DRAGONFLY MERCURY PROJECT

SAMPLING GUIDE FOR THE COLLECTION OF DRAGONFLY LARVAE SAMPLES FROM NATIONAL PARKS FOR MERCURY ANALYSIS

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Revised March 2018
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Scope and Application

Background

Mercury (Hg) is a globally distributed contaminant that can harm human and wildlife health. In its toxic methylated form, mercury bioaccumulates (builds up) in aquatic and terrestrial foodwebs. Effects can include reproductive and neurological impairment. Due in part to emissions from coal-burning power plants, even remote national park environments receive mercury deposition from the atmosphere. (See https://www.nps.gov/subjects/air/nature-toxics.htm for background on mercury in national parks.)

Mercury threatens natural resources the National Park Service is charged with protecting. This citizen science project encourages students and visitors in national parks to collect dragonfly larvae for mercury analyses. Dragonfly larvae (Odonata: anisoptera) could serve as indicators of ecosystem health by characterizing the risk and potential toxic effects of mercury. These aquatic macroinvertebrates are long-lived (up to 5 years as larvae) before emerging as adult dragonflies, widespread across the U.S., predatory (i.e., prone to higher concentrations of mercury), important prey for fish species, and they reflect the mercury sensitivity of a specific watershed. Moreover, they are relatively easy to collect!

The Dragonfly Mercury Project (DMP) connects people to parks, advances the NPS educational mission, fosters biodiversity discovery opportunities, and provides baseline data to better understand the spatial distribution of mercury contamination in national parks. The DMP expands the geographic scope of research previously conducted by scientists and citizen scientists, and provides data that can be compared across parks. Early data are being used to develop hypotheses regarding whether mercury varies with odonate larval body size or by family, or whether a site’s landscape setting drives variability in mercury in odonate larvae. See project webpage, available at http://go.nps.gov/dragonflymercury. Educational content and lesson plans for use by teachers and NPS staff are also available (e.g., http://go.nps.gov/dragonflyeducation, http://participatoryscience.org/project/mercury-watersheds), ultimately helping to raise awareness about mercury impacts. The DMP enlightens a new generation of citizen scientists about the connection of all living things and the influence humans have upon natural systems, and how environmentally-responsible decisions can protect our parks and the planet.

The citizen science effort to collect dragonfly larvae from national parks for mercury analysis started in 2011 with four NPS units in the Northeast and Great Smoky Mountains. It expanded in 2012 to include 11 units, and in 2013 to include 23 units across all seven NPS regions: Northeast, Southeast, National Capitol, Midwest, Intermountain, Pacific West, and Alaska. The project continued to grow from 33 units in 2014 to 49 units in 2015. In 2016, the year of the National Park Service Centennial, Dragonfly Mercury Project participants collected 2,475 dragonfly larvae from 169 sites in 65 NPS units. In 2017, participants collected 1,840 dragonfly larvae from 127 sites in 56 NPS units and National Natural Landmarks. More than 90 NPS units and 3,000 citizen scientists have participated in the DMP in its first seven years! In 2018, we are continuing the effort by sampling some new parks and supporting the resampling of some parks that have already participated. Citizen scientists involved include students ranging from elementary school to college-aged, park visitors, and BioBlitz participants.
Study Design
The optimal study design is 3 sites per park that represent a gradient in mercury-relevant chemistry; or a gradient in landscape conditions, such as a high and low elevation, amount of wetlands adjacent to or upstream from the site, or forested versus urban land cover. Consider scoping the sampling locations in advance of collecting samples. Does the proposed sampling location have a healthy population of dragonflies? (A sample size of 15 dragonfly larvae per site is preferred.) Will nearby riparian flora and fauna get trampled? Is the site spacious enough for a group?

Research Permit
A park research permit will be needed to conduct this project. In addition, in some cases, a state permit may be needed to collect dragonfly larvae. Example text for the permit application can be found in Appendix A of this sampling protocol. The DMP’s fact sheet and other materials will provide further assistance. (See URL’s above).

