# Qualitative Investigation and Identification of Odonate Larvae at Brookhaven National Laboratory

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Participant: _____
Signature

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## ABSTRACT

Qualitative Investigation and Identification of Odonate Larvae at Brookhaven Laboratory. SUSAN M. COSTA (Community College of Rhode Island, Warwick, RI 02886) DR. TIMOTHY M. GREEN (Brookhaven National Laboratory, Upton, New York 11973-5000).

The Order Odonata is believed to have appeared about 250 to 300 million years ago during the Carboniferous period, which was part of the Paleozoic Era. Based on fossil records dragonflies of this time period were huge with wingspans that measured up to 70 cm (27.5 in.). Over time they have diverged into two Suborders, Zygoptera (damselflies) and Anisoptera (dragonflies). The purpose for conducting the research at Brookhaven National Laboratory (BNL) was to identify and catalog the specimens found at the laboratory. The goal was to identify the specimens down to the species level whenever possible, and to compare the distribution of the various species across all ponds studied. Some of the ponds selected for the research have historically been used by the tiger salamander (*Ambystoma t. tigrinum*) and are designated as P-1, P-2, P-6, P-7, P-9, and P-10 (P standing for pond). Several of the ponds near the Relativistic Heavy Ion Collider (RHIC) were also sampled. One of the ponds is referred to as the "9 O'clock Pond" and the others are referred to as Recharge Ponds A6a, A6b, A6c and A6d. The Peconic River was also sampled specifically for Ebony Jewelwing larvae because it is the only location on the Lab where the adults have be found. Once a pond was selected, a dip net, seining net, or a minnow trap was used to collect specimens out in the field. Specimens were temporarily stored in Ziploc bags and placed in a cooler on ice. The purpose for the ice was to keep specimens alive and immobile while identifying them in the lab. Once specimens were brought into the lab, a dissecting microscope and taxonomic keys were used to identify specimens to genus and species. By project end, seven dragonfly larvae have been identified to the species level, two have been identified to the genus level and three damselflies have been identified to the species level. Anisoptera larvae identified at BNL include *Anax junius* found at P-10 and 9 O'clock Pond, and Recharge Basins A6a, A6b, A6c and A6d; *Aeshna umbrosa* found at A6d and the Peconic River; *Anax longipes* found at P-7, *Somatochlora williamsoni* found at P-10, *Symptrum janeae* and *Pachydiplax longipennis* found at P-1, *Libellula semifasciata* found at P-7; and *Dythemis sp.* found at P-2 and P-7, but could not be identified to the species level. Zygoptera identified at BNL were: *Enallagma durum* found at P-10, *Lestes eurinus* found at P-1, and *Lestes unguiculatus* were found at P-2 and P-1. Future research may expand to other ponds as part of a larger on going biotic inventory of the lab.

## INTRODUCTION

Research conducted during the summer of 2003 focused on the diversity of the aquatic insects in the Order Odonata, including the identification and cataloging of the different species found at Brookhaven National Laboratory (BNL). Research on both adults and larvae were conducted simultaneously as part of an ongoing larger investigation of the flora and fauna found at the Lab.

The Order Odonata is believed to have arisen about 250 to 300 million years ago during the Carboniferous period, which was part of the Paleozoic Era [1], this was about 1 million years before the dinosaurs walked the earth. Dragonflies of this time period were huge with wingspans that measured up to 70 cm (27.5 inches). As time passed, the odonates diverged into two suborders, Anisoptera (dragonflies) and Zygoptera (damselflies). The most noticeable difference between the two suborders are the adult wings, anisopterans having dissimilarities in the fore and hind wings and zygopterans having similar fore and hind wings. [2] As larvae, anisopterans are longer in length with larger abdomens, where as zygopterans are very slender, shorter in length and their gills resemble small feathers at the end of the abdomen.

Larval odonates fall into the macroinvertebrate category of benthic dwellers (bottom dwellers), are recyclers of nutrients, are an important part of the food web [3]. As part of the food web they are both predator and prey. As predator they are opportunists preying on most anything that is the same size as them or smaller [2]. Their diet consists of mosquito larvae, other insect larvae including other odonates, worms, tadpoles, snails, and even some small fish [2]. As prey they are eaten by, larger odonates, frogs, salamanders, and fish.

