REPORTS ON THE WALNUT CREEK WATERSHED MONITORING PROJECT, JASPER COUNTY, IOWA WATER YEARS 1995-2000

Geological Survey Bureau Technical Information Series 46





Iowa Department of Natural Resources Jeffrey R. Vonk, Director February 2002



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Geological Survey Bureau Technical Information Series 46

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INTRODUCTION

Keith E. Schilling Iowa Department of Natural Resources, Geological Survey Bureau

This report on the Walnut Creek Watershed Restoration and Water-Quality Monitoring Project consists of a series of articles that summarize the results of monitoring activities in Walnut and Squaw creek watersheds in Jasper County for water years 1995 to 2000. (A water year is a 12-month period, from October 1 through September 30, designated by the calendar year in which it ends.) As discussed in the introductory article of this report, the Walnut Creek project was established in 1995 to monitor the effects of large-scale prairie restoration occurring at the Neal Smith National Wildlife Refuge on water quality in the Walnut Creek watershed.

This report is the third comprehensive report prepared for the Walnut Creek project. In 1999, a summary report was published which reported on results from the first three years of monitoring (water years 1995 to 1997), including land use, discharge and suspended sediment, surface and groundwater quality, and biological monitoring (Schilling and Thompson, 1999). In 2000, results of discharge and suspended sediment monitoring in Walnut and Squaw creek watersheds for water years 1995 to 1998 were examined in detail (Schilling, 2000). Unlike previous reports, this compendium is organized as a series of articles in order to present more discussion associated with individual project components and make individual topics more accessible for viewing on the Iowa Department of Natural Resources, Geological Survey Bureau web page (www.igsb.uiowa.edu).

Following an introductory article describing the project background and land use changes, other articles in this report focus on specific aspects of surface water quality monitoring, including:

- Nitrate, chloride and sulfate concentrations and loads,
- Herbicide concentrations and loads,
- Fecal coliform concentrations,
- Common field parameters of pH, specific conductance, turbidity, etc.,
- Biological monitoring of macroinvertebrates and fish.

Discharge and suspended sediment data were not presented in this report. This information will be included in a later report in conjunction with presentation of a sediment erosion and delivery model for the Walnut and Squaw creek watersheds.

We hope that the monitoring topics discussed in this report are of interest to those following the Walnut Creek project as well as those interested in water quality monitoring in the State of Iowa. Lessons from this project, both positive and negative, offer valuable insight on strategies for monitoring watershed scale relationships between land use and water quality. Improved understanding of these relationships moves us closer to being able to track the effectiveness of best management practices to reduce nonpoint source pollution.

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WALNUT CREEK NONPOINT SOURCE MONITORING PROJECT: BACKGROUND AND LAND USE CHANGES

Keith E. Schilling Iowa Department of Natural Resources, Geological Survey Bureau

INTRODUCTION

Numerous programs employing a variety of best management practices (BMPs) have been implemented in Iowa to mitigate nonpoint source (NPS) pollution from agriculture. However, monitoring water quality improvements resulting from BMPs is not an easy task. Many projects implemented under Section 319 of the Clean Water Act have had little or no monitoring associated with them. Often project leaders assume water quality improvements will occur rather than measuring actual results, or estimate water quality improvements using field-scale or watershed models.

The Walnut Creek Watershed Restoration and Water-Quality Monitoring Project is providing a valuable opportunity to measure quantitatively, on a watershed scale, water quality improvements resulting from large-scale land use changes. The project was established in 1995 as a NPS monitoring program in conjunction with watershed habitat restoration and agricultural management changes implemented by the U.S. Fish and Wildlife Service (USFWS) at the Neal Smith National Wildlife Refuge and Prairie Learning Center (Refuge) in Jasper County Iowa (Figure 1). A large portion of the Walnut Creek watershed is being restored from row crop agriculture to native prairie and/or savanna (Drobney, 1994). Although it is not expected that large-scale prairie restoration will ever be used as an NPS management practice, the magnitude of the land use changes within the Walnut Creek watershed is large compared to other watershed projects. This project is forming a baseline against which to set expectations for other watershed improvement projects and helps establish the amount and location of non-agricultural land that might be placed in watersheds to reach a given water quality objective.

In 1996, the Walnut Creek Monitoring project



Figure 1. Location map of Walnut Creek and Squaw Creek watersheds.

was approved by the U.S. Environmental Protection Agency (EPA) as a Section 319 National Monitoring Program project. The project is supported, in part, by a Nonpoint Source Program (Section 319, Clean Water Act) grant from the EPA, Region VII. National Monitoring Program projects comprise a small subset of NPS pollution control projects funded under the Clean Water Act. The goal of the national program is to support 20-30 watershed projects nationwide that meet a minimum set of planning, implementation, monitor-

Basin Characteristics	Walnut	Squaw
	Creek	Creek
Total Drainage Area (sq mi)	20.142	18.305
Total Drainage Area (acres)	12,890	11,714
Slope Class:		
A (0-2%)	19.9	19.7
B (2-5%)	26.2	26.7
C (5-9%)	24.4	25.0
D (9-14%)	24.5	22.2
E (14-18%)	5.0	6.5
Basin Length (mi)	7.772	6.667
Basin Perimeter (mi)	23.342	19.947
Average Basin Slope (ft/mi)	10.963	10.981
Basin Relief (ft)	168	191
Relative Relief (ft/mi)	7.197	9.575
Main Channel Length (mi)	9.082	7.605
Total Stream Length (mi)	26.479	26.111
Main Channel Slope (ft/mi)	11.304	12.623
Main Channel Sinuosity Ratio	1.169	1.141
Stream Density (mi/sq mi)	1.315	1.426
Number of First Order Streams (FOS)	12	13
Drainage Frequency (FOS/sq mi)	0.596	0.710

Table 1. Basin characteristics of the Walnut and Squaw creek watersheds.

ing, and evaluation requirements designed to lead to successful documentation of project effectiveness with regard to water quality protection or improvement. Monitoring of both land treatment and water quality to document improvement is necessary to provide decision-makers with information on the effectiveness of NPS control efforts. Currently there are 22 projects, including Walnut Creek, in the national program.

The primary objectives of the Walnut Creek Monitoring project are to: 1) perform comprehensive, long-term NPS monitoring in the Walnut and Squaw Creek watersheds; 2) quantitatively document, over time, reduction in NPS pollution and associated environmental improvements resulting from watershed habitat restoration and land management changes; and 3) use the monitoring data to increase our understanding of what implementation measures are successful and expand public awareness of the need for NPS pollution prevention measures in the State of Iowa.

The purpose of this article is to summarize the project background, sample collection strategy and methods, and report on land use changes and reductions in chemical loading in the Walnut and Squaw creek watersheds for the period 1992 to 2000.

WATERSHED ATTRIBUTES

Walnut and Squaw creeks are warm-water streams located in Jasper County, Iowa (Figure 1). Walnut Creek drains 30.7 mi.² (19,500 acres) and discharges into the Des Moines River at the upper end of the Red Rock Reservoir. Only the upper part of the watershed (12, 890 acres) is included in the monitoring project because of possible backwater effects from the reservoir. The Squaw Creek basin, adjacent to Walnut Creek, drains 25.2 mi.² (16,130 acres) above its junction with the Skunk

Soil Characteristics	Walnut	Creek	Squav	v Creek
	Acres	Percent	Acres	Percent
Soil Parent Material:				
Alluvium	2043.87	15.86	2050.90	17.51
Eolian Sand			245.15	2.09
Weathered Shale	14.88	0.12		
Local Alluvium	192.79	1.50	383.34	3.27
Gray Paleosol	405.27	3.14	157.86	1.35
Loess	6155.89	47.75	6312.66	53.89
Loess and Local Alluvium	24.99	0.19	27.62	0.24
Loess-gray or gray mottles	2073.92	16.09	1245.56	10.63
Paleosol-reddish	13.27	0.10	7.96	0.07
Sandy Alluvium	168.52	1.31		
Till (pre-Illinoian)	1773.99	13.76	1255.80	10.72
Highly Erodible Land	6935.11	53.78	6226.13	53.57
Dominant Soil Taxa:				
Tama	2528.92	19.61	4018.23	34.29
Killduff	1889.72	14.66	1242.04	10.66
Muscatine	1038.25	8.05	548.54	4.68
Otley-Mahaska	1396.53	10.83	999.57	8.53
Shelby-Adair	508.47	3.94	986.67	8.42
Ackmore, Ackmore-Colo	1612.18	12.50	1309.69	11.17
Ladoga-Gara	1556.96	12.08	40.56	0.35

Table 2. Soil characteristics in the Walnut and Squaw creek watersheds.

River. The watershed included in the monitoring project is 18.3 mi.² (11,714 acres) and does not include the wide floodplain area near the intersection with the Skunk River. Basin characteristics of the Walnut Creek and Squaw Creek watersheds are very similar and make them well suited for a paired watershed design (Table 1).

The Walnut Creek and Squaw Creek watersheds are located in the Southern Iowa Drift Plain, an area characterized by steeply rolling hills and well-developed drainage (Prior, 1991). The soils and geology of the two watersheds are similar (Table 2). Soils within the Walnut and Squaw Creek watersheds fall primarily within four major soil associations: Tama-Killduff-Muscatine; Downs-Tama-Shelby; Otley-Mahaska; and Ladoga-Gara (Nestrud and Worster, 1979). Dominant soil taxa are indicated in Table 2; these soil taxa account for 82% of the soils found in the Walnut basin and 78% of the soils found in the Squaw basin. Tama and Muscatine soils are found primarily in upland divide areas, whereas Ackmore soils are associated with bottomlands. Killduff, Otley and Ladoga-Gara soils are found developed in slope areas. Most of the soils are silty clay loams, silt loams, or clay loams formed in loess and till. Moderate to high erosion potential characterizes many of the soils and both watersheds contain equal amounts of highly erodible land (Table 2).

Loess mantled pre-Illinoian till typifies much of the geology of the Walnut and Squaw creek watersheds. Both watersheds are mantled primarily by loess in upland areas. Outcrops of pre-Illinoian till and Late Sangamon paleosols are occasionally found in hillslope areas, whereas alluvium dominates the shallow subsurface of the main channels and second order tributaries. Pre-Illinoian till underlying most of the watersheds is 20 to 100 feet thick. Bedrock occurs at an approximate elevation of 850 to 700 feet above mean sea level and is primarily Pennsylvanian Cherokee Group shale, limestone, sandstone, and coal. In the drainageways of Walnut and Squaw creeks, Holocene alluvial deposits consist of stratified sands, silts, clays and occasional peat. In the Walnut Creek drainageway, post-settlement alluvial and colluvial materials deposited in the stream valley range from approximately two to six feet in thickness.

MONITORING PLAN DESIGN AND METHODS

The Walnut Creek Monitoring project utilizes a paired-watershed as well as upstream/downstream comparisons for analysis and tracking of trends. The Walnut Creek watershed is paired with the Squaw Creek watershed and a common basin divide is shared (Figure 1). Based on their similar basin characteristics (Tables 1 and 2), the watersheds are well suited to such a design. In addition, several subbasins are monitored in both watersheds to allow comparisons of differential implementation over time, and for analyzing their incremental contributions to the overall basin response. There are four basic components to the project: 1) tracking of land cover and land management changes within the basins, 2) stream gaging for discharge and suspended sediment at two locations on Walnut Creek and one on Squaw Creek, 3) surface water quality monitoring in the Walnut and Squaw creek watersheds, and 4) biomonitoring for aquatic macroinvertebrates and fish in Walnut and Squaw Creeks. A fifth project component, groundwater quality and hydrologic monitoring, was discontinued in water year 1999. Sampling stations located in the Walnut and Squaw Creek basins are shown on Figure 1.

Land Cover

Land cover in the Walnut and Squaw Creek basins has been tracked each year since 1994 using a Geographical Information System (GIS) at the Iowa Department of Natural Resources, Geological Survey Bureau (IDNR-GSB). Land cover data from both watersheds was compiled using a combination of plat maps, aerial photographs and field surveys. Data from 1994 and 1995 was derived primarily from plat maps and aerial photographs, whereas 1996 through 1998 data were compiled mainly from field surveys. However, annual field surveys have not proven especially effective for monitoring land use changes at a watershed scale due to inconsistencies in land use designations. For this report, a recently completed draft statewide inventory of land use for year 2000 was used to compile land use categories in the Walnut and Squaw Creek watersheds. Land cover data was interpreted from Landsat satellite imagery from 2000. USFWS personnel have tracked prairie planting areas and locations of cooperative farmer rental ground in the Walnut Creek watershed. Historical land use in the watersheds (pre-restoration) was compiled from 1:24,000 scale color infrared aerial photographs taken in 1992.

USGS Stream Gaging Stations

Standard U.S. Geological Survey (USGS) gaging facilities are present at three main stem sites (WNT1, WNT2, and SQW1; Figure 1). Stage is monitored continuously with bubble-gage sensors (fluid gages) and recorded by data collection platforms (DCP) and analog recorders (Rantz and others, 1982). The DCPs digitally record rainfall and stream stage at 15-minute intervals. The equipment is powered by 12 volt gel-cell batteries which are recharged by solar panels or battery chargers run by external power. Reference elevations for all USGS gage stations are established by standard surveys from USGS benchmarks. Stage recording instruments are referenced to outside staff plates placed in the streambeds, or to type-A wire-weights attached to the adjacent bridges. Rainfall is recorded using standard tipping bucket rain gages.

	-	-
Sampling Location	Parameters	Frequency
WNT1, WNT2, SQW2	Stage/Discharge, Suspended Sediment	Daily
WNT1, WNT2, WNT3, WNT5, WNT6, SQW1, SQW2, SQW3, SQW4, SQW5	Fecal Coliform, Anions, Phosphorus Temperature, Conductivity, Dissolved Oxygen, Turbidity, pH	April (2), May(4), June(4), July(2), August(2), September(2)
	Common herbicides	April, May (4), June (4), July, August, September
WNT1, WNT2, SQW1, SQW2	Fecal coliform, Anions, Phosphorus Temperature, Conductivity, Dissolved Oxygen, Turbidity, pH,	January, March, July, August, September, October, November
Biomonitoring stations	Biomonitoring	Annually (Aug)
Note: Number of samples colle	cted per month indicated under frequency column.	

Table 3. Summary of sampling locations, parameters and frequency.

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Stream discharge is computed from a rating developed for each site (Kennedy, 1983). The stream-gaging and calibration is performed by USGS personnel, using standard methods (Rantz and others, 1982; Kennedy, 1983). Current meters and portable flumes are used periodically to measure stream discharge and refine the station ratings.

Suspended Sediment

Suspended sediment samples are collected daily by local observers and weekly by water quality monitoring personnel. The observers collect depth integrated samples at one vertical section at one point in the stream using techniques described by Guy and Norman (1970). Samples are collected daily at all three stations. During storm events, suspended sediment samples are collected with an automatic water-quality sampler installed by the USGS at the gaging stations. Sampling is initiated

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by the DCP when the stream rises to a pre-set stage, and terminates when the stream falls below this stage. Suspended sediment concentrations are determined by the USGS Sediment Laboratory in Iowa City, Iowa, using standard filtration and evaporation methods (Guy, 1969). Discharge, rainfall, and sediment data are stored in the USGS Automatic Data Processing System (ADAPS) and published in the Iowa District Annual Water-Data Report.

Chemical Parameters

Table 3 shows the sampling sites, analytes, and frequency planned for each water year. Actual sample collection has occasionally varied from this schedule in response to field conditions and precipitation patterns. Temperature, pH, conductivity, dissolved oxygen, reduction-oxidation potential (redox), and turbidity are measured in the field; all other analyses are performed by the University of Iowa Hygienic Laboratory (UHL) using standard methods and an EPA-approved QA/QC plan (Thompson et al., 1995).

Biomonitoring

The purpose of the biomonitoring is to document changes in the aquatic vegetation, fish and macroinvertebrate populations in Walnut Creek as a result of the land use and management changes implemented in the watershed. Two biomonitoring sites are located in each watershed at downstream and midreach locations. In 1995 and 1996, two additional biomonitoring sites were funded through the USFWS Field Office. Locations are shown on Figure 1. Details regarding the biological monitoring component of the project are included in reports by Hubbard and Luzier (this issue).

LAND RESTORATION IMPLEMENTATION

Cropland Management Plan

A Cropland Management Plan was prepared by the USFWS in 1993 to guide the rapid conversion of traditional row crop areas to native, local ecotype habitat (USFWS, 1993). The goal has been to restore the land as rapidly as possible, although the rate of refuge development has varied with political, ecological and operational needs of the refuge. As refuge development takes place, various tracts of land currently in crops are removed from row crop production and converted to native habitat. The intent is to eliminate crop production on the refuge within approximately 10 years (USFWS, 1993).

Land currently owned by the Refuge but still farmed is rented to area cooperative farmers on a cash-rent basis. At the end of each crop year, a determination is made of which tracts to remove from row crop production. Farmers are notified of this decision and required to discontinue the farming practices on that particular tract. Refuge cropland is managed by conventional crop rotation of corn and beans. No-till production methods are mandatory whereas other management methods are more prescriptive, including soil conservation practices, nutrient management through soil testing, yield goals and nutrient credit records.

Herbicide and Fertilizer Management

It is the ongoing intent of the refuge to move towards a reduced chemical dependency for the cooperating farmers on refuge land. All chemicals and application rates are approved prior to application to minimize adverse impacts on non-target plants and animals. Use of chemicals not on the "pre-approved" list may be requested only after demonstrating that the intended use is consistent with an Integrated Pest Management Plan and crop scouting indicates a favorable cost/benefit ratio. All cooperative farmers are required to enter into a contract for crop scouting services for pest management. The following list of procedures for herbicide and fertilizer management are followed on Refuge-owned land (USFWS, 1993):

1. No fall application of fertilizer is allowed.

2. No anyhydrous ammonia has been allowed since 1993; only liquid fertilizer is permitted. Care in application is exercised to avoid runoff into wetlands or riparian areas.

3. A maximum of 100 pounds of liquid nitrogen per acre is allowed on conventional rotation corn ground.

4. Post emergent and banding application of fertilizer is required because this process increases the potential for immediate plant uptake and decrease leaching.

5. Post emergent herbicide is required and pre-emergent herbicide is not allowed. This decreases chances for leaching, encourages specific herbicide use for target species, and prevents broad-spectrum use.

6. As land use and vegetation type changes occur during restoration, the use of pesticides has decreased; however, there are some long term needs for certain pesticides to manage specific problem areas. These are addressed on a case-bycase basis as they become known.

