JOURNAL OF THE AMERICAN WATER RESOURCES ASSOCIATION
DECEMBER AMERICAN WATER RESOURCES ASSOCIATION 2003

# ECOSYSTEM DYNAMICS AND POLLUTION EFFECTS IN AN OZARK CAVE STREAM[1]

*Gary O. Graening and Arthur V. Brown*[2]

ABSTRACT: Subterranean ecosystems harbor globally rare fauna and important water resources, but ecological processes are poorly understood and are threatened by anthropogenic stresses. Ecosystem analyses were conducted from 1997 to 2000 in Cave Springs Cave, Arkansas, situated in a region of intensive land use, to determine the degree of habitat degradation and viability of endangered fauna. Organic matter budgeting quantified energy flux and documented the dominant input as dissolved organic matter and not gray bat guano (*Myotis grisescens*). Carbon/nitrogen stable isotope analyses described a trophic web of Ozark cavefish (*Amblyopsis rosae*) that primarily consumed cave isopods (*Caecidotea stiladactyla*), which in turn appeared to consume benthic matter originating from a complex mixture of soil, leaf litter, and anthropogenic wastes. Septic leachate, sewage sludge, and cow manure were suspected to augment the food web and were implicated in environmental degradation. Water, sediment, and animal tissue analyses detected excess nutrients, fecal bacteria, and toxic concentrations of metals. Community assemblage may have been altered: sensitive species – grotto salamanders (*Typhlotriton spelaeus*) and stygobromid amphipods – were not detected, while more resilient isopods flourished. Reduction of septic and agricultural waste inputs may be necessary to restore ecosystem dynamics in this cave ecosystem to its former undisturbed condition.
(KEY TERMS: aquatic ecosystems; endangered species; karst; nonpoint source pollution; stable isotopes; subterranean ecosystems; water quality.)

Graening, Gary O. and Arthur V. Brown, 2003. Ecosystem Dynamics and Pollution Effects in an Ozark Cave Stream. Journal of the American Water Resources Association (JAWRA) 39(6):1497-1507.

## INTRODUCTION

Subterranean habitats harbor 50 percent of North America's imperiled species (Culver *et al.,* 2000) and contain 25 percent of the world's freshwater resources (Ford *et al.,* 1988), yet are among the most poorly understood ecosystems. Little is known about the composition and distribution of ground water communities (Gounot, 1994; Strayer, 1994), their food webs (Culver, 1994), or fluxes through the ground water/surface water ecotone (Gibert *et al.,* 1994). This lack of knowledge is exacerbated by their inaccessibility: only 10 percent of cave complexes are accessible from the earth's surface (Curl, 1958), and human sized conduits (caves) represent only a fraction of subterranean drainage systems (White *et al.,* 1995). If ground water resources such as those of the Ozark Plateaus ecoregion are to be managed properly, these deficiencies must be addressed.

### *Organic Flux and Cave Community Dynamics*

Like many first-order streams, subterranean rivers are oligotrophic and rely almost entirely upon allochthonous sources of energy (Fisher and Likens, 1973; Culver, 1982). But unlike surface streams, photosynthetic production is absent, leaf and woody debris inputs are greatly reduced, and energy is so limited in subterranean habitats that energy may fundamentally structure community composition, trophic dynamics, and even evolutionary processes. Other trophic inputs, especially animal feces and cadavers, add complexity and diversity to cave food webs (Harris, 1970). Bat feces (guano) may supply sufficient food to relax the selective pressure of oligotrophy, with resulting changes in community structure, including the presence of species without

[1]Paper No. 03002 of the *Journal of the American Water Resources Association.* **Discussions are open until June 1, 2004.**
[2]Respectively, Ecologist, The Nature Conservancy, 601 North University Avenue, Little Rock, Arkansas 72205; and Associate Professor, Department of Biological Sciences, University of Arkansas, 601 Science-Engineering, Fayetteville, Arkansas 72701 (E-Mail/Graening: ggraening@tnc.org).

troglomorphic (cave adapted) characteristics (Culver, 1982). Bat guano is thought to be the trophic base of cave food webs (Poulson, 1972; Willis and Brown, 1985), but few studies have tested this hypothesis. The regional decline of many bat populations (Harvey, 1996) and the resulting loss of guano input may fundamentally alter cave food webs. Anthropogenic inputs, especially land application of human sewage and confined animal feeding operation wastes, are increasing and hypothetically augment cave stream food webs (Stewart, 1984; Sket, 1999) but also impoverish species richness (Wood *et al.,* 2002). This study investigated an Ozark cave stream ecosystem containing both guano and anthropogenic inputs.

### *Organic Pollution and Ozark Ground Water Ecosystems*

The Ozark Plateaus consist of fractured and dissolved carbonate bedrock (karst) that store significant quantities of ground water. These aquifers are a major water resource for northern Arkansas and are highly susceptible to organic pollution from land application of animal wastes and other waste disposal practices because karst landscapes allow rapid infiltration of surface pollutants to ground water (Smith and Steele, 1990; Pasquarell and Boyer, 1996). With ample annual rainfall (more than 1 m accumulation), extensive use of septic systems, and intensive agricultural animal production (e.g., 1 billion poultry produced per year in Arkansas), northern Arkansas' ground water is chronically contaminated, particularly with excess nutrients and coliform bacteria (Steele, 1985; Smith and Steele, 1990; National Agricultural Statistics Service, 2000).

Organic pollution alters the oligotrophic status of ground water ecosystems and impacts community composition and diversity (Sinton, 1984; Notenboom *et al.,* 1994; Simon and Buikema, 1997; Wood *et al.,* 2002). Such organic inputs may increase the food base and attract surface dwelling (epigean) species, which may invade these eutrophied habitats and prey upon or compete with cave limited (troglobitic) and ground water limited (stygobitic) fauna (Poulson, 1976; Brown *et al.,* 1994).

### *Objectives*

The loading of oligotrophic caves ecosystems with nutrient rich, anthropogenic wastes constitutes an experimental treatment (although inadvertent and uncontrolled) that deserves scientific observation. Certain cave ecosystems such as Cave Springs Cave are of special concern because they harbor many globally rare species and are situated in areas of intensive land use practices that may impact cave habitat quality and community assemblage. The purpose of this study was to describe ecosystem dynamics in an Ozark cave stream and specifically to determine the relative contribution of historic, and hypothetically dominant, organic matter inputs (i.e., guano) to novel organic matter inputs (animal and human wastes) and to determine any effects upon the cave community. The objectives of this investigation were to: determine environmental quality using water, sediment, and tissue analyses; describe energy flux using organic matter budgeting; and describe trophic structure and nutrient sources using stable isotope assays.