Unable to swim away, macroinvertebrates are often captive spectators in ponds and lakes and can be an indicator of water quality depending on which species are present [3]. Although

odonates are one of the indicator species of water quality, they are able to tolerate a minimal amount of environmental change such as water pollution, temperature or, pH change, unlike some aquatic species that are unable to tolerate any level of water contamination or change [3].

Many damselflies and a few dragonflies lay their eggs in the tissue of aquatic plants (endophytic egg layers [4]) due to their strong ovipositors (female genitalia found on the s9 of the abdominal segments), but many lay their eggs just on the surface of aquatic plants, in the mud at the edge of the water, or in the water itself (these are exophytic egg layers [4]) [2].

Odonates are hemimetabolous insects, which means they only have 3 stages of life, egg=> larvae =>adult; unlike butterflies that have 4 stages of life, egg=> larvae => pupa=> adult [5].

A week or so after eggs have been laid in the water; larvae hatches and they begin their lives as predatory feeders, which may last for several months or years in their aquatic realm [2]. Odonates are opportunistic hunters having keen eye site, due to a compound eye, which utilizes 80% of their brain function. The larvae have a hinged labium (part of their mouth) that in some cases stretches out to two-thirds the length of their body [5]. The name odonate comes from the Greek word “odon”, meaning tooth [5], which can be found in abundance within their mouths. Odonate predatory behavior is to either conceal themselves under mud and leaves, sprawled out under sediment, or clinging to aquatic vegetation as they lie in wait for some unsuspecting prey [5]. Odonate larvae growth rate depends mainly on two things, water temperature and abundance of food [2]. Dragonfly larvae generally do not grow any larger than 50-55mm in length and damselfly larvae being smaller, around 30-35 mm in length [9].

Because odonates are of the Phylum Arthropoda (which means they have an exoskeleton), in order for them to grow in size they must molt several times while in the larval

stage of life. These stages of molting are referred to as instars [6]. Odonates, because they do not have a pupa stage, grow their wings on the outside of their bodies as wing pads. These wing pads appear after about 6 or 7 instars, with most having a between 10-15 instars [5].

Some of the Anisoptera that are considered “sprawlers” have short antennae and a flatter wedged-shaped head. Often the middle pair of their six legs is modified and is much shorter than the anterior or posterior legs. They may also have a curved hook on the end, which is use for burrowing [5]. Sprawlers generally have longer legs; their bodies are covered in setae (hair) and they are most often vigilant hunters. The setae are used as a tactile organs aiding in the detection of prey. Their coloring is usually mottled green and brown, which helps them blend into their surroundings [5].

Most species the suborder Zygoptera and the remaining Anisoptera are “climbers” and perch camouflaged against aquatic vegetation, as they lie in wait for unsuspecting prey [4].

Because odonates are aquatic insects the larvae live in the water until they emerge as adults at the end of their final instar. During the larval stages, they have closed respiratory systems and breath via gills. Larval gills of Anisoptera are found in the anus, from which they respire by pumping water in and out. Within the anus, gasses are exchanged through a thin layer of skin in the rectal wall. This pumping of water through the anus is also used to propel them through the water quickly when needed [5]. Zygoptera respire mainly through three gills, which look like feathers extending past the end of the abdomen. In later instars, gas exchange also occurs within the wing pads [5]. Once odonates emerge and become adults, they respire through an open system found on their thoracic (spiracles) and an abdominal segments (auricles) [5].

## MATERIALS AND METHODS

Some of the ponds selected for the research have historically been used by the tiger salamander (*Ambystoma t. tigrinum*) and are designated as P-1, P-2, P-6, P-7, P-9 and P-10 (P standing for pond see figure 1). Several of the ponds found near the Relativistic Heavy Ion Collider (RHIC) were also sampled. One pond is referred to as the "9 O'clock Pond" (figure 1), while the other four bodies of water found in this enlarged section on the map are referred to as Recharge Basins A6a, A6b, A6c, and A6d. The Peconic River was also sampled specifically for Ebony Jewelwing larvae because it is the only known location on the Lab where the adults are found. Once a body of water was selected for sampling, chest waders were worn while wading out into the water and a dip net was used to collect odonate larvae from the bottom and the vegetation. Larger specimens were also successfully caught using seining nets and minnow traps. As specimens were collected from the water they were placed in various containers, including a small aquarium, magnifying specimen jar, and Ziploc bags. Specimens were placed in a cooler with ice to induce a state of torpor. A digital camera was used to take pictures of specimens before and after identification.