Watershed and Subwatershed	Basin Size (acres)	Year	Row Crop	Grass	Woods	Water	Artificial	Other
Walnut Creek	12,891.0	1992	68.7	27.1	2.8	0.1	0.8	0.5
		2000	61.1	30.3	7.2	0.1	1.3	0.0
WNT1	4,312.5	1992	75.1	23.3	0.2	0.1	1.2	0.2
		2000	82.0	16.1	0.1	0.0	1.8	0.0
WNT3	731.3	1992	74.3	23.2	0.3	0.00	1.9	0.3
		2000	52.2	39.8	4.3	0.0	0.8	0.0
WNT5	1,964.6	1992	72.8	23.3	1.0	0.00	0.5	2.
		2000	57.7	37.2	4.3	0.0	0.8	0.0
WNT6	497.8	1992	73.4	16.4	8.2	1.3	0.1	0.6
		2000	75.5	15.3	8.5	0.0	1.3	0.0
Squaw Creek	11,622.0	1992	70.9	27.3	0.7	0.0	0.8	0.2
		2000	79.4	18.6	0.6	0.0	1.4	0.0
SQW1	2,876.0	1992	85.1	14.2	0.3	0.0	0.2	0.4
		2000	88.2	10.6	0.1	0.0	1.2	0.0
SQW3	1,859.3	1992	66.8	29.8	0.6	0.0	2.7	0.1
		2000	71.3	25.6	0.7	0.0	1.1	0.0
SQW4	292.1	1992	37.2	62.6	0.0	0.0	0.3	0.0
		2000	63.3	34.1	1.5	0.0	1.9	0.0
SQW5	585.7	1992	47.2	50.3	0.1	0.2	0.3	1.9
		2000	76.3	21.2	0.2	0.0	1.4	0.0

Table 4. Summary of land use in 1992 and 2000.

LAND USE

Prior to land restoration activities in 1992, land use in the Walnut Creek watershed consisted of approximately 69 % row crop and 27 % grass (Table 4; Figure 2). These values were similar to row crop and grass percentages measured in Squaw Creek (71% and 27%, respectively). Also in 1992, land use in Walnut Creek subbasins (WNT1, WNT3, WNT5 and WNT6) ranged between 73 and 75 % row crop. Row crop land use in Squaw Creek subbasins in 1992 ranged from 37 % in SQW 4 to 85 % in SQW1 (Table 4).

From 1992 to 2000, two major changes in land use have occurred in both Walnut and Squaw creek watersheds. An obvious change in Walnut Creek watershed was the conversion of 2,341 acres of land to native prairie (Table 5; Figure 2). This will be discussed more completely later in this section. A second major change in land use was apparently a result of the passage of the Freedom to Farm Act in 1996 which seemed to have substantially increased row crop production in both watersheds. For example, land use in Squaw Creek watershed increased from 71 to 79 % row crop from 1992 to 2000 (Table 4). In subbasins, land use changes were especially noticeable, as row crop acreage increased approximately 30 % in SQW 4 and SQW5. Row crop acreage increased three to five percent in SQW1 and SQW3 (Table 4). Changing land use percentages suggest that the majority of land converted to row crop were previously grasslands enrolled in the Conservation Reserve Program (CRP). Land use in Walnut Creek was not immune to this trend in areas less affected by refuge activities. In upper Walnut Creek watershed (WNT1) from 1992 to 2000, row crop acreage increased from 75 to 82 % and subbasin WNT6 increased from 73 to 75 %.

In Walnut Creek watershed, row crop land use decreased from 69 to 61 % between 1992 to 2000 as a result of prairie restoration by the USFWS at the Neal Smith refuge (Table 4). Table 5 summarizes the annual acreage and the percentage of land



Figure 2. Land use in the Walnut Creek and Squaw Creek watersheds in 1992 and 2000.

in the watershed and subwatersheds planted in prairie. From 1992 to 2000, an average of approximately 260 acres of prairie have been planted each year, with areas planted in 1994 and 1995 exceeding 400 acres (Table 5). As of 2000, approximately 2,341 acres in Walnut Creek watershed were planted in native prairie, representing 18.2% of the watershed (Figure 3). In the subbasins, restored prairie accounted for 7 to 51 % of the land area (Table 5). Hence, from 1992 to 2000, row crop in subbasins WNT3 and WNT5 decreased by 22 and 15 %, respectively. The amount of land owned by the refuge but farmed on a cash-rent basis totaled 579 acres in 2000, or 4.5 % of the watershed. The remaining land within the refuge boundary in the watershed consists of cool season grass or woods and comprises approximately 1,418 acres (11 %). As of 2000, the USFWS controlled approximately 33.7% (4,343 acres) of the Walnut Creek watershed above the WNT2 gaging station.

	Walnu	t Creek						
	Watershed			NT3	W	NT5	W	NT6
Year	Acres	Percent	Acres	Percent	Acres	Percent	Acres	Percent
Prairie Re	storation:							
1992	87.0	0.7			87.0	4.46		
1993	290.5	2.2			50.7	2.6		
1994	512.4	4.0	43.9	6.0				
1995	408.9	3.2			202.3	10.3		
1996	148.5	1.1			112.7	5.7	35.8	7.2
1997	281.8	2.2	76.1	10.4	3.7	0.2		
1998	362.6	2.8	212.8	29.1	33.6	1.7		
1999	185.2	1.4	38.9	5.3				
2000	63.8	0.5			21.6	1.1		
Total	2340.7	18.2	371.7	50.8	511.6	26.0	35.8	7.2
USFWS La	ands Farm	ed on Casl	1-Rent B	asis:				
2000	772.8	6.0			180.1	9.2	162.5	32.6

Table 5. Summary of annual prairie plantings and refuge lands farmed on a cash-rent basis.

NITROGEN LOADING REDUCTIONS

Land use changes have significantly reduced nitrogen loading in Walnut Creek watershed. With the conversion of row crop to native prairie and mandatory reductions in nitrogen applications on refuge-owned cropland, reductions in nitrogen loading in Walnut Creek watershed have invariably occurred. However, accurately quantifying nitrogen reductions is problematic. A major confounding factor is changing land use in the control watershed (Squaw Creek), where row crop land use has increased nearly 8.5% since 1992. Interpreting nitrogen reductions in the treatment watershed is difficult with a moving baseline condition indicated by the control watershed. Earlier estimations of nitrogen load reductions in Walnut Creek watershed were based on land use conditions in 1992 as the baseline condition (Schilling and Thompson, 1999). However, we know that land use conditions in 1992 are unlike conditions in 2000 since row crop land use has increased substantially post-1996 with the passage of the Freedom to Farm Act.

Nevertheless, nitrogen load reductions in Walnut Creek watershed may be estimated using some of the same assumptions used previously (i.e., Schilling and Thompson, 1999) and some hypothetical scenarios of land use conditions. First, a control condition in Squaw Creek watershed must be established. In 1992, 70.9% of the land in Squaw Creek watershed consisted of row crop. Schilling and Thompson (1999) showed that corn is the predominant row crop approximately 57% of the time, corresponding to a frequency of nearly two out of every three years in corn rotation. Typical nitrogen application in farmland around Prairie City was estimated to be 150 lbs/acre (Schilling and Thompson, 1999). Thus the amount of nitrogen applied in Squaw Creek watershed in 1992 may be estimated by the following equation:

 $(11,622 \text{ acres}) \times (70.9\% \text{ RC}) \times (57\% \text{ corn}) \times (150 \text{ lbsN/ac}) = 704,520 \text{ lbs N}$ (1)

Using the same equation for 2000 land use conditions and substituting 79.4% row crop for the 70.9% value suggests that nitrogen application in 2000 was 788,983 lbs, or an 11.9% increase over 1992 nitrogen application.

Estimating nitrogen-loading changes in Walnut



Figure 3. Summary of prairie planting areas and locations of rental farm lands.

Creek watershed involves a variety of "what-if" scenarios to constrain the possible load reductions. A Walnut Creek baseline condition for 1992 was estimated in the manner described above:

 $(12,891 \text{ acres}) \times (68.7\% \text{ RC}) \times (57\% \text{ corn}) \times (150 \text{ lbsN/ac}) = 757,198 \text{ lbs N}$ (2)

In a simple case, without considering annual land use changes at Neal Smith refuge or otherwise increasing row crop trends in the area, row crop acreage in Walnut Creek watershed decreased to 61.1% in 2000. Simply substituting 61.1% row crop for the 1992 value of 68.7% suggests that land use changes have decreased N application to 673,432 lbs, a 11.1% decrease from 1992 to 2000.

However, this scenario is rather simplistic and does not consider several important factors. From 1992 to 2000, a total of 2,341 acres of land were restored to native prairie in Walnut Creek watershed. Assuming that the number of new prairie planting acres is equal to the amount of row crop acres taken out of production (a simplifying assumption that was more true with early plantings than later plantings), prairie restoration from 1992 to 2000 reduced N applications as follows:

 $(2,341 \text{ acres}) \times (57\% \text{ corn}) \times (150 \text{ lbs N}) = 200,155 \text{ lbs N}$ (3)

In addition, 597 acres of row crop land was owned by the refuge in 2000 but rented to area farmers on a cash-rent basis. In these areas, N applications were reduced from 150 lbs/acre to 100 lbs/acre. Assuming a 50% corn rotation in these areas mandated by the refuge, rental farmlands in 2000 reduced N applications by 14,475 lbs N (579 ac x 50% corn x 50 lbs N). Thus, combined actions taken by the Neal Smith refuge from 1992 to 2000 reduced N application in the Walnut Creek watershed by an estimated 214,630 lbs. Considering this estimate to represent reduced N application in the watershed due to the refuge, nitrogen loads were reduced from the 1992 baseline condition from 757,198 lbs N to 542,568 lbs N, a hypothetical reduction of 28.3%.

A confounding factor in the analysis of nitrogen reductions is the otherwise increasing trend of row crop land use on non-refuge lands in Squaw and Walnut Creek watersheds. Comparing nitrogen reductions in 2000 to "baseline" land use conditions in 1992 does not seem adequate, in light of increasing row crop trends in the area. Walnut Creek baseline conditions may be adjusted to reflect changing land use practices in the area by considering what nitrogen loading would be in the watershed if the Neal Smith refuge was not present. Provided that row crop land use in the Walnut Creek watershed increased by the same percentage as Squaw Creek from 1992 to 2000 (8.5%), nitrogen application loads in Walnut Creek watershed for 2000 would have been approximately 850,883 lbs N (assuming the row crop percentage is equal to 77.2% in equation 2). Comparing this N load to the current condition of 673,432 lbs N (61.1% row crop in 2000) suggests a hypothetical reduction of 26.5% rather than 12.4%. From this adjusted "baseline" condition, reduced N applications of 214,630 lbs by the Neal Smith refuge would account for a 25.2% reduction.

A final baseline scenario to consider would be the situation in which the Neal Smith refuge was not present and all lands currently planted in prairie were now in row crop as a result of the Freedom to Farm Act (not an entirely implausible scenario). In this case, an additional 2,341 acres would have been planted to row crop in the Walnut Creek watershed in 2000, increasing the percentage of row crop land in the watershed to 83.7%. Substituting this percentage in equation 2 suggests that 922,525 lbs N would have been applied in the watershed in 2000. Thus, comparing this N total to the current N total (673,432 lbs N) suggests that nitrogen loading may have been reduced by as much as 37% in the watershed as a result of land use changes. From this adjusted "baseline" condition, reduced N applications of 214,630 lbs by the Neal Smith refuge would account for a 23.5% reduction.

It is interesting to note that as the baseline condition for nitrogen applications increased with each scenario, the effect of reduced N applications on refuge-owned lands decreased from an estimated 28.3% to 23.5%. This was a function of a constant value of nitrogen reduction applied to an everincreasing amount of hypothetical nitrogen loads. This situation points to the possible range in error associated with estimating nitrogen reductions with transient land use conditions. Clearly, changing land use conditions in the Walnut and Squaw creek watershed make estimating nitrogen reductions difficult especially when the "control" situation changes nearly as much as the "treatment" case. Regardless of the true amount of nitrogen reduction provided by the refuge, reduced nitrogen loading appears to be quite substantial.

PESTICIDE REDUCTIONS

Pesticide use was substantially reduced in the Walnut Creek watershed following the purchase of land and adoption of the Cropland Management Plan. As Schilling and Thompson (1999) reported, pre-emergent pesticides were not applied on refugeowned land, including common Iowa herbicides, atrazine, cyanazine, metolachlor, alachlor, metribuzin and acetochlor. Schilling and Thompson (1999) reported that the adoption of the Cropland Management Plan on refuge-owned land in 1993 reduced pre-emergent pesticide application in Walnut Creek watershed by 28%. Since this estimate was derived from the proportion of row crop land that came under control of USFWS and their Cropland Management Plan in 1993 (which has not changed substantially), the estimated reduction in pesticide application in Walnut Creek watershed remains valid.

CONCLUSIONS

The Walnut Creek Monitoring Project began with the objective to establish a water quality monitoring program to document water quality improvements resulting from large-scale watershed restoration and management. From 1994 to 2000, the surface water monitoring program was performed in accordance with project workplans and reports and continues to meet the objectives established for the project.

With the aid of satellite imagery, and tracking of annual prairie plantings and land management conditions by the USFWS, land use changes in Walnut and Squaw Creek watersheds between 1992 and 2000 have been monitored. While row crop land use in Squaw Creek watershed has increased 8.5% during this period, row crop in Walnut Creek watershed decreased from 69% to 61%. Prairie plantings in 2000 comprised 2,341 acres and accounted for 18.2% of the watershed area.

Nitrogen applications in Walnut Creek watershed decreased substantially between 1992 and 2000 although actual reductions are difficult to quantify. In Squaw Creek watershed, increases in row crop land use suggest nitrogen loading has increased by 12%. In Walnut Creek watershed, row crop acreage has decreased due to the Neal Smith NWR. Estimates of nitrogen application reduction in Walnut Creek watershed ranged from 11 to 37%. Pesticide reductions are estimated to be 28% in Walnut Creek watershed.

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NO3-N, CL AND SO4 CONCENTRATIONS, LOADS AND TRENDS IN SURFACE WATER IN WALNUT AND SQUAW CREEK WATERSHEDS

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INTRODUCTION

Watershed restoration and agricultural management changes implemented by the U.S. Fish and Wildlife Service (USFWS) at the Neal Smith National Wildlife Refuge have resulted in largescale land use changes in the Walnut Creek watershed located in Jasper County, Iowa (Figure 1). Large portions of the Walnut Creek watershed are being restored from row crop to native prairie in an effort to rebuild a portion of the tallgrass prairie and savanna ecosystem that is large enough for long term viability (Drobney 1994). The Walnut Creek Watershed Monitoring Project was established in 1995 as a nonpoint source monitoring project related to the watershed restoration activities (Schilling and Thompson, 1999). Water quality conditions have been monitored to evaluate changes in chemical transport resulting from conversion of large tracts of land from row crop to native prairie in a treatment watershed (Walnut Creek) compared to a highly agricultural control watershed (Squaw Creek). Descriptions of the project and land use changes implemented in the Walnut and Squaw Creek watersheds are discussed elsewhere in this report.

Nitrate-nitrogen (nitrate), chloride and sulfate are common anions detected in surface and groundwater and these constituents have been monitored in Walnut and Squaw creek surface water since project startup. These constituents are of interest for the Walnut Creek project because of their sources and manner in which they are delivered to streams. Nitrate is a common agricultural pollutant that is primarily delivered to streams through baseflow groundwater discharge and tile drainage (Hallberg, 1987; Schilling and Wolter, 2001). Chloride and sulfate are naturally occurring constituents in groundwater that discharge into surface water. Chloride inputs in surface water have both natural and agricultural land use sources, with agricultural inputs associated with KCl fertilizer application in the watersheds. Sulfate probably provides the best marker for tracking groundwater inputs to surface water independent of land use. Schilling and Thompson (2000) noted that sulfate concentrations in surface water, unlike nitrate and chloride did not relate significantly to the percentage of row crop land use in Walnut and Squaw creek subwatersheds.

The purpose of this report is to present results of nitrate, chloride and sulfate monitoring in the Walnut and Squaw Creek watershed for water years 1995 to 2000 and evaluate whether changes in anion concentrations and chemical loads have occurred in the watersheds.

METHODS

Sample Collection and Analysis

Anion concentrations are monitored weekly to monthly at ten sites in the Walnut and Squaw creek watersheds (Figure 1). Upstream and downstream sites on the main stems and three tributary basins are monitored in each watershed. Sample collection is stratified by season, with greater sampling frequency during spring and early summer. Weekly monitoring is targeted for May and June when nitrogen transport is greatest following postapplication, whereas bimonthly sampling occurs in March, April, July, August and September. During late fall and winter, stream samples are collected on a monthly basis at upstream-downstream locations at main stem sites only. Laboratory analyses were performed by The University of Iowa Hygienic Laboratory (UHL) using standard methods.



Figure 1. Location map of Walnut Creek and Squaw Creek watersheds.

Statistical Methods

Statistical analyses were performed according to the guidelines of Grabow et al. (1998, 1999). To test for the gradual change in chemical concentrations over time a multiple linear regression analysis was performed. The equation is:

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 \tag{1}$$

where, Y is either the water quality variable or log of the variable for the treatment watershed (Walnut Creek), X is the same water quality variable (or log) for the control watershed (Squaw Creek), and X₂ is elapsed time (in weeks), and β_0 , β_1 , and β_2 are regression parameters. The estimate of β_2 indicates the magnitude of change over time in units per week. By having a control watershed (variable X₁), the analysis blocks out much of the hydrologic variability and the change should be isolated to treatment effects, which in this case is being modeled as time (X₂).

In some cases, seasonality was present even when data were paired with a control watershed. In this case a class variable denoting a season can be brought into the analysis. The resulting equation is:

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_{3i}$$
⁽²⁾

where β_{3i} is the parameter estimate for season i. If the year was broken down into months, there would be 12 values for β_3 if broken down into growing season and non-growing season, there would be 2 values for β_3 .

Chemical Loads

The USGS program ESTIMATOR was used to estimate loads of anions at the three stream gaging sites (Figure 1). The ESTIMATOR program utilizes a Minimum Variance Unbiased Estimator (MVUE) to implement a seven-parameter regression model based on the relationship between log-flow and log-concentration (Cohn et al, 1989; 1992; Gilroy et al., 1990). Daily chemical load data were tabulated and summarized by month and water year. Load data were normalized on a unit area basis by dividing the total annual load at each gaging site by the watershed area above the gage. In the case of Walnut Creek watershed, the load per unit area between the two gage sites was determined by subtracting the load estimated at WNT1 from WNT2.