## SITE DESCRIPTION

Cave Springs Cave (CSC) in Cave Springs, Benton County, Arkansas, is a joint-controlled, phreatic conduit with a surveyed length of 515 m (Figure 1) and was formed in Mississippian Period limestone (Boone Formation). The cave stream has a mean annual discharge of 100 l/s and a mean annual water temperature of 14.4°C, and it is recharged primarily by ground water from the unconfined Springfield Plateau aquifer. The ground water basin drainage area (recharge area) is at least 38 $km^2$ (Williams, 1991), situated in an area of intensive rural and urban land use practices and rapid land conversion. The recharge area contains at least 40 confined animal feeding operations (poultry and swine) and hundreds of septic systems; municipal sewage sludge was applied on pastures for at least six years (1990 to 1996). The surrounding Illinois River watershed has been declared an impaired water body by the State of Arkansas (due primarily to nutrient loadings), and most of the original land cover – oak/hickory forest and savannah – has been converted to cattle pasture, confined animal feeding operations, and housing developments.

Cave Springs Cave faunal community includes a maternity colony of approximately 3,000 gray bats (*Myotis grisescens*) (federally listed as endangered), the largest known population (*circa* 150 fish) of Ozark cavefish (*Amblyopsis rosae*) (federally listed as threatened), populations of stygobitic isopods (*Caecidotea stiladactyla*) (Arkansas State Species of Concern), and stygobitic amphipods (*Stygobromus onondagaensis* and *S. ozarkensis*) (Arkansas State Species of Concern). Other prominent fauna include the eastern pipistrelle bat (*Pipistrellus subflavus*), cave salamander (*Eurycea lucifuga*), darksided salamander (*Eurycea*

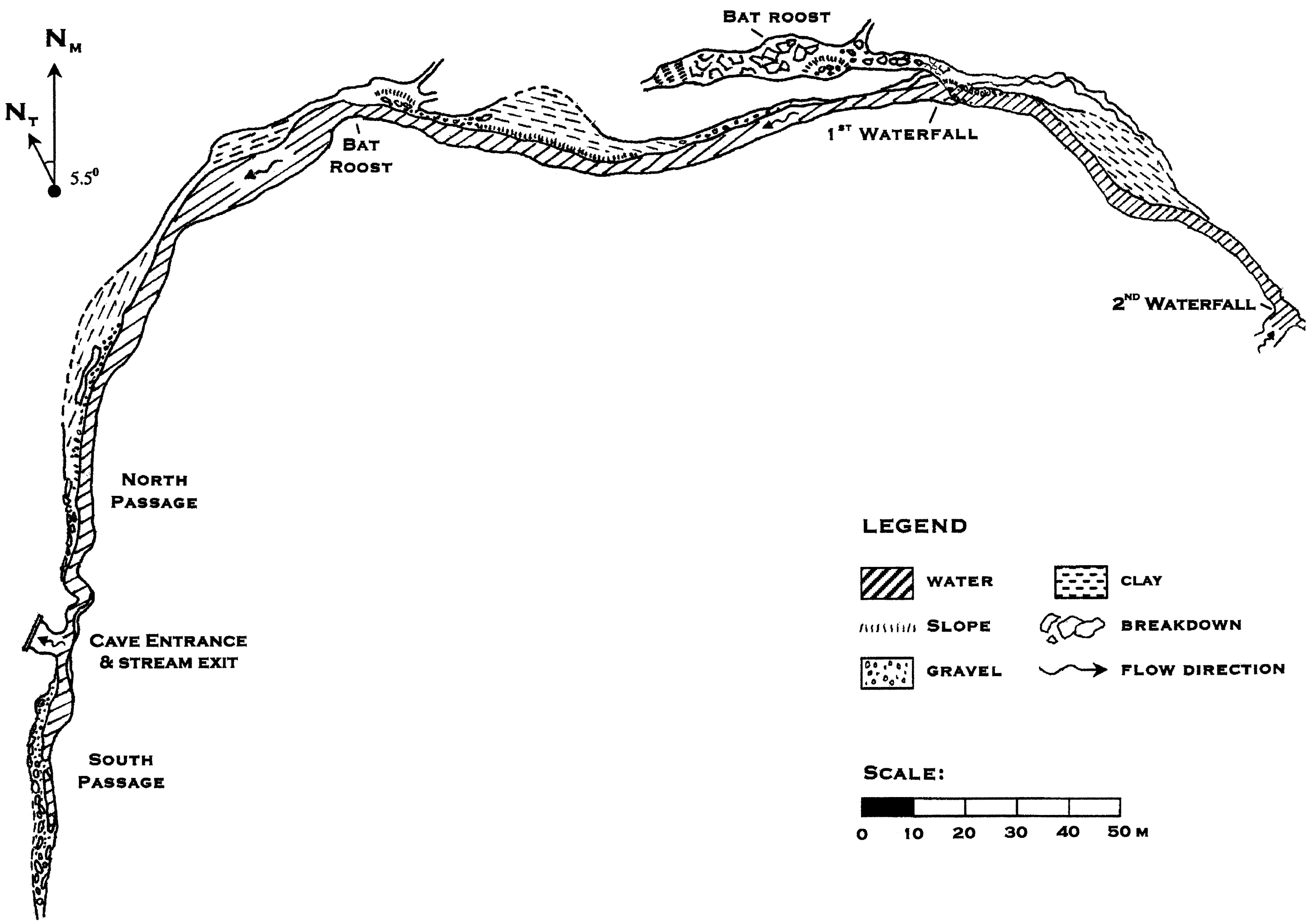


Figure 1. Map (Plan View) of Cave Springs Cave, Arkansas, Showing Passage Orientation and Substrate (water, chert gravel, clastic mud, or boulders from ceiling breakdown). Cartographic survey used tape and compass, with total length of 515 m, and a study reach (270 m) spanning the cave entrance to the first waterfall in the north passage.

*longicauda melanopleura*), and spot handed crayfish (*Orconectes punctimanus*).

## METHODS

### *General*

This study was conducted November 1997 to December 1999 and focused upon a 270 m stream reach with two sampling stations (a downstream station at the cave mouth where the stream resurges and a station inside the cave upstream of all bat roosts at the first waterfall) and a streambed area of 900 m$^2$, calculated from a cartographic survey performed using tape and compass (see cave map, Figure 1). This study was performed under U.S. Fish and Wildlife Service Permits PRT-834518 and TE834518-2, Arkansas Natural Heritage Commission Permit S-NHCC-99-005, and Arkansas Game and Fish Commission Permit 1082. Impact was minimized by restricting visits into the cave to times when gray bats were not present and by avoiding wading in the cave stream whenever possible. One individual Ozark cavefish that was found severely wounded in the stream, possibly from inadvertent trampling during a population census, was collected and used for stable isotope and heavy metals analyses with permission from the U.S. Fish and Wildlife Service.

*Organic Matter Budget*

**Budget Inputs.** Organic matter budget methods followed Webster and Meyer (1997), using grams of ash free dry mass (AFDM) and a conversion factor of 2 g AFDM/g C. Estimates of instream organic matter import and export were based on concentrations of total organic carbon (mg/l TOC) and dissolved organic carbon (mg/l DOC), measured by the Dumas combustion method during baseflow (monthly sampling) and storm flow conditions (daily sampling). Particulate organic carbon (mg/l POC) in transport was calculated as the difference between TOC and DOC. Daily load (kg/d) was determined by multiplying each concentration by discharge (l/s) converted to flux (g/m$^2$/d) using a streambed area of 900 m$^2$, and yearly load (g/m$^2$/y) was calculated by integration of daily load curve areas. Discharge was measured *in situ* using the stage/discharge relationship: stage (m) x 35.79 (m$^2$/min) - 110 = discharge (m$^3$/min) (J. Van Brahana, U.S. Geological Survey, unpublished data). Bat guano inputs (g/m$^2$/d) were measured by the method of Brown *et al.* (1994), in which guano was captured on plates at the beginning of the north passage over a span of 150 days (when the maternity colony was present), oven dried, and weighed; using a total plate surface area of 0.60 m$^2$ and a measured TOC content of 41.3 percent dry basis (d.b.), it was calculated as organic matter input (g AFDM/m$^2$/y).

