question-**

This block summarizes two additional questions asked of all parties:
(a) the average distance (miles) that members of each fishing party travelled to the lake.
(b) how they rated the success of their fishing trip on a scale of 1-10, where 10 is the highest rating.

The averages reported here are based on <u>All</u> interviews, rather than on just completed trip interviews.

This block also reports the number of parties with illegal harvests noted by the creel clerk.

(3) **# Interviews (and %) Per Species Sought -**

This summarizes the Fish Species or Species Group that fishing parties are targeting. "ANY" represents cases were no specific species/group was targeted. This breakdown gives the FAS fish code followed by the number of parties that were targeting that taxon, plus what percentage of all parties were targeting this taxon. This includes all interviews - both completed and incomplete trips.

(4) **Party Size vs. # Interviews-**

This presents a breakdown of all interviews based on the size of the fishing party interviewed for both boat and shore anglers. Party size is given in the left column - number of interviews in the right. All interviews are included in this summary.

(B.) # FISH HARVESTED/ RELEASED BY # ANGLER-COMPLETED TRIPS FOR DIFFERENT TAXA Table

This next set of supplementary pages report on all anglers from all <u>completed trip</u> interviews only. It examines each interview for the number of fish of a single species or species group reported as harvested and released . It then calculates the average harvest and catch per angler by dividing the total number harvested and the total released for that species by the number of anglers in the party. The table reports the #ANGLERS, broken down by their catch rate. Catch rates are given in the left column

under #SPP.

The table reports on up to three species or species groups (user-selected prior to beginning the analysis), with the number of fish/angler ranging from 1-15+ for the first taxon, and 1-31+ for the other two. The top half of the table applies only to harvested fish, the lower half just to released fish. Any catch rate greater than 14 fish/angler for the first species or 30 fish/angler for the second and third species is pooled together in the 15+ or 31+ category.

An example of this table, for walleye reported as harvested in 500 completed trip interviews, might be:

#WAE	#ANGLERS
0	651
1	50
2	7
3	0
....	....
15+	0

The 500 completed trip interviews actually cover the catch of 708 anglers in this case, since a number of angler parties had more than one fisherman. Of these 708 anglers, 651 completed trip anglers reported no Walleye harvested on their trip (or averaged less than 1 fish/angler/angler party), 50 fishermen were in parties that averaged 1 walleye harvested/angler, and 7 anglers were part of trips that averaged 2 walleye harvested/angler. No angler averaged more than two walleye harvested for their party's trip.

(C.) Fish Length Frequency Histograms for Harvested and Released Fish.

The final pages of the creel report present Length-Frequency histograms for Harvested and Released fish for all species or species groups reported in the #FISH/ COMPLETED TRIP ANGLER tables just described. Unlike that table, the fish reported in each plot are taken from ALL interviews, not just completed trip interviews. The number of fish reported for each centimeter total length group are the actual totals of fish reported in interviews rather than estimated totals as are reported in the Harvest and Catch tables. There will be a pair of plots per species analyzed, except in cases were fish either weren't harvested or weren't released. The first plot displays harvested fish only while the second released fish only.

Hopefully, this discussion has made interpreting your creel report an easier task. However, if you have any further questions on the contents of any INHS creel report or on any of the explanations given within this document, contact the creel project staff at the Illinois Natural History Survey (especially those personnel listed on the cover page of your report). They'll be happy to discuss any of your questions.

FAS Fish Species Codes

ABL - AMERICAN BROOK LAMPREY
ALE - ALEWIFE
ALG - ALLIGATOR GAR
ALS - ALABAMA SHAD
AME - AMERICAN EEL

BAD - BANDED DARTER
BAK - BANDED KILLIFISH
BAM - BRASSY MINNOW
BAS - BANDED SCULPIN
BBD - BLUEBREAST DARTER
BCF - BLUE CATFISH
BCS - BLACKCHIN SHINER
BGB - BIGMOUTH BUFFALO
BGC - BIGEYE CHUB
BGH - Hybrid BLUEGILL x GREEN SUNFISH
BGS - BIGEYE SHINER
BHC - BIGHEAD CARP
BHS - BLUEHEAD SHINER
BKB - BLACK BUFFALO
BKC - BLACKFIN CISCO
BKD - BLACKNOSE DACE
BKS - BROOK STICKLEBACK
BKT - BROOK TROUT
BLB - BLACK BULLHEAD
BLC - BLACK CRAPPIE
BLD - BLACKSIDE DARTER
BLG - BLUEGILL
BLO - BLOATER
BLR - BLACK REDHORSE
BLS - BLUNTNOSE MINNOW
BLT - BLACKSTRIPE TOPMINNOW
BMS - BIGMOUTH SHINER
BNS - BLACKNOSE SHINER
BOH - Hybrid BLUEGILL x ORANGESPOTTED SUNFISH
BOW - BOWFIN
BPS - BANDED PYGMY SUNFISH
BRB - BROWN BULLHEAD
BRH - Hybrid BLUEGILL x REDEAR SUNFISH
BRM - BRINDLED MADTOM
BRS - BROOK SILVERSIDE
BRT - BROWN TROUT
BSF - BANTAM SUNFISH
BSH - Hybrid BLACKSIDE x SLENDERHEAD DARTER
BSS - BLACK BASS spp.
BST - BLACKSPOTTED TOPMINNOW
BTS - BLACKTAIL SHINER
BUB - BURBOT
BUD - BLUNTNOSE DARTER
BUF - UNIDENTIFIED BUFFALO sp.
BUL - BULLHEAD spp.
BUM - BULLHEAD MINNOW
BUS - BLUE SUCKER
BWH - Hybrid BLACK x WHITE CRAPPIE

CAP - CARP
CAR - UNIDENTIFIED CARPSUCKER sp.
CCF - CHANNEL CATFISH
CCS - CREEK CHUBSUCKER
CEM - CENTRAL MUDMINNOW
CGH - Hybrid GOLDFISH x CARP
CGH - Hybrid CARP X GOLDFISH
CHL - CHESTNUT LAMPREY
CHO - COHO SALMON
CMS - COMMON SHINER
COS - CENTRAL STONEROLLER
CRC - CREEK CHUB
CRD - CRYSTAL DARTER
CRP - CRAPPIE spp.
CSC - CISCO
CYD - CYPRESS DARTER
CYM - CYPRESS MINNOW

DAR - DARTER spp.
DUD - DUSKY DARTER

EMS - EMERALD SHINER
ESD - EASTERN SAND DARTER

FAD - FANTAIL DARTER
FCF - FLATHEAD CATFISH
FCS - FALL CHINOOK SALMON
FHM - FATHEAD MINNOW
FLC - FLATHEAD CHUB
FLR - FLIER
FOS - FOURHORN SCULPIN
FRD - FRESHWATER DRUM
FRM - FRECKLED MADTOM

GAR - UNIDENTIFIED GAR sp.
GBH - Hybrid GRASS CARP x BIGHEAD CARP
GHS - GHOST SHINER
GLD - GILT DARTER
GOF - GOLDFISH
GOH - Hybrid GREEN SUNFISH x ORANGESPOTTED SUNFISH
GOL - GOLDEYE
GOR - GOLDEN REDHORSE

Fish Species Groups are listed in ***BOLD ITALIC***. Individual Species names are given in PLAIN text.

FAS Fish Species Codes sorted by FAS code

Last Updated: 4/11/95

GOS - GOLDEN SHINER
GRC - GRASS CARP
GRD - GREENSIDE DARTER
GRP - GRASS PICKEREL
GRR - GREATER REDHORSE
GSF - GREEN SUNFISH
GTH - Hybrid GIZZARD SHAD x THREADFIN SHAD
GVC - GRAVEL CHUB
GWH - Hybrid GREEN SUNFISH x WARMOUTH
GZS - GIZZARD SHAD

HAD - HARLEQUIN DARTER
HFC - HIGHFIN CARPSUCKER
HOC - HORNYHEAD CHUB
HSH Hybrid HORNYHEAD CHUB x STRIPED SHINER

INS - INLAND SILVERSIDES

JOD - JOHNNY DARTER

LAC - LAKE CHUB
LAM - UNIDENTIFIED LAMPREY sp.
LAS - LAKE STURGEON
LAT - LAKE TROUT
LAW - LAKE WHITEFISH
LBH - Hybrid LONGEAR SUNFISH x BLUEGILL
LBL - LEAST BROOK LAMPREY
LCS - LAKE CHUBSUCKER
LED - LEAST DARTER
LGD - LONGNOSE DACE
LGH - Hybrid LONGEAR SUNFISH x GREEN SUNFISH
LMB - LARGEMOUTH BASS
LNS - LONGNOSE SUCKER
LOG - LONGNOSE GAR
LOP - LOGPERCH
LOS - LONGEAR SUNFISH
LPH - Hybrid LONGEAR SUNFISH x PUMPKINSEED
LSS - LARGESCALE STONEROLLER

MAD - MADTOM spp.
MIN - NON-CARP MINNOW spp.
MMH - MEANMOUTH BASS (LARGEMOUTH x SMALLMOUTH BASS Hybrid)
MMS - MIMIC SHINER
MOF - MOSQUITOFISH
MOM - MOUNTAIN MADTOM
MOO - MOONEYE
MOX - UNIDENTIFIED REDHORSE sp.
MSS - MISSISSIPPI SILVERSIDES
MTS - MOTTLED SCULPIN
MUD - MUD DARTER
MUE - MUSKELLUNGE

NBL - NORTHERN BROOK LAMPREY
NHS - NORTHERN HOG SUCKER
NOM - NORTHERN MADTOM
NOP - NORTHERN PIKE
NOS - NORTHERN STUDFISH
NSS - NINESPINE STICKLEBACK

ORD - ORANGETHROAT DARTER
ORS - ORANGESPOTTED SUNFISH
OWD - IOWA DARTER
OZM - OZARK MINNOW

PAH - PADDLEFISH
PAS - PALLID STURGEON
PBH - Hybrid PUMPKINSEED x BLUEGILL
PBH - Hybrid BLUEGILL x PUMPKINSEED
PGH - Hybrid PUMPKINSEED x GREEN SUNFISH
PLM - PLAINS MINNOW
PLS - PALLID SHINER
PRP - PIRATE PERCH
PUD - PUMPKINSEED
PUM - PUGNOSE MINNOW
PUS - PUGNOSE SHINER
PWH - Hybrid PUMPKINSEED x WARMOUTH

RAD - RAINBOW DARTER
RAS - RAINBOW SMELT
RBS - RIBBON SHINER
RBT - RAINBOW TROUT
RCS - IRONCOLOR SHINER
RDS - REDFIN SHINER
RES - RED SHINER
RGH - Hybrid REDEAR SUNFISH x GREEN SUNFISH
RLH - Hybrid REDEAR SUNFISH x LONGEAR SUNFISH
ROB - ROCK BASS
ROS - ROSEFIN SHINER
ROW - ROUND WHITEFISH
RRC - RIVER CHUB
RSF - REDEAR SUNFISH
RSH - Hybrid RED SHINER x SPOTFIN SHINER
RUD - RUDD
RUH - Hybrid RED SHINER x Notropis sp.
RVC - RIVER CARPSUCKER
RVD - RIVER DARTER
RVR - RIVER REDHORSE
RVS - RIVER SHINER
RWH - Hybrid REDEAR SUNFISH X

Fish Species Groups are listed in ***BOLD ITALIC***. Individual Species names are given in PLAIN text.

WARMOUTH
RYS - ROSYFACE SHINER

SAB - SMALLMOUTH BUFFALO
SAR - SAUGER
SAS - SAND SHINER
SBH - Hybrid STRIPED BASS x WHITE BASS
SCP - SILVER CARP
SCS - SPRING CHINOOK SALMON
SDS - SPOTTED SUCKER
SEL - SEA LAMPREY
SES - STEELCOLOR SHINER
SFH - UNIDENTIFIED Hybrid SUNFISH
SFS - SPOTFIN SHINER
SGC - STURGEON CHUB
SGD - STARGAZER DARTER
SHA - SHAD spp.
SHD - SLENDERHEAD DARTER
SHG - SHORTNOSE GAR
SHR - SHORTHEAD REDHORSE
SHS - SHOVELNOSE STURGEON
SHT - STARHEAD TOPMINNOW
SJM - SILVERJAW MINNOW
SKC - SICKLEFIN CHUB
SKH - SKIPJACK HERRING
SLD - SLOUGH DARTER
SLM - SLENDER MADTOM
SLS - SLIMY SCULPIN
SMB - SMALLMOUTH BASS
SNC - SPRING CAVEFISH
SOS - SPOONHEAD SCULPIN
SPB - SPOTTED BASS
SPC - SPECKLED CHUB
SPD - SPOTTAIL DARTER
SPG - SPOTTED GAR
SPS - SPOTTAIL SHINER
SRD - SOUTHERN REDBELLY DACE
SSF - SPOTTED SUNFISH
STB - STRIPED BASS
STC - STONECAT
STD - STRIPETAIL DARTER
STO - UNIDENTIFIED STONEROLLER sp.
STS - STRIPED SHINER
SUM - SUCKERMOUTH MINNOW
SUN - SUNFISH spp., EXCLUDING BLACK BASS AND CRAPPIE
SVC - SILVER CHUB
SVL - SILVER LAMPREY
SVM - SILVERY MINNOW
SVR - SILVER REDHORSE
SVS - SILVERBAND SHINER

TGC - TRIPLOID GRASS CARP
TGM - TIGER MUSKIE
THS - THREADFIN SHAD
TIL - TILAPIA
TPM - TADPOLE MADTOM
TRP - TROUT-PERCH

ULL - QUILLBACK

WAE - WALLEYE
WAM - WARMOUTH
WBH - Hybrid WARMOUTH X BLUEGILL
WCF - WHITE CATFISH
WES - WEED SHINER
WHB - WHITE BASS
WHC - WHITE CRAPPIE
WHS - WHITE SUCKER
WSD - WESTERN SAND DARTER
WSH - SAUGEYE (Hybrid WALLEYE x SAUGER)
WSM - WESTERN SILVERY MINNOW

YEB - YELLOW BULLHEAD
YEP - YELLOW PERCH
YLB - YELLOW BASS

Fish Species Groups are listed in ***BOLD ITALIC***. Individual Species names are given in PLAIN text.

1995 Technical Report

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Cholinesterase Activity in Adult Bluegill (*Lepomis macrochirus*) from Mark Twain Refuge in Summer, 1995

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Cholinesterase Activity in Adult Bluegill (*Lepomis macrochirus*) from Mark Twain Refuge in Summer, 1995

Introduction

The risks associated with the use of modern agricultural insecticides are increasingly being questioned, yet to date we have relatively little information upon which to judge risk, especially based on field-derived data. Organophosphorus insecticides (OPs) represented 40% of the global insecticide market in 1989 (Racke, 1993). They were applied to about 32% of the corn acres in Iowa in 1990; 35.2% of the corn acres were treated with insecticides (Hartzler and Wintersteen, 1991). Terbufos (Counter), chlorpyrifos (Lorsban), fonofos (Dyfonate), and phorate (Thimet) were the most commonly used insecticides on corn in Iowa in 1990. On a national basis, Fairchild et al. (1992) reported the following percent of total corn acres on which each insecticide was applied: terbufos 43%, chlorpyrifos 21%, fonofos 12%, and phorate 8%.