RESULTS AND DISCUSSION

Concentrations

Nitrate concentrations have ranged between 0.8 to 13 mg/l at the Walnut Creek outlet (WNT2) and 2.1 to 15 mg/l at the downstream Squaw Creek outlet (SQW2) (Table 1). Mean nitrate concentrations were 1 mg/l higher in Squaw Creek (SQW2) than Walnut Creek (WNT2) and highest at the upstream monitoring sites in both watersheds, averaging 12.4 mg/l at SQW1 and 11.1 mg/l at WNT1. Maximum nitrate concentrations did not exceed 20 mg/l in either watershed (Figure 2). Widest range and lowest mean nitrate concentrations were measured in Squaw Creek subbasin SQW4 (Figure 2; Table 1). Both Walnut and Squaw Creek watersheds have shown a similar temporal pattern of detection, with higher concentrations observed in the spring and early summer months coinciding with periods of application, greater precipitation and higher stream flow (Figures 3 and 4).

Mean chloride and sulfate concentrations were approximately 3 mg/l higher at the Squaw Creek outlet (SQW2) than the Walnut Creek outlet (WNT2)(Table 1). Chloride concentrations ranged from 6.3 to 29 mg/l in Walnut Creek watershed and from 7.4 to 32 mg/l in Squaw Creek watershed, although data are mostly clustered between 10 and 20 mg/l for all sites (Figure 5). At Walnut Creek mean chloride showed a downstream decrease in concentration from WNT1 to WNT2, but mean chloride concentrations increased from upstream to downstream sites in Squaw Creek (Table 1). Lowest mean chloride concentrations were measured in WNT5 subbasin (10.1 mg/l) and SQW4 subbasin (10.8 mg/l). Chloride concentrations appeared to decrease in water years 1999 and 2000 relative to previous years (Figure 6).

Mean sulfate concentrations were lower in the Walnut Creek watershed (means ranging from 14.0 to 23.6 mg/l) compared to the Squaw Creek watershed (means ranging from 20.9 to 25.6 mg/l) (Table 1). With exception to samples collected from WNT5 and WNT6, sulfate concentrations typically ranged between 20 and 30 mg/l (Figure 7). Highest mean sulfate concentrations were observed at SQW2 (26.7 mg/l) and lowest values were consistently measured at WNT6 (14.0 mg/l) (Table 1). While no difference in sulfate concentrations exists between upstream monitoring sites in both watersheds (WNT1 and SQW1), subbasins in Squaw Creek watershed consistently showed higher sulfate concentrations than Walnut Creek subbasins (Table 1). Observed decreases in sulfate concentrations have occurred from water years 1997 through 2000 (Figure 8).

Temporal Changes

Box plots of annual nitrate concentrations at WNT2 and SQW2 indicated lower medians at WNT2 than SQW2, particularly in WY 1996, WY 1998 and WY 2000 (Figure 9). A t-test found a significant difference between the nitrate concentration means of the Walnut and Squaw creek data sets from 1995 to 2000 (n = 97, p < 0.05) with the overall mean nitrate concentration in Walnut Creek substantially lower than Squaw Creek (8.19 mg/l