**Budget Standing Crops.** Fine benthic organic matter (FBOM, g AFDM/m$^2$) of cave sediments was calculated from a measured TOC content of 4.7 percent (d.b), mean density of 1.5 g/cm$^3$, and volume of 35 m$^3$ (mean depth of 50 cm and sediment area of 700 m$^2$, calculated from cartographic survey). For streambed areas covered with cobble (200 m$^2$), epilithon standing crop was estimated by measuring biofilm surface area on randomly picked rocks, scraping the biofilm off rocks, then oven drying, weighing, and using a measured TOC content of 1.7 percent (d.b.) to calculate mean epilithon density (g AFDM/m$^2$). Standing crop of cave biota was calculated from the measured mass of each species (g AFDM) and the estimated population density (individuals/m$^2$) determined from ocular bioinventories performed annually using dive lights and standardized techniques and search effort.

**Budget Outputs.** Respiration (g AFDM/m$^2$/y) associated with the cave stream substrate community (benthos) was measured *in situ* by the oxygen uptake method (Bott *et al.,* 1978) using four replicates: representative proportions of rocks, gravel, and mud were placed in 8 liter plastic buckets, buried in the cave streambed for nine weeks to reestablish the epilithic community, then sealed with lids outfitted with ports for probes (YSI Model 57 stirring oxygen meter); change in oxygen concentration (mg/l O$^2$) was measured every 30 minutes for six hours, and respiration rate (g C/l/hr) was calculated by g C = g $O_2$ x 0.85 x 12/32; final output was calculated using the mean interstitial water volume (l) and the bucket base area (0.0415 m$^2$). Respiration rate for suspended cave stream community (seston) was determined in a similar manner using self stirring oxygen probes in darkened BOD bottles (300 ml).

*Stable Isotope Analyses (SIA)*

Seston was collected as per Voss and Struck (1997) by filtering large volumes of cave water (circa 1,200 l) through precombusted 0.45 mm glass fiber filters held in an inline filter housing, oven drying the filters, and scraping the residue into clean glass vials. Because animal size was small, composite samples were collected of whole isopods (*C. stiladactyla*) and of epigean crayfish abdominal muscle (*O. punctimanus*). Poultry litter and beef cattle manure were obtained from the University of Arkansas at Fayetteville (UAF) Experimental Farm. To increase the sample size and accuracy of poultry litter stable isotope ratios, isotope values from this study were combined with values from Kwak (1999), who obtained poultry wastes from the same source. Septic system sludge and leachate were collected from two residential septic systems near the study area. Municipal sewage sludge (press cake) was collected directly from the belt press of the Springdale Sewage Treatment Plant, Benton County, Arkansas. Samples of soil, pasture grass (*Festuca arundinacea*), and leaf litter (*Quercus* spp., *Platanus occidentalis*, and *Celtis occidentalis*) were collected randomly near the cave. Samples were replicated (n = 2 to 5) where possible.

All samples were oven dried, pulverized, acidified with 1 N HCl to remove inorganic carbon, redried and pulverized to pass through a 500 µm mesh sieve, and analyzed by Stable Isotope Ratio Facility for Environmental Research, University of Utah at Salt Lake City, to determine natural abundance of carbon and nitrogen isotopes, with analytical variability of 0.1 per mil. The ratios of heavy and light isotopes in a sample ($R_{sa}$) were compared to the ratios of the corresponding standard ($R_{std}$), and the difference was calculated using delta (δ) notation as $\delta$ (‰) = ($R_{sa}$ / $R_{std}$ - 1) x 1,000 (McKinney *et al.,* 1950).

*Environmental Quality Analyses*

Water samples were collected at the cave mouth monthly at baseflow conditions and during 10 storm events: March 5 to 11, 1998 (with 7.6 cm rain accumulation); June 8 to 10, 1998 (2 cm); September 13 and 14, 1998 (5.6 cm); March 8 to 20, 1999 (9.7 cm); April 2 to 5, 1999 (7.1 cm); May 2 to 5, 1999 (6.7 cm); September 7 to 8, 1999 (3 cm); October 29 to November 2, 1999 (6.7 cm); December 2 to 6, 1999 (5.8 cm), with record high flow (333 l/s) during the May 1999 spate. Samples were collected and analyzed using U.S. Environmental Protection Agency methods for total coliform and *Escherichia coli* densities (colony forming unit/100 ml); nitrate, nitrite, and total Kjeldahl nitrogen (TKN) (mg/l as N); orthophosphate and total phosphate (mg/l as P); and TOC and DOC (mg C/l). Cave stream discharge (l/s), specific conductivity (µSiemens/cm), turbidity (nephlometric turbidity unit), pH, temperature (°C), and dissolved oxygen concentration (mg/l) were measured *in situ*. For the metals analyses of tissue, sewage sludge, FBOM, and epilithon, samples were collected in prewashed glass containers, stored on ice, oven dried at 60°C, pulverized, and analyzed at Central Analytical Laboratory (UAF) using an inductively coupled plasma optical emission spectrometer.

## RESULTS AND DISCUSSION

*Organic Matter Flux in Cave Springs Cave and Other Subterranean Streams*

Results of organic matter budgeting (Table 1) documented a dissolved organic matter (DOM) influx of 4,730 $g/m^2/y$ and bat guano deposition rate of 10.4 $g/m^2/y$. Primary production was assumed negligible because of the absence of insolation and any evidence of chemolithotrophy. Direct and lateral inputs of organic matter such as leaf litter were also negligible because of the lack of karst windows such as collapsed sinkholes (dolines) or sinking (losing) streams in this cave complex. FBOM standing crop measured 1,209 $g/m^2$, and where sediment was scoured away, epilithon contributed approximately 2 $g/m^2$. No woody debris was present, and the only coarse benthic organic matter (CBOM) was bat guano, estimated at 1 $g/m^2$. Density of fauna was low (0.2 fish/$m^2$, 0.5 crayfish/$m^2$, and 5.0 isopods/$m^2$), and the resulting total standing crop was 0.8 $g/m^2$. Mean oxygen loss rate from benthic and sestonic respiration was 0.2 mg $O_2$/l/h, and the resulting organic matter export was 102 $g/m^2/y$ and 98 $g/m^2/y$, respectively. Mean yearly export was 5,330 kg/y as dissolved output and 280 kg/y as particulate output, with the majority of particulate transport occurring during storm events.