The contamination of surface waters by OPs is difficult to monitor because of the relatively low persistence of these compounds in water, sediment and aquatic organisms. OPs are easily hydrolyzed in aquatic systems and are readily metabolized by most vertebrates. In addition, they tend to have moderate to low water solubilities and moderate to high soil adsorption coefficients (K_{oc}). Thus, the parent compound rapidly disappears from the water

column resulting in concentrations generally near or below detection limits, making direct monitoring of the insecticides in water and aquatic organisms very difficult. In addition, these chemicals often reach aquatic systems mainly after storm events, which are generally not sampled in routine monitoring programs. Thus, these compounds are not often identified by monitoring programs. This does not mean, however, that they do not enter the water and pose a significant risk to aquatic life.

Currently there is no well established method to test for OP contamination. By the time water samples are taken, which they seldom are, the chemical has been diluted, degraded and adsorbed to settleable solids, resulting in concentrations in the water which are below detection limits. In addition, chemical analyses of water for a mixture of insecticides is expensive and time consuming. This tends to limit the number of samples and sample times in any monitoring program. Aquatic organisms may have been exposed to concentrations sufficient to cause negative effects, yet water samples are unlikely to identify that exposure.

One thing that is clear about these OPs, however, is that they are very toxic to fish. A survey of toxicity data published by the National Fisheries Contaminants Research Center (now the Midwest Science Center), Columbia, Missouri (Mayer and Ellersieck, 1986) described relative toxicity of OPs to fish and also showed that there is wide variation in sensitivity among fish species (Table 1). Note that acutely lethal concentrations are in the low µg/L levels for

the OPs currently used on Iowa corn. For instance, the 96-h LC_{50} (lethal concentration to 50% of the test animals in 96 hours) for bluegill exposed to chlorpyrifos is about 2.4 μg/L. Vittozzi and De Angelis (1991) reviewed comparative toxicity data on freshwater fish and found that there are large differences among species in levels of sensitivity to OPs. The fathead minnow (*Pimephales prcmelas*) is one of the most common laboratory test species, yet in general they are the least sensitive of the routinely used species. The rainbow trout (*Oncorhynchus mykiss*) does not inhabit warmwater streams and rivers in the western cornbelt ecoregion. However, the bluegill (*Lepomis macrochirus*), although not very abundant in small streams, is plentiful in backwaters of larger rivers. In choosing an appropriate sentinel species sensitivity, abundance and distribution are all important. Bluegill meet these criteria for many types of aquatic systems.

Most available toxicity data on fishes are from laboratory tests; little is known about the potential lethal effects of these OPs on organisms under field conditions. Most laboratory tests use constant exposure levels which do not represent what occurs in the field where relatively short pulses of exposure probably occur. Even less is known about the more difficult to detect sublethal effects; yet sublethal effects may lead to population declines and community changes over time that are more significant than acutely lethal effects. How does a chronic laboratory test with constant exposure levels relate to field conditions? We do not know.

The fate and effects of several OPs have been studied in mesocosms. For standing water systems these include fonofos (Fairchild *et al.*, 1992), phorate (Dieter *et al.*, 1995), chlorpyrifos (Hurlbert *et al.*, 1972; Hughes *et al.*, 1980, Brazner *et al.*, 1989; Brazner and Kline, 1990; Kersting and van Wijngaarden, 1992; Hanratty and Stay, 1994). For flowing water systems the following have been studied: diazinon (Arthur *et al.*, 1983), chlorpyrifos (Eaton *et al.*, 1985; Brock *et al.*, 1992a; Brock *et al.*, 1992b; Pusey *et al.*, 1994; Montanes *et al.*, 1995). In all of these cases, the chemical was directly applied to the system, generally at concentrations higher than expected from agricultural runoff. Again, there are many questions about how realistic these exposures and effects are compared to actual aquatic ecosystems and agricultural watersheds.

Durations of exposure in these mesocosm studies are much closer to reality than what would be seen in the laboratory studies. Lartiges and Garrigues (1995) demonstrated that OP degradation can be affected by temperature, pH, exposure to light and availability of particulate matter. For those OPs with high K_{oc} values, adsorption to particulates can lead to a longer persistence of the chemical in aquatic systems. In agricultural drainages, insecticides would likely enter the water adsorbed to soil particles. In some monitoring programs, the insecticide concentration is determined only on the filterable fraction of the water collected and would miss this adsorbed portion.

We have very little reliable information on persistence and bioavailability of these chemicals in streams and rivers draining agricultural landscapes.

Because the OPs themselves are difficult to monitor in the field, a biochemical indicator of exposure and effects would be useful as the monitoring endpoint (Habig and Di Giulio, 1991). OPs are nerve poisons and their mode of action is the inhibition of cholinesterase (ChE) activity at the nerve synapse in animals. Inhibition of ChE results in accumulation of the neurotransmitter, acetylcholine, in synapses, which disrupts normal neural transmission. Although substantial reduction in brain ChE activity may not be lethal to fish, the effect of this condition on such functions as feeding and reproduction in nature is not known (U.S. EPA, 1986). In general, fish acutely poisoned with OPs have symptoms of muscle paralysis, especially of the fins and gills, hyperactivity and loss of equilibrium; characteristic signs of poisoning also include stiffly flared pectoral fins and exaggerated opercular movements (Zinkl et al., 1991). Cholinesterases are widely distributed in the animal kingdom (Walker and Thompson, 1991; Kozlovskaya *et al.*, 1993) and fish primarily have acetylcholinesterase (AChE). Brain cholinesterase activity has been used to assay for exposure to OPs in many species, and it has been proposed that suppression of ChE activity could be used as a measure of exposure and effects in fish and other aquatic organisms (van der Wel and Welling, 1989; Bocquene *et al.*, 1990; de Bruijn *et al.*, 1991; Mayer *et al.*, 1992). With OPs, the inhibited ChE does not recover but the organism can synthesize new ChE over time. ChE

depression lasts for several weeks in fish (Carr *et al.*, 1995), so depression is much more persistent than the OP itself. This is why ChE activity is a good candidate as an indicator of exposure and effects.

Cholinesterase activity has been measured in fish in a number of studies (Coppage and Braidech, 1976; Jarvinen *et al.*, 1983; Clark *et al.*, 1985; Cripe *et al.*, 1985; Lockhart *et al.*, 1985; Morgan *et al.*, 1990; Carr *et al.*, 1995; and others below). The two major approaches to ChE analysis have been the colorimetric methods of Hestrin (1949) and Ellman *et al.* (1961) and the pH-stat method of Coppage (1971). Of the two approaches, the colorimetric methods have received much more attention. A rather detailed description of modifications of the Ellman *et al.* (1961) procedure for ChE activity was published by Hill and Fleming (1982) for use with samples of bird plasma and brain. Fairbrother *et al.* (1991) reviewed these techniques from an avian toxicology viewpoint. Marden *et al.* (1994) reported on an interlaboratory comparison of ChE assay measurements in birds; no such study has been done on ChE assays in fish.

The principle of the Ellman *et al.* (1961) method is rather simple. ChE hydrolyzes acetylthiocholine iodide into thiocholine and acetate. Thiocholine reacts with dithiodinitrobenzoic acid to form thionitrobenzoic acid, which has a yellow color that can be measured spectrophotometrically at 405 nm. The rate of color production represents ChE activity.

The Ellman *et al.* (1961) method has been applied to fish in several recent studies (Johnson and Wallace, 1987; Salte *et al.*, 1987; Pavlov *et al.*, 1992;

Richmonds and Dutta, 1992; Heath *et al.* 1993a and 1993b). Morgan *et al.* (1990) applied the Hill and Fleming method to a study of the response and recovery of brain acetylcholinesterase activity in Atlantic salmon (*Salmo salar*) exposed to fenitrothion. The modified method was also used on fish by Finlayson and Rudnicki (1985). However, these various researchers did not attempt to determine how well the method worked on exothermic fish compared to the endothermic birds. Cole (1995) addressed some questions of the effects of ambient temperature, fish size, and sample storage on ChE activity. This was done on fish raised in the laboratory. Questions about the effects of temperature and fish size under natural conditions still need to be addressed. Data on normal (uninhibited) ChE activities for adult bluegill in field settings are not readily available in the literature. A baseline data set would be very helpful in determining natural variability and the size of fish sample needed to determine significant inhibition. With data from field collected fish, effects of collection method, and fish age, size, sex and reproductive status, on ChE activity can also be determined.

The purpose of this study was to determine whether fish in backwater areas of the Mark Twain National Wildlife Refuge on the Mississippi River are exposed to and affected by insecticides from the adjacent agricultural landscape. The specific objectives were: 1) to determine whether fish in this area are exposed to organophosphorus insecticides at concentrations sufficient to inhibit cholinesterase; 2) to determine protocol for acceptable sampling methods and

sample size for using ChE to monitor for organophosphorus insecticide exposure; and 3) to develop a data base for future monitoring.

Study Area

The sample area was in North Lake in the Keithsburg backwater area of the Mark Twain National Wildlife Refuge on the upper Mississippi River (See Figure 1). The watershed adjacent to the site was primarily agricultural, and the land nearest the study area had been planted in corn before this study began. This land drained directly into North Lake through several ditches. Much of the bottomland was flooded throughout the sampling (May 1-June 30, 1995). A nearby reference site was selected in a mine pool in Snake Den Hollow in which no pesticide exposures were expected.

Methods

Bluegill Sample Collection

Adult bluegill were collected from North Lake and Snake Den Hollow at approximately two week intervals from May 1 through June 30, 1995. A goal was to collect ten fish at each site for each sample period. To minimize variance in cholinesterase activity due to individual differences, the target size range was 15-17 cm total length. It was not always possible to achieve these targets.

Bluegill were collected by USFWS with DC-pulse electrofishing, or by Illinois Department of Conservation by AC electrofishing. The first collection also included fish that were collected with a trap net by the Illinois Department of Conservation, to determine if cholinesterase activity collected this way would differ from those captured by electrofishing. Angling was used to catch several fish in the last sample collection.

At time of collection, the total length of each fish was measured to the nearest centimeter. Fish were bagged individually and placed on ice immediately; they were then shipped to the Department of Animal Ecology at Iowa State University, on dry ice for cholinesterase analysis as described below. Fish were kept frozen until analysis, which generally occurred within 12 hours of arrival at ISU.

At time of ChE analysis, each fish was partially thawed and removed from its freezer bag. Excess mucous was wiped from the exterior of the fish with paper towels and the fish's wet weight was obtained on a top-loading Ohaus Model CT-600S balance. The wet weight, the total length written on the freezer bag, and sex were recorded for each fish.

Cholinesterase Analysis

ChE activity was analyzed by a modification of the procedure of Ellman *et al.* (1961) and Hill and Fleming (1982). Brain tissue was used as it has a high concentration of ChE and is easy to work with. The brains were removed after

partially thawing the fish to the point that brain tissue was soft, and placed in 500 µL cold pH 7.4 trizma buffer. Brains were weighed to the nearest 0.1 mg on a Sartorius A200S analytical balance, added to a proportional volume of pH 7.4 trizma buffer to get a 100-fold dilution and then homogenized ultrasonically with a Cole-Parmer Model 4710 ultrasonic homogenizer. The homogenate was stored on ice or refrigerated until analysis. Great care was taken to keep the samples cool and to run the assay at a constant 25°C temperature because enzyme activity is temperature dependent (Fairbrother et al., 1991).

The ChE activity of homogenates was analyzed by mixing with the substrate, acetylthiocholine, which reacted with ChE in the same manner as acetylcholine. The subsequent hydrolysis created a negatively charged sulfur atom on the end of the thiocholine complex. The next step was to add 5,5'-dithiobis(2-nitrobenzoic acid) (DTNB), which reacted with the thiocholine complex to form a stable 5-thio-2-nitrobenzoate ion which absorbs light strongly at 405 nm. The formation of this yellow anion was quantified on a computerized 96-well THERMOmax kinetic microplate reader (Molecular Devices Corporation). The system was controlled by Molecular Devices Corporation Softmax software via a Zenith Z386/20 computer. This instrument worked the same as a spectrophotometer, which is generally used for such analyses, but analyzed 96 samples at one time and greatly improved the speed and precision of the analysis. The rate of formation of the yellow anion was used to determine the activity of the ChE in the sample. The increase in absorbance was monitored for

2 min at 25° C with readings every 7 sec. The software calculated the maximum reaction rate (Vmax) by doing a series of regressions on the 16 data points. The ChE activity was then calculated from Vmax and from the amount of sample, and reported as micromoles acetylthiocholine hydrolyzed per minute per gram of brain tissue (μM AThCh hydrolyzed/min/g brain tissue). See Appendix A for the detailed standard operating procedure.

Quality Assurance for ChE Assay

For quality assurance, a check standard made of pooled homogenates from the first sample group was incorporated into each run. Each sample, including the check standard, was run in triplicate sample wells to assure accurate pipetting. A coefficient of variation (c.v.) was calculated for each set of triplicates and if the c.v. was 10% or greater the sample was rerun. When the c.v. of the check standard was 10% or greater the entire plate was run again until the check standard was less than 10%.

For each species analyzed, the ChE has to be characterized, and the assay has to be optimized to assure that the ChE activity determination is as accurate and precise as possible. Initially the best acetylthiocholine concentration needs to be determined by running a series of concentrations of acetylthiocholine and selecting the concentration which produces the optimal optical density for the plate reader. Additionally, a test is run which analyzes differing dilutions of brain samples, and then those activities are plotted. That

plot should be linear, which indicates that the reagents are in sufficient concentrations and that the only limiting factor is the amount of ChE in the sample. Finally, using all the determined concentrations the assay is tested by running it for an extended time (~10 minutes) and monitoring how long the development of color remains linear. A period of linearity of at least 5 minutes is sufficient. The activity of the ChE is determined by using the slope of the color development. That slope needs to be linear throughout the length of the assay (2 minutes) to produce an accurate result. For this study, the reaction rate was optimized on a pooled sample from the first group of fish; optimal temperature was 25° C and optimal acetylthiocholine concentration was 2.51×10^{-3} M.

Statistics

All statistical analyses were performed using SAS (SAS Institute, 1989). Contrast results were considered significant at an α level of 0.05. A univariate analysis of variance (ANOVA) compared fish taken from the Keithsburg backwater area and from the mine pool in Snake Den Hollow, to confirm that cholinesterase was not significantly different between the two populations. A second univariate ANOVA compared cholinesterase among collection dates to look for significant differences. A multivariate analysis of variance (MANOVA) compared effects of body weight and brain weight on cholinesterase activity; a MANOVA was appropriate here because there is correlation between brain and body weight (i.e., brain weight could be considered an additional dependent variable with respect to body weight). A MANOVA was also done to compare cholinesterase activity and size between trapnetted and electrofished bluegill.

Finally, cholinesterase and size were compared between males and females among collection dates in a MANOVA.

The minimum sample size statistically required to detect exposure to cholinesterase inhibitors was determined from the 1995 bluegill cholinesterase results. Calculations were made with the following formula from Cochran (1977): $n = [tS/r\ \bar{Y})]^2$, where n is the sample size, t is obtained from the Student's t distribution, S is the standard deviation, r is the acceptable relative error, and $\bar{Y}$ is the population mean.

Results

Water Quality

Water depth, pH, temperature, conductivity and turbidity were measured by FWS personnel during March through June in North Lake (Table 2). The variable most likely to affect ChE activity is temperature, which increased from 10.5° C on March 23 to 25.5° C on June 27, 1995.