While in the lab, various tools were used including petri dishes, dissecting tweezers, metric ruler, and specimen trays in order to examine the larvae. A dissecting microscope was used to magnify features specified in the taxonomic keys ( [5], [7], and [8]) in order to identify collected specimens to genus and species. Once specimens were identified, digital images were taken and larvae were released back to their original ponds, basins, or river.

## RESULTS

Table 1 is a listing of the various species of odonate larvae identified at Brookhaven National Laboratory during the summer of 2003, and their locations. The various ponds surveyed (P-1, P-2, P-6, P-7, P-9, P-10, 9 O'clock pond at the RHIC Ring, as well as the Recharge Basins 6a, 6b, 6c,and 6d at the RHIC Ring, and the Peconic River) were distributed throughout the Lab property (see figure 1 for Lab layout). Several of the species were reaffirmed by the adult odonata research that was being conducted simultaneously at the Lab by Another intern. Pond 1 was found to have the highest species richness of odonates (as shown in figure 2) and 9 o'clock Pond was found to have the lowest number of larval odonate species (as shown in figure 3).

## DISCUSSION AND CONCLUSION

The purpose of the odonate larvae research was to identify as many of the species found at the Lab as possible in the given time allowed for the research project. Of the two suborders Zygoptera (damselfly) and Anisoptera (dragonfly), three species of Zygoptera were identified early on in the sampling of several of the ponds. They were the Big Bluet (*Enallagma durum*), Amber-winged Spreadwing (*Lestes eurinus*), and Lyre-tipped Spreadwing (*Lestes unguiculatus*), each was found to be common to New York [8]. All specimens appeared to be in their later instars, which meant within a short period of time they should have emerged as adults once they were returned to their original bodies of water. Only one of the Zygoptera larva identified matched adults being identified and cataloged at the Lab by another intern. Although Zygoptera were collected throughout the research project, many of the larvae found later were in very early instars, and were too small to clearly identify to genus and species. The Peconic River was also

sampled in late July specifically for the zygopteran larva of the Ebony Jewelwing (*Calopteryx maculata*). This species requires slow moving water, and the Peconic River is the only location on the Lab's property that fits this specification [8]. The search for larval *Calopteryx maculata* was unsuccessful due as no larvae were found. Adults were found and observed around the river in abundance flying and perching in tandem (mating formation) as well as laying eggs. This would suggest that if we were able to return later in the season that it would be possible to retrieve larvae of this species and positively identify them. Or arrived earlier in the season.

Seven dragonfly larvae were identified to species level, through the research period, many in late instars as well as several in early instars. Two dragonflies were identified to the Genus level, but the species were unclear. Of the unclear species, one was sent out to Dr. Michael L. May for his expert assistance in identifying odonate specimens, and he was able to identify it to a Spot-winged Glider (*Pantala hymenaea*), which is often easily mistaken for other species because of the crenulations (scalloped edging) on the labial palps (part of the mouth). Four of the Anisoptera identified matched adults that had been clearly identified during the summer at the same pond locations. These dragonflies were the Comet Darner (*Anax junius* ), Common Green Darner(*Anax longipes* ), Cherry-faced Meadowhawk ( *Symptrum internum*) and the Blue Dasher (*Pachydiplax longipennis* ). The latter three were a common site at many of the ponds sampled as well as being very common to New York [7]. Several of the larval specimens that were in their later instars, were kept in an aquarium to observe their emergence, confirming their identification once they became adults. After emergence and confirmation the adults were set free. Pond 1 was found to have the most diversity if all the bodies of water surveyed and 9 o'clock Pond had the least diversity (figure 3).

Continued odonate diversity research is being considered for future summer projects as part of the continued flora and fauna identification being conducted at the Lab. This is due to the fact that the summer is the time of year when the most mating and emergence takes place, making it the prime season to conduct such research.