Two years of organic matter flux analyses in CSC described an oligotrophic stream ecosystem reliant almost exclusively on allochthonous energy sources. The vast majority of organic matter input was in the dissolved form, with bat guano representing less than 1 percent of input, even though thousands of bats roost in the cave in summer and guano is rich in organic carbon (40 percent TOC). Brown *et al.* (1994) found no difference in TOC concentration of the water below and above bat roosts in a similar Ozark cave stream (Logan Cave) and concluded that bat guano was not a significant input into the food web even though approximately 9,000 bats occupy the cave every summer (Harvey, 1996). The organic matter standing crop, consisting mainly of clastic sediments of low organic matter content (5 percent TOC), supported a small invertebrate community in both our study cave and Logan Cave.

We further compared the CSC organic matter budget to that of Logan Cave, Benton County, Arkansas (Brown *et al.*, 1994), which occurs in the same watershed, geologic strata, and aquifer as CSC and has similar biota, stream chemistry and substrates, cave length, and ground water recharge area size. The organic matter dynamics of these two caves were remarkably similar (Table 1). Dissolved organic carbon accounted for more than 90 percent of carbon inputs in both cave complexes, and bat guano inputs were similar. Logan Cave differs from CSC by having a collapsed doline that allowed lateral input of particulates (56 $g/m^2/y$) and that contributed woody debris (25 $g/m^2$) to the standing crop.

To place these subterranean stream energy fluxes in perspective with surface streams, we compared budgets of these subterranean streams to a summary (Table 1) of 19 first-order and second-order surface stream budgets from North America, Australia, Antarctica, and Europe by Webster and Meyer (1997). This dataset was used because no other budget studies were known from within the region of our study stream. These cave streams differed markedly from surface streams by their greater dependence upon DOM, presence of bat guano inputs, absence of autotrophic production or respiration, paucity of CBOM, woody debris inputs, and standing crops, and relative richness of FBOM standing crop.

*Stable Isotope Analyses and Trophic Dynamics*

Results of SIA are presented graphically in Figure 2, and isotopic signatures were similar (less than 2 per mil difference) to published values for bat guano

TABLE 1. Comparison of Organic Matter Budgets of Cave Springs Cave (this study), Logan Cave (Brown *et al.,* 1994), and 19 First-Order and Second-Order Surface Streams Worldwide (Webster and Meyer, 1997), Summarized by Minimum, Median, and Maximum Values. All masses are expressed as grams of ash free dry mass. Parameters not applicable to surface or subterranean ecosystems denoted by N.A. and parameters not measured denoted by N.M.

| | Cave Springs | Logan Cave | Summary of 19 Surface Streams | | |
|---|---|---|---|---|---|
| | | | Minimum | Median | Maximum |
| **Physical Characteristics** | | | | | |
| Latitude | 36 | 36 | 18 | 45 | 78 |
| Watershed Area (ha) | 3,770 | 3,015 | 8 | 50 | 44,700 |
| Streambed Area ($m^2$) | 900 | 2,500 | 150 | 2,122 | 32,100 |
| Gradient (m/km) | 4 | 4 | > 1 | 53 | 450 |
| Mean Annual Water Temp. (°C) | 14 | 14 | 1 | 8 | 22 |
| Mean Annual Discharge (l/s) | 100 | 250 | 2 | 16 | 1,200 |
| Mean Annual Precipitation (cm) | 114 | 114 | 10 | 123 | 315 |
| Mean Stream Width (m) | 4 | 3 | > 1 | 2 | 6 |
| **Inputs** | | | | | |
| Gross Primary Product. ($g/m^2/y$) | N.A. | N.A. | 0 | 71 | 5,400 |
| Leaf-fall ($g/m^2/y$) | 0 | 0 | 0 | 448 | 736 |
| Lateral Movement ($g/m^2/y$) | 0 | 56 | 0 | 10 | 1,111 |
| DOM Input ($g/m^2/y$) | 4,370 | 3,250 | 0 | 95 | 36,037 |
| Bat Guano Deposition ($g/m^2/y$) | 10 | 3 | N.A. | N.A. | N.A. |
| **Standing Crops** | | | | | |
| FBOM ($g/m^2$) | 1,209 | 632 | 0 | 333 | 1,400 |
| Wood ($g/m^2$) | 0 | 25 | 0 | 2,988 | 28,993 |
| **Outputs** | | | | | |
| Autotrophic Respiration ($g/m^2/y$) | N.A. | N.A. | 0 | 50 | 2,700 |
| Heterotrophic Respiration ($g/m^2/y$) | 200 | N.M. | 44 | 222 | 2,500 |
| Particulate Transport (kg/y) | 280 | 1,360 | 37 | 1,071 | 13,571 |
| Dissolved Transport (kg/y) | 5,330 | 25,550 | 54 | 1,068 | 178,000 |

(Mitzutani *et al.,* 1992), hardwood litter (McArthur and Moorhead, 1996), septic and sewage waste (Kwak and Zedler, 1997), crayfish (Whitledge and Rabeni, 1997), and stream sediment (Kwak, 1999). Nitrogen stable isotopes can describe food chain length by the consistent enrichment of the isotope ratio ($^{15}N/^{14}N$) by a mean 3.4 per mil at each trophic level (DeNiro and Epstein, 1981; Peterson and Fry, 1987; Kwak and Zedler, 1997). Although omnivory, common in cave food webs, obscures discrete trophic level transfers (Polis and Strong, 1996), our SIA suggested that the CSC food web had three distinct trophic levels: a food base of benthic detritus; a guild of invertebrate consumers – isopods, crayfish, and historically, amphipods; and a top predator – Ozark cavefish. A food web study of a chemoautotrophic cave ecosystem (Movile Cave, Romania) reported three trophic levels as well: a base of chemoautotrophic microbial mats; invertebrate grazers; and invertebrate predators

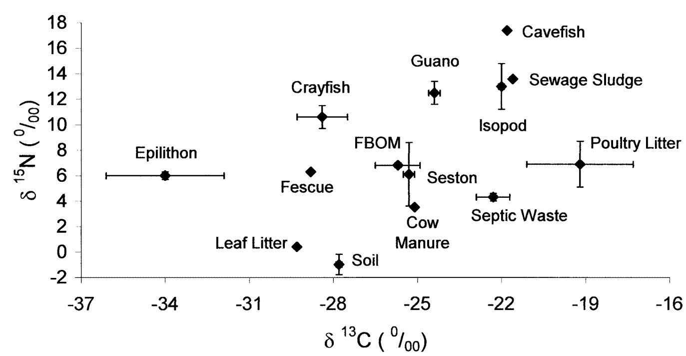


Figure 2. Plot of Mean Carbon and Nitrogen Stable Isotope δ Values (‰, ± 1 SE) of Cave Springs Cave Stream Ecosystem Components and Potential Organic Matter Inputs, Replicated as Noted: Fine Benthic Organic Matter (FBOM, n = 2); Epilithon (2); Seston (2); Spot Handed Crayfish (5); Stygobitic Isopods (2); Ozark Cavefish; Soil; Leaf Litter; Poultry Litter (5); Cow Manure; Municipal Sewage Sludge; Septic System Sludge (3); Gray Bat Guano (5); and Fescue Grass.