Cholinesterase

A total of 130 adult bluegill were collected and analyzed. The mean body and brain weights, and cholinesterase activities by date are given in Table 3. Complete results are in Appendix B.

Although fish weights were significantly different across collection dates ($F = 2.23$, $P = 0.03$), brain weights were not ($F = 1.42$; $P = 0.20$). Cholinesterase activity was significantly different across collection dates ($F = 6.20$; $P = 0.0001$).

Mean cholinesterase activities were higher in later collections (other than the June 17 sample, which was the only sample collected by angling). Increases in mean cholinesterase activities were probably due to temperature, though differences due to reproductive status cannot be ruled out. There was no evidence of cholinesterase inhibition in the means of these samples; activities (8.25-11.85 μM substrate hydrolyzed/min/gm brain tissue) were similar to a literature value for uninhibited bluegill cholinesterase (Richmonds and Dutta, 1992). One individual in each of the collections from May 1 and 2, from Snake Den Hollow as well as well as North Lake, had a very low cholinesterase activity (3.21 and 3.65, respectively; see Appendix A), and on these two dates several individuals had cholinesterase activities between 5 and 7. There was some indication of difference in cholinesterase by size, though some of the variation may be explainable by reproductive state and environmental temperature.

A comparison of the May 1 and 2 collections, comparing bluegill from North Lake to bluegill from Snake Den Hollow, showed no significant difference in cholinesterase activities between these populations ($F = 0.22$; $P = 0.64$; Table 4). Bluegill from North Lake were collected either by trap net or electrofishing; Snake Den Hollow fish were all collected by electrofishing. Snake Den Hollow bluegill had a higher mean brain weight, although they had a smaller overall body weight; neither difference was significant ($F = 1.48$ and $P = 0.23$; $F = 2.07$ and $P = 0.16$).

Sex determinations were not made on the May 1 collection, and were made on only part of the May 2 collection; sex was determined for all bluegill in subsequent collections. Of the 131 fish analyzed, 99 were sexed; 41 were female and 58 were male (Table 5). There were no significant differences by sex on bluegill for cholinesterase activity ($F = 0.02$; $P = 0.90$) or for brain weight ($F = 0.03$; $P = 0.86$). Males were significantly heavier than females ($F = 4.16$; $P = 0.04$).

On May 2, 28 bluegill from North Lake were collected by trap net, and 16 by electrofishing. Fish captured in the trap net tended to be heavier than those collected with electrofishing, with higher brain weights (Table 6). These differences were statistically significant (wet weight $F = 23.43$ and $P = 0.0001$; brain weight $F = 21.05$ and $P = 0.0001$). Differences in cholinesterase activity were not significant, however ($F = 1.58$ and $P = 0.22$).

Over a three-day interval, June 15-17, sixteen bluegill were electrofished from Snake Den Hollow, 9 were electrofished from North Lake and 7 were angled from North Lake. Angled fish tended to be heavier, with higher brain weights (Table 7), although the differences were not statistically significant (weight $F = 1.39$ and $P = 0.25$; brain weight $F = 2.97$ and $P = 0.09$). Cholinesterase activity, however, was significantly lower in angled fish ($F = 7.25$ and $P = 0.01$).

A final comparison of collection methods was made on bluegill; this compared ten fish that were collected from Spring Slough using AC

electrofishing on June 27 with 10 fish collected from North Lake using DC electrofishing on June 30. Although fish collected with AC electrofishing tended to have lower body and brain weights and lower ChE activity, these were not significantly different from fish collected with DC electrofishing (Table 8).

Discussion

Sample Size

The minimum sample size for this population was calculated after setting an acceptable value for r, the relative error of the mean. Gibson *et al.* (1969) found that freezing and thawing of bluegill brains increased variances and decreased mean cholinesterase for groups of 20 bluegill by as much as 11%. In other vertebrates such as birds, 20% inhibition has been commonly accepted as evidence of exposure to cholinesterase inhibitors (Ludke *et al.*, 1975; Grue and Hunter, 1984). For these reasons, sample size calculations were based on detection of 20% inhibition. For bluegill, mean cholinesterase of the 131 fish collected was 9.63; S was 2.45. The relative error was set at 20% of the mean, or 1.926. The minimum sample size was calculated to be 7. By this criterion, the sample sizes used in 1995 were sufficient; similar sample sizes are recommended for future sampling.

Cholinesterase Inhibition

There was no evidence of inhibition in the cholinesterase means, but inhibition may have occurred before the start of this study; planting occurred prior May 1, 1995. A few individuals in the May 1 and 2 collections had low cholinesterase activities, which fit the criterion of greater than 20% inhibition that is commonly accepted as evidence of exposure to a ChE inhibitor (Ludke *et al.* 1975). As these low activities occurred at both sites (Snake Den Hollow and North Lake), the cause is not obvious. Unlike fish collected from North Lake, fish in Snake Den Hollow were not expected to have been exposed to a ChE inhibitor. Perhaps there is an unknown source of contamination there, or there may be some unknown environmental factor responsible for the low ChE activities. This could also just be normal biological variability. We are certain that the low activities are not due to anything in handling or analysis at ISU.

Choice of Reference Site

There were no significant differences in cholinesterase activities, brain or body weights between bluegill collected in North Lake and those collected in Snake Den Hollow. The choice of Snake Den Hollow as a reference site for North Lake appears valid.

Differences by Sex

Although females tended to weigh less than males, and that weight difference was statistically significant, the differences in brain weight and cholinesterase activity were not statistically significant. This suggests that it is not necessary to determine the sex in samples of bluegill collected for cholinesterase analysis. However, controlling for sex in samples may decrease overall variability.

Differences by Collection Method

Cholinesterase activities were somewhat lower in bluegill collected by trap net than in electrofished bluegill. The difference, however, was not statistically significant, and may be due to the fact that fish collected by trap net were larger than those collected by electrofishing. Cholinesterase activity may also be affected by the collection method itself; if so, it is not affected enough to confound detection of exposure to cholinesterase inhibitors. The difference in cholinesterase activities between bluegill collected by AC and by DC electrofishing was not statistically significant. The use of any of these methods should be acceptable.

Cholinesterase activities tended to be lower in fish collected by angling than by electrofishing; this difference was statistically significant and fairly large: the angled fish had a mean cholinesterase activity that was 75 % of the electrofished bluegill. As this is greater than the 20% decrease in ChE

considered to be indicative of inhibition (Ludke *et al.*, 1975), angling is not recommended as a collection method.

Factors Which May Affect Cholinesterase Activity

Understanding of relationships between physiological factors and ChE activity are essential for reliable interpretation of ChE results. In studies of cholinesterase in several vertebrate species, ChE activity increased from birth through adulthood. There were also indications of declining ChE activity with senescence (Rattner and Fairbrother, 1991). Little is known about the effect of life stage on ChE in fish.

Environmental variables, especially temperature, may affect fish ChE activity. Bluegill ChE activity has been shown to increase with summer water temperatures (Zinkl *et al.*, 1991).

Recommendations for Monitoring Cholinesterase Activity

Sampling from summer, 1995, resulted in the creation of a baseline data set, and many of the procedures used in sample collection and processing should be maintained in any future effort. Bluegill should be collected by either trap net or by electrofishing, and fish should be individually bagged and placed on ice immediately. The minimum sample size should be at least seven, based on the variance in ChE activities of bluegill collected in summer, 1995. A larger sample size would be advisable, in case individual variability is higher under different conditions. Precautions for keeping samples frozen until analysis are

important, as are all quality assurance procedures. All information recorded during the sample collection and analysis, including total length, wet weight, sex, and brain weight, is needed for interpretation of results.

Future monitoring efforts may be improved by staying within the target size range, 15-17 cm. Also, collection of water quality data, especially temperature, consistently with each sample, is indicated from the differences in ChE activities across dates.

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Table 1. Summary of acute toxicity test results (96-h LC_{50} in mg/L) for organophosphorus insecticides conducted at the National Fisheries Contaminants Research Center (Mayer and Ellersieck, 1986). These are all for static tests but the size of fish used, temperatures, pH and hardness are variable: attempts were made to use the most consistent conditions available for these comparisons.

Chemical	Bluegill	Rainbow Trout	Fathead Minnow
azinphos-ethyl	1.1	20	--
azinphos-methyl	8.0	6.3	235
chlorpyrifos	2.4	7.1	--
diazinon	168	90	--
fenitrothion	2,700	2,000	3,200
fonofos	6.8	20	--
malathion	30	200	8,650
parathion-ethyl	400	864	2,350
parathion-methyl	4,380	3,700	8,900
phorate	2.0	13	--
terbufos	1.8	10	390

Table 2. Water quality data in North Lake, collected by Mike Coffey in summer, 1995.

Collection Date	Time	Water Depth (ft)	pH	Water Temp. (°C)	Conductivity (µS)	Turbidity (NTU)
March 23	2:51 PM	4.5	8.55	10.5	344.9	7.6
April 19	3:30 PM	6	8.48	15.8	297	15.8
May 15	9:30 AM	8	8.43	17.7	354.0	6.6
June 6	1:57 PM	6	8.45	25.0	286.5	15.6
June 27	noon	5	7.9	25.5	302.4	38.3

Table 3. Comparison of wet weight, brain wet weight, and cholinesterase activity between collection dates among adult bluegill collected in summer, 1995. Standard deviations are given in parenthesis behind each mean.

Collection Date	Number Collected	Mean Wet Weight (g)	Mean Brain Weight (g)	Mean Brain ChE Activity [a]
May 1	11	86.6 (24.05)	0.1128 (0.03314)	8.58 (2.732)
May 2	44	104 (37.74)	0.0998 (0.03141)	8.25 (1.918)
May 19	11	115 (33.61)	0.0884 (0.02838)	10.2 (1.659)
June 5	13	105 (38.41)	0.1131 (0.03077)	10.6 (2.407)
June 15	16	77.1 (17.88)	0.0882 (0.02123)	11.8 (3.052)
June 16	9	75.8 (38.47)	0.0982 (0.03908)	11.0 (1.753)
June 17	7	90.8 (35.54)	0.1138 (0.03438)	8.66 (1.855)
June 27	10	93.9 (34.79)	0.1003 (0.03206)	10.7 (2.296)
June 30	10	107 (18.79)	0.1149 (0.03301)	11.1 (2.192)
F value[b]		2.23	1.42	6.20
Probability of F[c]		0.03	0.20	0.0001

[a] Cholinesterase activities are reported as micromoles acetylthiocholine hydrolyzed per minute per gram of brain tissue (μM AThCh hydrolyzed/min/g brain tissue).
[b] F value obtained in analysis of variance of means in column.
[c] Probability of F (P value) obtained in analysis of variance of means in column; differences between means are considered significant if P value is less than 0.05.

Table 4. Comparison of wet weight, brain wet weight, and cholinesterase activity between adult bluegill collected by electrofishing in Snake Den Hollow and by electrofishing and trap net in North Lake, on May 1 and 2, 1995, respectively. Standard deviations are given in parenthesis behind each mean.

Site	Number Collected	Mean Wet Weight (g)	Mean Brain Weight (g)	Mean Brain ChE Activity [a]
Snake Den Hollow	11	85.6 (24.05)	0.1128 (0.03314)	8.58 (2.73)
North Lake	44	104 (37.74)	0.0998 (0.03141)	8.25 (1.92)
F value[b]	--	2.07	1.48	0.22
Probability of F[c]	--	0.16	0.23	0.64

[a] Cholinesterase activities are reported as micromoles acetylthiocholine hydrolyzed per minute per gram of brain tissue (μM AThCh hydrolyzed/min/g brain tissue).
[b] F value obtained in analysis of variance of means in column.
[c] Probability of F (P value) obtained in analysis of variance of means in column; differences between means are considered significant if P value is less than 0.05.

Table 5. Comparison of wet weight, brain wet weight, and cholinesterase activity between male and female adult bluegill collected in summer, 1995, by electrofishing and trap net in North Lake. Standard deviations are given in parenthesis behind each mean.

Sex	Number Collected	Mean Wet Weight (g)	Mean Brain Weight (g)	Mean Brain ChE Activity [a]
Female	41	89.2 (35.55)	0.1039 (0.03083)	10.23 (2.800)
Male	58	103.1 (33.83)	0.1027 (0.03228)	10.29 (2.146)
F value [b]		4.16	0.03	0.02
Probability of F [c]		0.04	0.86	0.90

[a] Cholinesterase activities are reported as micromoles acetylthiocholine hydrolyzed per minute per gram of brain tissue (μM AThCh hydrolyzed/min/g brain tissue).
[b] F value obtained in analysis of variance of means in column.
[c] Probability of F (P value) obtained in analysis of variance of means in column; differences between means are considered significant if P value is less than 0.05.

Table 6. Comparison of wet weight, brain wet weight, and cholinesterase activity between adult bluegill collected on May 2, 1995, by electrofishing and trap net in North Lake. Standard deviations are given in parenthesis behind each mean.

Collection Method	Number Collected	Mean Wet Weight (g)	Mean Brain Weight (g)	Mean Brain ChE Activity [a]
Trap net	28	121 (30.88)	0.1133 (0.02949)	7.98 (2.022)
Electrofishing	16	74.2 (30.07)	0.0760 (0.01784)	8.73 (1.675)
F value [b]		23.43	21.05	1.58
Probability of F [c]		0.0001	0.0001	0.22

[a] Cholinesterase activities are reported as micromoles acetylthiocholine hydrolyzed per minute per gram of brain tissue (μM AThCh hydrolyzed/min/g brain tissue).
[b] F value obtained in analysis of variance of means in column.
[c] Probability of F (P value) obtained in analysis of variance of means in column; differences between means are considered significant if P value is less than 0.05.

Table 7. Comparison of wet weight, brain wet weight, and cholinesterase activity between adult bluegill collected on June 15-17, 1995, by electrofishing and angling. Standard deviations are given in parenthesis behind each mean.

Collection Method	Number Collected	Mean Wet Weight (g)	Mean Brain Weight (g)	Mean Brain ChE Activity [a]
Angling	7	90.8 (34.54)	0.1138 (0.03438)	8.66 (1.855)
Electrofishing	25	76.6 (26.33)	0.0918 (0.02854)	11.5 (2.6484)
F value [b]		1.39	2.97	7.25
Probability of F [c]		0.25	0.09	0.01

[a] Cholinesterase activities are reported as micromoles acetylthiocholine hydrolyzed per minute per gram of brain tissue (μM AThCh hydrolyzed/min/g brain tissue).
[b] F value obtained in analysis of variance of means in column.
[c] Probability of F (P value) obtained in analysis of variance of means in column; differences between means are considered significant if P value is less than 0.05.

Table 8. Comparison of wet weight, brain wet weight, and cholinesterase activity between adult bluegill collected on June 27, 1995, by AC electrofishing and on June 30, 1995, by DC-pulse electrofishing. Standard deviations are given in parenthesis behind each mean.

