ABSTRACT
This semi-guided inquiry activity explores the macroinvertebrate fauna in water sources affected by different levels of pollution. Students develop their ability to identify macroinvertebrates, compare aquatic fauna from different sources of water samples, evaluate water quality using an index, document and analyze data, raise questions and hypotheses, and discuss other possible issues that could be investigated at a later time. These sets of activities were designed for freshman high school students but are applicable to middle school students as well.

Inquiry is a process of interaction between teacher and students whereby the teacher engages students in generating questions and pursuing answers through careful observation and reflection (Llewellyn, 2004). The inquiry cycle begins with a question that has to be comparative, time-wise, simple, and exciting. In this cycle (question → action → reflection → question), students help decide what to compare, what to measure (compare at least two, measure one), and how to measure and collect the data (action). In the reflection process, students explain their results and deliberate about what to do differently next time, which generates new questions. These are the stepping stones to reaching a global understanding of the inquiry process and involving students in doing science, both of which are conducive to more sophisticated comparisons later on.

One way to help students understand the scientific inquiry process is by encouraging them to investigate the quality of different types of water samples. This is important because the water quality in streams and rivers had been declining since the 1960s, becoming a serious concern (Feminella & Flynn, 1999). Because of the release of harmful pollutants in the water, such as heavy metals, sewage, and other chemical wastes, the Clean Water Act (CWA) was enacted in 1977. The goal of the CWA was to “restore and maintain the chemical, physical and biological integrity of the nation’s waters” (CWA, 1977).

In order to identify which water sources were becoming polluted, a water-quality monitoring approach was needed. The traditional water-quality monitoring approach became a collection of water samples and laboratory analysis for suspended physical and chemical pollutants. However, a biological approach to water-quality monitoring was less costly and incorporated water organisms as a basis for pollution detection. The basis of biomonitoring is that certain types of water animals, such as macroinvertebrates, occur or thrive only under certain water-quality conditions (Lenat, 1988). When conditions change, such as when a stream receives a significant chemical run-off, the abundance and distribution of invertebrates in the affected site change as well (Feminella & Flynn, 1999).

To introduce students to the cycle of inquiry and how to conduct real-world science, the ideal situation would be to bring students outdoors to stimulate their curiosity. However, when this is not possible, an alternative is to bring the outside world into the classroom. This is the case with water samples taken from rivers and ponds that are filled with macroinvertebrates and microscopic organisms. They are easily identified and categorized using magnifying glasses, portable scopes, ID books, and pictures.

Macroinvertebrates are small animals, usually >1 mm long, that do not have backbones and live on the bottom of a pond, lake, stream, or river for at least part of their lives (Feminella & Flynn, 1999). They can be found in crevices between submerged stones, in organic debris, on aquatic vegetation, or within sediments. Most of the species are aquatic insects, represented by immature stages of mayflies, dragonflies, damselflies, stoneflies, caddisflies, flies, and beetles. However, other types of organisms can be commonly found in the water, such as crustaceans (side swimmers,
crayfish, scuds), oligochaetes (earthworms, leeches), molluskus (snails, mussels, clams), and arachnids (aquatic mites).

We developed a semi-guided inquiry-based activity in the classroom to compare abundances of macroinvertebrate faunas from water samples that were assumed to have different pollution levels. Students related these macroinvertebrate abundances to pollution levels using macroinvertebrate tolerance levels and a key. The students processed the samples, analyzed and interpreted the data, and discussed the effects of pollution on macroinvertebrate populations. The students' discussion of their findings generated more questions that could be used for new inquiry investigations.

○ Materials

3-gallon plastic buckets for water samples
3x magnifying lenses (http://www.amazon.com)
Dual plastic magnifiers (3x and 6x) (http://www.carolina.com)
Battat two-way microscopes (http://www.amazon.com)
Illuminated zoom microscopes (60–100x) (http://www.dealextreme.com)
ID cards (Voshell, 2001)
ID-water-quality sorting sheet (Figure 1)
Petri dishes, one for each student (http://www.enasco.com)
Dropper (http://www.amazon.com)

Figure 1. Sorting sheet to evaluate water quality using macroinvertebrates. Each group represents organisms that are (1) very sensitive to pollution (high water quality; Group 1); (2) somewhat sensitive to water quality but could survive in waters that aren’t quite clean (mid-level pollution; Group 2); and (3) tolerant to high pollution (poor water quality; Group 3).