(Sarbu *et al.,* 1996). A trophic study of an anchialine cave ecosystem in Mexico reported 3 to 3.5 trophic levels: producers (algae, bacteria, and detritus); invertebrate consumers; and invertebrate predators/ scavengers (Pohlman *et al.,* 1997). These limited results suggest that three trophic levels may be typical for aquatic cave ecosystems.

Stable carbon isotopic compositions of animals reflect those of their diets within about 1 per mil (Peterson and Fry, 1987), and our results suggested that the primary dietary component of Ozark cavefish was stygobitic isopods, a prey item reported by Eigenmann (1909) and Poulson (1963). Our SIA also suggested that sewage derived organic matter contributed to cavefish diet because of almost identical carbon isotopic signatures. Gray bat guano, although isotopically separate, might be another food source for the Ozark cavefish, which is known to ingest guano (Poulson, 1963).

Isotopic signatures of stygobitic isopods suggested that bat guano was not the primary constituent of their diet. Sewage derived organic matter was a likely source because of its similar isotopic signature, but FBOM and animal wastes could also contribute to isopod diet (Figure 2). Similarly, spot handed crayfish did not appear to rely exclusively upon bat guano: isotopic signatures of four out of five composite samples (each composed of five to 10 crayfish) were trophically separate from guano, while one sample was almost identical to gray bat guano. Crayfish diet may have consisted of a mixture of FBOM and epilithon. While restricted in organic matter content, subterranean sediments are documented to be an important food of stygobitic crustaceans (Dickson, 1975; Mathieu *et al.,* 1991). Yet what constituted FBOM in CSC was not clear. FBOM was isotopically indistinguishable from seston, suggesting that particulate in transport was the source of FBOM – components of leaf litter, guano, septic waste, and cow manure could constitute the signature. In a Mexican cave stream, Pohlman *et al.* (1997) concluded that cave FBOM derived from surface soil and that it supplied the majority of organic matter to the food web. Because many organic matter resources were involved and most of the animals were omnivorous, stable isotope methods were not able to definitively detail each trophic component and linkage in our study. Further investigation using additional techniques is needed to trace surface inputs into subsurface trophic webs.

*Evidence of Degraded Environmental Quality in Cave Springs Cave*

Results of environmental quality analyses (Tables 2 and 3) indicated that the CSC stream ecosystem was contaminated with excess nutrients, fecal bacteria, and metals. Mean total coliform density (3,200 CFU/100 ml at baseflow, 10,800 CFU/100 ml at storm flow) and mean nitrate concentration (5.6 mg/l at base flow, 6.4 mg/l at storm flow) in CSC were both twice as high as regional levels reported by National Water Quality Assessment program (Petersen *et al.,* 1998), and the largest bacterial loadings (up to 83,100 CFU/100 ml) occurred during storms and during the summer growing seasons when confined animal wastes were applied to pastures. Mean baseflow concentrations of orthophosphate (35 µg/l) exceeded background concentrations for U.S. ground waters (USGS, 1999), and several storm flow samples exceeded the State of Arkansas limit for total phosphorous of 100 µg/l (APCEC, 1998). Total coliform and *E. coli* densities were significantly higher during storm events than during baseflows (*E. coli*: $t$ = 2.323, $P$ = 0.012; total coliforms: $t$ = 2.914, $P$ = 0.003). Both total coliform and *E. coli* densities were positively correlated to TOC, TKN, total phosphorous, and orthophosphate ($P$ < 0.001 for all pairwise correlations). A negative binomial regression model (Type III) determined that season was significantly related to both bacterial metrics, with microbial densities significantly lower in late winter/early spring (*E. coli*: $\chi^2$ = 23.69, $P$ < 0.001; total coliforms: $\chi^2$ = 35.32, $P$ < 0.001). Metals analyses (Table 2) detected beryllium, copper, lead, selenium, and zinc in concentrations in cave water that exceeded state of Arkansas limits for chronic and acute toxicity to aquatic life (APCEC, 1998). Toxic metals were also detected in FBOM and in tissues of crayfish, isopods, and cavefish (Table 3).

Bat guano did not appear to be the primary source of microbes because no significant difference was detected between mean bacterial counts upstream and downstream of bat roosts. Total coliform densities were significantly correlated ($P$ < 0.001) with stream discharge, turbidity, and orthophosphate and nitrate concentrations, strengthening the conclusion that fecal bacteria originated from the surface and were flushed in during storm events, rather than originating solely from gray bat defecation. Williams (1991) discounted bat guano as a possible fecal coliform source for CSC because surface streams in the recharge area had similarly high levels of coliforms. Water samples collected in Logan Cave upstream of bat roosts often had higher coliform densities than downstream samples (Means and Johnson, 1995).

TABLE 2. Summary Statistics (sample size = n, minimum, median, mean, and maximum value) in Cave Springs Cave Water Samples Collected at the Cave Mouth From November 1997 Through December 1999 During Baseflow and Storm Flow Conditions. Parameters not detected denoted by N.D. Chromium, molybdenum, and silver were not detected in any sample, and antimony, beryllium, cadmium, cobalt, and nickel were never detected at greater than 3 µg/l.

| | Unit | n | Minimum | Median | Mean | Maximum |
|---|---|---|---|---|---|---|
| Water temperature | °C | 43 | 13.4 | 14.4 | 14.4 | 15.2 |
| Discharge | l/s | 59 | 17.0 | 83.0 | 100.0 | 333.0 |
| Spec. Conductivity | µS/cm | 51 | 240.0 | 330.0 | 326.0 | 400.0 |
| Turbidity | NTU | 44 | < 1.0 | 4.0 | 5.0 | 48.0 |
| Field pH | pH unit | 30 | 6.2 | 6.8 | 6.8 | 7.5 |
| Dissolved Oxygen | mg/l | 37 | 8.3 | 9.4 | 9.6 | 11.6 |
| Aluminum | µg/l | 34 | N.D. | 24.0 | 66.0 | 275.0 |
| Arsenic | µg/l | 34 | N.D. | N.D. | 3.0 | 14.0 |
| Barium | µg/l | 18 | 54. | 65.0 | 65.0 | 95.0 |
| Boron | µg/l | 18 | N.D. | N.D. | 3.0 | 33.0 |
| Calcium | µg/l | 18 | 30.1 | 51.0 | 50.7 | 64.0 |
| Chloride | µg/l | 17 | 6.1 | 7.7 | 7.8 | 9.5 |
| Copper | µg/l | 34 | N.D. | 11.0 | 20.0 | 230.0 |
| Iron | µg/l | 34 | N.D. | 11.0 | 80.0 | 640.0 |
| Lead | µg/l | 34 | N.D. | 4.0 | 10.0 | 39.0 |
| Magnesium | mg/l | 18 | 1.7 | 2.2 | 2.1 | 2.5 |
| Manganese | µg/l | 34 | N.D. | 2.0 | 8.0 | 45.0 |
| Selenium | µg/l | 34 | N.D. | 12.0 | 9.0 | 32.0 |
| Sulfate | mg/l | 16 | 2.4 | 3.7 | 3.7 | 7.1 |
| Vanadium | µg/l | 18 | N.D. | N.D. | 1.0 | 8.0 |
| Zinc | µg/l | 33 | N.D. | 13.0 | 38.0 | 288.0 |
| TOC | mg/l | 43 | N.D. | 1.2 | 1.6 | 5.1 |
| DOC | mg/l | 10 | 0.6 | 1.6 | 1.7 | 3.4 |
| Ammonia - N | µg/l | 35 | N.D. | 9.0 | 21.0 | 323.0 |
| Nitrite - N | µg/l | 44 | N.D. | N.D. | 6.0 | 104.0 |
| Nitrate - N | mg/l | 53 | N.D. | 6.0 | 6.2 | 8.8 |
| Total Kjeldahl N | mg/l | 22 | N.D. | 0.1 | 0.2 | 1.3 |
| Total Phosphorous | µg/l | 37 | N.D. | 43.0 | 54.0 | 193.0 |
| Orthophosphate | µg/l | 37 | 17.0 | 26.0 | 38.0 | 103.0 |
| *Escherichia coli* | CFU/100 ml | 56 | < 10.0 | 215.0 | 1,826.0 | 20,050.0 |
| Total Coliforms | CFU/100 ml | 56 | < 200.0 | 5,310.0 | 8,904.0 | 83,100.0 |