Collection Method	Number Collected	Mean Wet Weight (g)	Mean Brain Weight (g)	Mean Brain ChE Activity [a]
AC Electrofishing	10	93.3 (34.79)	0.1003 (0.03206)	10.7 (2.30)
DC-pulse Electrofishing	10	107 (18.79)	0.1149 (0.03301)	11.1 (2.19)
F Value [b]	--	1.09	1.01	0.15
Probability of F [c]	--	0.31	0.33	0.70

[a] Cholinesterase activities are reported as micromoles acetylthiocholine hydrolyzed per minute per gram of brain tissue (μM AThCh hydrolyzed/min/g brain tissue).
[b] F value obtained in analysis of variance of means in column.
[c] Probability of F (P value) obtained in analysis of variance of means in column; differences between means are considered significant if P value is less than 0.05.

Figure 1. Aerial photograph of Keithsburg backwater area of the Mark Twain National Wildlife Refuge on the upper Mississippi River. Sample sites are marked with arrows. Water levels were higher during sampling than when photograph was taken.

Appendix A

Standard Operating Procedure

for the Determination of Cholinesterase Activity

in Bluegill (*Lepomis macrochirus*) Brain Tissue

I. Introduction - Justification

Cholinesterase activity is a measure of the amount of effective cholinesterase in tissues. Cholinesterase (ChE) is an essential enzyme in the central and peripheral nervous systems of vertebrates, where it hydrolyzes acetylcholine, a primary neurotransmitter. The determination of ChE activity can be used as a biomarker to determine if organisms have been exposed to organophosphorus or carbamate insecticides; both types of insecticides inhibit ChE activity as their primary mode of action.

A spectrophotometric assay using a plate reader is used to determine ChE activity in fish brain tissue (Ellman *et al.*, 1961; Hill and Fleming, 1982; Corvallis Environmental Research Laboratory, 1987; The Institute for Wildlife and Environmental Toxicology, 1991). ChE activity is determined from the result of two reactions occurring in the assay solution: acetylthiocholine hydrolysis and reaction of the thiocholine product with a colorimetric reagent. The assay solution consists of a portion of the brain sample containing the ChE enzyme, acetylthiocholine (AThCh substrate), and 5,5-dithiobis(2-nitrobenzoic acid) (DTNB colorimetric reagent).

AThCh is an analogue of the natural ChE substrate, acetylcholine; the analogue has a sulfur atom which replaces the esteric oxygen of acetylcholine. Hydrolysis of AThCh results in the formation of a negatively charged thiocholine complex and an acetate ion.

The thiocholine complex reacts with DTNB to generate a stable, yellow-colored anion (5-thio-2-nitrobenzoate) which absorbs light strongly at 412 nm. For every molecule of AThCh hydrolyzed, approximately one molecule of the anion is generated. The rate of formation of the yellow-colored anion can be measured and subsequent calculations can determine the ChE activity for the sample.

II. Materials

A. Chemicals

1. Acetylthiocholine iodide (AThCh)
2. 5,5-dithiobis-2-nitrobenzoic acid (DTNB)
3. Sodium bicarbonate
4. Trizma 7.4 pH pre-set crystals
5. Trizma 8.0 pH pre-set crystals
6. 1.0 N HCl
7. 1.0 N NaOH

B. Equipment

1. Spectrophotometer: e.g., automated kinetic microplate reader, Molecular Devices Corporation, Thermo max interfaced with a desk top computer (e.g. Zenith z-386/20) loaded with appropriate software package to run spectrophotometer (e.g. Softmax).

2. Constant temperature water bath set a 25°C.

3. Ice bucket and/or ice chest.

4. Crushed ice.

5. Disposable test tubes (13x100 mm).

6. Multi-aliquot, variable volume pipette (e.g., Eppendorf Combitip Pipette) with disposable tips 10µl, 50 µl, 100 µl and 1000 µl (e.g., Eppendorf Combitips).

7. Single aliquot, variable volume pipette, 10-1000 µl range, with disposable tips.

8. Vortex mixer.

9. Magnetic stirrer and stir bars.

10. pH meter and standards.

11. 96 multi-well microplates, e.g., Dynatech Microtiter.

12. Analytical balance.

13. Volumetric flasks, 5-50 ml and 1000 ml.

14. Weigh boats, glass and plastic.

III. Preparation of buffers, reagents and substrate.

Nanopure or distilled water is used to mix solutions. Bottles containing solutions are labeled with chemical name, date, and preparer's name. Solutions are prepared according to the following procedures:

A. Trizma 7.4 pH buffer solution

1. Weigh 7.58 g Trizma 7.4 pre-set crystals in a weigh boat and transfer to a 1-liter volumetric flask.

2. Make a quantitative transfer of chemical by rinsing the weigh boat with water at least 3 times, until no crystals are seen in weigh boat.

3. Dilute to the mark with water. Stopper flask or cover with Parafilm and invert flask 20 times to mix.

4. Check pH and adjust to pH 7.4 with HCl or NaOH.

5. Store in the refrigerator (4°C). Buffer solution will be good for three weeks. Before each use, check pH and adjust to pH 7.4 with HCl or NaOH.

B. Trizma 8.0 pH buffer solution

1. Weigh 8.02 g Trizma 8.0 pre-set crystals in a weigh boat and transfer to a 1-liter volumetric flask.

2. Make a quantitative transfer of chemical by rinsing the weigh boat with water at least 3 times, until no crystals are seen in weigh boat.

3. Dilute to the mark with water. Stopper flask or cover with Parafilm and invert flask 20 times to mix.

4. Check pH and adjust to pH 8.0 with HCl or NaOH.

5. Store in the refrigerator (4°C). Buffer solution will be good for three weeks. Before each use, check pH and adjust to pH 7.4 with HCl or NaOH.

C. **AThCh substrate**

1. Weigh 0.4512 g AThCh in a weigh boat and transfer to a 10 ml volumetric flask

2. Make a quantitative transfer of crystals by rinsing weigh boat with water at least 3 times, until no crystals are seen in weigh boat.

3. Dilute to the mark with water. Stopper flask or cover with Parafilm and invert flask 20 times to mix.

4. Transfer to a labeled amber bottle, or cover flask with aluminum foil and store in the refrigerator (4°C). Substrate solution will be good for up to 3 days.

D. **DTNB reagent**

1. Weigh 0.198 g of DTNB in a glass weigh boat and transfer to a labeled amber bottle.

2. Make a quantitative transfer of crystals by rinsing weigh boat with Trizma 7.4 pH buffer solution at least 3 times, until no crystals are seen in weigh boat.

3. Weigh 0.075g sodium bicarbonate in a glass weigh boat and transfer to the same amber bottle. Again, make a quantitative transfer of crystals by rinsing weigh boat with Trizma 7.4 pH buffer solution at least 3 times, until no crystals are seen in weigh boat.

4. Add the remaining buffer solution to the bottle and mix until dissolved. Store in the refrigerator (4°C). Solution will be good for 3 days.

IV. Analysis procedure:

A. Turn on ice machine and water bath $\geq$ 1 h prior to analysis.

B. Place appropriate volume of Trizma 8 pH buffer in water bath. If Trizma is cold (4°C) allow appropriate time in water bath for it to come to temperature (25°C).

C. Turn on the spectrophotometer (Thermo Max) and control computer. Run the controling software (double click the Softmax icon). Turn the incubator on and set the temperature to 25° C under the control heading. Open the appropriate file (bgche) with the analysis parameters as listed below.

1. wavelength: 405 nm
2. run time: 2:00 min
3. read interval: 7 s
4. OD limit: 0.500 OD
5. lag time: 0.00 s
6. auto mix ON

D. Remove check standards from liquid nitrogen freezer and place in ice to thaw.

E. Transfer fish to refridgerator for partial thawing; fish will be somewhat flexible, but still have ice crystals on surface when it has adequately thawed. This will give brain tissue that is soft enough to distinguish from skull; if fish is thawed too much, brain tissue will be liquid. Remove brain tissue by cutting away the top of the skull, severing the optic nerves and then lifting out the brain. Keep the brain tissue in iced pH 7.4 Trizma buffer until analysis. Homogenize tissue in pH 7.4 Trizma buffer with a motorized teflon pestle and glass tube. Dilute tissue homogenate using Trizma 7.4 pH to an activity appropriate for the spectrophotometer (usually 100-fold). Record the fish size data on form #1 and the weights of the brain tissue and appropriate dilutions on form #2.

F. Prepare cholinesterase assay plate reader set-up form (#3) indicating the positions of the various samples and check standards and their respective dilution factors.

G. Mark microplate to indicate where particular samples will be placed.

H. Pipette appropriate amounts of reagent into each well for each determination to be performed. Place the DTNB and AThCh on ice next to the analysis station. All samples should be assayed in triplicate.

Volumes of reagents for the various wells are as follows (in µl):

	Blank	ChE
Trizma 8.0 pH	200	170
DTNB	20	20
Enzyme	0	30
AThCh	30	30

I. Add compounds to wells in the order shown in the table. Once the AThCh is added the reaction begins. Immediately select read under the control heading in the software. The drawer will then open for a few seconds to allow for locking of the plate into place.

J. After the analysis is complete, type in comments on the data screen and save the file under an appropriate name. Print off a hardcopy of the file.

K. Check the data for any signs of error. Samples with a coefficient of variance (CV) greater then 10 % should be rerun. Also check if the check standards are in control.

L. Convert mOD output units into international units of enzyme activity using the following equation:

(((enzyme mOD/min)-(blank mOD/min))/1000) x 0.817 x dilution factor = (µmoles AThCh hydrolyzed/min) / gram tissue.

The above equation is derived from Ellman et al. (1961).

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Appendix B

Full Data Set from Summer, 1995

Column Headers:

OBS: observation number, sequential, assigned by SAS.

SAMPLE: identifier assigned to each sample before analysis at Iowa State.

PLACE: location from which fish was collected. S = Snake Den Hollow, K = North Lake, Keithsburg Refuge.

METHOD: how fish was collected. E = AC electrofishing, T = trap net, A = angling, F = DC electrofishing

SEX: N = not determined, M = male, F = female.

COL_DATE: date collected, given by year, month, day.

COL_NUM: sequential numbering of collection dates.

AN_DATE: date analyzed, given by year, month, day.

TL: total length, in cm.

WT: wet weight of fish, in grams.

BRAIN: wet weight of brain, in grams.

VMAX: rate of color change reaction in cholinesterase analysis, mean of three microplate wells.

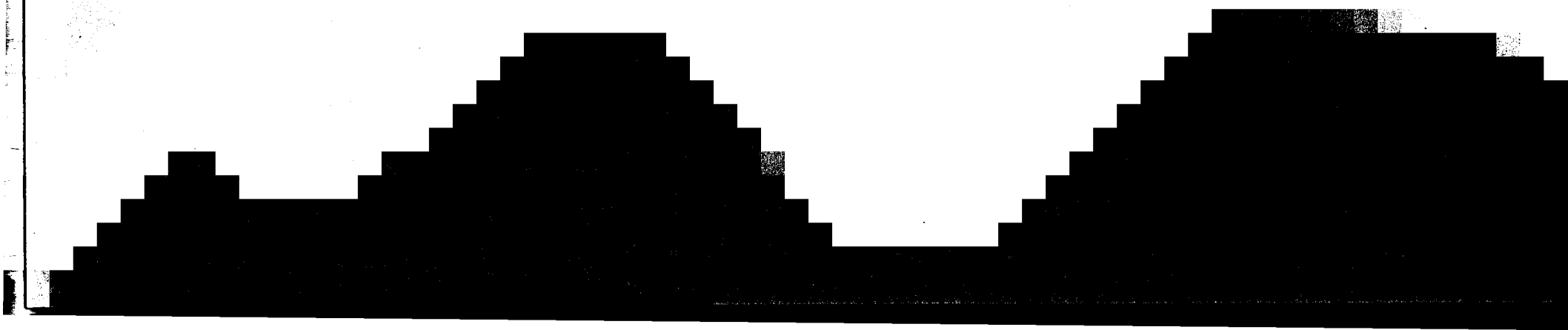
CV: coefficient of variance in VMAX.

ACTIVITY: cholinesterase activity, calculated according to formula given in Appendix A.

OBS	SAMPLE	PLACE	METHOD	SEX	COL_DATE	COL_NUM	AN_DATE	TL	WT	BRAIN	VMAX	CV	ACTIVITY
1	M1	S	E	N	950501	1	950504	163	85.1	0.0814	104.50	7.431	8.54
2	M2	S	E	N	950501	1	950504	172	94.6	0.1735	87.44	4.214	7.14
3	M3	S	E	N	950501	1	950504	154	66.2	0.0583	178.40	2.612	14.58
4	M4	S	E	N	950501	1	950504	159	78.1	0.1462	82.83	5.811	6.77
5	M5	S	E	N	950501	1	950504	177	116.4	0.0917	110.10	3.522	9.00
6	M6	S	E	N	950501	1	950504	186	129.9	0.1364	91.99	1.385	7.52
7	M7	S	E	N	950501	1	950504	164	87.3	0.1175	117.30	7.970	9.58
8	M8	S	E	N	950501	1	950504	140	52.9	0.1055	39.34	3.784	3.21
9	M9	S	E	N	950501	1	950504	142	52.5	0.0873	120.50	4.756	9.84
10	M10	S	E	N	950501	1	950504	168	89.3	0.1084	110.40	2.491	9.02
11	M11	S	E	N	950501	1	950504	171	99.3	0.1347	112.30	3.797	9.17
12	E1	K	E	N	950502	2	950504	155	87.9	0.1102	100.50	2.472	8.21
13	E2	K	E	N	950502	2	950504	157	92.1	0.1131	80.60	1.565	6.59
14	E3	K	E	N	950502	2	950504	164	92.2	0.0690	81.78	2.839	6.68
15	E4	K	E	N	950502	2	950504	189	161.1	0.0704	94.96	4.948	7.76
16	E5	K	E	N	950502	2	950504	167	93.8	0.0782	136.30	2.072	11.14
17	E6	K	E	N	950502	2	950504	161	99.9	0.0923	81.70	3.483	6.67
18	E7	K	E	N	950502	2	950504	129	38.9	0.0658	104.70	11.130	8.55
19	E8	K	E	N	950502	2	950504	136	50.0	0.0531	136.00	4.422	11.11
20	E9	K	E	N	950502	2	950504	141	52.8	0.0694	129.00	0.947	10.54
21	E10	K	E	N	950502	2	950504	142	61.3	0.0615	100.10	1.177	8.18
22	E11	K	E	N	950502	2	950504	138	55.3	0.0522	101.70	9.892	8.31
23	E12	K	E	F	950502	2	950505	138	58.5	0.0765	126.30	2.222	10.32
24	E13	K	E	M	950502	2	950505	123	44.5	0.0751	137.10	2.810	11.20
25	E14	K	E	M	950502	2	950505	145	64.4	0.0798	93.81	3.927	7.66
26	E15	K	E	M	950502	2	950505	144	68.2	0.0607	116.10	9.881	9.49
27	E16	K	E	F	950502	2	950505	146	66.4	0.0894	88.63	11.340	7.24
28	T1	K	T	N	950502	2	950504	160	102.1	0.1487	63.93	5.827	5.22
29	T2	K	T	N	950502	2	950504	156	100.3	0.1258	44.67	6.284	3.65
30	T3	K	T	N	950502	2	950504	188	153.8	0.0699	140.60	7.701	11.49
31	T4	K	T	N	950502	2	950504	184	142.6	0.1344	98.68	7.144	8.06
32	T5	K	T	N	950502	2	950504	186	131.8	0.1124	81.57	3.805	6.66
33	T6	K	T	N	950502	2	950504	188	132.8	0.0678	74.64	9.672	6.10
34	T7	K	T	N	950502	2	950504	167	95.2	0.0693	78.11	6.182	6.38
35	T8	K	T	N	950502	2	950504	184	129.9	0.0936	82.24	0.929	6.72
36	T9	K	T	N	950502	2	950504	168	108.1	0.1022	105.80	9.783	8.64
37	T10	K	T	N	950502	2	950504	182	144.0	0.1007	91.24	2.214	7.45
38	T11	K	T	M	950502	2	950505	167	105.0	0.1280	97.19	5.004	7.94
39	T12	K	T	F	950502	2	950505	179	137.7	0.1437	96.26	1.485	7.86
40	T13	K	T	M	950502	2	950505	192	167.4	0.1212	103.00	1.962	8.42
41	T14	K	T	F	950502	2	950505	187	157.5	0.1772	88.57	2.257	7.24
42	T15	K	T	M	950502	2	950505	180	133.8	0.1184	98.74	7.611	8.07
43	T16	K	T	F	950502	2	950505	191	168.6	0.1293	71.36	4.041	5.83
44	T17	K	T	M	950502	2	950505	184	167.9	0.1598	91.16	9.963	7.45
45	T18	K	T	F	950502	2	950505	147	87.4	0.0717	101.00	2.160	8.25
46	T19	K	T	F	950502	2	950505	177	153.2	0.1229	84.23	6.312	6.88
47	T20	K	T	M	950502	2	950505	152	89.0	0.0865	108.30	4.714	8.85
48	E17	K	E	F	950519	3	950523	178	124.3	0.0831	123.50	2.314	10.09
49	E18	K	E	F	950519	3	950523	197	191.6	0.1202	99.69	6.252	8.14
50	E19	K	E	F	950519	3	950523	153	78.7	0.0578	68.10	2.612	10.82
51	E20	K	E	M	950519	3	950523	189	153.0	0.0746	105.80	3.056	8.64
52	E21	K	E	M	950519	3	950523	168	120.4	0.0623	121.10	3.930	9.89
53	E22	K	E	M	950519	3	950523	159	98.7	0.0522	162.00	2.351	13.24
54	E23	K	E	M	950519	3	950523	162	86.2	0.0901	96.72	5.172	7.90
55	E24	K	E	F	950519	3	950523	173	112.4	0.1202	123.50	7.059	10.09
56	E25	K	E	F	950519	3	950523	176	124.8	0.1400	136.30	5.944	11.14