Safety
Field work carries some inherent risks, and the safety of study participants is a primary concern. The varied and spectacular resources of our national parks, and the engagement of citizen scientists who are potentially less skilled in field work and the associated hazards, present a challenge to safely conduct this project. Further, travel in remote areas may be required to conduct science that has meaningful inference to the parks’ diverse landscapes, increasing risks. Be attentive of the possible risks and refer to a detailed Safety Standard Operating Procedure (SOP) or to a Job Hazard Analysis (JHA) that is developed specific to surface water sampling, backcountry work, traveling in high elevations, poison ivy, tickborne disease, etc. (Contact colleen_flanagan_pritz@nps.gov for draft JHA.) Riparian edges can be very slippery, as are stream and lake bottoms. High water levels and turbulent flows may cause an individual to lose balance in the water. Collectively define, communicate, and enforce your park’s safety philosophy, standards, and guidelines; follow the tenets of Operational Leadership (e.g., brief your team prior to sampling on mission elements: Communication, Contingency Resources, Environment, Planning, Task Complexity, Supervision, Team Fitness, and Team Selection) and other relevant safety trainings such as swift water rescue; allow sufficient time for oversight and interaction with your group; and follow Departmental guidelines on the use of personal protective equipment, such as personal flotation devices (PFDs). We highly recommend citizen scientists use PFDs when sampling in lentic (still) or lotic (moving) water bodies, and in fact, USGS scientists are required to wear PFDs. Nothing in this program’s mission, project goals, or day-to-day objectives is so important as to compromise the health and well-being of those participating in this project.
Overview

This protocol has several sections. Each sampling procedure is fairly straightforward, after you’ve gotten the hang of it. You should read each section’s specific instructions on the following pages. The general structure is as follows:

1. **Prepare by gathering field gear.** Some gear is provided in the sampling kit, but you’ll need to round up a few items. Check what’s needed several weeks before sampling (in case you need to order something).

2. **Collect!**

   **First, collect dragonfly larvae samples.** Many hands sampling makes this go quicker. Larvae are collected using nets and temporarily stored in a small tote or dishpan with lake water until being sorted. Once at least 10 larvae (but ideally 15) have been collected, samples are sorted, and up to 15 individuals are selected for mercury analysis. These are measured, identified to family, then individually bagged to prevent contamination.

   **Second, observations about the site.** Many are qualitative/visual, but they are still very useful data. You know your site best and you can help the project scientists understand your watershed. We also encourage observations through [iNaturalist.org](https://www.inaturalist.org).

3. **Ship your samples.** You’ll ship dragonfly samples overnight in a cooler with dry ice. Dragonflies should go in a freezer after the sampling trip is over. They can stay in your freezer for several weeks, if you have multiple sites to sample over extended time periods.
1. Prepare

The following sampling supplies are needed to collect dragonfly larvae samples from each site. All supplies will be provided in the sampling kit, with the exception of those materials in **bold**. Participating parks must provide those items separately.

**Materials**

**Dragonfly sampling:**
- **Nets** – D Nets or dip nets
- **Clean, white dishpan, bucket, or ice cube trays**
- **Clean, new plastic spoons**
- **Brand new zipper-seal bags – 30 small size + 3 large size**
- **Powder free Nitrile gloves (blue or purple color)**
- **A plastic ruler with mm scale – clear is best**
- **Tags for outer bag, pre-printed with ID codes**

**General field supplies:**
- **Sharpie marker and pencil/pen**
- **Field sheets**
- **Dragonfly identification cards**
- **One cooler**
- **Trash bag**
- **Optional – hand lens, GPS, camera, macroinvertebrate field guide, waders**
- **Citizen science groups must provide their own PFDs (personal flotation devices)**

**Shipping:**
- **Dry ice for dragonflies**
- **Dry ice label and pre-paid shipping labels for return shipping**

**Resources:**
- See the project web site for educational tools, information about mercury and dragonflies, curriculum activities, and news about previous sampling efforts.
- Watch the training videos available at: [http://www.schoodicinstitute.org/what-we-offer/educational-scientific-partnerships/dragonfly-mercury-project/](http://www.schoodicinstitute.org/what-we-offer/educational-scientific-partnerships/dragonfly-mercury-project/)
- Follow “Six-Legged Scouts in the National Parks” on Facebook for updates and news.