						Quartile	
	n	range	mean	sd	25 th	50 th	75 th
			Nitrate-Nitr	ogen (mg/l)		
WNT1	99	2.3-17	11.1	3.3	9.1	11	14
WNT2	99	0.8-13	8.2	3.1	5.9	8.8	11
WNT3	66	2.9-15	9.7	3.3	7.0	9.7	13
WNT5	65	0.6-15	10.0	3.0	8.8	10	12
WNT6	65	0.5-13	7.0	3.2	5.1	7.6	9.6
SQW1	90	5.2-17	12.4	2.5	10	13	14
SQW2	97	2.1-15	9.2	3.0	7.2	9.5	11
SQW3	64	4.7-15	10.7	2.4	9.4	11	13
SQW4	65	0.6-11	3.7	2.2	2.3	3.4	4.5
SQW5	65	1.0-12	7.4	2.7	5.2	8.2	9.7
			Chlorid	· (//)			
WANT 1	02	11.20	Life 2	e (mg/l)	12	1 /	17
WN11 WNIT2	95	11-29	15.5	3.0	13	14	17
WN12	93 50	7.8-24	12.2	2.6	10	12	13
WN13	59	7.8-24	12.4	2.4	11	12	14
WN15	58 50	6.3-16	10.1	1.6	9.2	9.8	11
WN16	58	7.9-19	11.8	2.8	9.4	11	14
SOW1	86	8 9-19	14 7	8.0	12	14	15
SOW2	93	9 1-30	15.2	2.7	14	15	16
SOW3	58	11-25	16.8	2.7	15	17	18
SOW4	60	7 4-19	10.8	2.4	9.0	10	12
SOW5	60	11-32	16.5	3.9	14	16	18
~~~~~							
			Sulfate	(mg/l)			
WNT1	93	11-33	20.5	4.1	18	20	23
WNT2	93	15-50	23.6	6.2	19	22	26
WNT3	59	14-30	20.0	3.1	18	20	21
WNT5	58	15-31	18.6	3.4	16	18	20
WNT6	58	4.9-30	14.0	4.9	11	13	15
SOW1	86	15 21	20.0	35	10	20	22
SQW1	00	15-51	20.9	5.5 5.7	10	20	25 20
SQW2	93 50	13-44	20.7	J./ 1 1	22	∠0 25	<u>∠</u> 9 27
SQW3	50 60	17-41	25.0	4.4	23 10	∠ <i>3</i> 25	∠ / 2 2
SQW4	00	12-02 19-41	25.0	9.3 5 7	19	23 25	32 20
SUWS	60	18-41	23.6	5.7	21	25	30

 Table 1. Summary of nitrate, chloride and sulfate concentrations in surface water for water years 1995 to 2000.



**Figure 2.** Box plot of nitrate-N concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000. Box plots illustrate the 25th, 50th and 75th percentiles; the whiskers indicate the 10th and 90th percentiles; and the circles represent data outliers.

and 9.20 mg/l, respectively). Regression analysis was performed to determine if a change had occurred over time in the relationship of nitrate concentrations in the treatment watershed (Walnut) and the control watershed (Squaw) (Table 2). The parameter  $\beta_2$  is the parameter for elapsed time and therefore indicates the magnitude of change. A negative value indicates a decrease and a positive value an increase over time. Nitrate has decreased by 0.0028 mg/l/week over 326 weeks, equivalent to 0.912 mg/l over the entire sampling period. Using a mean value of the control watershed (9.20 mg/l) for X₁ in Equation (1), nitrate has decreased from 9.19 mg/l to 8.28 mg/l in the treatment watershed over the entire sampling period during the growing season (May-August), and from 8.06 mg/l to 7.15 mg/l during the non-growing season.

When analyzed as an upstream/downstream design, Equations (1) and (2) are still used. In this case Y is the downstream station value and  $X_{\perp}$  is the upstream value. This analysis on the Walnut Creek watershed for nitrate indicated that the downstream concentration was decreasing over time while considering the upstream value, however it was only significant at the 80% confidence

interval(p=0.194).

Statistically significant changes in concentrations of chloride and sulfate have also occurred between WY 1995 and WY 2000 (Table 2). Ttests indicated that the means of Walnut and Squaw creek data sets for chloride and sulfate were significantly different (p<0.0001) and means of upstream and downstream samples in Walnut Creek for chloride and sulfate were also significantly different (p<0.0001). Regression analysis indicated that chloride concentrations in Walnut Creek have decreased by 0.0086 mg/l/week over a 285 week sampling period, equivalent to a reduction of 2.45 mg/l (Table 2). Analyzed as an upstream/ downstream design, downstream chloride concentrations were decreasing by 0.0094 mg/l/week over 285 weeks (2.70 mg/l) while adjusting for the upstream value. Sulfate concentrations at WNT2 have decreased by 5.67 mg/l and 7.44 mg/l over the 285-week sampling period, while adjusting for the controls of Squaw Creek and WNT1, respectively (Table 2).

#### **Chemical Loads**

Total export loads of NO3-N, Cl and SO4 from both watersheds were similar, averaging 125 to 134 Mg/yr of NO₃-N, 166 to 183 Mg/yr Cl and 310 to 314 Mg/yr SO4 (Table 3). Annual losses of NO3-N from the watersheds were greatest in 1998 ranging from 228 to 265 Mg/yr. Export loads for most constituents were generally lower in Walnut Creek compared to Squaw Creek despite greater precipitation and higher stream flows in Walnut Creek. Chemical loads strongly followed a pattern dictated by the amount of discharge in any given month or year. Maximum monthly export loads of NO3-N and Cl exceeded 20 Mg in most years, with load peaks typically occurring in February and May/ June of any given year (Figure 10). Peak loads for all constituents occurred in February and May 1996, May 1997 and June 1998 (Figure 10).

On a unit area basis, major differences in chemical loss rates emerge between Walnut and Squaw Creek watersheds, particularly with respect to the lower portion of Walnut Creek (Table 4). Nitrate and Cl losses from Walnut Creek



Figure 3. Nitrate-N concentrations at upstream and downstream sampling sites in Walnut and Squaw creeks for water years 1995 to 2000.



Figure 4. Nitrate-N concentrations versus discharge at WNT2 and SQW2 sites for water years 1996 to 2000.

watershed are lower than Squaw Creek, most of which can be traced to substantially lower losses emanating from lower Walnut Creek. Losses of NO₃-N and Cl from lower Walnut Creek containing the prairie restoration sites are approximately one-half the mass lost from upper Walnut Creek and Squaw Creek watershed areas dominated by row crop. Nitrate and Cl losses from upper Walnut Creek were similar or slightly higher than Squaw Creek (Table 4). Few consistent differences are noted in SO₄ loads among watershed areas (Table 4). The lack of consistent trends in sulfate losses among watershed areas suggests that differences in NO₃-N and Cl are related mainly to land use changes in Walnut Creek watershed.

While NO₃-N losses are clearly less in the lower Walnut Creek watershed compared to Squaw Creek and upper Walnut Creek, NO₃-N losses remain considerably higher than those reported for pristine tallgrass prairie watersheds (Dodds et al., 1996). Observations of surface water flowing from four watersheds on Konza Prairie Research Natural Area in Kansas indicated annual NO₃-N losses <0.2 kg/ha. Previous research at Walnut



**Figure 5.** Box plot of chloride concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000.

Creek watershed, consisting of a single sampling event completed during a baseflow period in May 1999 showed that some small (<100 ha) interior watersheds containing nearly 100% restored prairie had NO₃-N and Cl in surface water < 1 mg/l and <3 mg/l, respectively (Schilling and Wolter, 2001). Extrapolating from a single sampling event, annual NO₃-N losses for restored prairie watersheds in Walnut Creek watershed were <1-2 kg/ha (Schilling and Wolter; unpublished data).

Average annual losses of NO₃-N from Squaw Creek and upper Walnut Creek (28-34 kg/ha) are similar to those reported for a different Walnut Creek in Story County, Iowa (Jaynes et al., 1999). Although wide variability in annual losses was observed, average NO3-N loss from Walnut Creek (Story) was 29 kg/ha for the period 1992 to 1995 (Jaynes et al., 1999). Comparable NO₃-N losses are noteworthy considering that Walnut Creek in Story County is located in a heavily agricultural and tile-drained watershed developed on poorly-drained, recent glacial materials (Wisconsinan Des Moines Lobe). In contrast, Walnut and Squaw Creek watersheds in Jasper County are located on older glacial sediments with well-developed drainage. However, land use in Squaw Creek and Walnut Creek (Story) is similar (73% and 83% row crop,

respectively; Walnut (Story) data from Hatfield et al., 1999) and NO₃-N losses to Iowa streams have been linked to row crop agriculture (Schilling and Libra, 2000)

Flow-weighted concentrations followed a similar pattern exhibited by chemical mass losses (Table 5). Flow-weighted concentrations of NO₃-N and Cl were higher in Squaw Creek and upstream Walnut Creek than lower Walnut Creek. Average flow-weighted concentrations of NO₃-N were >10 mg/l in Squaw Creek and upper Walnut Creek but were 6.6 mg/l in lower Walnut Creek (Table 5). Similarly, concentrations of Cl were 14.4 mg/l in Squaw Creek and 15.1 mg/l in upper Walnut Creek but 9.2 mg/l in lower Walnut Creek. Sulfate concentrations were slightly higher at SQW2 compared to WNT2, and concentrations were higher in lower Walnut Creek compared to upper Walnut Creek (Table 5). Discharge of groundwater from Pennsylvanian bedrock in lower reaches of Walnut Creek is believed to contribute to SO₄ differences within the Walnut Creek watershed (Schilling and Wolter, 2001). In general, average flow-weighted concentrations compared favorably to the mean of all analyses measured during the same five-year period (WY 1996-2000) (Table 5).

Paired T-tests were performed to evaluate statistical differences in discharge and chemical loads among watershed areas (Table 6). Total monthly NO₃ and Cl loads were significantly lower in lower Walnut Creek compared to upper Walnut Creek and Squaw Creek (P<0.1) (Table 6). Discharge and SO₄ did not exhibit statistically significant differences among WNT2, SQW2, WNT1 and WNT2-1 data sets.

#### SUMMARY AND CONCLUSIONS

Results from surface water monitoring in Walnut and Squaw Creek watersheds for nitrate, chloride and sulfate indicate that changes in anion concentrations have occurred during monitoring for water years 1995 to 2000. Mean nitrate-N concentrations were 1 mg/l higher in Squaw Creek (9.2 mg/l) than Walnut Creek (8.2 mg/l) and highest at the upstream monitoring sites in both watersheds. Mean chloride and sulfate concentrations



**Figure 6.** Chloride concentrations at upstream and downstream sampling sites in Walnut and Squaw creeks for water years 1995 to 2000.

were approximately 3 mg/l higher at the Squaw Creek outlet than the Walnut Creek outlet.

Multiple linear regression analysis suggested that concentrations of nitrate, chloride and sulfate have gradually decreased in Walnut Creek while adjusting for the control watershed (Squaw Creek). Nitrate has decreased from 9.19 mg/l to 8.28 mg/l in the treatment watershed over the entire sampling period during the growing season (May-August), and from 8.06 mg/l to 7.15 mg/l during the nongrowing season. Chloride and sulfate have decreased in the treatment watershed approximately 2-3 mg/l and 5-7 mg/l, respectively, while adjusting for the control watersheds of Squaw Creek and upstream Walnut Creek (WNT1).

Land use differences among the watershed areas evaluated in this study are believed respon-

sible for major differences in chemical loading rates of NO₃-N and Cl. The lower portion of Walnut Creek watershed subject to large-scale prairie restoration exported on average 18.8 kg/ha of NO₃-N and 26.1 kg/ha of Cl. Upper Walnut Creek watershed and Squaw Creek watershed, consisting of 80% row crop, averaged 28-34 kg/ha NO₃-N and 39-43 kg/ha Cl. Average flow-weighted concentrations of NO₃-N exceeded 10 mg/l in upper Walnut Creek and Squaw Creek, but averaged 6.6 mg/l in lower Walnut Creek containing the restored prairie. Statistical analyses of total monthly export of NO₃-N, Cl and SO₄ indicated that lower Walnut Creek exported significantly less NO₃-N and Cl than upper Walnut Creek and Squaw Creek.

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	$\beta_0$	р	$\beta_1$	р	$\beta_2$	р	β ₃	р	Comments
Nitrate ¹	.0545	0.405	.8697	.0001	-0.0028	.034	1.132	.0001	$\beta_3$ , growing season
Nitrate ²	.626	.233	.7673	.0001	-0.00198	.194	1.157	.0003	$\beta_3$ , growing season
Chloride ¹	5.165	0.001	0.5469	0.000	-0.0086	0.002			Equation 1
Chloride ²	7.256	0.000	0.4197	0.000	-0.0094	0.001			Equation 1
Sulfate ¹	9.708	0.003	0.6339	0.000	-0.0199	0.004			Equation 1
Sulfate ²	13.20	0.001	0.6945	0.000	-0.0261	0.001			Equation 1

 Table 2.
 Summary of regression parameter estimates.

¹ paired watershed design ² upstream/downstream design (Walnut)

Table 3. Summary of precipitation,	flow and chemical loads measured at	Walnut and Squaw creek watershed outlets.

			Total	NO3-N	Chloride	Sulfate
	Water	Precip	Q	Load	Load	Load
Watershed	Year	(mm)	$(m^3x10^6)$	(	Mg	)
Walnut	1996	833.9	15.7	112.2	184.3	373.9
Creek	1997	645.2	11.0	93.9	135.6	261.8
	1998	1055.9	24.5	227.5	275.4	493.7
	1999	811.3	15.3	136.9	165.0	298.0
	2000	580.9	6.8	54.4	67.6	123.8
	Average	785.4	14.7	125.0	165.6	310.2
Squaw	1996	596.6	14.5	138.5	207.2	394.7
Creek	1997	677.9	7.8	76.5	125.7	221.9
	1998	873.8	22.7	265.0	336.9	547.4
	1999	800.6	11.7	128.1	167.6	275.3
	2000	380.0	6.2	61.2	75.5	131.6
	Average	665.8	12.6	133.8	182.6	314.2



**Figure 7** Box plot of sulfate concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000.

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Figure 8. Sulfate concentrations at upstream and downstream sampling sites in Walnut and Squaw creeks for water years 1995 to 2000.



**Figure 9.** Box plot of annual nitrate-N concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000.



**Figure 10.** Monthly mean discharge and total chemical loads of nitrate, chloride and sulfate measured at WNT2 (solid line) and SQW2 (dashed line).

<b>Table 4.</b> Discharge and loss of nitrate, chloride and sulfate from various W alnut and Squaw creek waterships	ed areas.
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		Total	NO3-N	Chloride	Sulfate
	Water	Q	Load	Load	Load
Watershed	Year	(meters)	(kg/ha per year)		
WNT2	1996	0.30	21.51	35.31	71.64
	1997	0.21	17.99	25.99	50.17
	1998	0.47	43.58	52.76	94.60
	1999	0.29	26.23	31.62	57.10
	2000	0.13	10.43	12.96	23.73
	Average	0.28	23.95	31.73	59.45
SQW2	1996	0.31	29.44	44.05	83.92
	1997	0.17	16.25	26.73	47.17
	1998	0.48	56.33	71.62	116.39
	1999	0.25	27.22	35.64	58.53
	2000	0.13	13.01	16.06	27.97
	Average	0.27	28.45	38.82	66.80
WNT1	1996	0.29	32.53	46.26	65.60
	1997	0.18	22.42	29.47	40.13
	1998	0.47	62.15	74.03	91.86
	1999	0.30	37.05	44.79	54.80
	2000	0.15	16.59	19.75	24.72
	Average	0.28	34.15	42.86	55.42
WNT2-	1996	0.30	15.97	29.81	74.68
WNT1	1997	0.22	15.76	24.24	55.21
	1998	0.47	34.26	42.08	95.98
	1999	0.29	20.80	25.00	58.26
	2000	0.12	7.34	9.54	23.23
	Average	0.28	18.82	26.13	61.47

		NO3-N	Chloride	Sulfate
Watershed	Water Year	(concentrations in mg/L)		
WNT2	1996	7.2	11.8	23.9
	1997	8.6	12.4	23.9
	1998	9.3	11.2	20.1
	1999	8.9	10.8	19.5
	2000	8.0	9.9	18.1
	Average	8.4	11.2	21.1
	5-yr monitoring	8.2	12.2	23.6
SQW2	1996	9.5	14.3	27.2
	1997	9.8	16.1	28.4
	1998	11.7	14.8	24.1
	1999	11.0	14.3	23.6
	2000	9.9	12.3	21.4
	Average	10.4	14.4	24.9
	5-yr monitoring	9.2	15.2	26.6
WNT1	1996	11.1	15.8	22.4
	1997	12.3	16.2	22.1
	1998	13.1	15.7	19.4
	1999	12.2	14.7	18.0
	2000	11.1	13.2	16.5
	Average	12.0	15.1	19.7
	5-yr monitoring	11.1	15.3	20.5
WNT2-WNT1	1996	5.3	9.8	24.6
	1997	7.0	10.8	24.6
	1998	7.3	9.0	20.5
	1999	7.2	8.7	20.2
	2000	6.0	7.8	19.1
	Average	6.6	9.2	21.8

**Table 5.** Summary of flow-weighted concentrations measured at various watershed areas, water years 1996 to 2000. The five-year average of concentration data collected at stream gaging sites is provided for comparison.
		WNT2	SQW2	WNT1
Total Q	SQW2	0.829		
	WNT1	0.991	0.833	
	WNT2-1	0.996	0.829	0.988
Total NO ₃	SQW2	0.491		
	WNT1	0.148	0.117	
	WNT2-1	0.334	0.000	0.022
Total Cl	SQW2	0.355		
	WNT1	0.170	0.645	
	WNT2-1	0.378	0.081	0.031
Total SO ₄	SQW2	0.587		
	WNT1	0.740	0.390	
	WNT2-1	0.874	0.699	0.626

Table 6. Summary of t-test results comparing discharge and chemical losses from various watershed areas.

# HERBICIDE CONCENTRATIONS, LOADS AND TRENDS IN SURFACE WATER IN WALNUT AND SQUAW CREEK WATERSHEDS

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### **INTRODUCTION**

Herbicides are common surface water contaminants in the agricultural Midwest (Thurman et al., 1991, 1992). Herbicide losses in runoff and subsurface drainage tiles contribute the majority of herbicides detected in streams. Herbicide concentrations in surface water are usually greatest after spring application, with peak levels often occurring in the first runoff event following application (Thurman et al., 1991). In many watersheds, concentrations of atrazine [6-chloro-N-ethyl-N'-(1-methylethyl)-1,3,5-triazine-2,4-diamine] in surface water may exceed the U.S. Environmental Protection Agency (USEPA) maximum contaminant level (MCL) of 3.0 µg/L during post-application periods. Recent estimates of atrazine losses in an intensely farmed watershed in Story County, Iowa ranged from 0.2 to 7.5 g/ha which represented 0.18 to 5.6% of the mass of the herbicide applied in the watershed (Jaynes et al., 1999).

In the Walnut and Squaw Creek watersheds in Jasper County Iowa, herbicide concentrations have been monitored in surface water since 1994 as part of the Walnut Creek Watershed Monitoring Project (Schilling and Thompson, 1999; 2000; this issue). Herbicide applications were significantly reduced in the Walnut Creek watershed following acquisition of land by the U.S. Fish and Wildlife Service (USFWS) for the Neal Smith National Wildlife Refuge in Jasper County Iowa (Figure 1). The USFWS currently owns 33.7% of the Walnut Creek watershed, including 18.2% of the watershed converted from row crop to native prairie and another 4.5% of the watershed owned by the refuge but rented to area farmers (see this issue).

In 1993, the USFWS adopted a Cropland Management Plan for the refuge which banned the use of pre-emergent herbicides on refuge-owned lands, including atrazine, cyanazine [2-(4-chloro-6-

(ethylamino)-1,3,5-triazin-2-yl)-2-methylpropionitrile], metolachlor [2-chloro-N-(2-ethyl-6methylphenol)-N-(2-methoxy-1-methylethyl)acetamide], alachlor [2-chloro-2'-6'-diethyl-N-(methoxymethyl)-acetanilide], metribuzin [4amino-6-(1,1-dimethylethyl)-3-(methylthio)-1,2,4triazin-5(4H)-one] and acetochlor [2-chloro-N-(ethoxymethyl)-N-(2ethyl-6-methylphenyl)-acetamide]. Schilling and Thompson (1999) estimated that herbicide applications were reduced by 28% in the Walnut Creek watershed following adoption of the Cropland Management Plan on refuge-owned lands.

The purpose of this report is to present results of herbicide monitoring in the Walnut and Squaw creek watersheds for water years 1995 to 2000. Herbicide concentrations and chemical loads measured in the treatment watershed (Walnut Creek) are compared to the control watershed (Squaw Creek) to determine whether changes in herbicide transport have occurred between the two watershed areas. Results from a one-time sampling of streams in the Walnut Creek watershed are presented to determine sources of elevated atrazine concentrations in the watershed and isolate the effects of refuge management activities on surface water quality.

## **METHODS**

#### **Sample Collection and Analysis**

Herbicide concentrations are monitored weekly to monthly at ten sites in the Walnut and Squaw Creek watersheds (Figure 1). Upstream and downstream sites on the main stems and three tributary basins are monitored in each watershed. Sample collection is stratified by season, with greater sampling frequency during spring and early summer. Weekly monitoring is targeted for May



Figure 1. Location map of Walnut Creek and Squaw Creek watersheds.

and June when herbicide transport is greatest following post-application, whereas bimonthly sampling occurs in March, April, July, August and September. During late fall and winter, stream samples are collected on a monthly basis at upstream-downstream locations at main stem sites only. Laboratory analyses were performed by the University of Iowa Hygienic Laboratory (UHL) using standard methods.

## **Statistical Methods**

Statistical analyses were performed according to the guidelines of Grabow et al. (1998, 1999). To test for the gradual change in chemical concentrations over time a multiple linear regression analysis was performed. The equation is:

$$\mathbf{Y} = \boldsymbol{\beta}_0 + \boldsymbol{\beta}_1 \boldsymbol{X}_1 + \boldsymbol{\beta}_2 \boldsymbol{X}_2 \tag{1}$$

)

where, Y is either the water quality variable or log of the variable for the treatment watershed (Walnut Creek), X1 is the same water quality variable (or log) for the control watershed (Squaw Creek), and  $X_2$  is elapsed time (in weeks), and  $\beta_0$ ,  $\beta_1$ , and  $\beta_2$  are regression parameters. The estimate of  $\beta_2$  indicates the magnitude of change over time in units per week. By having a control watershed (variable X1), the analysis blocks out much of the hydrologic variability, and the change should be isolated to treatment effects which in this case is being modeled as time (X2). For statistical analyses of atrazine concentration data, concentrations reported as  $<0.1 \mu g/L$  were considered to be one-half the detection limit (0.05 µg/L). Atrazine data were highly skewed and required log transformation before regression analyses were conducted.

A second method of evaluating trends in atrazine concentrations was conducted by treating time as a discrete variable rather than a continuous variable. This was done by treating year as a class variable:

$$\mathbf{Y} = \boldsymbol{\beta}_0 + \boldsymbol{\beta}_1 \, \boldsymbol{X}_1 + \boldsymbol{\beta}_{2i} \tag{2}$$

where the terms are the same as in Equation (1) except that  $\beta_2$  is now a class variable related to time rather than a continuous variable. B₂ has i values one for each year.