OBS	SAMPLE	PLACE	METHOD	SEX	COL_DATE	COL_NUM	AN_DATE	TL	WT	BRAIN	VMAX	CV	ACTIVITY
57	E26	K	E	M	950519	3	950523	157	93.7	0.0962	127.40	0.691	10.41
58	E27	K	E	M	950519	3	950523	152	85.0	0.0757	152.00	3.882	12.42
59	T21	K	T	F	950502	2	950601	154	82.0	0.1633	66.89	6.462	5.46
60	T22	K	T	M	950502	2	950601	163	104.9	0.1185	120.60	9.804	9.85
61	T23	K	T	M	950502	2	950601	162	90.1	0.0802	117.90	2.283	9.63
62	T24	K	T	F	950502	2	950601	142	65.4	0.1020	142.50	8.166	11.64
63	T25	K	T	F	950502	2	950601	149	63.2	0.1065	114.10	3.170	9.32
64	T26	K	T	M	950502	2	950601	168	101.1	0.1283	92.01	3.288	7.52
65	T27	K	T	M	950502	2	950601	180	139.0	0.1039	153.60	9.618	12.55
66	T28	K	T	M	950502	2	950601	175	123.5	0.0872	125.10	1.804	10.22
67	E28	K	E	F	950605	4	950607	118	32.2	0.0765	116.20	6.601	9.49
68	E29	K	E	F	950605	4	950607	141	58.2	0.0783	112.50	4.328	10.11
69	E30	K	E	M	950605	4	950607	176	128.3	0.1785	119.60	4.424	9.77
70	E31	K	E	M	950605	4	950607	171	130.8	0.0973	197.80	5.126	16.16
71	E32	K	E	F	950605	4	950607	155	79.9	0.0947	126.40	4.660	10.33
72	E33	K	E	M	950605	4	950607	169	108.4	0.1118	109.90	4.956	8.98
73	E34	K	E	F	950605	4	950607	144	62.2	0.0802	155.80	8.602	12.73
74	E35	K	E	M	950605	4	950607	182	146.7	0.1540	152.90	7.341	12.49
75	E36	K	E	F	950605	4	950607	164	105.7	0.1317	161.60	9.493	13.20
76	E37	K	E	M	950605	4	950607	189	168.0	0.1356	92.36	7.119	7.55
77	E38	K	E	M	950605	4	950607	174	131.5	0.1245	100.30	3.223	8.19
78	E39	K	E	M	950605	4	950607	173	115.9	0.1071	108.60	4.727	8.87
79	E40	K	E	M	950605	4	950607	165	103.1	0.1001	119.10	6.005	9.73
80	M12	S	E	F	950615	5	950630	132	47.9	0.0717	140.50	7.946	11.48
81	M13	S	E	F	950615	5	950630	146	59.4	0.0997	92.57	3.618	7.56
82	M14	S	E	F	950615	5	950630	148	56.7	0.0714	120.80	2.321	9.87
83	M15	S	E	M	950615	5	950630	168	102.7	0.1039	131.00	2.769	10.70
84	M16	S	E	F	950615	5	950630	165	94.4	0.0917	132.40	6.487	10.82
85	M17	S	E	M	950615	5	950630	152	82.7	0.0676	165.70	4.396	13.54
86	M18	S	E	M	950615	5	950630	156	81.4	0.1091	170.50	4.811	13.93
87	M19	S	E	M	950615	5	950630	173	115.8	0.0999	122.50	3.324	10.01
88	M20	S	E	F	950615	5	950630	156	67.9	0.0736	100.20	8.337	8.19
89	M21	S	E	F	950615	5	950630	150	66.7	0.0772	148.20	3.603	12.11
90	M22	S	E	F	950615	5	950630	149	65.8	0.0888	148.00	3.831	12.09
91	M23	S	E	M	950615	5	950630	165	84.4	0.0767	181.50	9.534	14.83
92	M24	S	E	F	950615	5	950630	148	68.2	0.0482	243.30	8.278	19.88
93	M25	S	E	F	950615	5	950630	164	90.4	0.1241	139.40	1.658	11.39
94	M26	S	E	F	950615	5	950630	152	68.6	0.1259	106.10	3.869	8.67
95	M27	S	E	F	950615	5	950630	157	79.9	0.0820	177.60	3.488	14.51
96	E41	K	E	M	950616	6	950630	130	46.3	0.0669	168.20	9.789	13.74
97	E42	K	E	M	950616	6	950630	109	25.6	0.0647	136.30	8.800	11.14
98	E43	K	E	M	950616	6	950630	123	38.5	0.0460	159.70	4.413	13.05
99	E44	K	E	F	950616	6	950630	140	58.9	0.0844	117.50	2.187	9.60
100	E45	K	E	M	950616	6	950630	175	116.2	0.1445	111.10	2.656	9.08
101	E46	K	E	M	950616	6	950630	173	125.1	0.1019	135.80	4.946	11.09
102	E47	K	E	M	950616	6	950630	140	60.9	0.0850	115.60	7.902	9.44
103	E48	K	E	M	950616	6	950630	174	124.5	0.1605	153.30	3.477	12.52
104	E49	K	E	M	950616	6	950630	158	86.6	0.1296	115.40	7.143	9.43
105	A1	K	A	F	950617	7	950630	149	71.4	0.0961	90.82	6.009	7.42
106	A2	K	A	M	950617	7	950630	164	113.7	0.1400	87.80	4.614	7.17
107	A3	K	A	M	950617	7	950630	173	151.2	0.1625	141.30	3.412	11.54
108	A4	K	A	F	950617	7	950630	163	95.1	0.1104	100.60	2.084	8.22
109	A5	K	A	M	950617	7	950630	153	77.7	0.0926	82.51	7.424	6.74
110	A6	K	A	F	950617	7	950630	152	84.6	0.1343	106.30	1.599	8.68
111	A7	K	A	M	950617	7	950630	124	42.0	0.0605	132.60	4.254	10.83
112	F1	K	E	M	950630	8	950707	169	116.4	0.1189	139.30	7.055	11.38

OBS	SAMPLE	PLACE	METHOD	SEX	COL_DATE	COL_NUM	AN_DATE	TL	WT	BRAIN	VMAX	CV	ACTIVITY
113	F2	K	E	F	950630	8	950707	152	76.2	0.0737	167.90	3.080	13.72
114	F3	K	E	F	950630	8	950707	156	90.7	0.1086	153.60	2.164	12.55
115	F4	K	E	F	950630	8	950707	185	139.3	0.1498	154.10	3.553	12.59
116	F5	K	E	M	950630	8	950707	170	103.5	0.1713	100.40	5.039	8.20
117	F6	K	E	F	950630	8	950707	176	118.5	0.1537	88.46	3.424	7.23
118	F7	K	E	M	950630	8	950707	158	101.1	0.0823	160.10	4.115	13.08
119	F8	K	E	M	950630	8	950707	164	92.9	0.1021	125.80	4.098	10.28
120	F9	K	E	M	950630	8	950707	178	127.9	0.1008	116.70	8.342	9.53
121	F10	K	E	M	950630	8	950707	161	102.8	0.0880	151.10	3.909	12.34
122	KH1	K	F	M	950627	9	950707	174	114.3	0.0675	88.24	6.982	7.21
123	KH2	K	F	M	950627	9	950707	160	88.0	0.0662	130.60	4.160	10.67
124	KH3	K	F	M	950627	9	950707	165	117.4	0.0609	102.40	4.082	8.37
125	KH4	K	F	M	950627	9	950707	193	176.0	0.0991	112.50	5.961	9.19
126	KH5	K	F	M	950627	9	950707	144	63.0	0.1321	142.70	2.504	11.66
127	KH6	K	F	F	950627	9	950707	151	73.9	0.0862	186.20	3.585	15.21
128	KH7	K	F	M	950627	9	950707	162	95.9	0.1182	141.20	6.744	11.54
129	KH8	K	F	M	950627	9	950707	143	62.8	0.0859	113.30	8.868	9.26
130	KH9	K	F	F	950627	9	950707	153	73.3	0.1421	176.00	3.108	11.50
131	KH10	K	F	M	950627	9	950707	154	73.9	0.1447	151.60	9.115	12.39



1996 Technical Report
Addendum to 1995 Report

To:

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April 7, 1997

Cholinesterase Activity in Adult Bluegill (*Lepomis macrochirus*) from Mark Twain Wildlife Refuge in Summer, 1996

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Four collections of bluegill were made from the Keithsburg backwater area of the Mark Twain National Wildlife Refuge on the upper Mississippi River during the summer of 1996. Below is a summary of means for each collection. As in 1995, there was no evidence of inhibition of brain cholinesterase activity.

Table 1. Summary Statistics for Each Sampling in 1996 (means ± SD)

	April 23	May 21	July 17	August 12
Number fish	22	17	24	27
Mean TL (mm)	144 ± 20	134 ± 22	152 ± 16	152 ± 17
Mean Wet Weight (g)	61.5 ± 22.8	58.1 ± 30.3	77.7 ± 25.7	78.4 ± 29.9
Mean ChE Activity	10.91 ± 2.74	9.27 ± 1.66	11.67 ± 1.42	12.79 ± 2.06

Below is an overall means comparison between 1995 and 1996. Fish tended to be smaller in 1996, but also tended to have higher brain cholinesterase activities.

Table 2. Summary Statistics of Bluegill for 1995 and 1996 (means ± SD)

	1995	1996	1995 and 1996
Total number fish	131	90	221
Mean TL (mm)	161 ± 17	147 ± 20	155 ± 20
Mean Wet Weight (g)	97.06 ± 34.4	70.2 ± 28.3	86.1 ± 34.6
Mean Brain Weight (g)	0.1017 ± 0.0315	0.0925 ± 0.0309	0.0979 ± 0.0315
Mean ChE Activity	9.73 ± 2.56	11.38 ± 2.35	10.40 ± 2.60

Attached is a table listing raw data for bluegill sampled in 1996. Symbols have the same meanings as in 1995.