## ACKNOWLEDGEMENTS

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## References

[1] N.A. Campbell, J.B. Reece, "Biology" 6th ed., San Francisco: Benjamin Cummings, 2002, pp. 487.

[2] Terry T, Dragonflies of the Athabasca River Basin. 2002 June. Available from: Science Outreach-Athabasca. http://scienceoutreach.ab.ca/documents/brochure_dragonflies_1.doc

[3] Macroinvertebrates as Water Quality Indicators. 2003 June. Available from: http://www.lakecountyohio.org/soil/monitoring_information.htm

[4] Silsby J., A Beginner's Guide to Dragonflies. 2002 June. Available from: http://powell.coalgate.edu/wda/beginners_guide.htm

[5] R.W. Merritt, K.W. Cummins, “An Introduction to the Aquatic Insects of North America”, 3rd. ed., Iowa: Kendall/Hunt, 1996, pp.164-167

[6] An Introduction to Insect Life Cycles. 2003 June. Available from: http://www.earthlife.net/insects/lifecycles.html

[7] J.G. Needham, M.J. Westfall, Jr., M.L. May, “Dragonflies of North America”, Gainesville: Scientific Publishers, 2000,

[8] M. J. Westfall, Jr., M. L. May, “Damselflies of North America”, Gainesville: Scientific Publishers, 1996,

[9] G. Zieman, “Dragonfly”, 2003 June. Available from: http://www.bv229.k12.ks.us/biophilia/wsc/pondanimal/dragonfly.html

**Table 1**

| Family | Scientific Name | Common Name | Pond # | Date |
|---|---|---|---|---|
| Zygoptera | | | | |
| Coenagrionidae | *Enallagma durum* | Big Bluet | P-10 | 6/24/2003 |
| Lestidae | *Lestes eurinus* | Amber-winged Spreadwing | P-1 | 7/1/2003 |
| | *Lestes unguiculatus* | Lyre-tipped Spreadwing | P-2 | 6/19/2003, |
| | | | P-1 | 7/1/2003 |
| Anisoptera | | | | |
| Aeshnidae | *Anax longipes* | Comet Darner | P-7 | 7/10/2003 |
| | *Anax junius* | Common Green Darner | P-10, | 6/24/2003 |
| | | | 9 O'clock pond | 7/15/2003, |
| | | | A6a (middle of RHIC) | 7/23/2003 |
| | | | A6b | 7/23/2003 |
| | | | A6d, | 7/25/2003 |
| | *Aeshna umbrosa* | Shadow Darner | Peconic River | 7/25/2003 |
| | | | A6d | 7/25/2003 |
| Libellulidae (Subfamily Corduliidae) | *Somatochlora williamsoni* | Williamson's Emerald | P-10 | 6/24/2003 |
| | *Pantala hymenaea* | Spot-winged Glider | A6d | 7/25/2003 |
| Libellulidae | *Dythemis ?* | Setwing | P-7 | 6/19/2003 |
| | | | P-2 | 6/19/2003 |
| | *Symptrum internum* | Cherry-faced Meadowhawk | P-1 | 7/1/2003 |
| | *Pachydiplax longipennis* | Blue Dasher | P-1 | 7/1/2003 |
| | *Libellula semifasciata* | Painted Skimmer | P-7 | 7/10/2003 |