Septic system leachate, livestock manures, and sewage sludge were probable sources of the high fecal coliform loading in these ground water ecosystems.

*Effects of Pollution Upon Cave Streams Ecosystems*

Organic pollution has adverse effects on cave ecosystems, including alteration of community assemblages, impoverishment of biodiversity, and increased risk of predation by epigean fauna. Septic pollution was implicated in the eradication of invertebrates from other Ozark caves (Aley, 1976). Faunal inventories performed during the course of our study in CSC failed to detect either stygobitic amphipod species (*S. onondagaensis*, *S. ozarkensis*) where they were formerly reported (Holsinger, 1972; Holsinger, pers. comm. 1999). However, stygobitic isopods were found in abundance in CSC, paralleling the findings of other studies of polluted cave systems. Banners Corner Cave, Virginia, for example, had direct septic system leachate input for at least 30 years, and stygobitic isopods (*Caecidotea recurvata*) thrived, using sewage-fed bacteria as a food source, while stygobitic amphipods (*Stygobromus mackini*) and flatworms (*Sphalloplana* sp.) were extirpated (Simon and Buikema, 1997). The similar dearth of amphipods and abundance of isopods (i.e., change in amphipod to isopod ratio) and the detection of organic pollution in CSC implicate human and animal wastes disposal in

TABLE 3. Solids Concentrations (mg/kg, d.b.) of Aluminum (Al), Arsenic (As), Barium (Ba), Boron (B), Cadmium (Cd), Chromium (Cr), Cobalt (Co), Copper (Cu), Iron (Fe), Lead (Pb), Manganese (Mn), Molybdenum (Mo), Nickel (Ni), Vanadium (Va), and Zinc (Zn) in Cave Springs Cave Ecosystem Components and Nutrient Sources, replicated as noted. Parameters not detected denoted by N.D. and not measured by N.M. Antimony, beryllium, and selenium were never detected at greater than 6 mg/kg.

| | **Al** | **As** | **Ba** | **B** | **Cd** | **Cr** | **Co** | **Cu** | **Fe** | **Pb** | **Mn** | **Mo** | **Ni** | **V** | **Zn** |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Sewage Sludge | 7,574 | 2.0 | 237.0 | 21.5 | 1.1 | 23.1 | 3.7 | 190.3 | 6,995 | 25.5 | 192.5 | 10.4 | 57.6 | 20.4 | 454.0 |
| FBOM No. 1 | 42,616 | 20.3 | 162.0 | N.D. | 5.0 | 39.1 | 18.2 | 34.4 | 32,977 | 45.0 | N.M. | 0.7 | 86.5 | 138.0 | 384.7 |
| FBOM No. 2 | N.M. | 6.6 | 72.5 | 1.0 | N.D. | 20.3 | 4.8 | 17.0 | N.M. | 9.3 | N.M. | 0.4 | 27.7 | 116.3 | 114.0 |
| Epilithon No. 1 | 21,651 | 10.1 | N.M. | N.D. | 1.0 | 24.6 | 9.2 | 18.0 | 16,400 | 17.3 | N.M. | 0.4 | 27.1 | 74.0 | 162.3 |
| Epilithon No. 2 | N.M. | 2.7 | 28.8 | 3.8 | 0.2 | 23.0 | 4.3 | 10.1 | N.M. | 10.3 | N.M. | 0.4 | 24.7 | 24.2 | 87.5 |
| Guano No. 1 | 2,294 | 1.4 | 163.7 | 1.0 | 9.4 | 5.0 | 6.7 | 434.9 | 5,044 | 19.9 | 112.9 | 8.3 | 18.2 | 27.2 | 904.0 |
| Guano No. 2 | N.M. | 1.3 | 159.2 | N.D. | 9.6 | 0.6 | 6.7 | 501.0 | N.M. | 20.0 | N.M. | 14.7 | 6.1 | 23.1 | N.D. |
| Septic Waste | N.M. | 6.4 | 146.9 | N.D. | N.D. | 12.7 | 15.8 | 10.5 | N.M. | 17.6 | N.M. | N.D. | 30.5 | 149.6 | 71.3 |
| Cow Manure | N.M. | N.D. | 94.8 | N.D. | 0.1 | 1.1 | 0.8 | 22.0 | N.M. | N.D. | N.M. | 1.5 | 1.5 | 2.1 | 47.8 |
| Poultry Litter | N.M. | 18.5 | 31.9 | 11.5 | 0.3 | 3.2 | 0.8 | 330.4 | N.M. | N.D. | N.M. | 4.7 | 13.9 | 6.1 | 607.5 |
| Isopods | 24,785 | N.D. | 451.0 | N.D. | 47.6 | 27.0 | 6.0 | 174.0 | 19,730 | 48.4 | 539.0 | 4.7 | 28.2 | 122.0 | 539.0 |
| Crayfish No. 1 | N.M. | N.D. | 13.3 | N.D. | 0.3 | 4.7 | 0.1 | 64.2 | N.M. | N.D. | N.M. | 0.9 | 7.6 | 1.8 | 91.6 |
| Crayfish No. 2 | 773 | N.D. | 129.8 | N.D. | 1.7 | 1.9 | 0.6 | 118.0 | 340 | 2.4 | 51.2 | N.D. | 2.3 | 1.8 | 100.5 |
| Cavefish | N.M. | N.D. | 54.6 | N.D. | 0.3 | 2.3 | N.D. | 14.4 | N.M. | N.D. | N.M. | 1.5 | 1.7 | N.D. | 69.2 |

the recharge area in the alteration of its community assemblage. Furthermore, the grotto salamander (*Typhlotriton spelaeus*), another troglobite historically inhabiting CSC (Brown *et al.,* 1994), was not observed during our study period. Other studies have documented the disappearance of the grotto salamander from caves disturbed by habitat alteration (Schwartz, 1976) and chemical pollution (Crunkilton, 1984). However, our most recent census of the Ozark cavefish population produced a record high count (164 fish in November 2000), and any impact from habitat degradation was not yet evident.