○ Methods

The teachers used 3-gallon plastic buckets to scoop and bring water samples from each of three locations (see Appendix): the Des Plaines River, Salt Creek, and Bison Pond. Some floating vegetation was added to increase the chances of capturing macroinvertebrates. Because the purpose of the inquiry method is to encourage students to think and make their own conjectures through questions, observations, classification, communication, measures, predictions, inference, and experimentation – as opposed to merely learning facts, concepts, and theories someone else has concluded (Yoon & Ari Onchwari, 2006) – the teachers did not explain to the students how the samples were collected. This was important for later reflections about the results. Even though the sources were a river, a creek, and a stagnant pond, the goal was to ensure that the water samples had different pollution levels. Bison Pond is located in an isolated area in the middle of the Brookfield Zoo, near Chicago. It has no tributaries, and neither animals nor humans have access to it. Therefore, it had no source of pollution (turbidity: 2.61 NTU, pH: 8.1, nitrogen: 0.03 mg/L, and phosphorus: 0.09 mg/L; collected by students several weeks prior to this activity). The Des Plaines River is near factories and has many tributaries (turbidity: 59 ppm, pH: 6.7–8.4, nitrogen: 0.65 mg/L, and phosphorus: 0.169 mg/L; Environmental Protection Agency [EPA], 1993, Nairn & Mitsch, 2000; Illinois Environmental Protection Agency, 2009). Therefore, there was a higher risk of pollution. Salt Creek is a tributary of the Des Plaines River (turbidity: 4–39 NTU, pH: 7.8, nitrogen: 0.44–0.84 mg/L, and phosphorus: 0.09–0.122 mg/L; Figueroa-Nieves et al., 2006; Heatherly et al., 2007). Therefore, there was a lower risk of pollution than in the Des Plaines River but a higher risk of pollution than in Bison Pond. All water samples with their respective organisms were returned after the activity to the exact locations where they were collected.

A total of 59 freshman students (20 students per class) participated in the study. Using 3x plastic box magnifying lenses, dual plastic magnifiers (3x and 6x), Battat two-way microscopes, and illuminated zoom microscopes (60–100x Lumagny illuminated pocket microscope), the students, in groups of 4–6, placed aliquots of water samples on Petri dishes, observed for 12 minutes for each location (2 evaluations per location [3] per group [4] per class [3]; N = 72 observations), and identified the organisms that were present in each water sample. ID books, ID cards, and a new ID-water-quality sorting sheet (Figure 1) based on tolerance levels for each type of organism were used for identification of macroinvertebrates (see timeline for additional provisions). Certain macroinvertebrates are less tolerant to pollution (Lenat, 1988). Therefore, they will be found more in pristine waters (like
Bison Pond). Other macroinvertebrates are more tolerant to pollution (Heatherly et al., 2007), and those will be found mostly in polluted waters (such as the Des Plaines River or Salt Creek). In this inquiry-based activity, the simplest premise of “measure one, compare at least two” was used, and the students measured water quality through an index (“measure one”) and compared three locations.

We designed a sorting sheet (Figure 1) based on Macro Mania (LaMotte Company, 2004), a classroom game that introduces the use of stream macroinvertebrates to determine water quality. Our sorting sheet presented three groups of organisms. Group 1 represented macroinvertebrates that were very sensitive to pollution and therefore could survive only in water of high quality. Group 2 represented a group of macroinvertebrates that were somewhat sensitive to water quality but could survive in waters that weren’t quite clean. Group 3 represented macroinvertebrates that were tolerant of poor water quality and could therefore survive in more polluted waters than invertebrates in Group 1 and Group 2 (Lenat, 1988).