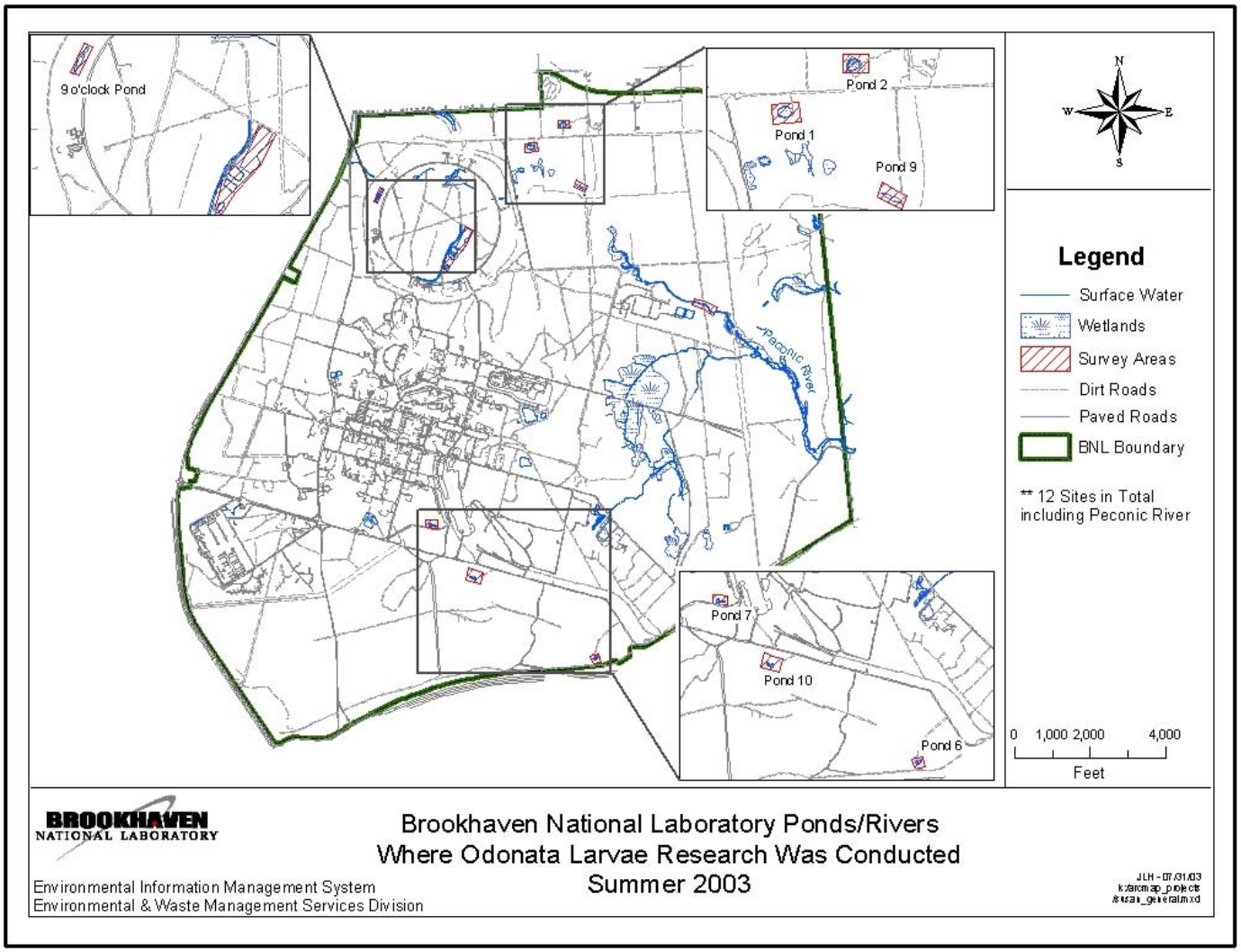
9 o'clock Pond
Pond 2
Pond 1
Pond 9
Peconic River
Pond 7
Pond 10
Pond 6
N
W
E
S
Legend
Surface Water
Wetlands
Survey Areas
Dirt Roads
Paved Roads
BNL Boundary
** 12 Sites in Total including Peconic River
0 1,000 2,000 4,000
Feet
BROOKHAVEN NATIONAL LABORATORY
Brookhaven National Laboratory Ponds/Rivers
Where Odonata Larvae Research Was Conducted
Summer 2003
Environmental Information Management System
Environmental & Waste Management Services Division
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**Figure 1**