Some investigators argue that nutrient enrichment of oligotrophic, subterranean ecosystems may have positive effects. Stewart (1984) suggested that land-applied animal waste was not a threat to Ozark cavefish habitat but may in fact augment the food supply. Sket (1977) reported that stygobites were abundant in a cave stream enriched with organic pollution, and Sket (1999) suggested that moderate organic pollution might benefit stygobites in oligotrophic habitats provided epigean fauna do not invade. However, the majority of relevant studies suggest that organic pollution disrupts cave community structure and function. Further research is needed to more clearly determine the biological ramifications of augmenting cave stream trophic webs with anthropogenic wastes.

Organic pollutants such as municipal sewage sludge and poultry litter often contain considerable quantities of toxic metals, and this could explain the toxic concentrations of metals detected in CSC water samples taken during this study. Furthermore, CSC stream sediment and tissues of isopods, crayfish, and one cavefish had greater concentrations of toxic metals than cave water samples. Because of the longevity of stygobitic organisms, even low concentrations of dissolved metals are of concern because they could bioaccumulate to lethal concentrations (Dickson *et al.,* 1979).

## CONCLUSIONS

Contrary to current hypotheses, bat guano did not appear to be the dominant component in energy flux or the food web in this cave stream ecosystem. Energetically, cave stream DOM was the most important input. Trophically, cave FBOM was the most important, sustaining a guild of crustacean detritivores, which in turn sustained a deme of cavefish. The origin of FBOM was complex and probably consisted of a mixture of natural inputs (leaf litter, soil, and occasionally guano) and anthropogenic inputs (septic and animal wastes). Evidence gathered here suggested that these organic pollutants altered the ecosystem dynamics of CSC; isotopic analyses detected these pollutants in trophic linkages, and environmental quality analyses detected excessive nutrients, bacteria, and metals in cave water, sediment, and fauna. The community assemblage may also have been altered: sensitive stygobitic fauna (gammarid amphipods and grotto salamanders) were absent, while resilient fauna (asellid isopods) flourished – a pattern reported from other studies that documented impacts of organic pollution upon cave ecosystems. The reduction of anthropogenic pollution input into this ground water basin is necessary to restore ecosystem dynamics in CSC to its former undisturbed condition. It is imperative that cave streams be managed with respect to their tributary ground water basins and historic energy fluxes, with a special focus on indicator species such as stygobitic crustaceans and sensitive areas such as sinkholes, losing streams, fissures, or other karst features.

## ACKNOWLEDGMENTS

This research was funded by grants from the Arkansas Natural Heritage Commission and a Cave Conservancy Foundation fellowship awarded to Graening. Richard Thompson, Arkansas Department of Environmental Quality, contributed laboratory analyses. The field work was performed with the assistance of many colleagues, including Casey Pevey and Dennis Lichtenburg for cartography and Brian Wagner, Stan Todd, Mike Slay, and Dr. Larry Willis for faunal inventories. Lynette Duncan was consulted for statistical analyses. Dr. Thomas Kwak and anonymous reviewers provided constructive criticism in the preparation of this manuscript.

## LITERATURE CITED

Aley, T. J., 1976. Hydrology and Surface Management. *In:* The National Cave Management Symposium Proceedings. Speleobooks, Albuquerque, New Mexico, pp. 44-45.

APCEC (Arkansas Pollution Control and Ecology Commission), 1998. Regulation 2, as Amended: Regulation Establishing Water Quality Standards for Surface Waters of the State of Arkansas. Little Rock, Arkansas.

Bott, T. L., J. T. Brock, C. E. Cushing, S. V. Gregory, D. King, and R. C. Peterson, 1978. A Comparison of Methods for Measuring Primary Productivity and Community Respiration in Streams. Hydrobiologia 60:3-12.

Brown, A. V., W. K. Pierson, and K. B. Brown, 1994. Organic Carbon Resources and the Payoff-Risk Relationship in Cave Ecosystems. *In:* Second International Conference on Ground Water Ecology. U.S. Environmental Protection Agency and the American Water Resources Association, pp. 67-76.

Crunkilton, R., 1984. Subterranean Contamination of Meramec Spring by Ammonium Nitrate and Urea Fertilizer and Its Implication on Rare Cave Biota. Proceedings of the 1984 National Cave Management Symposium. Journal of the Missouri Speleological Society 25:151-158.

Culver, D. C., 1982. Cave Life: Evolution and Ecology. Harvard University Press, Cambridge, Massachusetts.

Culver, D. C., 1994. Species Interactions. *In:* Groundwater Ecology, J. Gibert, D. Danielopol, and J. A. Stanford (Editors). Academic Press, San Diego, California, pp. 271-286.

Culver, D. C., L. L. Master, M. C. Christman, and H. H. Hobbs III, 2000. The Obligate Cave Fauna of the 48 Contiguous United States. Conservation Biology 14:386-401.

Curl, R. L., 1958. A Statistical Theory of Cave Entrance Evolution. Bulletin of the National Speleological Society 20:9-22.

DeNiro, M. J. and S. Epstein, 1981. Influence of Diet on the Distribution of Nitrogen Isotopes in Animals. Geochimica et Cosmochimica Acta 45:341-351.

Dickson, G. W., 1975. A Preliminary Study of Heterotrophic Microorganisms as Factors in Substrate Selection of Troglobitic Invertebrates. National Speleological Society Bulletin 37:89-93.

Dickson, G. W., L. A. Briese, and J. P. Giesy, Jr., 1979. Tissue Metal Concentrations in Two Crayfish Species Cohabiting a Tennessee Cave Stream. Oecologia 44:8-12.

Eigenmann, C. H., 1909. Cave Vertebrates of North America, a Study in Degenerative Evolution. Carnegie Institution of Washington Publications 104:1-241.

Fisher, S. G. and G. E. Likens, 1973. Energy Flow in Bear Brook, New Hampshire: An Integrative Approach to Stream Ecosystem Metabolism. Ecological Monographs 43:421-439.

Ford, D., A. Palmer, and W. White, 1988. Landform Development; Karst. *In:* The Geology of North America, Volume O-2, Hydrogeology. W. Back, J. Rosenshein, and P. Seaber (Editors). Geological Society of America, Boulder, Colorado, Chapter 43.

Gibert, J., J. A. Stanford, and M. Dole-Olivier, 1994. Basic Attributes of Groundwater Ecosystems and Prospects for Research. *In:* Groundwater Ecology, J. Gibert, D. Danielopol, and J. A. Stanford (Editors). Academic Press, San Diego, California, pp. 8-37.

Gounot, A., 1994. Microbial Ecology of Groundwaters. In: Groundwater Ecology, J. Gibert, D. Danielopol, and J. A. Stanford (Editors). Academic Press, San Diego, California, pp. 189-215.

Harris, J., 1970. Bat-Guano Cave Environment. Science 169:1342-1243.

Harvey, M. J., 1996. Status and Management of Endangered Bats in Arkansas. 1996 Proceedings of the Annual Conference of the Southeastern Association of Fish and Wildlife Agencies 50:246-253.