The students counted the number of organisms of each type within each of the three groups on the sorting sheet (Figure 1). The total number of types of macroinvertebrate in each group was multiplied by a number. Group 1 was multiplied by 3, Group 2 was multiplied by 2, and Group 3 was multiplied by 1 (LaMotte Company, 2004). These values corresponded to ranges of tolerance levels (Fore et al., 1996). The sum of the three types of macroinvertebrate in each group that was previously multiplied by each respective number was assigned to a water-quality category: excellent, good, fair, or poor (see Figure 1). The hypothesis was that the Des Plaines River water would have the poorest water quality, with macroinvertebrates characteristic of polluted waters, and that Bison Pond would have the best.

Statistical Analysis: The students detected differences among sites (high, medium, low), and the teachers tested the data for normality and homoscedasticity to corroborate the students’ findings prior to performing analysis of variance (ANOVA). Because normality was not reached after any transformation, a Kruskal-Wallis ANOVA (Statview; SAS Institute, Cary, NC) was used to determine whether group results differed among sites. Statistical differences were detected by a nonparametric multiple comparison between treatments (NP_MCBT, P < 0.05; Siegel & Castellan, 1988). These results were later presented to the students for discussion.

Timeline
This type of activity is best conducted from spring through fall, when aquatic organisms are available and active. Water samples were collected 2 days prior to the activity. Several Petri dishes with macroinvertebrates from each site were prepared 2 hours before the activity, anticipating that students might not be able to find macroinvertebrates in their water samples. That way, students have an alternative resource to see and identify these organisms.

Results
Students found that Bison Pond’s water quality ranged from 5 to 24 (Figure 2). The maximum value of 24 corresponds to excellent water quality (Figure 1). Discrepancies in evaluations were due to students’ inexperience at identifying macroinvertebrates and to the short time available for identification. However, given that one group was able to obtain a water-quality level of 24 (excellent; see Figure 1), the sample may contain little if any pollution. The samples from Bison Pond were statistically different from the Des Plaines River and Salt Creek samples (Kruskal-Wallis ANOVA, N = 72, df = 2, P = 0.0030). The water quality of the Des Plaines River ranged from 1 to 12 (Figure 2). These values correspond to waters that are of poor to fair quality (Figure 1). Even though we assumed that this dark brown river had more pollution than its tributary, the Salt Creek, the students found more organisms in the Des Plaines River samples; however, these values were not statistically different between sites (NP_MCBT, P > 0.05). The Salt Creek samples ranged in quality from 1 to 8 (poor; Figure 2).

Discussion & Implications for Teaching
The objective of this investigation was to promote a semi-guided inquiry activity in the classroom and encourage students to think critically, make their own conjectures, and go through the inquiry cycle using macroinvertebrates to assess water quality. The original question was: “Is the quality of a water sample with no source of pollution different from samples that have been exposed to chemical run-off and human activity?” Another goal was to meet the National Science Education Standards that require students to understand the processes of scientific inquiry to “investigate questions, conduct experiments and solve problems” (Illinois State Board of Education, 2007). Furthermore, this experience was essential to increase the students’ awareness of threats to the community and broaden their perspective on how to identify and deal with environmental problems.

The students found that a water source that is protected from contamination had increased populations of mayflies, caddisflies, and stoneflies, corroborating that Bison Pond had the highest water quality. By contrast, water samples that were assumed to have pollutants from human activity such as fertilizers from lawns, pet excrement, motor oil, and litter negatively affected these organisms.
Accordingly, other organisms less vulnerable to pollution could thrive, such as midges, water fleas, leeches, and mosquitoes, which were the macroinvertebrates that the students found in samples from Salt Creek and the Des Plaines River.