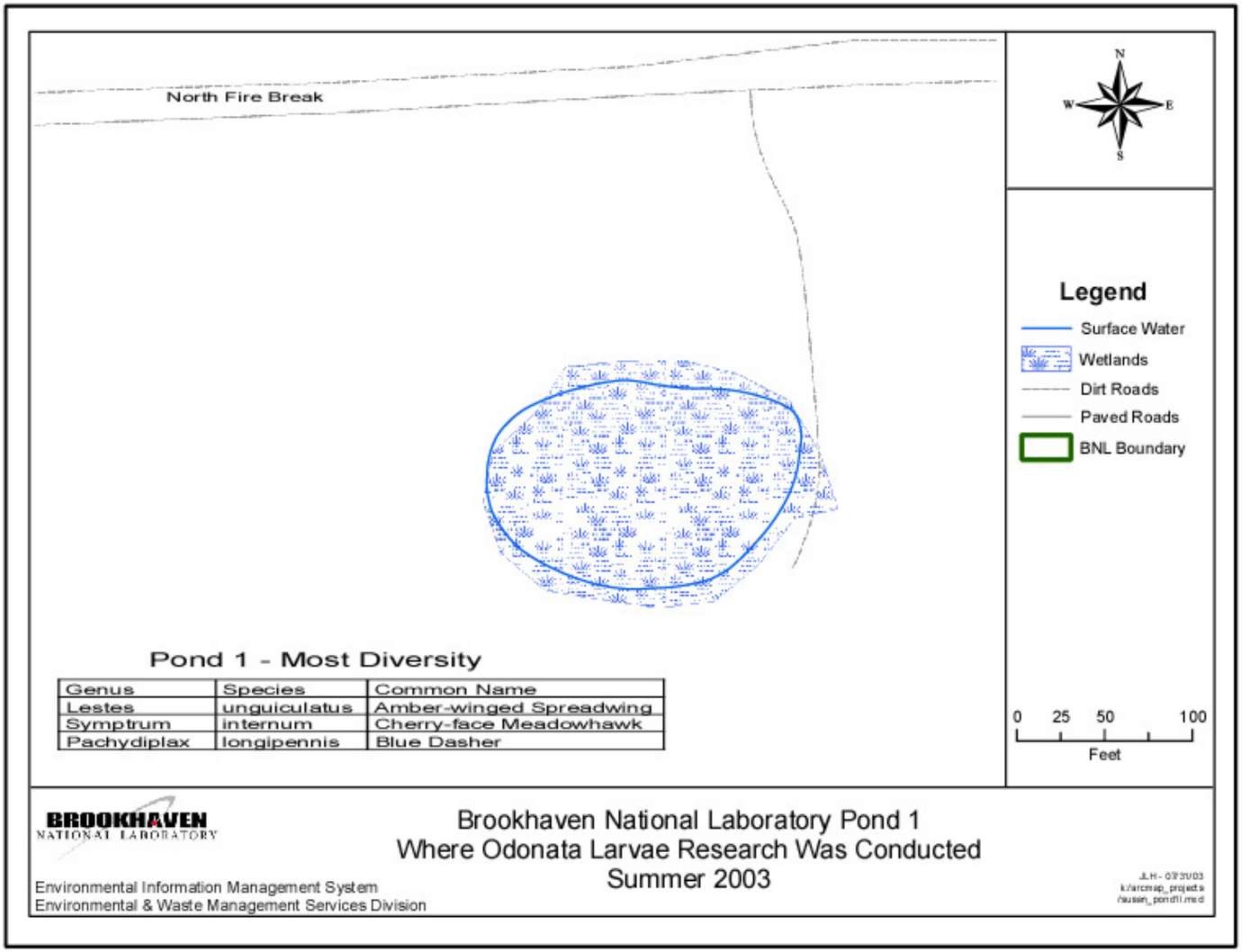
North Fire Break
N
W
E
S
Legend
Surface Water
Wetlands
Dirt Roads
Paved Roads
BNL Boundary
0 25 50 100
Feet
Pond 1 - Most Diversity
Genus | Species | Common Name
Lestes | unguiculatus | Amber-winged Spreadwing
Symptrum | internum | Cherry-face Meadowhawk
Pachydiplax | longipennis | Blue Dasher
BROOKHAVEN NATIONAL LABORATORY
Brookhaven National Laboratory Pond 1
Where Odonata Larvae Research Was Conducted
Summer 2003
Environmental Information Management System
Environmental & Waste Management Services Division