### **Chemical Loads**

The U.S. Geological Survey (USGS) program ESTIMATOR was used to estimate daily loads of atrazine at the three stream gaging sites. Stream gaging stations are located at the bottom of the Walnut and Squaw creek watersheds and an additional gage is located at the upper end of the Walnut Creek watershed where the majority of refuge land begins. The ESTIMATOR program utilizes a Minimum Variance Unbiased Estimator (MVUE) to implement a seven parameter regression model based on the relationship between log-flow and logconcentration (Cohn et al, 1989; 1992; Gilroy et al., 1990). Daily chemical load data were tabulated and summarized by month and water year. Load data were normalized on a unit area basis by dividing the total annual load at each gaging site by the watershed area above the gage. In the case of Walnut Creek watershed, the load per unit area between the two gauge sites was determined by subtracting the load estimated at WNT1 from WNT2.

The accuracy of the modeling approach was evaluated by comparing actual concentration data with estimated concentrations generated by the ESTIMATOR model (Figure 2). There was good correlation between the modeled concentrations and measured values. In general, estimated concentrations tended to be higher than actual values at lower concentration ranges, but lower at higher concentration ranges.

#### **One-Time Sampling Event**

Discharge rate and water chemistry was measured during baseflow conditions at 81 tributary creeks and drainage tiles over a two-day period in May 1999. On May 7, 1999, discharge and water chemistry was measured at 18 creeks and 19 tiles along the main channel of Walnut Creek (Schilling and Wolter, 2001). On May 8, 1999, with the assistance of refuge staff and local volunteers, additional water sampling was conducted to subdivide several watersheds into smaller drainage basins. An additional 28 creeks and 16 tiles were sampled during this portion of the study (Schilling, 2001). Sampling methodology and results from the



**Figure 2.** Comparison of atrazine concentrations estimated with ESTIMATOR model with actual concentration data (data for WNT2 shown).

one-time sampling event are reported in Schilling and Wolter (2001) and Schilling (2001). Previously unreported results of atrazine monitoring in 45 tributary streams sampled during this study are included in this report.

### **RESULTS AND DISCUSSION**

#### Concentrations

Atrazine was the most frequently detected herbicide compound in both Walnut and Squaw creek watersheds with the frequency of detection ranging between 78% to 85% in the main channels (Table 1). Concentrations ranged between <0.1 to  $5.0 \mu g/L$  at Walnut Creek and <0.1 to  $8.1 \mu g/L$  at Squaw Creek, with median concentrations at the downstream stations nearly equal (0.33  $\mu g/L$  vs.  $0.34 \mu g/L$ , respectively) (Table 1). Atrazine and two atrazine degradates, deethylatrazine (DEA), and deisopropylatrazine (DIA), were detected in more than 50% of the water samples collected at all sample sites except SQW4 and SQW5 subbasins (Figures 3 and 4). Detection frequencies of DEA far exceeded detection frequencies of DIA, with DEA detections exceeding 80% at most sample sites. Median concentrations of DEA were generally one-half to one-third less than the parent compound atrazine.

Median cyanazine concentrations exceeded the detection at only one site (WNT1) (Figure 5). In general, cyanazine concentrations tended to be higher in Walnut Creek watershed than Squaw Creek. Acetochlor detections were similar to cyanazine with median concentrations less than  $0.35 \mu g/L$ . Alachlor and metolachlor were rarely detected in surface water samples in either watershed.

Atrazine concentrations were highest in May and June of each year during periods of high stream flow associated with rainfall runoff (Figure 6). Following peak events, atrazine concentrations decreased in the late summer and fall. The timing of peak concentrations in the late spring/early summer with high streamflow events is consistent with the "spring flush" described by Thurman et al. (1991). Jaynes et al. (1999) reported similar patterns at stream sites, county drain sites and field tile sites in Story County Iowa.

### **Temporal Changes**

Box plots of annual atrazine concentrations at WNT2 and SQW2 show similar temporal patterns (Figure 7). Median concentrations generally increased from WY1995 to WY1997, decreased slightly in WY1998, increased in WY1999, then decreased in WY2000.

Multiple linear regression analysis (Equation 1) indicated a general decrease in atrazine at the outlet of Walnut Creek (WNT2) while adjusting for the control (SQW2). The parameter of  $\beta_2$  in the regression equation (elapsed time) was negative (-0.000387) which indicated a decrease in concentration over time. However, the trend over time was not significant at P=0.05 (95% significance). It was nominally significant at a 90% significance level (P=0.10). The mean decrease over the entire sampling period was -0.126 log units. Using the mean log value of Squaw Creek (-0.551) as X₁, and taking the antilog to obtain an untransformed an-

	Atraz	zine	Cyana	zine	Acetoo	chlor	Deethylatrazine			
Sample Site	Detection Frequency (%)	Median Conc. (ug/l)	Detection Frequency (%)	Median Conc. (ug/l)	Detection Frequency (%)	Median Conc. (ug/l)	Detection Frequency (%)	Median Conc. (ug/l)		
WNT1	84.8	0.29	41.7	0.17	13.3	0.24	81.0	0.19		
WNT2	78.3	0.33	33.3	0.27	21.7	0.20	80.7	0.16		
WNT3	78.9	0.23	20.8	0.13	7.6	0.31	73.0	0.13		
WNT5	82.4	0.30	21.2	0.16	23.1	0.16	82.0	0.15		
WNT6	82.7	0.28	22.6	0.18	20.8	0.19	82.4	0.19		
SQW1	84.6	0.37	23.1	0.21	30.8	0.26	88.5	0.21		
SQW2	83.3	0.34	31.7	0.18	33.3	0.27	80.0	0.16		
SQW3	86.3	0.34	27.5	0.14	23.5	0.17	80.4	0.17		
SQW4	56.9	0.29	5.9	0.16	25.5	0.29	9.8	0.16		
SQW5	68.0	0.21	20.0	0.26	38.0	0.32	32.0	0.12		

Table 1. Summary of herbicide concentrations in surface water for water years 1995 to 2000.



**Figure 3.** Box plot of atrazine concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000. Box plots illustrate the  $25^{th}$ ,  $50^{th}$  and  $75^{th}$  percentiles; the whiskers indicate the  $10^{th}$  and  $90^{th}$  percentiles; and the circles represent data outliers.



**Figure 4.** Box plot of deethylatrazine (DEA) and deisopropylatrazine (DIA) concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000.



**Figure 5.** Box plot of cyanazine concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000.

swer, atrazine decreased from 0.3080 to  $0.230 \,\mu\text{g}/$ L over the entire 326-week sampling period while adjusting by the control watershed.

To visualize this approach, a box plot of residuals grouped by year is shown in Figure 8. (For this analysis, concentration data were grouped by calendar year rather than water year.) These residuals are from a simple regression of treatment versus control (without a time factor). A change in this relationship over time would be evident in the residuals from the simple regression. Figure 8 suggests that 1994 and 2000 were relatively low years as adjusted by the control, compared to 1997. Incorporating this idea into a statistical analysis based upon Equation (2) showed that 1994 and 2000 were statistically different than 1997, while no other years were statistically different from each other (Table 2). This probably agrees with the regression analysis results with time as a continuous variable, as the significance wasn't particularly strong, however the values in the year 2000 probably "pulled" or "leveraged" the line on time such that it had a negative slope (indicating a reduction over time).

When analyzed as an upstream/downstream design there was no statistically significant trend in Walnut Creek atrazine concentrations over time in the downstream station while adjusting for the upstream control.

## **Atrazine Loads**

Annual export of atrazine from the Walnut and Squaw creek watersheds ranged from 10.57 kg at SQW2 in WY 1998 to 1.90 kg at WNT2 in WY 2000 (Table 3). Except for WY 1997, export loads were lower in Walnut Creek compared to Squaw Creek. Peak loads occurred in May 1998 and May 1996 when export of atrazine from Squaw Creek exceeded 4 kg (Figure 9). Atrazine loads often exceeded 1 kg in May and June in both watershed areas each year (Figure 9). Over a fiveyear period, the months of May and June accounted for approximately 70% of the export load of atrazine, and the period of April through July accounted for 85 to 95% of the annual atrazine load each year (Table 4).

On a unit area basis, atrazine loads ranged from 0.47 to 2.25 g/ha in Squaw Creek watershed and 0.36 to 1.43 g/ha in Walnut Creek watershed (Table 3). Average annual atrazine loads in Squaw Creek (1.32 g/ha) are considerably higher than Walnut Creek (0.76 g/ha). Within Walnut Creek watershed, atrazine loading rates tended to be higher in upper Walnut Creek (WNT1) compared to lower Walnut Creek (WNT2-1), although little difference in average annual loads exists (Table 3). A plot of annual atrazine loading rates suggests that the rates are influenced by the amount of discharge



**Figure 6.** Atrazine concentrations versus discharge at WNT2 and SQW2 sites for water years 1996 to 2000.

emanating from the watershed areas (Figure 10).

# Atrazine Losses in Walnut Creek Watershed

Atrazine loading rates measured in 45 subwatersheds during a two-day period in May 1999 are shown in Figure 11. Atrazine loads from the single sampling event were annualized to provide comparable units to the annual export loads. Major differences in atrazine loading rates are evident within the Walnut Creek watershed (Figure 11). Highest atrazine loads per ha are located in many headwater areas. Of the 45 subwatersheds sampled in this study, nine subwatersheds (20%) showed annualized atrazine loads greater than 0.72 g/ha, six of which are located in the area above WNT1. In contrast, the core of the watershed occupied by the Neal Smith National Wildlife Refuge showed annualized atrazine loads less than 0.007 g/ha (Figure 11). In many areas, the approximate boundary of the refuge can be traced by following the subwatershed areas exhibiting low atrazine loads. Results of this one-time sampling event suggest that differences in atrazine loading rates within Walnut Creek watershed are more pronounced than implied by the average annual loads reported in Table 3. Atrazine loads vary by more than two orders of magnitude in the Walnut Creek watershed suggesting that caution should be used when attributing atrazine loads at a watershed scale by simply sampling at watershed outlets.

## SUMMARY AND CONCLUSIONS

Results from surface water monitoring at Walnut and Squaw Creek watersheds indicate that atrazine and DEA are commonly detected herbicides in both watersheds. Water samples collected predominantly in spring and summer suggest that



**Figure 7.** Box plot of annual atrazine concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000.

detection frequencies for these compounds are near 80%. Cyanazine and acetochlor are occasionally detected (up to 40%) whereas alachlor and metolachlor were rarely detected (less than 5%). Concentrations of atrazine often exceeded 1  $\mu$ g/L during high streamflows in late spring/early summer; however, median concentrations of all detected herbicides were less than 0.4  $\mu$ g/L. Atrazine loads estimated using the USGS model ESTIMA-TOR suggest that 70% of the annual loss of atrazine occurred primarily in May and June and up to 95% of the atrazine loss occurred between April and July.

Multiple linear regression analysis suggests that atrazine concentrations have decreased from 0.31 to 0.23  $\mu$ g/L since the project began while adjusting by the control watershed (Squaw Creek). However, this trend was not highly significant and may have been biased by the relatively lower concentration values measured during Water Year 2000. An upstream-downstream analysis of Walnut Creek did not indicate a statistically significant trend over time.

Export of atrazine from Walnut and Squaw creek watersheds averaged 4.0 kg/yr and 6.2 kg/yr, respectively. Average atrazine losses were also higher in Squaw Creek (1.32 g/ha) compared to

Walnut Creek (0.76 g/ha). Results from a one-time sampling of 45 creeks in Walnut Creek watershed indicated major differences in atrazine loading rates within the watershed. Many headwater subwatersheds dominated by row crop land use appeared to contribute more than 100 times greater atrazine loading than subwatersheds dominated by restored prairie and refuge-owned farm lands.

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**Figure 8.** Box plot of regression residuals of atrazine concentrations at WNT2 adjusted for control (SQW2) grouped by calendar year.

**Table 2.** Least squared means of WNT2 atrazineconcentrations by year.

Year	LS Mean ¹ Log (ug/L)	Untransformed ug/L
1994	-0.866 A ²	0.136
1995	-0.538 AB	0.289
1996	-0.564 AB	0.273
1997	-0.495 B	0.319
1998	-0.6611 AB	0.218
1999	-0.600 AB	0.251
2000	-0.7137 A	0.193

¹LS or least squared mean is the predicted value of the treatment watershed evaluated at the mean value of the control watershed.

²Similar letters indicate no statistical difference between least squared means, i.e., A is different than B and AB is not different from either A or B

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**Table 3.** Atrazine losses from various Walnut and Squaw creek watershed areas.

Water				
Year	SQW2	WNT2	WNT1	WNT2-WNT1
		Atrazine Ex	port Load	(kg)
1996	7.05	3.23	1.64	1.59
1997	2.20	3.07	0.92	2.15
1998	10.57	7.44	2.31	5.12
1999	5.48	4.19	1.39	2.79
2000	5.67	1.90	0.93	0.97
Average	6.20	3.96	1.44	2.52
	A	trazine Load	ding Rate	(g/ha)
1996	1.50	0.62	0.94	0.46
1997	0.47	0.59	0.53	0.62
1998	2.25	1.43	1.32	1.48
1999	1.17	0.80	0.80	0.80
2000	1.20	0.36	0.53	0.28
Average	1.32	0.76	0.82	0.73



Figure 9. Monthly atrazine loads at various watershed areas.

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	Atrazine Lo	ad by Mont	th, WY96-00	(kg)
Month	WNT2	WNT1	WNT2-1	SQW2
Jan	0.02	0.03	0.00	0.05
Feb	0.17	0.19	-0.01	0.82
Mar	0.50	0.23	0.27	0.88
Apr	1.56	0.50	1.05	2.11
May	6.43	2.11	4.32	11.94
Jun	7.00	2.56	4.44	11.08
Jul	3.10	0.99	2.11	2.90
Aug	0.84	0.42	0.42	0.91
Sep	0.10	0.06	0.04	0.09
Oct	0.05	0.05	0.00	0.09
Nov	0.03	0.04	-0.01	0.06
Dec	0.02	0.02	-0.01	0.04
Total	19.82	7.20	12.62	30.98
Jan	0.1%	0.4%	0.0%	0.2%
Feb	0.9%	2.6%	-0.1%	2.7%
Mar	2.5%	3.2%	2.1%	2.8%
Apr	7.8%	7.0%	8.3%	6.8%
May	32.4%	29.3%	34.2%	38.6%
Jun	35.3%	35.5%	35.2%	35.8%
Jul	15.7%	13.8%	16.7%	9.4%
Aug	4.3%	5.9%	3.3%	2.9%
Sep	0.5%	0.8%	0.3%	0.3%
Oct	0.2%	0.6%	0.0%	0.3%
Nov	0.2%	0.6%	-0.1%	0.2%
Dec	0.1%	0.3%	-0.1%	0.1%

**Table 4.** Summary of atrazine losses by month from various Walnut and Squaw creek watershed areas.



Figure 10. Annual atrazine loads from various watershed areas relative to discharge at WNT2 and SQW2.



Figure 11. Atrazine losses from various Walnut Creek subwatersheds measured in May 1999.

# FECAL COLIFORM CONCENTRATIONS AND TRENDS IN SURFACE WATER IN WALNUT AND SQUAW CREEK WATERSHEDS

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#### **INTRODUCTION**

The sanitary quality of water is often assessed using bacteriological methods that detect the presence of certain bacteria that indicate the presence of fecal material from warm-blooded animals (USEPA, 1986). Concentrations of the fecal coliform bacteria, such as Escherichia coli and Aerobacter aerogenes, do not necessarily present risks for waterborne disease, such as gastroenteritis, bacillary dysentery, or others, but are found in association with pathogenic microorganisms (Salmonella, Shigella, etc.) that do present a risk of infection. The origin of fecal coliform contamination can be from point sources, such as outfalls from sewage treatment plants, or nonpoint sources. Nonpoint sources include a variety of diffuse sources, including agricultural animal waste storage and manure application, agricultural runoff from manure-applied fields, failed septic systems, urban and construction runoff, landfill leakage and wildlife waste. The standard for fecal coliform

bacteria in Iowa Class (A) surface waters is 200 colony forming units per 100 ml of water (200 CFUs or counts/100 ml).

The purpose of this report is to present results of fecal coliform bacteria monitoring in the Walnut and Squaw creek watersheds for water years 1995 to 2000. Fecal coliform concentrations measured in the treatment watershed (Walnut Creek) were compared to the control watershed (Squaw Creek) to determine whether a change in fecal coliform transport has occurred between the two watershed areas.

#### **METHODS**

#### **Sample Collection and Analysis**

Fecal coliform concentrations are monitored weekly to monthly at ten sites in the Walnut and Squaw creek watersheds (Figure 1). Upstream and downstream sites on the main stems and three tributary basins are monitored in each watershed.



**Figure 1.** Box plot of fecal coliform concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000. Box plots illustrate the 25th, 50th and 75th percentiles; the whiskers indicate the 10th and 90th percentiles; and the circles represent data outliers.

					a c th	Quartile	a cth
	<u>n</u>	range	mean	sa	25	50	/5
WNT1	98	<10-7,600,000	104,226	776,336	403	1,200	3,675
WNT2	98	<10-150,000	7,988	24,992	288	980	2,875
WNT3	65	<10-6,700	800	1,407	130	290	640
WNT5	64	<10-82,000	2,289	10,806	98	350	780
WNT6	64	<10-8,900	565	1,262	88	215	405
SQW1	91	<10-250,000	9048	39,400	165	570	1,100
SQW2	99	<10-13,000,000	138425	1,306,093	350	870	2,250
SQW3	63	<10-22,000	1409	3,293	140	350	965
SQW4	64	<10-49,000	2318	8,386	100	355	750
SQW5	64	<10-4,100,000	72060	512,200	338	530	1,325

Table 1. Summary of fecal coliform concentrations in surface water for water years 1995 to 2000.

Sample collection is stratified by season, with greater sampling frequency during spring and early summer. Weekly monitoring is targeted for May and June when herbicide transport is greatest following post-application, whereas bimonthly sampling occurs in March, April, July, August and September. During late fall and winter, stream samples are collected on a monthly basis at upstream-downstream locations at main stem sites only. Laboratory analyses were performed by The University of Iowa Hygienic Laboratory (UHL) using standard methods.

## **Statistical Methods**

Statistical analyses were performed according to the guidelines of Grabow et al. (1998, 1999). To test for the gradual change in chemical concentrations over time a multiple linear regression analysis was performed. The equation is:

$$\mathbf{Y} = \boldsymbol{\beta}_0 + \boldsymbol{\beta}_1 \, \boldsymbol{X}_1 + \boldsymbol{\beta}_2 \, \boldsymbol{X}_2 \tag{1}$$

where, Y is either the water quality variable or log of the variable for the treatment watershed (Walnut Creek), X₁ is the same water quality variable (or log) for the control watershed (Squaw Creek), and X₂ is elapsed time (in weeks), and  $\beta_0$ ,  $\beta_1$ , and  $\beta_2$  are regression parameters. The estimate of  $\beta_2$  indicates the magnitude of change over time in units per week. By having a control watershed (variable  $X_1$ ), the analysis blocks out much of the hydrologic variability, and the change should be isolated to treatment effects which in this case is being modeled as time ( $X_2$ ). For statistical analyses of fecal coliform concentration data, concentration data were highly skewed and required log transformation before regression analyses were conducted.

A second method of evaluating trends in fecal coliform concentrations was conducted by treating time as a discrete variable rather than a continuous variable. This was done by treating month as a class variable:

$$Y = \beta_0 + \beta_1 X_1 + \beta_{2i}$$
⁽²⁾

where the terms are the same as in Equation (1) except that  $\beta_2$  is now a class variable related to time rather than a continuous variable. B₂ has i values one for each month.

#### **RESULTS AND DISCUSSION**

#### Concentrations

Fecal coliform counts varied widely among sampling sites and water years, ranging from less than 10 counts/100 ml to 13 million counts/100 ml at



**Figure 2.** Box plot of annual fecal coliform concentrations measured at WNT2 and SQW2 sites for water years 1995 to 2000.

SQW2 (Table 1). Figure 1 shows box plots for fecal coliform counts detected in water samples collected from various sample sites. Highest median values occurred at WNT1 where the median fecal coliform count was 1,200 counts/100ml (Table Median values at downstream watershed 1). outlets WNT2 and SQW2 were similar (980 and 870 counts/100ml) whereas all other subbasin sites were less than 570 counts/100ml. All median fecal coliform values exceeded the water quality criterion of 200 counts/100ml. The 25th percentile of fecal coliform values exceeded the criterion at WNT1, WNT2, SQW2 and SQW5 (Table 1). Annually, median fecal coliform counts were higher at SQW2 than WNT2 in water years 1995 and 1999, higher at WNT2 in WY96, and similar in WY97, WY98 and WY00 (Figure 2).

Highest levels of fecal coliform bacteria tended to occur in spring and early summer months during high stream flow periods associated with rainfall runoff (Figure 3). This pattern is consistent with fecal coliform detections monitored at other Iowa sites (Langel et al., 2001; IDNR-GSB, 2001). Long-term monitoring data across Iowa indicate highest fecal coliform concentrations typically occur in the period between June and September (IDNR-GSB, 2001). In Walnut and Squaw Creek watersheds, highest fecal coliform counts tended to occur in the period between May and August (Figure 4). This may be viewed as a period corresponding to both primary grazing activity by local ranchers and a period of greater rainfall intensity (Schilling, 2000).