OBS	SAMPLE	PLACE	METHOD	SEX	COL_DATE	COL_NUM	AN_DATE	TEMP	TL	WT	BRAIN	DIL	VMAX	CV	ACTIVITY
1	KHA1	NL	F	M	960423	1	960511	13.0	100	18.6	0.0469	100	134.40	8.786	10.98
2	KHA2	NL	F	F	960423	1	960511	13.0	130	44.6	0.1090	100	137.70	7.543	11.25
3	KHA3	NL	F	F	960423	1	960511	13.0	145	69.3	0.0993	100	151.30	0.913	12.36
4	KHA4	NL	F	F	960423	1	960511	13.0	150	69.8	0.1012	100	151.20	3.324	12.35
5	KHA5	NL	F	M	960423	1	960511	13.0	160	87.5	0.1065	100	131.00	6.144	10.70
6	KHA6	NL	F	F	960423	1	960511	13.0	95	15.2	0.0627	100	143.40	0.594	11.72
7	KHA8	NL	F	F	960423	1	960511	13.0	162	80.2	0.1427	100	107.00	9.160	8.74
8	KHB1	SS	F	F	960423	1	960525	16.2	163	90.6	0.0786	98	76.33	4.899	6.11
9	KHB2	SS	F	M	960423	1	960525	16.2	164	86.2	0.0765	100	122.50	3.163	10.01
10	KHB3	SS	F	M	960423	1	960426	16.2	160	81.4	0.1468	100	97.28	2.707	7.95
11	KHB4	SS	F	F	960423	1	960426	16.2	162	90.3	0.1522	100	122.70	1.489	10.02
12	KHB5	SS	F	F	960423	1	960426	16.2	153	71.1	0.1030	100	137.40	2.891	11.23
13	KHB6	SS	F	M	960423	1	960511	16.2	162	83.0	0.0467	100	216.10	7.497	17.66
14	KHB7	SS	F	F	960423	1	960426	16.2	151	65.0	0.1255	100	126.30	11.030	10.32
15	KHB8	SS	F	F	960423	1	960426	16.2	153	69.6	0.1155	100	152.60	1.000	12.47
16	KHB9	SS	F	F	960423	1	960525	16.2	133	41.7	0.0400	100	211.30	8.413	17.26
17	KHB10	SS	F	F	960423	1	960511	16.2	144	58.0	0.0502	100	135.90	2.138	11.10
18	KHB11	SS	F	F	960423	1	960525	16.2	149	58.9	0.0903	100	92.61	4.607	7.57
19	KHB12	SS	F	F	960423	1	960525	16.2	134	41.3	0.0783	100	100.80	2.930	8.24
20	KHB13	SS	F	F	960423	1	960525	16.2	143	55.0	0.0810	100	108.80	2.668	8.89
21	KHB14	SS	F	F	960423	1	960426	16.2	133	46.4	0.0554	100	157.10	9.452	12.84
22	KHB15	SS	F	F	960423	1	960511	16.2	113	28.3	0.0925	100	123.40	4.045	10.08
23	KH1	NL	E	M	960521	2	960524	19.5	171	112.9	0.1092	100	81.13	7.037	6.63
24	KH2	NL	E	M	960521	2	960524	19.5	145	73.4	0.0947	100	114.60	5.908	9.36
25	KH3	NL	E	M	960521	2	960524	19.5	162	107.6	0.0957	100	148.80	5.773	12.16
26	KH4	NL	E	M	960521	2	960524	19.5	145	68.7	0.0699	100	94.24	4.913	7.70
27	KH5	NL	E	M	960521	2	960524	19.5	115	32.5	0.0312	100	103.90	3.862	8.49
28	KH6	NL	E	M	960521	2	960524	19.5	114	33.2	0.0966	100	104.40	8.901	8.53
29	KH7	NL	E	M	960521	2	960524	19.5	115	31.1	0.0431	100	138.50	3.483	11.32
30	KH8	NL	E	M	960521	2	960524	19.5	112	26.4	0.0422	100	126.00	12.500	10.29
31	KH9	NL	E	F	960521	2	960524	19.5	110	29.1	0.0485	100	92.42	6.373	7.55
32	KH10	NL	E	M	960521	2	960524	19.5	95	18.3	0.0589	100	119.30	7.616	9.75
33	KH11	SS	E	F	960521	2	960524	20.5	143	70.8	0.0731	100	88.93	2.874	7.27
34	KH12	SS	E	F	960521	2	960524	20.5	163	96.4	0.0900	100	93.44	1.439	7.63
35	KH13	SS	E	F	960521	2	960524	20.5	154	88.2	0.0934	100	117.60	6.841	9.61
36	KH14	SS	E	M	960521	2	960524	20.5	139	58.4	0.0665	100	112.40	3.223	9.18
37	KH15	SS	E	M	960521	2	960524	20.5	138	55.2	0.0796	100	149.90	6.232	12.25
38	KH16	SS	E	F	960521	2	960524	20.5	134	57.4	0.0600	100	116.80	5.169	9.54
39	KH17	SS	E	F	960521	2	960524	20.5	112	27.6	0.0554	100	126.80	2.627	10.36
40	KH1	USS	E	M	960717	3	960724	25.1	152	84.7	0.1127	100	144.60	4.065	11.81
41	KH2	USS	E	F	960717	3	960724	25.1	179	122.6	0.1458	100	143.00	1.829	11.68
42	KH3	USS	E	M	960717	3	960724	25.1	142	63.4	0.1276	100	154.90	2.682	12.66
43	KH4	USS	E	M	960717	3	960724	25.1	129	41.6	0.0947	100	171.90	5.215	14.04
44	KH5	USS	E	M	960717	3	960724	25.1	147	69.2	0.0930	100	149.10	3.240	12.18
45	KH6	USS	E	M	960717	3	960724	25.1	161	98.4	0.1150	100	143.10	6.303	11.69
46	KH7	USS	E	M	960717	3	960724	25.1	132	47.8	0.0779	100	131.00	5.238	10.70
47	KH8	USS	E	M	960717	3	960724	25.1	147	65.3	0.0666	100	134.10	1.922	10.96
48	KH9	USS	E	M	960717	3	960724	25.1	155	87.8	0.1017	100	158.80	5.476	12.97
49	KH10	USS	E	M	960717	3	960724	25.1	145	65.9	0.1233	100	148.80	4.066	12.16
50	KH11	USS	E	F	960717	3	960724	25.1	175	112.3	0.0948	100	154.70	6.716	12.64
51	KH12	USS	E	F	960717	3	960724	25.1	146	61.1	0.1085	100	99.65	9.019	8.14
52	KH23	LSS	E	M	960717	3	960724	24.5	156	75.2	0.0940	100	130.10	2.557	10.63
53	KH24	LSS	E	F	960717	3	960724	24.5	131	42.9	0.0722	100	145.70	5.900	11.90
54	KH25	LSS	E	M	960717	3	960724	24.5	139	56.0	0.0758	100	128.40	5.748	10.49
55	KH26	LSS	E	M	960717	3	960724	24.5	156	81.8	0.0903	100	132.80	6.235	10.85
56	KH27	LSS	E	F	960717	3	960724	24.5	173	114.4	0.1215	100	116.80	2.550	9.54

OBS	SAMPLE	PLACE	METHOD	SEX	COL_DATE	COL_NUM	AN_DATE	TEMP	TL	WT	BRAIN	DIL	VMAX	CV	ACTIVITY
57	KH28	LSS	E	F	960717	3	960724	24.5	172	100.2	0.0978	100	127.90	8.013	10.45
58	KH29	LSS	E	F	960717	3	960724	24.5	170	98.2	0.1425	100	152.00	7.925	12.42
59	KH30	LSS	E	M	960717	3	960724	24.5	129	42.6	0.0667	100	167.40	0.658	13.68
60	KH31	LSS	E	M	960717	3	960724	24.5	148	66.0	0.1027	100	142.10	6.091	11.61
61	KH32	LSS	E	M	960717	3	960724	24.5	135	52.5	0.0981	100	165.80	5.720	13.55
62	KH33	LSS	E	F	960717	3	960724	24.5	176	125.9	0.1495	100	162.20	2.490	13.25
63	KH34	LSS	E	F	960717	3	960724	24.5	163	87.7	0.1511	100	123.30	9.240	10.07
64	KH1	LSS	F	M	960812	4	960822	23.5	184	154.4	0.1594	100	131.30	2.365	10.73
65	KH2	LSS	F	M	960812	4	960822	23.5	172	134.3	0.1177	100	165.60	2.756	13.53
66	KH3	LSS	F	M	960812	4	960822	23.5	154	92.3	0.1049	100	137.10	9.694	11.20
67	KH4	LSS	F	M	960812	4	960822	23.5	164	89.1	0.1407	200	92.80	1.761	15.16
68	KH5	LSS	F	M	960812	4	960822	23.5	165	95.4	0.0971	100	112.70	0.460	9.21
69	KH6	LSS	F	F	960812	4	960822	23.5	156	86.0	0.0813	100	184.40	6.455	15.07
70	KH7	LSS	F	M	960812	4	960822	23.5	161	83.3	0.0852	200	94.71	3.453	15.48
71	KH8	LSS	F	M	960812	4	960822	23.5	160	99.8	0.0999	100	179.70	4.818	14.68
72	KH9	LSS	F	M	960812	4	960822	23.5	148	70.0	0.1024	100	172.40	2.115	14.09
73	KH10	LSS	F	M	960812	4	960822	23.5	161	79.9	0.0763	100	117.40	5.368	9.59
74	KH11	LSS	F	M	960812	4	960822	23.5	129	43.3	0.0454	200	103.30	0.630	16.88
75	KH12	LSS	F	M	960812	4	960822	23.5	137	55.6	0.0589	100	163.60	2.664	13.37
76	KH13	LSS	F	F	960812	4	960822	23.5	139	49.0	0.0797	100	155.60	2.077	12.71
77	KH14	LSS	F	M	960812	4	960822	23.5	144	67.7	0.0662	100	162.40	3.879	13.27
78	KH20	NL	F	M	960812	4	960822	25.9	165	93.1	0.0879	100	154.70	3.451	12.64
79	KH21	NL	F	F	960812	4	960822	25.9	176	118.9	0.1158	100	141.70	3.555	11.58
80	KH22	NL	F	M	960812	4	960822	25.9	173	102.4	0.1251	100	151.80	2.069	12.40
81	KH23	NL	F	F	960812	4	960822	25.9	143	58.0	0.1216	100	159.60	5.575	13.04
82	KH24	NL	F	M	960812	4	960822	25.9	165	95.2	0.1477	100	130.50	3.477	10.66
83	KH25	NL	F	F	960812	4	960822	25.9	153	76.7	0.1130	100	130.30	6.147	10.65
84	KH26	NL	F	F	960812	4	960822	25.9	153	75.5	0.0787	100	137.00	3.805	11.19
85	KH27	NL	F	F	960812	4	960822	25.9	150	84.8	0.1356	100	126.50	1.164	10.34
86	KH28	NL	F	M	960812	4	960822	25.9	148	55.0	0.1205	100	147.10	2.737	12.02
87	KH29	NL	F	M	960812	4	960822	25.9	130	46.1	0.0777	100	180.60	3.400	14.76
88	KH30	NL	F	F	960812	4	960822	25.9	129	41.2	0.0367	200	88.84	4.277	14.52
89	KH31	NL	F	M	960812	4	960822	25.9	111	19.3	0.0541	100	170.90	7.580	13.96
90	KH32	NL	F	F	960812	4	960822	25.9	135	49.7	0.0598	100	173.60	1.404	14.18

APPENDIX B

Quality Control and Quality Assurance Plan
for
Water and Sediment Chemical Analyses

TITLE: **Quality Assurance Program Plan**
(Revision -05/94)

CONTROL NUMBER: 89-1

University Hygienic Laboratory
The University of Iowa
Oakdale Hall
Iowa City, Iowa 52242

W. J. Hausler, Jr., Ph.D. Director
J. P. Getchell, Dr. P.H., Associate. Director

This plan covers the analytical activities of the following laboratory programs:

Bureau of Environmental Quality and Control

Organic Analysis Division
Inorganic Analysis and Limnology Division
Environmental Monitoring and Radiologic Health Division

Bureau of Disease Control

Microbiology Division
Viral and Rickettsial Diseases Division
Metabolic & Genetic Disease Screening Program
Immunology Division

QUALITY ASSURANCE PROGRAM PLAN

ANNUAL REVIEW AND UPDATE

Date	Reviewed By	Revisions By	Approved by Director
6/15/83	[illegible]	staff RLC	
3/11/84	RLC		
2/20/85	RL Cada	RLC	
20 Sept 1985	D. Mayer	D. Mayer	
2 Jan 1986	D. Mayer	D Mayer	
20 Mar 1987	D. Mayer	D. Mayer	
20 Oct 1987	D. Mayer	UHL Staff & Dr. Hawker	[signature]
18 Oct. 1988	C. Segan	UHL STAFF	
20 Feb 1990	C. Segan	QA Group	
26 March 1991	C Segan	None made	
21 Jan 1992	C Segan		

QUALITY ASSURANCE STANDARD OPERATING PROCEDURE MANUAL

ANNUAL REVIEW AND UPDATE

Date	Reviewed By	Revisions By	Approved by Director
2/27/90	C. Saxon	C. Saxon	
3/29/91	C. Saxon	C. Saxon	
5/13/91	Stacy Freeburg	Stacy Freeburg	

QUALITY ASSURANCE PROGRAM PLAN

ANNUAL REVIEW AND UPDATE

Date	Reviewed By:	Revisions By:	Approved by Director
5/14/94	QA Group	QA Group	Jane P. [illegible]

Introduction

As a service laboratory for public and private agencies, the University Hygienic Laboratory is asked to provide analytical expertise in a variety of modes and matrices. Correct interpretation of these data demand that the information reflects the true identification and quantitation of organisms and components of biological and environmental matrices. This document outlines the general quality assurance procedures covering all analytical activities performed by the University Hygienic Laboratory.

Policy Statement

The University Hygienic Laboratory holds firmly that the usefulness of medical or environmental decisions made by data users is to a major extent determined by the reliability of laboratory data. In the absence of accurate, precise information, all comments or decisions about health or environmental impacts due to physical, chemical, or biological incidents are subjective. To enforce this policy, the University Hygienic Laboratory has designated a Quality Assurance Group to oversee and support the program activities, that provide decision-makers with data and information, to properly monitor and recommend public health and environmental changes.

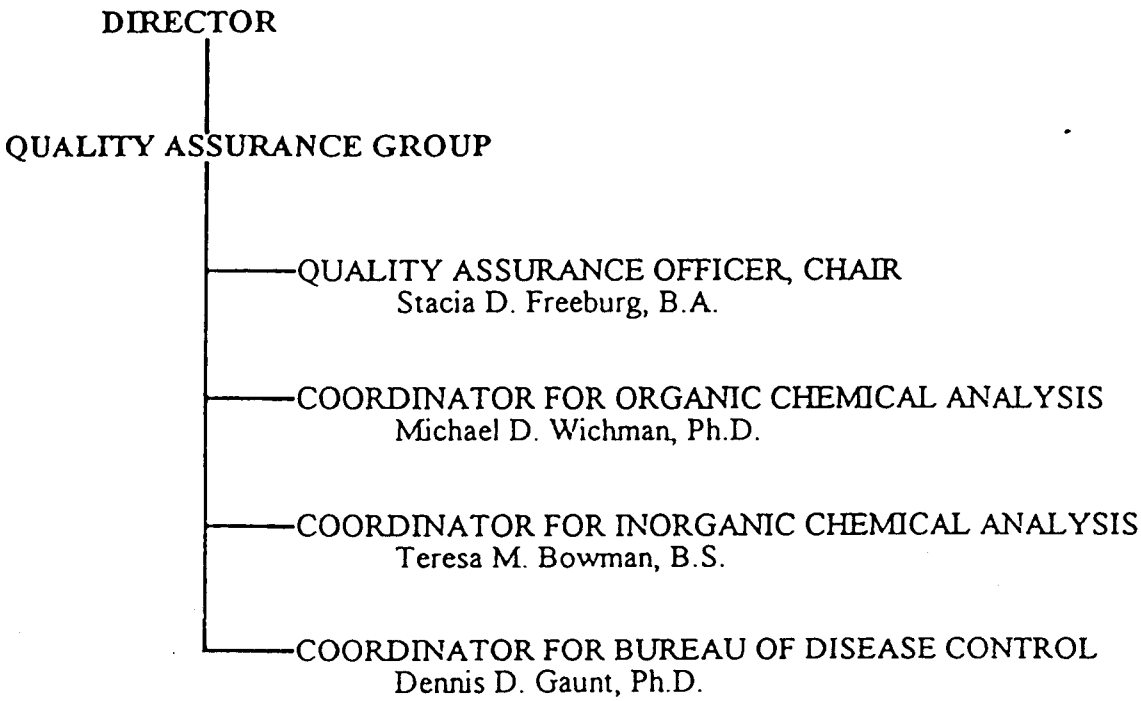
Quality Assurance Group

The Quality Assurance Group reviews and approves all Quality Assurance Project Plans developed and implemented by each organizational unit within the University Hygienic Laboratory.

The Quality Assurance Group provides periodic reports as well as a detailed semi-annual written report to the Laboratory Director.

The staff of the Quality Assurance Group serves as the clearinghouse for information relative to recommendations and requirements in quality assessment. They assist in the implementation of new techniques and improved methodology through training and consultation with Bureau Chiefs, Associate Director, and program managers.

Figure I: Quality Assurance Group



On April 1 and October 1 of each year, the Quality Assurance Group submits a report to the Director of the Laboratory. These reports contain at least the following information:

1. Status of Quality Assurance Program
2. Status of Quality Assurance Project Plans or Work/Quality Assurance Plans
3. Results of performance audit/proficiency testing
4. Results of systems audits
5. Significant problems, corrective actions, plans and recommendations
6. Quality assurance training needs and accomplishments

Quality Assurance Management

The quality assurance activities (those activities which assess the quality of the Laboratory's various outputs) of the University Hygienic Laboratory are assigned to the organizational entity titled the Quality Assurance Group. This group is independent of the analytical divisions of the laboratory and reports to the Director. The organization chart is shown in Figure I.

Quality assurance data generated by the analytical units are reviewed on a systematic basis and summary reports are generated. Maintenance of generally-accepted or UHL-specific quality control measures and implementation of corrective action are the responsibility of the individual program managers and supervisors with guidance and assistance from the Quality Assurance Group.