Holsinger, J. R., 1972. The Freshwater Amphipod Crustaceans (*Gammaridae*) of North America. Biota of Freshwater Ecosystems Identification Manual No. 5. U.S. Environmental Protection Agency. Washington, D.C.

Kwak, T. J., 1999. Animal Waste Nutrient Dynamics in the Stream Food Web: Evaluation of Multiple Stable Isotopes for Detection. Final Report to the Arkansas Game and Fish Commission. Little Rock, Arkansas.

Kwak, T. J. and J. B. Zedler, 1997. Food Web Analysis of Southern California Coastal Wetlands Using Multiple Stable Isotopes. Oecologia 110:262-277.

Mathieu, J., K. Essafi, and S. Doledec, 1991. Dynamics of Particulate Organic Matter in Bed Sediments of Two Karst Streams. Archiv fur Hydrobiologie 122:199-211.

McArthur, J. V. and K. K. Moorhead, 1996. Characterization of Riparian Species and Stream Detritus Using Multiple Stable Isotopes. Oecologia 107:232-238.

McKinney, C. J., J. McCrea, S. Epstein, H. Allen, and H. Urey, 1950. Improvements in Mass Spectrometers for the Measurement of Small Differences in Isotope Abundance Ratios. Review of Scientific Instruments 21:724-730.

Means, M. and J. Johnson, 1995. Movement of Threatened Ozark Cavefish in Logan National Wildlife Refuge, Arkansas. The Southwestern Naturalist 40(3):308-313.

Mitzutani, H., D. A. McFarlane, and Y. Kabaya, 1992. Carbon and Nitrogen Isotopic Signatures of Bat Guanos as Record of Past Environments. Mass Spectroscopy 40:67-82.

National Agricultural Statistics Service, 2000. Agricultural Statistics 2000. U. S. Department of Agriculture, U.S. Governmental Publications Office, Washington, D.C.

Notenboom, J., S. Plenet, and J. Turquin, 1994. Groundwater Contamination and Its Impact on Groundwater Animals and Ecosystems. *In:* Groundwater Ecology, J. Gibert, D. Danielopol, and J. A. Stanford (Editors). Academic Press, San Diego, California, pp. 477-503.

Pasquarell, G. C. and D. G. Boyer, 1996. Herbicides in Karst Groundwater in Southeast West Virginia. Journal of Environmental Quality 25:755-765.

Petersen, J. C., J. C. Adamski, R. W. Bell, J. V. Davis, S. R. Femmer, D. A. Freiwald, and R. L. Joseph, 1998. Water Quality in the Ozark Plateaus. U.S. Geological Survey Circular 1158.

Peterson, B. J. and B. Fry, 1987. Stable Isotopes in Ecosystem Studies. Annual Review of Ecology and Systematics 18:293-320.

Pohlman, J. W., T. M. Iliffe, and L. A. Cifuentes, 1997. A Stable Isotope Study of Organic Cycling and the Ecology of an Anchialine Cave Ecosystem. Marine Ecology Progress Series 155:17-27.

Polis, G. A. and D. R. Strong, 1996. Food Web Complexity and Community Dynamics. American Naturalist 147:813-846.

Poulson, T. L., 1963. Cave Adaptation in Amblyopsid Fishes. American Midland Naturalist 70:257-290.

Poulson, T. L., 1972. Bat Guano Ecosystems. National Speleological Society Bulletin 34(2):55-59.

Poulson, T. L., 1976. Management of Biological Resources in Caves. *In:* Proceedings of the First National Cave Management Symposium. Speleobooks, Albuquerque, New Mexico, pp. 46-52.

Sarbu, S. M., T. C. Kane, and B. K. Kinkle, 1996. A Chemoautotrophically Based Cave Ecosystem. Science 272:1953-1955.

Schwartz, J., 1976. A Biological Study of Cathedral Cave, Crawford County, Missouri. Journal of the Missouri Speleological Society 16(4):1-21.

Simon, K. S. and A. L. Buikema, Jr., 1997. Effects of Organic Pollution on an Appalachian Cave: Changes in Macroinvertebrate Populations and Food Supplies. American Midland Naturalist 138:387-401.

Sinton, L. W., 1984. The Macroinvertebrates in a Sewage-Polluted Aquifer. Hydrobiologia 119:161-169.

Sket, B., 1977. Gegenseitige Beeinflussung Der Wasserpollution und des Hohlenmilieus. Proceedings of the 6th International Congress of Speleology 5:253-262.

Sket, B., 1999. The Nature of Biodiversity in Hypogean Waters and How it is Endangered. Biodiversity and Conservation 8:1319-1338.

Smith, C. R. and K. F. Steele, 1990. Nitrate Concentrations of Ground Water, Benton County, Arkansas. Miscellaneous Publication No. 73, Arkansas Water Resources Research Center, University of Arkansas, Fayetteville, Arkansas.

Steele, K. F., 1985. Groundwater in Northwest Arkansas. Arkansas Naturalist 3(7):5-10.

Stewart, J., 1984. Endangered and Threatened Wildlife and Plants; Proposed Threatened Status for the Ozark Cavefish (*Amblyopsis rosae*). Federal Register 49(21):3889-3891.

Strayer, D., 1994. Limits to Biological Distributions in Groundwater. *In:* Groundwater Ecology, J. Gibert, D. Danielopol, and J. A. Stanford (Editors). Academic Press, San Diego, California, pp. 287-310.

USGS (U.S. Geological Survey), 1999. The Quality of Our Nation's Waters – Nutrients and Pesticides. U.S. Geological Survey Circular 1225. Reston, Virginia.

Voss, M. and U. Struck, 1997. Stable Nitrogen and Carbon Isotopes as Indicator of Eutrophication of the Oder River (Baltic Sea). Marine Chemistry 59:35-49.

Webster, J. R. and J. L. Meyer (Editors), 1997. Stream Organic Matter Budgets. Journal of the North American Benthological Society 16:3-161.

White, W. B., D. C. Culver, and J. E. Mylroie, 1995. Karst Lands: The Dissolution of Carbonate Rock Produces Unique Landscapes and Poses Significant Environmental Concerns. American Scientist 83(5):450-459.

Whitledge, G. W. and C. F. Rabeni, 1997. Energy Sources and Ecological Role of Crayfishes in an Ozark Stream: Insights From Stable Isotopes and Gut Analysis. Canadian Journal of Fisheries and Aquatic Sciences 54:2555-2563.

Williams, R. D., 1991. Water Quality and Groundwater Recharge for the Cave Springs Complex and Associated Streams Near Cave Springs, Arkansas. Thesis, University of Arkansas, Fayetteville, Arkansas.

Willis, L. D. and A. V. Brown, 1985. Distribution and Habitat Requirements of the Ozark Cavefish, *Amblyopsis rosae*. American Midland Naturalist 114:311-317.

Wood, P. J., J. Gunn, and J. Perkins, 2002. The Impact of Pollution on Aquatic Invertebrates Within a Subterranean Ecosystem – Out of Sight Out of Mind. Archiv fur Hydrobiologie 155(2):223-237.