### **Temporal Changes**

When looking for a trend in time as per equation (1) there was no statistically significant trend in fecal coliform over time between treatment and control watersheds (p=0.2466). However when using equation (2) and using month as a class variable, there was a statistically significant decrease in fecal coliform over time while adjusting for the control and elapsed time in weeks (Table 2).

Seasonal variability was introduced into the regression analysis using equation (2) with months 5-8 (May-August) and months 9-12; 1-4 grouped into grazing versus non-grazing seasons. This was nominally significant (p=0.07). Parameter estimates for months 5-8 were higher than the other months and generated higher predictions of fecal coliform.

These two methods resulted a reduction of 0.39 and 0.35 log units of fecal coliform respec-



Figure 3. Fecal coliform concentrations versus discharge at WNT2 and SQW2 sites for water years 1996 to 2000.

tively over the sampling period (326 weeks). For the second method, this translates to a average change (using average value of Squaw Creek for X) from 3.444 to 3.095 log units during the "grazing season" and a change from 2.828 to 2.468 during the "non-grazing" season; or 2780 mpn/100 to 1245mpn/100 and 673 mpn/100 to 294 mpn/100 respectively.

When evaluated as an upstream/downstream design using Equation (1) there was no statistically significant trend in fecal coliform over time (p=0.60 on time unit). The same holds true when month was introduced into the analysis.

#### **Relation to Potential Sources**

In the Walnut and Squaw creek watersheds, primary sources of fecal coliform include pastures and manure application to cropped fields. Outfalls from sewage treatment plants from cities of Prairie City or Colfax do not enter into Walnut or Squaw creek watersheds, respectively (Figure 5). Outfall from the Prairie Learning Center at the Neal Smith NWR does enter the Walnut Creek watershed. However, prior to discharge, wastewater is directed through wetlands where bacteria and nutrients are removed.



**Figure 4.** Box plot of fecal coliform concentrations measured at Walnut and Squaw creek sampling sites for water years 1995 to 2000 grouped according to grazing and non-grazing seasons.

Locations of pastures depicted in Figure 5 include areas where animals, primarily cattle, appear to be located permanently throughout the year. Most permanent pastures have access to a waterway for direct contact of livestock with surface water supplies. Pasture sites located immediately upstream of WNT1 and WNT2, and SQW2 may help explain why elevated fecal coliform concentrations have been detected at these sites. Median and maximum fecal coliform levels were not particularly elevated in the WNT5 subbasin despite a large proportion of the watershed consisting of pasture (primarily bison pasture in restored prairie) (Figure 5). The low population density of the bison in the prairie area may contribute to the lower than expected concentrations. In many areas, higher fecal coliform concentrations detected at sampling sites during summer grazing season would be consistent with pasture sources.

### SUMMARY AND CONCLUSIONS

Results from surface water monitoring at Walnut and Squaw creek watersheds indicate that fecal coliform bacteria are detected frequently above the Iowa water quality standard of 200 count/100ml in both watersheds. Highest median concentrations were typically noted at WNT1, WNT2 and SQW2, but nearly all monitored watersheds had occasional elevated detections. Highest levels of fecal coliform bacteria tended to occur in spring and early summer months during high stream flow periods associated with rainfall runoff.

Multiple linear regression analysis suggested that fecal coliform concentrations have decreased from 2780 mpn/100 to 1245 mpn/100 in Walnut Creek during the "grazing season" while adjusting by the control watershed (Squaw Creek). Potential fecal coliform sources appear to be nonpoint in nature and related to pastures and concentrated livestock in riparian corridors.

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β ₀	р	$\beta_1$ Control	р	β ₂ Elapsed time	р	β ₃	р	Comments
1.137	.0024	.5724	.0001	-0.00128	.047	varies	.0001	Log values. $\beta_{3,12}$ values one for each month
0.834	.0002	.65079	.0001	-0.00107	.077	.626	.0001	$\beta_3$ month 5-8, other 0.0

**Table 2**. Regression parameter estimates for fecal coliform concentrations.



**Figure 5.** Locations of continuous pasture areas and other potential sources of fecal coliform concentrations in Walnut and Squaw creek watersheds and surrounding areas.

## SUMMARY OF FIELD MEASUREMENTS IN SURFACE WATER IN WALNUT AND SQUAW CREEK WATERSHEDS

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#### INTRODUCTION

The Walnut Creek Watershed Monitoring Project was initiated as a nonpoint source monitoring project related to large-scale land use changes implemented by the U.S. Fish and Wildlife Service (USFWS) at the Neal Smith National Wildlife Refuge in Jasper County, Iowa (Schilling and Thompson, 1999). Water quality has been monitored since 1995 to detect changes in chemical transport resulting from conversion of large tracts of land from row-crop to native prairie in a treatment watershed (Walnut Creek) compared to a highly agricultural control watershed(Squaw Creek) (Figure 1).

Several field parameters have been measured since project startup, including temperature, pH, turbidity, specific conductance, dissolved oxygen, alkalinity and reduction-oxidation potential (redox). Collection of field data assists in characterizing surface water quality in the two watersheds and lends support to conclusions drawn about other chemical constituents. Field measurements refer to analytical determinations that document water conditions at the time of sample collection. Field measurements are also important for parameters that may be altered during storage and shipment to the laboratory (e.g. pH, alkalinity). This report presents results of field parameters measured in the Walnut and Squaw creek watersheds during water years 1995 through 2000.

# **METHODS**

Field measurements were made at ten sites in the Walnut and Squaw creek watersheds (Figure 1). The monitoring sites in both watersheds consisted of upstream and downstream sites on the main stem and three tributaries. Weekly monitor-



**Figure 1.** Location map of Walnut and Squaw Creek surface water monitoring sites.

ing was completed in May and June, and bimonthly sampling occurred in March, April, August, and September. During late fall and winter, water samples were collected monthly at upstream and downstream locations only. A Hydrolab multiprobe water analyzer was used to measure temperature, pH, turbidity, specific conductance, redox, and dissolved oxygen. Alkalinity was measured using a direct titration method with sulfuric acid of pH 4.5. Turbidity was measured using a Hach 2100P turbidimeter. All field equipment was calibrated on a regular basis prior to use.

T-tests were conducted to determine statisti-

		SQW1	SQW2	SQW3	SQW4	SQW5	WNT1	WNT2	WNT3	WNT5	WNT6
Temperat	ure (deg C)										
	۲	93	98	62	62	61	97	95	61	60	59
	Range	-0.13 - 23.4	-0.18 - 24.4	2.4 - 19.02	1.5 - 19.8	3.6 - 18.76	-0.13 - 28.6	-0.1 - 29.3	4 - 25.8	4.2 - 26.4	3.7 - 27.1
	Mean	12.2	12.6	12.7	12.9	12.6	13.7	14.2	14.0	16.5	17.4
	Standard Deviation	5.3	6.0	3.8	4.1	3.6	6.2	6.9	3.8	4.9	5.3
Hq											
	E	84	06	56	56	55	87	87	54	54	53
	Range	6.2 - 8.54	6.49 - 8.56	6.12 - 8.41	6.41 - 8.29	6.34 - 8.34	6.28 - 8.71	6.33 - 9.02	6.39 - 8.9	6.86 - 9.1	6.5 - 9.08
	Mean	7.8	7.8	7.7	7.6	7.3	7.9	7.9	7.8	8.1	7.9
	Standard Deviation	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
Turbidity	(NTU)										
	L	90.0	96	59	60	59	95.0	93	58	57	55
	Range	2 - 293	5 - 1000	1 - 361	0.8 - 165	2 - 189	3.4 - 526	2.1 - 1000	4.6 - 76	5 - 183	0.8 - 323
	Mean	24.9	6.99	29.0	21.8	14.1	70.3	80.5	19.2	37.0	32.9
	Standard Deviation	36.6	160.9	51.1	29.9	27.2	94.6	150.9	15.3	37.7	47.5
Specific C	Sonductance (umhos/	cm)									
	Е	89	96	59	58	58	95	93	59	57	57
	Range	339 - 649	395 - 665	439 - 634	441 - 648	431 - 693	314 - 630	336 - 599	244 - 640	374 - 557	308 - 570
	Mean	568.9	534.8	545.2	518.0	529.3	539.0	495.3	515.1	478.5	453.5
	Standard Deviation	46.8	58.3	35.1	49.4	48.4	51.6	50.7	61.3	41.4	56.2
Redox (m	()										
	c	18	18	8	8	80	18	16	80	7	7
	Range	108 - 371	106 - 377	163 - 377	154 - 351	99 - 345	138 - 437	156 - 418	254 - 378	275 - 435	183 - 349
	Mean	283.4	278.7	304.4	291.3	269.1	303.4	317.9	315.0	334.4	292.3
	Standard Deviation	74.2	88.2	63.9	61.0	76.6	80.5	63.6	38.6	59.6	57.4
Dissolved	1 Oxygen (mg/L)										
	Ę	91	97	60	60	59	93	93	59	58	56
	Range	5.61 - 17.2	5.53 - 16.2	7.5 - 16.5	4.04 - 13.7	1.2 - 17.8	2.97 - 18	3.7 - 18.1	4.3 - 14.4	7.68 - 16	3.83 - 13.1
	Mean	10.6	10.4	10.3	0.6	7.6	10.1	10.0	9.6	10.2	9.3
	Standard Deviation	2.3	2.1	1.7	2.1	3.3	2.6	2.4	2.0	1.9	1.8
Alkalinity	(mg/L)										
	L	63	70	45	45	44	67	69	45	45	44
	Range	132 - 246	124 - 228	128 - 248	160 - 260	135 - 220	128 - 264	108 - 296	102 - 234	102 - 260	98 - 250
	Mean	190.9	183.4	176.6	209.8	172.3	187.4	176.5	180.7	171.2	163.0
	Standard Deviation	25.2	25.8	21.6	24.6	20.4	27.4	31.6	24.2	32.3	32.8

Table 1. Summary of field parameter measurements.

cally significant differences among sampling sites using Microsoft Excel. Statistical analyses were performed for upstream/downstream sites within a watershed (i.e. SQW1 versus SQW2) and upstream/downstream sites between the two watersheds (i.e. SQW2 versus WNT2).

## **RESULTS AND DISCUSSION**

The mean water temperature in Squaw Creek ranged between 12.2 and 12.6 °C, whereas temperature values in Walnut Creek averaged 13.7 and 14.2 °C (Table 1). Temperature differences were likely related to the timing of routine sample collection in the watersheds since water samples were typically collected in Squaw Creek before Walnut Creek. The weekly and bimonthly data for temperature revealed expected seasonal variability in both watersheds. The box plot in Figure 2 displays the wide range of water temperatures encountered in both creeks.

The hydrogen-ion activity of the water is measured as pH. Field measurements of pH are more likely to represent the natural conditions of the water than laboratory results of pH (Hem, 1989). Walnut and Squaw creeks exhibited similar pH values over the sampling period, with mean annual pH values nearly the same (7.8 to 7.9; Table 1).

Turbidity measures the optical properties of water that cause light to be reflected and is closely related to concentrations of total suspended solids contained in the water. Mean turbidity was higher at WNT1 (70.3 nephelometric turbidity units or NTU) and WNT2 (80.5 NTU) compared to SQW1 (24.9 NTU) and SQW2 (66.9 NTU) (Table 1). Turbidity values ranged from 0.8 NTU measured at WNT6 and SQW4 to values greater than 1,000 NTU measured at the downstream sites SQW2 and WNT2 after precipitation events (Figure 3). A t-test found that mean annual turbidity at WNT1 was significantly higher than SQW1 and WNT2 was significantly higher than SQW2 (p<0.05). Turbidity in Walnut Creek appears to be decreasing over time (Figure 3). In 1995, both upstream and downstream sites on Walnut Creek had turbidity annual means of 130.4 and 81.8 NTU respectively. These turbidity levels decreased over time to 33.6



**Figure 2.** Box plots of temperature and pH measured at Walnut and Squaw creek monitoring sites.



**Figure 3.** Box plot and mean annual turbidity measured at Walnut and Squaw creek monitoring sites.

and 39.9 NTU in Water Year 2000. Turbidity means at the Squaw Creek upstream and down-stream sites did not show such a trend.

Specific conductance measures the ability of water to conduct electrical current and is directly related to the amount of dissolved ions in the water. Higher dissolved ion concentrations correspond to higher specific conductance. Both Walnut and Squaw creek watersheds had statistically higher specific conductance in upstream samples compared to downstream samples (Table 1, p<0.05). Mean annual specific conductance at SQW1 (568.9 µmhos/cm) was significantly higher than the mean at WNT1 (539.0 µmhos/cm), and mean specific conductance at SQW2 (534.8 µmhos/cm) was significantly higher than the mean at WNT2 (495.3 µmhos/cm) (Figure 4). All specific conductance values were within the normal range of specific conductance for surface water in Iowa and did not appear to change over time in either watershed.

Reduction-oxidation potential (redox) reflects the intensity of the oxidizing or reducing conditions in the water. Positive potentials indicate the solution is oxidizing, whereas negative potentials indicate the solution is reducing. Mean annual redox at SQW1 (283.4 mV) was lower than at WNT1 (303.4 mV) and mean annual redox at SQW2 (278.7 mV) was less than at WNT2 (317.9 mV) (Table 1). The redox values were within the range found in natural surface waters, however data on specific redox values in Iowa was not available for comparison. One of the tributaries of Squaw Creek (SQW5) had several characteristically low redox values (Figure 4). These low values and a noticeable white coloring to the creek prompted an investigation in October 2001 by regulatory personnel in the Department of Natural Resources. This investigation determined that a local property owner had dumped a large quantity of milk into the upstream portion of the tributary that is sampled at SQW5. The farmer has since been ordered to discontinue the discharge of milk to the creek, but the dumping may have occurred in past years. If so, anaerobic conditions caused by the discarded milk may have influenced the low redox values measured at SQW5 and may have contributed to lower values at SQW2 over the last several years. Walnut



**Figure 4.** Box plots of specific conductance, redox, dissolved oxygen, and alkalinity measured at Walnut and Squaw creek monitoring sites.

Creek sites WNT1 and WNT2 appear to have a decreasing trend over time with respect to redox. However, two years of data are not sufficient to draw conclusions about this downward trend.

Dissolved oxygen is a measure of the oxygen concentration in water. This parameter is often influenced by water temperature because warmer water holds less oxygen than colder water. Effects of water temperature were evident in the dissolved oxygen data. The box plot in Figure 4 displays the variability in dissolved oxygen concentrations at these sites. Mean annual dissolved oxygen was higher at SQW1 and SQW2 (10.6 and 10.4 mg/L, respectively) compared with WNT1 and WNT2 (10.1 and 10.0 mg/L, respectively) (Table 1). These differences in dissolved oxygen are likely related to the water temperature at the time of sample collection. Thus, in Squaw Creek, surface water tended to be cooler and the dissolved oxygen higher in comparison to Walnut Creek. Dissolved oxygen concentrations at SQW5 have decreased in recent years possibly due to the effects of discarded milk in the upstream portion of the tributary. Bacterial growth from the discarded milk may have lowered dissolved oxygen concentrations in the tributary and contributed to lower dissolved oxygen concentrations detected at the downstream monitoring site SQW2. Dissolved oxygen concentrations appeared to be relatively stable at the Walnut Creek sites over time.

Alkalinity measures the capacity of solutes contained in a solution to react with and neutralize acids. The principal sources of alkalinity are dissolved carbon dioxide species such as bicarbonate and carbonate. A t-test (p<0.05) determined that mean annual alkalinity was significantly higher at the upstream sites within the Walnut and Squaw creek watersheds compared with the downstream sites. Mean annual alkalinity at SQW1 (190.9 mg/ L) was higher than at SQW2 (183.4 mg/L), and mean annual alkalinity at WNT1(187.4 mg/L) was higher than at WNT2 (176.5 mg/L) (Table 1). Mean alkalinity at SQW2 (183.4 mg/L) was significantly higher than the mean value at WNT1 (176.5 mg/L) (p<0.05; Table 1). Alkalinity means varied little over time in both creeks and were within the normal range for natural surface waters (Figure 4).

## SUMMARY AND CONCLUSIONS

Based on results of field parameters measured in Walnut and Squaw creek surface water, Walnut Creek sites tended to have higher turbidity values than the Squaw Creek sites. However, a decreasing trend in mean annual turbidity was evident in the Walnut Creek upstream and downstream site data (WNT1 and WNT2). Specific conductance and alkalinity were higher in Squaw Creek than Walnut Creek, whereas redox was higher in Walnut Creek. Differences in water temperature and dissolved oxygen appear related to the timing of routine sample collection in the watersheds. All field measurements were within the normal range of values measured in Iowa surface water (IDNR-GSB, 2002).

The effort expended to measure field parameters at Walnut and Squaw creeks has generated a profile of the water quality in these watersheds. The analysis of how field parameters vary in intensity and with time describes the character and variability of surface water quality and can reinforce conclusions drawn regarding other differences in water chemistry, such as nitrate, sulfate and chloride.

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# SUMMARY OF BENTHIC MACROINVERTEBRATES IN WALNUT AND SQUAW CREEK WATERSHEDS: 1995-2000

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# **INTRODUCTION**

The Walnut Creek Watershed Restoration and Water Quality Monitoring Project began in 1995 to evaluate changes in water quality and biological communities as large portions of the Walnut Creek watershed are restored from row crop to native prairie at the Neal Smith National Wildlife Refuge in Jasper County, Iowa. A paired watershed approach is being used to provide comparative information as prairie/savanna restoration and agricultural management practices are implemented in the Walnut Creek watershed (USEPA 1993). The Squaw Creek watershed is being used as the control watershed where land use is primarily agricultural. Walnut and Squaw creek watersheds have similar basin characteristics and soils and, prior to restoration, had similar land use (see Background and Land Use Changes, this issue). Squaw Creek is approximately 18.3 mi² and drains to the South Skunk River whereas Walnut Creek is approximately 20.1 mi² and drains to the Des Moines River. Both Walnut and Squaw creek watersheds are located within the Southern Iowa Rolling Loess Prairies Ecoregion (Ecoregion 47f; Figure 1). The Southern Iowa Rolling Loess Prairies ecoregion is characterized by irregular plains to open low hills with moderate to thick loess (Griffith et. al. 1994).

Biological monitoring was done in Walnut and Squaw creek watersheds to provide an integrated assessment of changes that are occurring through



time that may be reflected within the resident biological community. This report summarizes the benthic macroinvertebrate communities sampled in Walnut and Squaw creek watersheds from 1995 to 2000.

# BENTHIC MACROINVERTEBRATE INDEX OF BIOTIC INTEGRITY (BM-IBI)

Macroinvertebrates were collected from Walnut Creek and Squaw Creek and measures of community were evaluated through twelve numeric metrics. The benthic macroinvertebrate metrics were integrated to provide a biological summation, or BM-IBI. The BM-IBI is a composite index of 12 individual metrics that provides an assessment of biological community integrity. A metric is an attribute or characteristic of the aquatic community that is quantifiable and has ecological relevance. Useful metrics share the following characteristics: a) measured relatively easily and economically; b) exhibit relatively low natural variability; c) respond predictably to changes in stream quality; d) do not duplicate information supplied by other metrics.

The twelve BM-IBI metrics described below quantify various attributes of the benthic macroinvertebrate community that relate to taxon richness, community balance, pollution tolerance, and trophic (feeding) guild composition. The metrics vary in how they are quantified (i.e. integer, proportion, real number); therefore, the ranges of possible values are not equivalent. In order to construct a multi-metric index, in which each metric is assigned equal weigh, it is first necessary to convert the raw metric values into a standard scoring range (Karr et al. 1986). The procedures described by Hughes et. al (1998) were used to convert the metric values into scores ranging from 0 to 10.

# **Taxa Richness Metrics**

**1. Multi-habitat Taxa Richness (MHTR).** In Iowa's warmwater streams, benthic macroinvertebrate taxon richness decreases with decreasing water quality and habitat complexity. The highest levels of taxa richness are generally found in streams that have good water quality and a diversity of benthic habitats (e.g. detritus, heterogeneous sediments, root mats, streamside vegetation, woody debris). Conversely, low taxa richness is found in streams that have extreme flow fluctuations, monotonous habitat characteristics, and poor water quality.

The MHTR metric represents the total number of benthic macroinvertebrate taxa collected by handpicking organisms from all types of benthic habitat found in the sampling reach. As stream size increases from headwater stream to middle order stream, the optimum level of benthic macroinvertebrate taxon richness generally increases and then levels off.