We are uncertain why the abundance of macroinvertebrates was lower in Salt Creek than in the Des Plaines River; we had predicted the opposite pattern. Salt Creek is a tributary of the Des Plaines River, and its watershed is protected for recreation and land use (Northeastern Illinois Planning Commission, 2004). Therefore, we assumed that pollution would be lower and that macroinvertebrates of Groups 1 and 2 (Figure 1) would be more abundant here than in the river. The Des Plaines River transforms from a prairie creek to a suburban stream and a large urbanized river, flowing 150 miles through Wisconsin before crossing into Illinois, at which point it flows northwest of Chicago to become a major industrial waterway (EPA, 2004). Given this information, we assumed that the Des Plaines River had higher pollution levels than Salt Creek. Therefore, we hypothesized that organisms from Group 3 (Figure 1) were going to be more abundant in these samples than in Salt Creek water samples. Therefore, this experiment would need to be repeated in order to identify whether our conclusion always applies.

When the students reflected about the macroinvertebrate abundances in the creek and the river, they wondered whether water samples from these two sites were taken at different depths. Scooping water from the top of the river could have affected the efficiency of macroinvertebrate collection, because many macroinvertebrates are found in the bottom of the river within sediments or between submerged stones (Feminella & Flynn, 1999). This collection procedure might explain discrepancies in our results and expectations. In spite of these discrepancies, our goal was to promote students’ reflection and new questioning about the results, completing the inquiry cycle. Furthermore, students recorded and represented data in charts and graphs, discussed the importance of averages, and analyzed data using the calculated indexes to identify the quality of the waters, all of which are actions used in scientific research.

This inquiry-based activity also promoted reflection about pollution. After the students were shown a map of the sample-collection sites (Appendix), they realized that it would have been interesting to carry out several trials with different samples along the river. They hypothesized that samples near a golf course (where pollutants from fertilizers and pesticides might be higher) would have lower quality than samples taken at the forest preserve, where pollution might be lower. Reflecting about these possible outcomes increased the students’ awareness of threats to the community. They also discussed connections to the real world by considering how to deal with environmental problems. If chemical run-off was affecting water quality near golf courses, this could be used to promote new legislation for pollution control to fine the culprit(s) (Bajaj, 2010, Elks, 2010).

The students also postulated other new questions to modify the investigation, such as “What is the water quality on the north part of the river, compared to samples on the south which are near a dam?” and “Will the macroinvertebrates in one place differ if we compare collections in the spring to those in the summer?”

Suggestions & Further Applications
Let the students investigate macroinvertebrates on the Web: the types of macroinvertebrates expected in waters with different pollution levels, the roles of macroinvertebrates in their ecosystems, their habitat needs, their tolerance of pollution levels, and what they eat (are they carnivores or herbivores?). By investigating these organisms, students will realize the type of pre-research activities that scientists need to do for their investigations. In addition, they will become familiar with the animals they might encounter, get excited when they find them, and make a connection between macroinvertebrates and aquatic food webs.

As an alternative, use a monitor connected to a microscope to let students view the discoveries of other students or your pre-prepared Petri dishes with macroinvertebrates; this will give them a visualization of what might be found if they are unable to find macroinvertebrates in their samples.

Conclusions
Studying and discussing the quality of water using macroinvertebrates is a quick and fun way to encourage students’ interest in scientific discovery. It is also a way to fill in the gap between the formal education system and the students’ real life, bringing perspective on how to identify environmental problems and insight into dealing with them and taking action. Incorporation of inquiry-based classes in the curriculum is the key to promote students’ critical thinking and habits of mind, to empower them to become independent and life-long learners, to teach them to confront problems, to generate and test ideas for themselves, and to question everyday values and their understanding of the world. We hope that you and your students will find exploring the aquatic macroinvertebrate fauna as exciting as we do.

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References


HELENA PUCHE is Adjunct Assistant Professor at the University of Illinois at Chicago, 815 West Taylor St., Chicago, IL 60607; e-mail: hpuche@uic.edu.

JAME HOL'T is a ninth-grade Biology Teacher at Riverside Brookfield High School, 160 Ridgewood Road, Riverside, IL 60546-2908; e-mail: holtj@rbhs208.org.
Appendix. Detailed map of the Des Plaines River, Bison Pond, and Salt Creek sampling locations. Students’ reflections included sampling the Des Plaines River near the Riverside Golf Club (where chemical runoffs from fertilizer and pesticides might be higher than at other sites) and the Cook County Forest Preserve (where pollutants are assumed to be lower).