**Figure 2**

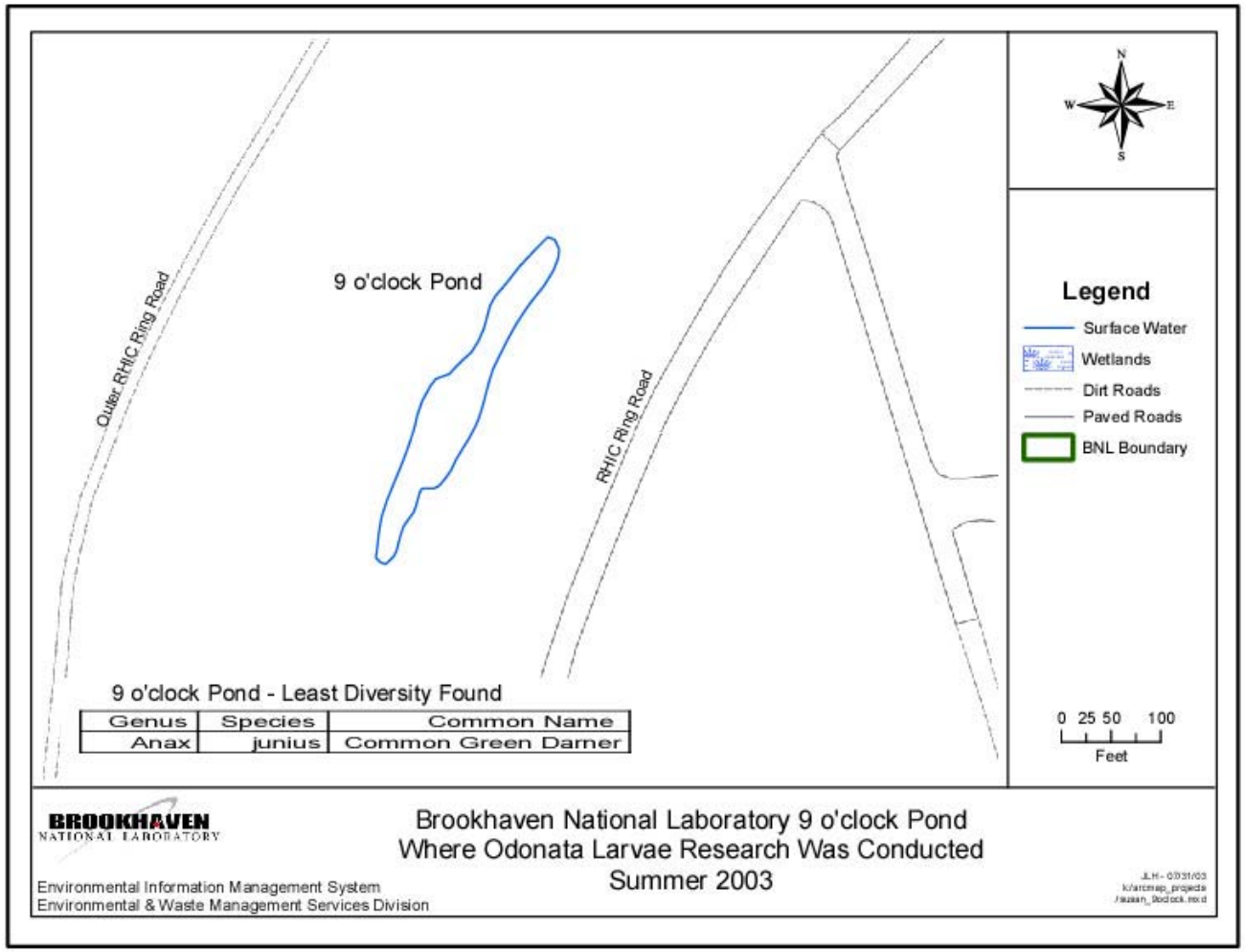
9 o'clock Pond
Outer RHIC Ring Road
RHIC Ring Road
N
W
E
S
Legend
Surface Water
Wetlands
Dirt Roads
Paved Roads
BNL Boundary
0 25 50 100
Feet
9 o'clock Pond - Least Diversity Found
Genus Species Common Name
Anax junius Common Green Darner
BROOKHAVEN NATIONAL LABORATORY
Brookhaven National Laboratory 9 o'clock Pond
Where Odonata Larvae Research Was Conducted
Summer 2003
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**Figure 3**