**2. Standard-habitat Taxa Richness (SHTR).** The SHTR metric represents the total number of taxa identified in a single, 100-organism standard-habitat subsample. There are two types of standard-habitat samples: (1) coarse rock substrates in riffle/shallow run habitat; (2) artificial wood-plate substrates in shallow run habitat (streams lacking riffles). In healthy streams, wood or rock substrates situated in flowing water support an abundant and diverse benthic macroinvertebrate community. Iowa's wadeable streams can support twenty or more taxa in a relatively small area (~ 0.1 m²). As water quality declines, the benthic macroinvertebrate community becomes simplified and fewer taxa are supported.

**3. Multi-habitat EPT richness (MHEPT).** EPT taxa are benthic macroinvertebrates that belong to the pollution-sensitive aquatic insect orders: Ephemeroptera, Plecoptera, and Trichoptera. Pollution sensitivities of EPT taxa range from extremely sensitive to moderately tolerant. Many EPT taxa are adversely impacted by toxic contaminants, such as heavy metals and insecticides. High quality streams support relatively high numbers of EPT taxa. As stream quality declines, the number of EPT taxa also declines. The MHEPT metric represents the total number of EPT taxa collected by handpicking organisms from various types of benthic habitat in the sampling reach. MHEPT has a broad range of response to varying water quality and habitat conditions.

**4. Standard-habitat EPT richness (SHEPT).** This metric represents the number of EPT taxa identified from a 100-organism subsample of the standard habitats described above. Many EPT taxa have a strong affinity for coarse substrates situated in flowing water. In healthy streams, relatively high numbers of EPT taxa are expected to colonize this type of habitat. A reduction or absence of EPT taxa suggests a water quality problem since habitat is not limited. An unusually low number of EPT taxa might also indicate the food resource base is unbalanced and favors organisms of a particular functional feeding group (e.g., collector-filterer organisms) to the exclusion of other organisms.

It might seem unnecessary to measure taxa richness and EPT richness metrics from both multihabitat and standard-habitat samples. However, there are important differences in the scale of the measurements which ensure the metrics are not redundant and contributes to a stronger biological assessment. Multi-habitat taxa richness metrics reflect water quality as well as habitat availability and suitability at the stream reach scale. Standard habitat samples are designed more to reflect water quality differences alone since the habitat sampled is standardized across sites.

When both types of samples are included, there are several possible assessment outcomes. A healthy stream with good water quality and benthic habitat diversity will ordinarily support high total numbers of taxa and EPT taxa in both the standard habitat and multi-habitat samples. Conversely, a stream with poor habitat and poor water quality will yield relatively few taxa in both types of samples. In streams where water quality is acceptable but benthic habitat is limited, taxon richness might be relatively high in the standard-habitat sample.

5. Multi-habitat Sensitive Taxa Richness (MHSTR). The number of sensitive taxa declines as stream water quality declines. For the purposes of this metric, sensitive taxa are defined as taxa that

have a biotic index tolerance value of three or less on the Hilsenhoff scale from 0 (no organic enrichment) to 10 (severe organic pollution). This group includes the most pollution-sensitive of the EPT taxa, as well as several non-EPT taxa. With increasing nutrient availability and organic enrichment, sensitive benthic macroinvertebrate taxa are replaced by more tolerant, facultative organisms.

# **Proportional Abundance Metrics**

6. Percent abundance of 3-dominant taxa (P3DOM). The proportion of the total number of organisms represented by the three most-abundant taxa is an indicator of benthic macroinvertebrate community balance. P3DOM is inversely related to stream biological integrity. Healthy warmwater streams have diverse benthic macroinvertebrate assemblages in which the majority of organisms are distributed somewhat evenly among numerous taxa. As stream conditions degrade, an increasingly higher proportion of the total number of benthic macroinvertebrate organisms is represented by just a few opportunistic taxa.

**7. Biotic Index (BINDX).** This metric is adapted from the Hilsenhoff Biotic Index, which was developed as an indicator of stream organic enrichment (Hilsenhoff 1977, 1987). The BINDX metric responds inversely to increased levels of organic waste and nutrient loading. The proportional abundance of each taxon in the sample is multiplied by its tolerance value. The products are then summed to obtain a weighted-average pollution tolerance score for the entire sample. BINDX metric values can range from 0 (no organic pollution) to 10 (severe organic pollution).

8. Percent abundance of EPT taxa (%EPT). In healthy streams, EPT taxa are usually abundant and often dominate stable rock or wood substrates that are situated in flowing water. EPT organisms tend to be replaced by tolerant organisms as water quality impacts or siltation problems become severe. Many EPT taxa are particularly sensitive to toxic contaminants such as ammonia, metals, and insecticides. Their absence or rare occurrence in standard habitat samples is strong evidence of a water quality problem. In Iowa streams, the %EPT metric seems to have a narrow range of response that is mostly observed in streams that experience acute or chronic water quality impacts. Concentrated animal feeding operations and wastewater discharges are common pollution sources.

9. Percent abundance of Chironomidae taxa (%CHR). Aquatic dipterans of the Chironomidae family (midges) are a normal component of healthy benthic macroinvertebrate communities. Several chironomid taxa are intolerant of pollution impacts. Other chironomids are very tolerant of pollution impacts, such as organic enrichment, sedimentation, and toxic metal loading. Ordinarily chironomids represent a relatively small proportion of the organisms in standard-habitat samples. Where significant water quality impacts occur, the abundance of tolerant and opportunistic chironomids often increases dramatically, while other benthic macroinvertebrates are eliminated or reduced in number. The %CHR metric has a relatively narrow range of response that is mostly concentrated in the lower end of the stream quality spectrum.

10. Percent abundance of Ephemeroptera taxa (%EPHM). Ephemeroptera (mayflies) are normally abundant and diverse in healthy Iowa streams. As a group, they are pollution-sensitive, and several taxa disappear quickly as stream disturbance increases. Mayflies compete with many other benthic macroinvertebrates for food resources and limited space on coarse substrates such as rocks or wood. They are often replaced by filter-feeding caddisflies at intermediate levels of organic enrichment.

11. Percent abundance of scraper organisms (%SCR). The proportion of organisms belonging to the scraper functional feeding group generally decreases as streams become more organically enriched. The main food sources of scraper organisms are periphyton and organic matter contained in the thin bio-film that is present on coarse substrates. As streams become more enriched, filter-feeding organisms (e.g., Diptera: Simuliidae; Trichoptera: Hydropsychidae) often become dominant in response to greater availability of fine particulate organic matter (FPOM). There is also a shift in the periphyton community in favor of filamentous algae, which is not efficiently utilized by scrapers as a food resource. Filamentous algae provides a good substrate for filter-feeder colonization and is a source of additional FPOM.

12. Percent abundance of dominant functional feeding group (%DFFG). This metric is a measure of the degree of balance among benthic macroinvertebrate functional feeding groups. As stream disturbance increases, the %DFFG also increases. It is based on the assumption that extreme dominance by one functional feeding group indicates the stream food web is unbalanced, probably due to an overabundance of a particular food item.

In healthy Iowa warmwater streams, most benthic macroinvertebrates occupying coarse substrates in flowing water belong in one of three functional feeding groups: (a) scrapers; (b) collector-filterers; (c) collector-gatherers. Other functional feeding groups, such as macrophyte (herbivore) piercers, predators, and shredders, are often present in much smaller numbers. As stream disturbance increases, one functional feeding group, typically collector-filterers or collector-gatherers, tends to dominate the benthic macroinvertebrate community and trophic diversity is reduced.

The BM-IBI has a possible scoring range from 0 to 100. The index is obtained by averaging the scores of the 12 individual metrics and then multiplying by 10. BM-IBI scores are divided into four categories; 0 to 30, poor; 31 to 55, fair; 56 to 75, good; 76 to 100, excellent.

#### METHODS

Artificial substrates were placed in-stream in early July and collected in the latter part of August of each year (1995-2000). In this manner temporal variability in community structure was constrained by reducing seasonality among years. Four substrates were placed at each location (SQW2 and



**Figure 2.** Monitoring sites in Walnut and Squaw Creek watersheds.

WNT2, Figure 2). After six weeks of colonization the samples were collected and preserved independently in the field using a 10% formalin solution. Samples remained in 10% formalin for at least two weeks and were rinsed and changed into a solution of 70% ethanol. Laboratory processing consisted of subsampling 100 organisms (when possible) from a Pyrex pan divided into 100 quadrats starting at a randomly selected square (UHL, 2001). Qualitative sampling, as opposed to the quantitative artificial substrates, consisted of sampling multiple types of substrate to include habitat not represented by quantitative sampling. Qualitative sampling was done for 60 minutes each time the substrates were collected.

Organisms were identified to the lowest practical taxonomic level. The confirmation of taxonomy work was performed by several experts outside of the University Hygienic Laboratory (UHL). Twelve data metrics were calculated and a numeric measure of the benthic macroinvertebrate community was constructed. A minimum of 90 organisms was necessary before metrics were calculated. In cases where 90 organisms were not present on one substrate, samples were pooled to reach the threshold. Final metric values were the result of the three most populous substrates. Metrics were calculated for each substrate and averaged to determine the BM-IBI.

### RESULTS

Data for each of the twelve metrics and the total BM-IBI score for a particular site and year are summarized in Table 1. BM-IBI scores for 1995 were not included because qualitative sampling was not done that year, however the nine quantitative metrics were calculated and listed.

BM-IBI scores were calculated for all years where data were available. For some years artificial substrates were aggregated to meet the numeric criteria of 90 organisms. Walnut Creek in 1999 and 2000 and Squaw Creek in 1996, 1997, and 1999 had adequate colonization on individual substrates. For all remaining years data were aggregated. Although BM-IBI scores have varied substantially over the course of the project (Figure 3), the scores have remained within the "good" classification (Table 1). The variability of BM-IBI scores was not statistically significant ( $p \le 0.05$ ). Consistent trends in BM-IBI scores have not emerged over the course of the biological monitoring project.

The level of macroinvertebrate colonization is a continuing issue at both Walnut and Squaw creek sites, thus comparison with ecoregion reference sites using the BM-IBI remains problematic. Biological reference sites are stream reaches located within the Southern Iowa Rolling Loess Prairies ecoregion that have minimally disturbed conditions. Ecoregion reference sites usually have extensive colonization of artificial or natural substrates; thus subsampling is often necessary to contend with the quantity of organisms to be identified. In eight vears of biological sampling at ecoregion reference sites, low colonization levels at Walnut and Squaw creeks have not been encountered, thus some discretion may be necessary when attaching a qualitative classification to the BM-IBI score (since classification is based on ecoregion reference site data). Additional information can be obtained by



**Figure 3.** BM-IBI scores for Walnut Creek and Squaw Creek.

reducing the data to the twelve component metrics and comparing between Walnut and Squaw creeks.

A Heptageniidae mayfly, Heptagenia diabasia, generally dominates community structure in both Walnut and Squaw creeks. This species has a low tolerance value (3 on a scale of 0 to 10) which is a measure of its ability to exist in organically enriched or disturbed streams (Hilsenhoff 1987). As a result of *H. diabasia*'s predominance coupled with its low tolerance value, four metrics (Biotic index, % EPT, % Ephemeroptera, and % Scraper) were influenced in a directly disproportional, positive manner and one (% Chironomidae) metric was influenced indirectly in a positive direction. Alternatively, two metrics (% three dominant taxa, % dominant functional feeding group) were influenced in a disproportionately negative direction. As the data in Table 1 show, if the % Ephemeroptera metric is large enough to generate a metric score of "10" then the four other metrics that were previously mentioned as responding positively nearly always have values of "9" or "10". The high degree of apparent correlation between these metrics strongly influences the total BM-IBI score. This has not been a problem in other biological assessments; thus it appears to be an atypical confluence of community characteristics at Walnut and Squaw creek watersheds that result in the inflation of total **BM-IBI** scores.

The metrics associated with qualitative sampling were not constrained by the colonization

issues that plagued the artificial substrates. The qualitative samples were collected from multiple habitats and consequently serve as crude surrogates for in-stream habitat. Walnut Creek has generally exhibited movement toward higher metric scores for two out of three metrics (MH-taxa richness, MH-EPT taxa), while Squaw Creek has remained largely stable over the course of the monitoring project (Table 1). Coarse substrate added at the WNT2 site in 1995 likely contributed to the increase in EPT and total taxa collected since many EPT taxa have an affinity for coarse substrate (e.g., riffle) environments. Comparable changes in habitat structure at Squaw Creek have not occurred and may account for its stable metric values. Coarse substrate is not present within the assessed reach of Squaw Creek.

# **CONCLUSIONS**

BM-IBI scores for Walnut and Squaw creeks have shown a large amount of variability over the course of the biological monitoring project. Differences between creeks have not been sufficient to indicate any statistically significant changes either within or between streams. Relying upon current classification criteria for the BM-IBI both sites are rated "good". However, the quantitative component of the BM-IBI may need some modification to appropriately evaluate sites that have chronic colonization issues such as Walnut and Squaw creeks. The qualitative component of the BM-IBI suggests habitat variability may be a factor among the sites being assessed. It appears that Walnut Creek has shown some positive movement of two qualitative metrics, although this may be a result of habitat modification work that was done in 1995.

Additional work is needed to address the dynamics of nutrients, habitat, and resident macroinvertebrate community density to more clearly understand how, and if, changes may be expected to occur as a result of terrestrial revegetation that is occurring within the Walnut Creek watershed.

Metrics	WNT 95	Score	WNT 96	Score	WNT 97	Score	WNT 98	Score	WNT 99	Score	WNT 00	Score	SQW 95	Score	SQW 96	Score	SQW 97	Score	SQW 98	Score	SQW 99	Score	SQW 00	Score
1. MHTR		0	19	5	21	5	23	6	27	7	22	5	1	0	11	3	13	3	16	4	13	3	15	4
2. SHTR	8.3	5	13	8	10	6	13	8	13.7	9	12.7	8	7.6	5	7.3	5	9.7	6	7	5	13	8	8	5
3. MH-EPT		0	6	3	7	4	13	7	13	7	11	6	1	0	4	2	7	4	5	3	7	4	8	4
4. SH-EPT	6.7	6	10	9	7	7	5	5	10	9	9	8	6	6	5.3	5	7	7	5	5	9	9	5	5
5. MH-STR		0	1	1	2	2	1	1	1	1	1	1	1	0	2	2	2	2	3	4	2	2	3	4
6. % 3Dom	73.9	6	77.6	5	90.6	2	81	4	68.8	7	36.6	10	92.5	2	94.8	1	54.9	10	89.5	2	64	8	88.9	2
7. BINDX	3.61	10	4.1	10	6.49	2	4.39	10	5.26	6	4.72	8	3.96	10	3.31	10	4.3	10	3.7	10	5.3	6	3.88	10
8. % EPT	97.6	10	80.7	8	93.6	10	69.5	7	90.4	9	78.4	8	90.2	9	97.1	10	92.2	10	88.7	9	88	9	90.8	10
9. % CHR	0.7	10	13.4	9	3.5	10	21.5	8	4.3	10	10	9	4.6	10	1.3	10	5.9	10	10.5	9	8.6	9	3.7	10
10. % EPHM	89.2	10	74.2	9	93.6	10	69.5	9	42.9	5	42.3	5	89.1	10	95.4	10	85.6	10	86	10	75	10	90.8	10
11. % Scraper	80.2	10	64.2	10	90.6	10	53.7	10	19	4	32.9	7	88	10	90.5	10	56.1	10	80.6	10	56	10	84.3	10
12. % Dom FFG	80.2	3	64.2	6	90.6	2	53.7	8	48.9	9	36	10	88	2	90.5	2	56.1	7	80.6	3	56	7	84.3	3
Annual BM-IBI				70		58		69		70		73				58		74		61		72		63

**Table 1.** BM-IBI metric results – individual and total metric scores (raw scores and adjusted scores, 0-10). Metric descriptions provided in text.

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## SUMMARY OF FISH COMMUNITIES IN WALNUT AND SQUAW CREEK WATERSHEDS: 1995-2000

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### **INTRODUCTION**

Fish community data collection for the Walnut Creek Watershed Restoration and Water Quality Monitoring Project began in 1995 to evaluate changes in biological health that occur during the large-scale restoration of native prairie/savanna in the Walnut Creek watershed by the Neal Smith National Wildlife Refuge in Jasper County, Iowa. The biological monitoring relies upon a paired watershed approach to provide comparative data (USEPA 1993). The Squaw Creek watershed is the control watershed where land use is primarily agricultural.

Walnut and Squaw Creek watersheds are located within the Southern Iowa Rolling Loess Prairies Ecoregion (Ecoregion 47f; Figure 1). Ecoregion 47f is characterized by irregular plains to open low hills with moderate to thick loess (Griffith et. al. 1994). Land use in the ecoregion is a mixture of cropland, grassland, and woodland. Historical land use for Walnut and Squaw Creek watersheds was similar prior to the restoration work occurring in Walnut Creek watershed and averaged approximately 70% row crop and 27% grassland in 1992 (See Background and Land Use Changes, this issue). Walnut Creek watershed is approximately 20.1 mi² and Walnut Creek flows into Lake Red Rock as a 3rd order stream. Squaw Creek watershed is approximately 18.3 mi² and Squaw Creek





**Figure 2.** Monitoring sites in Walnut and Squaw Creek watersheds.

enters its confluence with the South Skunk River as a 4th order stream.

The characteristics of the fish community can be used to evaluate and compare the biological health of Walnut and Squaw creeks. The Iowa Department of Natural Resources (IDNR) has constructed an Index of Biotic Integrity (IBI) for wadeable Iowa streams (Tom Wilton, IDNR, personal communication). The IBI is a multimetric biological indicator that uses 11 data metrics (Table 1), such as number of sensitive species and proportion of individuals as omnivores, to describe the fish community. IBIs calculated for Walnut and Squaw creeks can be compared to IBIs from stream ecoregion reference sites. This report summarizes the fish communities sampled in the Walnut and Squaw creek watersheds from 1995 to 2000.

#### **METHODS**

Electrofishing was conducted during mid summer with a single backpack electroshocker (Coffelt Manufacturing, Inc.). In 1995, only one site at the bottom of each watershed (near the outlet) was sampled (sites WNT2 and SQW2, Figure 2). In 1996, mid-watershed sites (WNTBM2 and SQWBM2) were added to provide a more heterogeneous sampling environment and to increase the distance each creek was sampled. To compare the fish communities in the two watersheds, the annual fish data from the two sites on each creek were combined. Combining the fish community data from the individual sites provided a better overall picture of stream health. This was possible for all years except 1995, when fish were collected from only one site on each creek.

A segment of each stream at least 35 times the average width was sampled at all sites (Lyons 1992). This distance was used to ensure sampling was performed on all major habitats present and several riffle/pool series. Fish were identified in the field; difficult specimens were preserved in 10% formalin and returned to the laboratory for identification. This method was in accordance with the sampling protocol recommended in the EPA Rapid Bioassessment Protocol V (Plafkin et al. 1989).