Results of external audits and proficiency testing are reviewed by the Quality Assurance Group and copies provided to the analytical units. A copy of each audit is retained by the Quality Assurance Coordinator and/or Officer. Requests for reports and corrective actions are addressed to appropriate supervisors. All response plans and "corrective actions completed" reports are submitted to and retained by the Quality Assurance Officer and/or respective Quality Assurance Coordinator.

Personnel

The staff of the Quality Assurance Group are required to have sufficient education and experience as follows:

1. Sufficient professional status to work effectively with program managers.
2. Education and training in a physical, chemical or biological science discipline.
3. Knowledge of descriptive and inferential statistics.
4. Knowledge of appropriate laws, regulations, and guidelines for quality assurance activities

5. Adequate communication skills.

Professional staff of the group are reviewed annually under general guidelines of The University of Iowa and specific performance guidelines of the Hygienic Laboratory, and the evaluations are recorded in the personnel record.

Facilities, Equipment and Services

As part of the systems evaluation process, the Quality Assurance Group reviews facilities, equipment, and services used in diagnostic and environmental monitoring programs.

Facilities must be safe and of adequate size for the intended purpose. Satisfactory lighting, ventilation, temperature, noise levels, and humidity are maintained to protect the safety and health of staff and to maintain proper operating performance of equipment. Personnel are provided with adequate protective equipment for their personal health and safety as well as periodic health screening and immunizations where appropriate. A copy of the Facilities Committee report concerning facilities deficiencies or recommended modifications to the administration is available to the Quality Assurance Group for review. The Quality Assurance Group works with the Facilities Committee to resolve facilities deficiencies. The University Hygienic Laboratory Safety Committee periodically reviews, and at least annually generates a report that is also available to the Quality Assurance Group for review concerning the status of health and safety monitoring of laboratory facilities and personnel.

Utility services, such as electricity, gas, water and air are appropriately available for the generation and processing of laboratory information. Disaster and fire plans are readily available to laboratory staff by the Safety Committee.

General laboratory equipment is present in sufficient quantity and condition, operationally consistent for the intended use and provides for the generation and

processing of data possessing the required levels of quality and integrity required by this Quality Assurance Program Plan. Examples of general laboratory equipment are air conditioners, ovens, furnaces, generators, refrigerators, incubators, laboratory hoods, sinks, benches, etc.

Routine maintenance of laboratory equipment is a procedural and administrative requirement and may:

* extend the usable life of the instrument
* reduce the number of wearout failures and instrument replacement costs
* predict and minimize down-time
* reduce error of laboratory data generation.

The necessary components of preventive maintenance systems are:

* Equipment identification - name, location, manufacturer, serial number, purchase date, etc.
* Responsibility assignment - whether maintenance can be performed by technical/maintenance personnel in-house, or name and phone number of person(s) to contact.
* Task definition - those action items which are to be performed by designated responsible personnel.
* Frequency schedule - the intervals at which the defined tasks are to be performed.
* Documentation and Review - recording of date and type of maintenance performed and evidence of periodic review by supervisory staff.

Maintenance and calibration intervals are as frequent as recommended by the method(s) utilized, regulatory process, supervisor, and the manufacturer.

General field equipment is present in sufficient quantity and is operationally consistent with its intended use. This includes equipment such as thermometers, sampling apparatus, pH meters, flow meters, etc.

Data Generation

Laboratory analytical activities include all field and laboratory investigations which generate data and data-processing activities such as data entry, storage, retrieval, and analysis. As appropriate, Quality Assurance Project Plans, Work/Quality Assurance Plans, and Procedure Manuals are developed and implemented for all diagnostic and environmental monitoring activities such that all data generated and processed will be accurate, precise, reliable, complete, defensible, and comparable.

Quality Assurance Project Plans, Work/Quality Assurance Plans, and Procedure Manuals contain the following items:

1. Title page, with provision for approval signatures
2. Table of contents
3. Project description/objectives
4. Project organization and designated responsibilities
5. Quality assurance objectives for measurement data, including specific limits for detection/quantitation, precision, accuracy, completeness, representativeness, and comparability
6. Sampling procedures
7. Sample transportation and custody
8. Calibration procedures, references and frequency
9. Parameter table and analytical methods
10. Data reduction, analysis, validation, and reporting
11. Internal quality control checks and frequency

12. Quality Assurance Performance Audits (proficiency tests, proficiency analytical testing), systems audits and frequency
13. Quality Assurance Reports - type and frequency
14. Preventive maintenance procedures and schedules
15. Specific procedures to be used in routinely assessing data precision and accuracy, representativeness, comparability and completeness of the specific parameters involved
16. Corrective action procedures and responsibilities

The Quality Assurance Group reviews Quality Assurance Project Plans and Work/Quality Assurance Plans, provides input, recommends changes where necessary and approves final plans of work/quality assurance plans that are generated by the University Hygienic Laboratory. The Group has ready access to all approved Project Plans as well as all Procedure Manuals for all programs within the University Hygienic Laboratory.

Quality Assurance Project Plans or Work/Quality Assurance Plans contain some portions which are consistent and do not vary with different studies. All such routine tasks are written as standard operating procedures for the Laboratory. Standard operating procedures are detailed documents describing who performs what, when, where, how, and why in a step-wise manner. They are sufficiently complete and detailed to ensure:

1. Data of quality and integrity are collected to meet the analytical objectives.
2. Minimal loss of data due to out-of-control conditions.

Quality Assurance Project Plans or Work/Quality Assurance Plans are:

1. Adequate to establish traceability of standards, instrumentation, samples, and data;
2. Sufficiently clear such that a user with basic required education, experience and/or training can properly use them;

3. Amply complete such that the user/reader can follow the directions in a step-wise manner through the sampling, analysis and data-handling process;
4. Consistent with sound scientific/engineering principles;
5. Consistent with current governmental regulations and guidelines;
6. Consistent with the instrument manufacturer's specific instruction manuals.

Procedure Manuals provide that documentation is sufficiently complete to:

1. Guide the performance of all tasks such that a user with basic required education, experience and/or training can properly perform given tasks.
2. Explain the cause for questionable data.
3. Validate the accuracy of data each time they are recorded, calculated, or transcribed.

To accomplish these objectives, Procedure Manuals address the following:

1. Sampling and analytical methodology;
2. Appropriate probes, collection devices, storage and shipping containers, and sample additives or preservatives;
3. Special precautions, such as holding times and protection from temperature extremes, light, reactivity, and combustibility;
4. Federal reference, equivalent, and alternate test methods;
5. Instrumentation selection and use;
6. Calibration and standardization;
7. Frequency of blank analyses;
8. Preventive and remedial maintenance;
9. Replicate sampling and analysis;
10. Blind and spiked samples;
11. Quality control procedures such as inter- and intra- laboratory activities;

12. Documentation;
13. Sample custody and handling procedures;
14. Sample transportation;
15. Special safety considerations;
16. Data handling and evaluation, including expected quantitation/detection limits, precision, accuracy, completeness, representativeness, and comparability;
17. Service contracts;
18. Document control.

All diagnostic and environmental monitoring must adhere to established published regulations, methods and guidelines. When deviations occur they must be justified and documented.

Data Processing

Data processing includes all aspects of data acquisition, reduction, storage and transfer. Each Quality Assurance Project Plan or Work/Quality Assurance Plan contains instructions for monitoring and verifying the reliability of data processing and handling systems, either automated or manual.

1. Collection - Each Quality Assurance Project Plan or Work/Quality Assurance Plan specifies the checks to be used in avoiding or minimizing errors in the data collection process;
2. Validation - Defined as the process whereby data are reviewed and accepted or rejected based on a defined set of criteria. Each Quality Assurance Project Plan or Work/Quality Assurance Plan specifies the criteria used in validating the data completeness and accuracy;
3. Storage - Procedures are established to ensure data integrity and security. The Quality Assurance Project Plans or Work/Quality Assurance Plans specify how

data will be stored, including media, conditions, location, retention time and access;

4. Data Transfer - Each Quality Assurance Project Plan or Work/Quality Assurance Plan specifies procedures to be used to ensure that data transfer, e.g., copying raw data from notebook onto data form, or copying from computer tape to disk, is error-free;

5. Reduction - Each Quality Assurance Project Plan or Work/Quality Assurance Plan identifies the processes used to change the form, in terms of size or dimensions, of the data set. Each type of reduction processing contains methods permitting the review and validation of the reduction procedure.

Data Quality Assessment

The quality of all data generated and processed is assessed for accuracy, precision, completeness, comparability and representativeness. Where available, approved methodology is used; if not available, other published methods are reviewed and validated by the program staff. The results of each assessment are documented in the Quality Assurance reports and in progress and final project reports.

Specific areas of assessment in the Quality Assurance Project Plans or Work/Quality Assurance Plans are:

1. Accuracy assessment - Methods by which the reported values are comparable to the "true value." Included are:

   a. Instrument requirements - specification of equipment and documentation of maintenance and calibration;

   b. Standard traceability - all calibration materials traceable to appropriate standards when available;

c. Sample tracking - system to ensure uniqueness of sample from collection through processing;

d. Data traceability - documentation to allow complete reconstruction from field records through data storage and retrieval;

e. Methodology - strict adherence to approved standard operating procedures;

f. Reference to spiked samples - use of standard material in comparable matrices to unknown samples (*Note: simple sample spiking may result in poor accuracy assessment due to sample matrix effects*);

g. Daily control limits and those activities performed when out-of-control situations exist.

h. Performance audit - participation in interlaboratory comparison programs supplementing intralaboratory performance audits.

2. Precision assessment - The reproducibility of the measurement process may be assessed by:

a. Replicate sampling - aliquots of the sample are within specified limits;

b. Duplicate spike analyses, duplicate analyses, and/or long term spike recovery data for given analyses.

c. Collocated monitors - sample data from devices in collocation are within predetermined acceptance limits;

d. Interlaboratory testing - results of performance are within the precision limits calculated from the participant laboratories;

e. Instrument checks - routine checks, such as zero and span, noise levels, drift, flow rate and linearity are performed and documented to demonstrate that variables are within predetermined acceptance limits.

3. Completeness - The quality of data needed to support a planning or enforcement action. Each Quality Assurance Project Plan or Work/Quality Assurance Plan considers the potential for environmental change with respect to time.

4. Comparability - Each Quality Assurance Project Plan or Work/Quality Assurance Plan assures the comparability of data produced by the University Hygienic Laboratory. Comparability involves the standardization of sampling analysis data, formatting, reporting units, and expression or interpretation of results in a manner to allow comparison with applicable standards.

5. Representativeness - Procedures are included in the Quality Assurance Project Plan or Work/Quality Assurance Plan to ensure that each sample collected is representative of the milieu from which it is derived. Assessment of representativeness include:

   a. Site Selection - preidentified, documentably logical location for the variable studied;

   b. Site Description - specific, coordinate identification including photo documentation;

   c. Sampling Conditions - physical descriptors of the sampling location, which may include such parameters as humidity, wind speed and direction, temperature and barometric pressure.

6. Standards and Sample Analysis - Procedures are included in the Quality Assurance Project Plan or Work/Quality Assurance Plan to ensure that the results generated are valid. Specifically:

   a. Standard curves must contain an appropriate minimum of data points plus a zero value (calibration blank) when appropriate;

   b. Standard curves are not extrapolated beyond the highest standard;

   c. When sample results fall outside calibration control limits, the sample must be diluted and reanalyzed or the instrument must be recalibrated over a greater linear dynamic range, whichever is appropriate.

Corrective Action

A comprehensive quality assurance management system provides for the contingency of rejectable data. The realization that conditions exist necessitating the withholding and/or review of data or measurement information may come from a variety of sources.

Each Quality Assurance Project Plan or Work/Quality Assurance Plan establishes and describes the system for corrective actions to be taken when necessary. Each analyst is responsible for applying the quality control procedures that are given in the respective standard operating procedure manuals. Any deviations or out-of-control situation must be resolved or reported to the immediate supervisor before analyzing samples. Program supervisors are responsible for implementation of corrective actions. The Quality Assurance Group is available for consultation and assistance in determining appropriate actions.

1. Quality assurance audits that most likely will detect problems and need for corrective actions are:

a. Transcendence of predetermined analytical limits,

b. Performance audits, including interlaboratory comparisons,

c. Systems audits,

d. Deviation from procedure manual,

e. Facility or equipment malfunction.

2. Specific audits which require immediate corrective action:

a. If control sample results exceed method specific QC acceptance criteria, the supervisor must be notified for corrective action.

b. If duplicate sample analyses is required by the determinative method, and the duplicates deviate by more than the method specific QC acceptance criteria, the supervisor must be notified for corrective action.

c. Spiked samples, duplicates or control samples, as appropriate, must be run with each batch of specimens. If any deviate by more than the acceptance criteria listed in the determinative method utilized, the supervisor must be notified for corrective action.

d. Method or system blanks must demonstrate that interferences from the analytical process are within method specific acceptance limits.

Resolution of potentially error-producing events are documented in a report prepared by the program supervisor and submitted to the Quality Assurance Group. The process will be reviewed and approved by appropriate senior management. When appropriate, the Quality Assurance Project Plan, Work/Quality Assurance Plan, or Procedure Manual is revised to reflect policies and procedures appropriate to minimize a repeat of the corrective action event.

History of Quality Assurance Program

ITEM	DATE
Designation of Quality Assurance Unit	11/80
Development of Quality Assurance Program Plan	12/80
Quality Assurance Program Plan	06/82
Reviewed and Revised:	06/83
Reviewed:	06/84
Reviewed and Revised:	02/85
Reviewed and Revised:	09/85
Reviewed and Revised:	01/86
Reviewed and Revised:	03/87
Major Revision:	10/87
Reviewed and Revised:	10/88
Reviewed and Revised:	02/90
Reviewed:	03/91
Reviewed:	01/92
Reviewed and Revised:	05/94
Quality Assurance Project Plans and Work/Quality Assurance Plans	As Required (continual)

ETHICS STATEMENT

University Hygienic Laboratory

It is explicitly and without exception the policy of the University Hygienic Laboratory that all work performed by the Laboratory - *including all employees in whatever capacity* - must be performed with the highest ethical standards. Falsification or inappropriate manipulation of data is not to be tolerated.

The Laboratory recognizes that workload may be sporadic and at times extremely heavy. Efforts of employees to meet these heavy workloads are appreciated on behalf of the agencies, individuals and corporations utilizing our services; under no circumstances, however, should unethical "shortcuts" be taken. Every effort should be made to provide services of the highest quality, but if quality control criteria cannot be met, no deceit or concealment may be made.

Employees are reminded, for their own protection and the protection of the Laboratory, that failure to report known unethical behavior may result in the employee being implicated in that behavior. Avenues are available to report violations through your supervisor, the Office of the Director, the Quality Assurance Officer, or in some cases through the requesting or supporting agency.

Unethical behavior hurts the people depending on our work, the employees involved as well as their colleagues, the reputation of the Laboratory, and indeed the fabric of society. We must not and will not tolerate it.

The University Hygienic Laboratory

QUALITY ASSURANCE PROGRAM PLAN AND ETHICS STATEMENT DECLARATION

I declare that I have received and reviewed all aspects of the Quality Assurance Program Plan including the Ethics Statement. Furthermore, I understand that any violation of the Ethics Statement is just cause for disciplinary action including suspension and/or dismissal from my position within the University Hygienic Laboratory and liability to civil and criminal penalties.