The IBI for wadeable streams consists of 11 data metrics (Table 1). When calculating the IBI score for a stream, each metric result receives a score from 0 to 10, based on a comparison to the population of reference stream data gathered by the IDNR. The IBI score is the sum of the 11 data metric scores, adjusted to range from 0 to 100. The greater the IBI score, the "healthier" the stream. As the IBI score increases, the stream biological health rating also increases.

The total IBI score can receive a penalty (adjustment) if less than 100 fish per 500 feet of stream are collected. If less than 25 fish are collected, proportional metrics (e.g. proportion of omnivores) receive a score of 0. If 26 to 50 fish are collected, proportional metrics receive a maximum score of 2.5. If 51 to 75 fish are collected, proportional metrics receive a maximum score of 5. If 76 to 100 fish are collected, proportional metrics

Table 1. D	Data Metrics	Used in the	Calculation	of an Index	of Biotic	Integrity (IBI)	).
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Metric	Explanation/Desired Condition
Native Fish Species Richness	More species implies greater habitat complexity and favorable stream conditions.
Proportion of Simple Lithophilic Spawners	Simple lithophilic spawners are sensitive to sedimentation, and require clean gravel/cobble substrate for reproduction. Therefore, a greater percentage of lithophilic spawners implies better benthic conditions.
Proportion of Fish as Omnivores	Omnivores consume a variety of plant and animal material, and therefore are less sensitive to environmental degradation that causes changes in the food base. A higher percentage of omnivores implies unfavorable environmental conditions.
Proportion of Benthic Invertivores	Benthic invertivores are often specialists, and therefore are sensitive to environmental degradation that causes a change in the food base. A higher percentage implies favorable stream conditions.
Proportion of Three Dominant Species	A higher percentage implies unfavorable stream conditions.
Proportion of Top Carnivores	Top carnivores are generally longer lived fish that need stable environmental conditions and food base. A higher percentage implies favorable stream conditions.
Number of Sucker Species	Suckers are generally longer-lived species that are intolerant of degraded conditions. A greater number implies favorable stream conditions.
Number Benthic Invertivore Species	Benthic invertivores are often specialists, and therefore are sensitive to environmental degradation that causes a change in the food base. A greater number of benthic invertivore species implies favorable stream conditions.
Number of Sensitive Species	A greater number of species sensitive to degraded environmental implies favorable water quality and habitat conditions.
Fish Assemblage Tolerance Index	Each fish species receives a tolerance value, the index is based on the sum of tolerance values for a site. A higher index value implies unfavorable environmental conditions.
Adjusted Catch per Unit Effort	A greater density of fish implies more favorable stream conditions. Tolerant fish species are not included in this metric

Species	<u>1995</u> ¹	<u>1996</u>	<u>1997</u>	<u>1998</u>	<u>1999</u>	<u>2000</u>
Catostomidae: Suckers						
Redhorse sp. Moxostoma sp.	-	-	-	-	1 (0.4)	-
Shorthead redhorse Moxostoma macrolepidotum	-	-	-	1 (0.5)	-	-
Bigmouth buffalo Ictiobus cyprinellus	-	6 (3.5)	-	1 (0.5)	-	-
carpsucker (juvenile) <i>Carpiodes sp.</i>	1 (0.6)	-	-	-	-	-
Northern hog sucker <i>Hypentelium nigricans</i>	-	-	-	1 (0.5)	-	-
River carpsucker Carpiodes carpio	+ (2.5)	1(06)	$\frac{9}{0.0}$	-	-	- 1 (0 5)
Smallmouth buffalo <i>Ictiobus bubalus</i>	-	-	1(0.7) 1(0.7)	-	2(0.8)	-
White sucker Catostomus commersoni	7 (4.1)	1 (1.8)	7 (4.7)		1 (0.4)	6 (2.9)
Centrarchidae: Sunfishes						
Bluegill Lepomis macrochirus	-	1 (0.6)	1 (0.7)	67 (32.5)	10 (4.0)	1 (0.5)
Green sunfish Lepomis cyanellus ³	18 (10.5)	19 (11.1)	2 (1.3)	7 (3.4)	6 (2.4)	4 (1.9)
Largemouth bass Micropterus salmoides	7 (4.1)	10 (5.8)	1 (0.7)	24 (11.7)	4 (1.6)	3 (1.5)
Clupeidae: Herrings						
Gizzard shad Dorosoma cepedianum ³	-	3 (1.8)	5 (3.4)	49 (23.8)	160 (63.7)	3 (1.5)
Cyprinidae: Minnows						
Bigmouth shiner Notropis dorsalis ³	-	7 (4.1)	30 (20.1)	1 (0.5)	6 (2.4)	37 (18.0)
Bluntnose minnow Pimephales notatus'	84 (48.8)	25 (14.6)	8 (5.4)	16 (7.8)	12 (4.8)	12 (5.8)
Central stoneroller <i>Campostoma anomalum</i>	-	36 (21.1)	11 (7.4)	2 (1.0)	6 (2.4)	46 (22.3)
Common carp Cyprinus carpio ⁵	- 10 (5 8)	1(0.6)	2(1.3)	-	2(0.8)	-
Eathead minnow Pimenhales promelas ³	10(3.8)	27 (13.8)	01 (40.9)	15 (7.5)	20 (11.2)	1(0.5)
Golden shiner Notemigonus crysoleucas ³	-	1(0.6)	-	$\frac{-1}{(0.5)}$	$\frac{-}{3(1.2)}$	-
Red shiner Cyprinella lutrensis ³	33 (19.2)	15 (8.8)	3 (2.0)	19 (9.2)	1(0.4)	3 (1.5)
Sand shiner Notropis stramineus	-	8 (4.7)	5 (3.4)	-	-	3 (1.5)
Spotfin shiner Notropis spiloptera	-	1 (0.6)	-	1 (0.5)	-	2 (1.0)
Suckermouth minnow Phenacobius mirabilis	-	4 (2.3)	-	3 (1.5)	7 (2.8)	22 (10.7)
Ictaluridae: Catfishes						
Black bullhead <i>Ameiurus melas</i> ³	7 (4.1)	-	2 (1.3)	-	-	-
Yellow bullhead Ameiurus natalis	-	-	-	-	-	1 (0.5)
Percichthyidae: Temperate Bass						
White bass Morone chrysops	-	-	-	-	1 (0.4)	-
Percidae: Perches						
Slenderhead darter <i>Percina phoxociphala²</i> Walleye <i>Stizostedion vitreum</i>	-	- 1 (0.6)	-	-	1 (0.4)	-
Sciaenidae: Drums						
Freshwater drum Aplodinotus grunniens	1 (0.6)	-	-	-	-	1 (0.6)
<ol> <li>¹ Includes data from the WNT2 site only.</li> <li>²Sensitive to degraded environmental conditions.</li> <li>³Tolerant to degraded environmental conditions.</li> </ol>						

Number of Fish Collected (Relative Frequency, %) from Walnut Creek: 1995-2000.

receive a maximum score of 7.5.

The total IBI score can also receive a penalty (adjustment) if the proportion of deformities, eroded fins, lesions, or tumors (DELTs) exceeds 2%. If the proportion of DELTs exceeds 2% but is less than 4%, 5 points are subtracted from the IBI score. If the proportion of DELTs exceeds 4%, 10 points are subtracted from the IBI score.

The IDNR IBI is calculated in a way to avoid over-penalizing a stream that has a high percentage of DELTs and less than 100 fish. If less than 100 fish are collected and the proportion of DELTs is greater than 4%, 5 points are subtracted from the IBI score. If less than 100 fish are collected and the proportion of DELTs is between 2% and 4%, 2.5 points are subtracted from the IBI score.

## **RESULTS AND DISCUSSION**

Since 1995, 28 species of fish from eight families were collected from Walnut Creek (Table 2). The fish community at Walnut Creek is usually dominated by minnows (Cyprinidae) and sunfishes (Centrarchidae) with some exceptions. In 1998 and 1999, gizzard shad *Dorosoma cepedianum* comprised a large proportion of the Walnut Creek fish population (24% and 64% respectively; Clupeidae). Gizzard shad are considered tolerant of degraded environmental conditions (Tom Wilton, IDNR, personal communication).

During all years, species tolerant of degraded environmental conditions made up a large proportion of the Walnut Creek fish community. Sensitive species, such as the northern hog sucker (*Hypentelium nigricans*) or the slenderhead darter (*Percina phoxocephala*), were rarely found. Seven species of suckers (Catostomidae) have been collected from Walnut Creek, although they usually are collected in small numbers. Suckers generally indicate favorable stream conditions because they are long lived and many sucker species are habitat specialists.

Twenty species of fish from five families have been collected from Squaw Creek since 1995 (Table 3). Similar to Walnut Creek, minnows usually comprise the majority of the population and the majority of the species collected are tolerant of degraded environmental conditions. One sensitive species, northern hog sucker, has been found in Squaw Creek. Three species of suckers (Catostomidae), usually in low numbers, have been collected from Squaw Creek since 1995.

An IBI score was calculated for all sampling events (Tables 4 and 5). The IBIs ranged from 15 at Walnut Creek in 1995 to 40 at Walnut Creek sites in 1996. The low score for Walnut Creek in 1995 is likely because only one site, WNT2, was sampled that year. Walnut Creek would have had its highest IBI score (43) in 2000 if it had not received a tenpoint penalty for lesions found on approximately 5% of the fish. A Wilcoxon signed rank test was used to compare the IBI scores for both watersheds; no significance difference was found between Walnut Creek and Squaw Creek IBI scores (p = 0.84). IBI scores for Walnut or Squaw Creek did not show any noticeable improvement or decline since 1995.

The IDNR divides IBI scores into quartiles and designates a score of 0 to 25 as poor, 26 to 50 as fair, 51 to 75 as good, and 76 to 100 as excellent. In streams that are classified as poor, lower than average numbers of fish are present and the species found are usually short-lived or pioneering species that are tolerant of degraded stream conditions. A higher proportion of fish with deformities, eroded fins, lesions, and tumors is often found. Few species tolerant of degraded environmental conditions are present. In streams that are classified as fair, the fish community is usually dominated by species tolerant of degraded environmental conditions. Sucker species, sensitive species, top carnivores, and habitat specialists are often present, but in low numbers, and omnivores are usually more dominant. Most IBIs calculated for Walnut Creek and Squaw Creek were in the fair category. Walnut Creek received a poor IBI score in 1995 and Squaw Creek received a poor IBI score in 2000.

The IDNR has calculated IBI scores for more than 200 wadeable streams in Iowa (Tom Wilton, IDNR, personal communication). Ten reference stream sites in Ecoregion 47f are comparable to Walnut Creek and Squaw Creek. These are sites in the Mississippi River drainage with minimally disturbed stream habitat that lack stable and abundant

Species	<u>1995</u> ¹	<u>1996</u>	<u>1997</u>	<u>1998</u>	<u>1999</u>	<u>2000</u>
Catostomidae: Suckers						
Bigmouth buffalo <i>Ictiobus cyprinellus</i> Northern hog sucker <i>Hypentelium nigricans</i> ² White sucker <i>Catostomus commersoni</i>	1 (0.2) 15 (2.9)	- -	6 (2.3)	2(1.1) 1(0.6) 2(1.1)	- 7 (5.3)	- - 1 (0.4)
Centrarchidae: Sunfishes					· · ·	
Bluegill <i>Lepomis macrochirus</i> Green sunfish <i>Lepomis cyanellus</i> ³ Largemouth bass <i>Micropterus salmoides</i>	1 (0.2) 4 (0.8) 2 (0.4)	- 1 (0.7) 1 (0.7)	- -	7 (3.9	2 (1.5) 7 (5.3)	1 (0.4) 1 (0.4)
Clupeidae: Herrings						
Gizzard shad Dorosoma cepedianum ³	2 (0.4)	-	-	1 (0.6)	-	-
Cyprinidae: Minnows						
Bigmouth shiner Notropis dorsalis ³ Bluntnose minnow Pimephales notatus ³ Brassy minnow Hybognathus hankinsoni Central stoneroller Campostoma anomalum Common carp Cyprinus carpio ³ Common shiner Luxilus cornutus Creek chub Semotilus atromaculatus ³ Fathead minnow Pimephales promelas ³ Red shiner Cyprinella lutrensis ³ Sand shiner Notropis stramineus Suckermouth minnow Phenacobius mirabilis	54 (10.5) 191 (37.2) 11 (2.1) 27 (5.3) 2 (0.4) 1 (0.2) 88 (17.1) 21 (4.1) 29 (5.6) 48(9.3)	23 (16.7) 15 (10.9) 8 (5.8) 10 (7.2) 4 (2.9) - 47 (34.1) - 5 (3.6) 16(11.6) -	40 (15.2) 38 (14.4) 9 (3.4) 34 (12.9) 2 (0.8) - 50 (19.0) 6 (2.3 3 (1.1) 36(13.7) 2 (0.8)	- 33 (18.2) 20 (11.1) 19 (10.5) 27 (14.9 2 (1.1) 31 (17.1) 5 (2.8 23 (12.7) 7 (3.9) 1(0.6)	2 (1.5) 16 (12.0) 22 (16.5) 12 (9.0) 6 (4.5) - 43 (32.3) 7 (5.3 1 (0.8) 5 (3.8	1 (0.4) 87 (35.2) 15 (6.1) 43 (17.4) - 3 (1.2) 60 (24.3) - 17 (6.9) - -
Percidae: Perches						
Johnny darter Etheostoma nigrum	17(3.3)	8(5.8)	37(14.1)	2(1.1)	3(2.3)	5(2.0)

Number of Fish Collected (Relative Frequency, %) from Squaw Creek: 1995-2000

¹ Includes data from the WNT2 site only.

²Sensitive to degraded environmental conditions.

³Tolerant to degraded environmental conditions.

amounts of riffles and coarse substrate. Walnut and Squaw Creek IBI scores within the 95% confidence interval (CI) for this mean reference site IBI (Table 6) are statistically similar to the mean reference site IBI. (Because of the small number of reference sites (N=10), the 95% CIs are fairly large.) Four of the six IBI scores for Walnut Creek (1996, 1997, 1998, and 2000) were within the 95% CI for the mean reference site IBI. Two of the six Squaw Creek IBI scores (1997, 1998) were within than the 95% CI. None of the IBIs measured in Walnut or Squaw Creeks exceeded the mean IBI score from similar reference sites.

The mean and 95% CI were also calculated for the individual metrics for the ten reference streams in Ecoregion 47f similar to Walnut Creek (Table 6). In 1995, ten of the eleven metric scores from Walnut Creek were less than the 95% CI for the mean reference site metric scores. In 1995, only one site was sampled on Walnut Creek; scores were probably low because a shorter stream segment with less habitat variety was sampled. Midwatershed sites were added in 1996 to increase the

Table 4.	IBI s	ummary	for	Wa	lnut	Creek.
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Data Metric Results (IBI score), Unadjusted IBI Score, and Adjusted IBI Score for Walnut Creek 1995- 2000.							
Metric	<u>1995</u>	<u>1996</u>	<u>1997</u>	<u>1998</u>	<u>1999</u>	2000	
Native Fish Species Richness	8 (3.7)	17 (7.7)	12 (5.5)	13 (5.9)	14 (6.4)	15 (6.9)	
Number of Sucker Species	2 (4.7)	3 (7.0)	4 (9.4)	3 (7.0)	3 (7.0)	2 (4.7)	
Number of Sensitive Species	0 (0)	0 (0)	0 (0)	1 (1.3)	1 (1.4)	0 (0)	
Number of Benthic Invertivore Species	1 (1.2)	1 (1.3)	1 (1.3)	3 (3.8)	3 (3.8)	3 (3.8)	
Proportion of Three Dominant Species (%)	78.5 (4.2)	51.4 (9.5)	72.3 (5.4)	67.3 (6.4)	79.7 (4.0)	68.9 (6.1)	
Proportion of Fish as Benthic Invertivores (%)	0.6 (0.2)	2.3 (0.7)	3.5 (1.1)	2.4 (0.8)	3.6 (1.1)	12.1 (3.9)	
Proportion of Fish as Omnivores (%)	75 (0.7)	29.2 (7.3)	16.3 (9.1)	40.9 (5.6)	72.1 (1.1)	12.6 (9.6)	
Proportion of Fish as Top Carnivores (%)	0 (0)	0.6 (3.6)	0 (0)	0 (0)	0.4 (3.0)	0 (0)	
Proportion of Fish as Simple Lithophilus Spawners (%)	0 (0)	2.3 (1.5)	0 (0)	2.4 (1.5)	2.8 (1.8)	10.7 (6.8)	
Fish Assemblage Tolerance Index	9.4 (0.9)	7.9 (3.3)	8.5 (2.3)	7.6 (3.9)	9.3 (1.1)	7.9 (3.4)	
Adjusted CPUE	10(1)	15.8 (1.6)	9.1 (0.9)	22.2 (2.2)	7.3 (0.7)	19.3 (1.9)	
Unadjusted IBI Score:	15	40	32	35	29	43	
Proportion (%) of Fish with DELTs (Adjustment)	0 (0)	0 (0)	0 (0)	0.5 (0)	0.8 (0)	4.9 (-10)	
Low Numbers of Fish Adjustment	No	No	No	No	No	No	
Adjusted IBI Score:	15	40	32	35	29	33	
DNR Rating*	Poor	Fair	Fair	Fair	Fair	Fair	

*0-25 = poor, 26-50 = fair, 51-75=good, 76-100=excellent

length of stream and types of habitat sampled.

From 1996 to 2000, metric scores for Walnut Creek compared favorably to the 95% CI for the mean reference site metric scores. Seventy-one percent of all the metric scores for Walnut Creek were within the 95% CI and 15% of all the metric scores at Walnut Creek were greater than the 95% CI. Fewer than 30% of the metric scores were less than the 95% CI. Examination of the metric scores did not reveal any trend of improvement or degradation.

Individually, several metric scores calculated for Walnut Creek also compared favorably to the 95% CI for the mean reference site metric scores. The native fish species richness metric was within the 95% CI during all years except 1995. The number of sucker species metric was greater than or within the CI during all years. The proportion of three dominant species metric was greater than or within the CI four out of the six years. The proportion of fish as omnivores metric scores were

within or greater than the CI in all years except 1995. Scores for these metrics imply habitat complexity and stream conditions comparable to similar streams in Ecoregion 47f (Table 1).

Ignoring the 1995 results, no metrics for Walnut Creek scored consistently less than the 95% CI for the mean reference site metric score. Scores for two metrics, the number of sensitive species and the proportion of top carnivores, were less than the 95% CI three of the last five years. Low scores for these metrics imply less favorable environmental conditions and habitat than similar streams in Ecoregion 47f (Table 1). The remaining metrics were less than the CI during one or two of the last five years.

Overall, metric scores for Squaw Creek did not compare to the mean reference site metric scores as favorably as Walnut Creek. Since 1996, 53% of the metric scores were within or greater than the 95% CI for the mean reference site metric scores. Eleven percent of the scores were greater than the

Table 5. IBI summary for Squaw Creek.

Metric	1995	<u>1996</u>	<u>1997</u>	1998	1999	2000
Native Fish Species Richness	14 (6.8)	9 (4.4)	11 (5.3)	15 (7.3)	11 (5.3)	10 (4.8)
Number of Sucker Species	2 (5.0)	0 (0)	1 (2.5)	3 (7.4)	1 (2.5)	1 (2.5)
Number of Sensitive Species	0 (0)	0 (0)	0 (0)	2 (2.8)	0 (0)	0 (0)
Number of Benthic Invertivore Species	1 (1.3)	1 (1.3)	2 (2.7)	3 (4.0)	1 (1.3)	1 (1.3)
Proportion of Three Dominant Species (%)	64.8 (7.3)	62.3 (7.8)	48.7 (10)	49.7 (10.0)	60.9 (8.1)	76.9 (4.8)
Proportion of Fish as Benthic Invertivores (%)	3.3 (1.1)	5.8 (2.0)	14.9 (5.0)	2.2 (0.7)	2.3 (0.8)	2.0 (0.7)
Proportion of Fish as Omnivores (%)	50.6 (4.3)	17.4 (9.2)	18.6 (9.1)	60.1 (2.9)	27.8 (7.7)	47.8(4.8)
Proportion of Fish as Top Carnivores (%)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Proportion of Fish as Simple Lithophilus Spawners (%)	0 (0)	0 (0)	0.8 (0.5)	1.1 (0.7)	0 (0)	0 (0)
Fish Assemblage Tolerance Index	8.8 (1.9)	8.4 (2.5)	7.5 (3.9)	7.9 (3.3)	8.1 (3.0)	8.4 (2.6)
Adjusted CPUE	44.7 (4.5)	10.1 (1.0)	30.6 (3.1)	13.2 (1.3)	12 (1.2)	19.1 (1.9)
Unadjusted IBI Score:	29	26	38	37	27	21
Proportion (%) of Fish with DELTs (Adjustment)	0 (0)	0 (0)	0 (0)	2.7 (-5)	0 (0)	0 (0)
Low Numbers of Fish Adjustment	No	No	No	No	No	No
Adjusted IBI Score:	29	26	38	32	27	21
DNR Rating	Fair	Fair	Fair	Fair	Fair	Poor

Data Metric Results (IBI score), Unadjusted IBI Score, and Adjusted IBI Score for Squaw Creek 1995- 2000.

*0-25 = poor, 26-50 = fair, 51-75=good, 76-100=excellent

95% CI while 47% of the metric scores were less than the 95% CI.

Individually, some Squaw Creek metric scores compared well to the reference site mean metric scores. The proportion of three dominant species metric score was within or greater than the 95% CI during all years. The native fish species metric and number of sucker species metric scores were within the 95% CI in all years except one (2000 and 1996, respectively). The proportion of omnivores and the adjusted CPUE metric scores were greater than or within the 95% CI during four of the six years.

Three metrics calculated for Squaw Creek did not compare well with the mean reference site metric scores. The proportion of fish as top carnivores was less than the 95% confidence interval during all years. The number of benthic invertivores and number of sensitive species metrics were less than the 95% confidence interval in all years except 1998. The proportion of fish as simple lithophilus spawners metric scores were less than the 95% CI in all years except 1997 and 1998. The remaining two metrics compared well during some years and not during others.

### CONCLUSIONS

Twenty-eight species of fish from eight families were collected from Walnut Creek and 25 species of fish from five families were collected from the Squaw Creek. Both fish communities were usually dominated by minnows (Cyprinidae) and species considered tolerant of degraded environmental conditions. IBIs calculated for most years received a reference site rating of fair compared to other warmwater, wadable streams in Iowa. When comparing IBIs for all years, no significant difference was found between the two watersheds (Wilcoxon rank-sum, p = 0.84). During four of the last six years, (1996, 1997, 1998, and 2000), the Walnut Creek IBI was similar (within the 95% CI) to the mean reference site IBI scores from comparable reference streams. The Squaw Creek IBI **Table 6.** Comparison of Walnut Creek mean IBIscore with Ecoregion 47F reference sites.

Mean Score (0 to 10 possible) and 95% Confidence Interval (CI) for the Index of Biotic Integrity (IBI) and 11 Metrics Scores for Ten Reference Streams in Ecoregion 47f.

Metric	Mean Score	<u>95% CI</u>
Native Fish Species Richness	6.7	5.2 - 8.2
Number of Sucker Species	4.6	2.3 - 6.9
Number of Sensitive Species	1.8	0.8 - 2.8
Number of Benthic Invertivore Species	4.8	3.5 - 6.1
Proportion of Three Dominant Species (%)	6.3	4.8 - 7.8
Proportion of Fish as Benthic Invertivores (%)	2.5	1.0 - 4.0
Proportion of Fish as Omnivores (%)	6.9	4.8 - 9.0
Proportion of Fish as Top Carnivores (%)	3.4	1.1 - 5.7
Proportion of Fish as Simple Lithophilus Spawners (%)	1.9	0.4 - 3.4
Fish Assemblage Tolerance Index	3.9	2.7 - 5.1
Adjusted CPUE	3.7	1.3 - 6.1
Overall IBI score	42.1	31.4 - 52.9

was similar to the reference site IBIs during two of the last six years (1997 and 1998). IBI scores since 1995 showed no noticeable improvement or degradation for Walnut Creek or Squaw Creek. Examination of individual metric scores did not reveal any particular pattern of improvement or decline. Walnut Creek had more (71%) individual metric scores within or greater than the 95% CI of the mean reference site metric scores than Squaw Creek (53%). During most years, the Walnut Creek fish community was comparable in most respects to other reference stream sites in Ecoregion 47f.

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