Signature _____ Date _____

This document will be placed in your personnel file.

APPENDIX C

Water Chemistry Data

South End WQ

	Temp (C)	pH	TDS (uS)	Secchi (c	Depth (ft)	DO (ppm)	Time
4/30/94	11.8	8.8	280	27.5			
5/31/94	24.7	8.23	400	36		12.73	1:35p
6/30/94	26.3	8.42	388	19	4.5	8.94	
7/28/94	24.3	7.19			4	6.56	11:00a
8/31/94	21	7.45	325	24	4	4.57	11:00a
9/30/94	16.2	7.84	403.8	27	4	8.09	11:30a
11/1/94	9.1	8.3	405.6	43	4		
3/21/95	11.3	7.69	333.3	35	5	9.31	11:00a
4/17/95	12.8	8.55	338.9	45.5	6	17.1	11:32a
5/15/95	16.7	9.2	420.5	55	8.5	23.16	12:00a
6/26/95	26.8	8.29	333.9	50.2	5	9.9	pm
7/31/95	29.7	7.65	300.1	17	3	8.66	11:30a
8/30/95	30.2	8.41	334.2	33	3	9.5	
10/3/95		9.11	296.5			26.42	3:00p
12/7/95	2.5	9.2	366.7	39	5.5		

North End WQ

	Temp (C)	pH	TDS (uS)	Secchi (c	Depth (ft)	DO (ppm)	Time
4/30/94	13.7	8.26		20 5/8"	4	12.15	am
5/31/94	24.7	5.57	320	43.9	5	24+	3:51p
6/30/94	28	7.4	370	42.8	3	4.8	
7/28/94	23	7.24					
8/31/94	18.9	7.46	320	36	3.75	6.2	1:30p
9/30/94	16.6	7.59	410.9			10.4	2:00p
11/1/94	9.5	7.6	391.7	58	4		
12/1/94	4.7	7.82	411.3	53	4		
3/21/95	10.5	8.55	344.9	48	4.5	16.67	2:51p
4/28/95	16	8.91	332.9	35	6	17.4	4:15p
5/15/95	17.7	8.43	354		8	13.69	9:30a
6/26/95	25.5	7.9	302.4	37	5	29.06	12:00p
7/31/95	24	7.63	358.5	20.5	3	11.15	11:30a
9/11/95	19.2	8.07	394.6	43	3.5	17.16	
10/3/95	17.6	8.1	350.8	44	3	16.38	3:03p
12/7/95	3.8	8.3	498.3	67			

Nitrate

	South End	North End		Trib 1	Trib 2	Trib 3	Trib 4		
4/30/94	0.3			9.8					
5/31/94	0.1			40					
6/30/94	ND	1.3		9.2	2.1	4.1	ND		
7/28/94	ND			9.4	1	2.3			
8/16/94	2.9	2.9		9.4	2.9	3.5			
8/31/94	ND	3.4							
9/30/94	ND	5.2		9.4	7	0.6			
11/1/94	ND	4.7		9.3	5.7	0.1	0.5		
12/1/94		8.5		8.8	7	3.7	2		
2/15/95	0.4	6.2		9.2	7.6	3.4			
3/21/95	ND	3.4		8.8	3.4	1.7			
4/17/95	ND	2.5	0.7	7.8	3.5	2.6	1		
4/24/95	ND	ND	0.5						
4/28/95	ND	ND		8.5	3	3.3	0.9		
5/4/96	8.5	2	2	0.5	ND	2	2.3		
5/11/95	4.6	0.2							
5/15/95	5	0.1	4.7	0.4	0.1	0.2	0.1		
5/31/95	0.5	0.2		4.4	0.3	1.5	0.4		
6/9/95				0.4	0.3	0.1	0.2		
6/26/95	ND	0.7		8.3	0.9	3	ND		
7/31/95	0.2	4.1	2.4	9.2	1.6	3.3	ND		
8/30/95	0.6								
9/11/95	0.3	5.9	3.9						
10/3/95	0.2	6.2							
12/7/95	ND	6.2							
ND = Below established analytical detection limits.									
Missing data indicates that a sample was not collected for that location and date.									

Phosphates

	South End	North End	North Lake (N)	Trib 1	Trib 2	Trib 3	Trib 4	
5/31/94	200		200					
6/30/94	400	200	500	700	600	400		
7/28/94	200		200	200	200			
8/16/94	100	100	100	200	200			
8/31/94	200	100						
9/30/94	100	0	100	100	600			
11/1/94	ND	100	ND	100	600	300		
12/1/94		0	ND	ND	ND	ND		
2/15/95	ND	100	100	100	ND			
3/21/95	100	0	ND	500	100			
4/17/95	ND	100	100	200	300	600		
4/28/95	100	100	200	1800	100	200		
5/4/96	ND	200	300	200	200	400		
5/15/95	ND	100	100	200	200	200		
5/31/95	200	100	100	100	300	300		
6/9/95			200	100	100	200		
6/26/95	200	200	200	300	200			
7/31/95	300	ND	200	300	200	200		
8/30/95	300							
9/11/95	200	ND						
10/3/95	200	100						
12/7/95	100	100						
ND = Below established analytical detection limits.								
Missing data indicates that a sample was not collected for that location and date.								

Alachlor

	South End	North End	Trib 1	Trib 2	Trib 3	Trib 4	
4/30/94	0.25	0.34	0.15				
5/31/94	0.3		0				
6/30/94	0.24	0.33	0	0.25	0.16	0.58	
7/28/94	0.18		0	0.19	0.15		
8/16/94	0.19	0.16	0	0.16	0		
8/31/94	0.15	0.13					
9/30/94	0.19	0	0	0.13	0.35		
11/1/94	0.13	0.12	0	0.13	0.16	0.19	
12/1/94		0	0	0	0.15	0.25	
2/15/95	0.39	0.39	0	0.13	0.19		
3/21/95	0.18	0.32	0	0.31	0.39		
4/17/95	0.17	0.47	0	0.2	1.41	1.61	
4/28/95	0.16	0.17	0.14	0.26	0.22	0.65	
5/4/96	0.29	0.3	0.27	0.3	0.31	0.86	
5/15/95	0.9	0.65	0.47	1.02	1.36	3.34	
5/31/95	0.42	2.4	0.19	2.4	0.9	2.6	
6/9/95			0.82	1.3	2.5	3	
6/26/95	0.55	0.76	0.13	0.4	0.39		
7/31/95	0.34	0.34	0.18	0.25	0.27	0.34	
8/30/95	0.47						
9/11/95	0.33	0.33					
10/3/95	0.34	0.38					
12/7/95	0.28	0.24					
ND = Below established analytical detection limits.							
Missing data indicates that a sample was not collected for that location and date.							

Metolachlor

	South End	North End	Trib 1	Trib 2	Trib 3	Trib 4	
4/30/94	0.3	0	0				
5/31/94	0.34	0	0				
6/30/94	0.17	0	0	0	0	0	
7/28/94	0.16		0	0	0		
8/16/94	0	0	0	0	0		
8/31/94	0	0					
9/30/94	0	0	0	0	0		
11/1/94	0	0	0	0	0		
12/1/94		0	0	0	0	0	
2/15/95	0	0	0	0	0	0	
3/21/95	0	0	0	0	0		
4/17/95	0	0	0	0	0	0.123	
4/28/95	0	0	0	0	0	0	
5/4/96	0.37	0.2	0.18	0.19	0.31	0.12	
5/15/95	1.73	0.31	0.22	0.29	0.26	0.37	
5/31/95	1.4	3.5	3.9	3.1	1.6	2.1	
6/9/95			2.2	2.1	2.4	2.1	
6/26/95	0.76	0.73	0.36	0.5	0.4		
7/31/95	0.44	0.34	0.3	0.31	0.33	0.32	
8/30/95	0.43						
9/11/95	0.37	0.2					
10/3/95	0.24	0.17					
12/7/95	0.34	0.4					
ND = Below established analytical detection limits.							
Missing data indicates that a sample was not collected for that location and date.							

Triazines

	South End	North End	Trib 1	Trib 2	Trib 3	Trib 4		
4/30/94	0.16	0.4	0					
5/31/94	0.18	0.32	0					
6/30/94	0.24	0.26	0	0.12	0.11	0.1		
7/28/94	0.14		0	0	0.1			
8/16/94	0	0	0	0	0			
8/31/94	0	0						
9/30/94	0	0	0	0	0			
11/1/94	0	0	0	0	0.15	0		
12/1/94		0	0	0	0	0		
2/15/95	0	0	0	0	0			
3/21/95	0	0	0	0	0			
4/17/95	0	0	0	0.11	0.18	0.19		
4/28/95	0	0	0	0.12	0	0.15		
5/4/95	0.19	0	0.1	0.1	0.11	0.11		
5/15/95	1.34	0.6	0.21	1.31	1.18	4.69		
5/31/95	2.1	2.5	1.5	3.1	1.2	2.4		
6/9/95			1.4	1.8	4.7	3.6		
6/26/95	1.05	0.83	0	0.76	0.12			
7/31/95	0.48	0.11	0	0	0.12	0		
8/30/95	0.15							
9/11/95	0.11	0						
10/3/95	0	0						
12/7/95	0	0						
ND = Below established analytical detection limits.								
Missing data indicates that a sample was not collected for that location and date.								

APPENDIX D

Sediment Chemistry Data

Excel Spreadsheet - Kelthsburg Division, Sediment Quality Data for 1994 (Metals mg/Kg dry wgt)																			
Location																			
		% H2O	mg NH3-N/K	ug NH3-N/L	mg PO4-P/Kg	% TOC	% Clay	mg As/kg	mg Cd/kg	mg Cr/kg	mg Cu/kg	mg Pb/kg	mg Ni/kg	mg Zn/kg	Volatile Hydrocarbons (mg/kg)	Total Hydrocarbons (mg/kg	Total Organochlorines (mg/kg	Total PCB (mg/kg
Miss R. levee	KH-1	46	75	1.9	590	2.2	24.8	1.6	ND	25	12	13	18	77	ND	43	ND	ND
Lower Spring Slough	KH-3	66	170	4.4	1100	3.9	30.8	6.4	ND	28	18	17	22	90	ND	93	ND	ND
Middle Spring Slough	KH-5	72	160	6.2	1700	6.4	25.5	7.2	ND	18	14	14	17	67	ND	130	ND	ND
North Lake	KH-7	70	150	4.4	1500	3.8	30.3	5	ND	32	17	17	22	96	ND	76	ND	ND
Edwards R. Spillway	KH-9	60	100	3.8	950	2.3	19.1	3.1	ND	25	14	13	20	80	ND	88	ND	ND
Moist Soil Area	KH-11	38	15		490	1.8	19.8	1.7	ND	19	11	14	15	57	100		ND	ND
Dup KH-5	KH-21	72	160	6.2	1600	7.1	21.8	5.8	ND	17	14	17	17	67	ND	130	ND	ND
Dup KH-11	KH-22	36	18		530	1.4	20.1	1.8	ND	22	11	14	16	60	ND			
ND = Below established analytical detection limit.																		
Missing data indicates that a sample was not collected for that location and date.																		

Sediments 95

North Lake Line Transect										
Ref No.	% Sand	% Co. Silt	% Fi. Silt	% Clay	%TOC	mg PO4-P/Kg	mg NH3-N/Kg	ug Alachlor/Kg	mg NH3-N/L	ug Alachlor/L
KH-1	43.6	11.1	22.7	22.6	3.6	940	98	0.12	11	1.6
KH-2	4.7	18.8	47.5	29	4	1700	200	ND	15	0.98
KH-3	7.3	10.3	47.1	35.4	6.6	2600	220	ND	9	0.97
KH-4	5.6	20.1	46.4	27.9	3.4	1500	190	0.24	12	1.6
KH-5	6.6	20.6	45.4	27.4	3.9	1800	210	ND	8	ND
KH-6	6.6	28.1	37.2	28.1	3.7	1500	190	ND	8.7	ND
KH-7					4.1	2000	260	ND	11	ND
ND = Below established analytical detection limit.										
Missing data indicates that the sample was not collected for that location and date.										

APPENDIX E

Fish Tissue Data

Fish

	Wet Wgt	mg As/kg	mg Cd/kg	mg Cr/kg	mg Cu/kg	mg Pb/kg	mg Hg/kg	mg Ni/kg	mg Se/kg	mg Zn/kg
Carp	KH-94-1	ND	ND	ND	1.2	ND	ND	ND	ND	51
Carp	KH-94-2	ND	ND	ND	ND	ND	ND	ND	ND	42
Carp	KH-94-3	ND	ND	ND	1.2	ND	ND	ND	ND	80
Carp	KH-94-4	ND	ND	ND	1.2	ND	ND	ND	ND	52
Carp	KH-94-5	ND	ND	ND	1.1	ND	ND	ND	ND	41
Carp	KH-94-6	ND	ND	ND	1.3	ND	ND	ND	ND	59
Carp	KH-94-7	ND	ND	ND	ND	ND	ND	ND	ND	60
Carp	KH-94-8	ND	ND	ND	1.2	ND	ND	ND	ND	56
Carp	KH-94-9	ND	ND	ND	ND	ND	ND	ND	ND	64
Carp	KH-94-10	ND	ND	ND	1.5	ND	ND	ND	ND	22
Carp	BT-94-1	ND	ND	ND	ND	ND	ND	ND	ND	83
Carp	BT-94-2	ND	ND	ND	ND	ND	ND	ND	ND	99
Carp	BT-94-3	ND	ND	ND	ND	ND	ND	ND	ND	48
Carp	BT-94-4	ND	ND	ND	ND	ND	ND	ND	ND	28
Carp	BT-94-5	ND	ND	ND	ND	ND	ND	ND	ND	66
Bass	KH-94-1	ND	ND	ND	ND	ND	0.14	ND	ND	8.1
Bass	KH-94-2	ND	ND	ND	ND	ND	0.17	ND	ND	17
Bass	KH-94-3	ND	ND	ND	ND	ND	0.14	ND	ND	14
Bass	KH-94-4	ND	ND	ND	ND	ND	0.16	ND	ND	9.4
Bass	KH-94-5	ND	ND	ND	ND	ND	0.11	ND	ND	15
Bass	BT-94-6	ND	ND	ND	ND	ND	ND	ND	ND	12
Bass	BT-94-7	ND	ND	ND	ND	ND	ND	ND	ND	16
Bass	BT-94-8	ND	ND	ND	ND	ND	ND	ND	ND	9.9
Bass	BT-94-9	ND	ND	ND	ND	ND	ND	ND	ND	15
Bass	BT-94-10	ND	ND	ND	ND	ND	0.2	ND	ND	9
Bluegill	KH-96-13						0.434			
Bluegill	KH-96-14						0.347			
Bluegill	KH-96-15						0.125			
Bluegill	KH-96-16						ND			
Bluegill	KH-96-17						0.661			
Bluegill	KH-96-18						ND			
Bluegill	KH-96-19						0.674			
Bluegill	KH-96-20						0.597			
Bluegill	KH-96-21						0.521			
Bluegill	KH-96-22						0.597			
ND = Below established analytical detection limits.										
Missing data indicates that a sample was not analyzed for that specific heavy metal.										