*More information on nets in Appendix A.*
2. Collect samples

- Collect dragonfly larvae.
- Repeat the sampling for 3 water bodies per park when available.
- We highly encourage you to take photos of the sampling sites and of the collection efforts. Images of engaged NPS employees (in uniform) with citizen scientists are especially preferred.
- Take observations using iNaturalist (see Appendix D for instructions).
- Reference Appendix E on “Sacrificing Dragonfly Larvae for Science” in order to address potential ethical concerns raised by your team of citizen scientists.

Procedure

1. **Look around.** Locate likely habitats for dragonflies: vegetated bank margins, snags and logs, aquatic vegetation and decaying organic matter, silt/sand/gravel substrate. (See Appendix F on how to find dragonfly larvae.)

2. **Note observations and finish completing the field sheet.** Take a site photo!

3. **Rinse the nets and dishpans** with water from the site you are sampling.

4. **Partly fill dishpan** or bucket with water from the site. Ice cube trays are nice sorting containers too – they can be filled with stream/pond water as well.

5. **Jab** your net into the likely habitats. Start downstream first, and work upstream if in flowing water. Jab a few times then sweep the net up to the surface. Empty the net into the dishpan or bucket – either by tipping it into the bucket or plucking (with a Nitrile-gloved hand or a plastic spoon) larvae directly from the net. Spend a few minutes jabbing and emptying the net. If you don’t find much after a few minutes, move to another area within the same waterbody. See a video demonstration of collecting dragonfly larvae: [https://youtu.be/YXXCOU2MxHg](https://youtu.be/YXXCOU2MxHg)

6. If your team consists of multiple hands, **record** dragonfly specimens with iNaturalist (App. D).

7. **Disinfect** waders, nets, and any other gear that has been in the water before moving to your next site. **Consult your permit** since some parks have different protocols for disinfection. Most (but not all) will specify a 5% or 10% bleach solution, which is a mixture of household bleach and water.

*Citizen scientists search for dragonfly larvae at Lewis & Clark National Historical Park, OR.*

*Dishpan and ice cube tray ‘holding tanks’ streamside with larvae that have been collected, and selection and bagging of specimens to be submitted (at Great Smoky Mountains National Park, NC/TN).*

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Keep it Clean! Dragonfly Larvae Sampling Protocol

It’s easy to contaminate samples with mercury because it’s all around us: in our hair, in soil, leaves, etc. So we need to keep everything ultra-clean.

- Do not touch the samples (individual dragonfly larvae) with anything except a clean plastic spoon or powder-free nitrile glove (purple gloves).
- Never touch the inside of the inner bag – the first bag you put the sample into.
- Think of each individual sample as a fresh start – everything needs to be clean between samples and be treated just like the first sample you took.
- Work in pairs and use the “Clean Hands–Dirty Hands” method:
  1. One person is in charge of the sample bags (“Dirty Hands”) and the other will pick up and place the samples in the bags (“Clean Hands”).
  2. Clean Hands puts on a glove (one gloved hand should suffice to pick and bag larvae). Once the glove is on, Clean Hands touches nothing but the sample and any pre-cleaned supplies. Dirty Hands does not need to wear gloves.
  3. Choose, measure, & bag up to 15 individuals per waterbody/site.
  4. If your team consists of multiple hands, record specimens using iNaturalist (App. D).
  5. Using a nitrile-gloved hand, or plastic spoon, Clean Hands places one dragonfly into a sample bag while Dirty Hands holds the bag. Nothing should touch the inside of the bag except the dragonfly larva. Clean Hands seals the bag.
  6. Dirty Hands uses the plastic ruler, held up to the individual in the bag, and measures the body length in millimeters (mm) – from the front of head to end of tail spine. Most larvae will range between about 10–40 mm, as a reference.
  7. Dirty Hands fills out a tag for the bagged individual and slips it and the inner bag into the outer bag. Your samples have an ID code pre-printed on the labels. Use these codes to record the lengths on the field sheet and to refer to a particular sample in your notes.
  8. Dirty Hands seals the outer bag.
  9. Be ‘clean’ with your spoon and/or gloves. If they have been tossed on the ground or contaminated – say, by brushing back your hair – start with a fresh one. Swish your spoon or gloved hands in pond water (in the dishpan) between samples to ensure there’s no carryover from one sample to the next.
  10. Go to the next individual and repeat. Place all of the small double bags into one or more of the larger zipper seal bags for storage and to keep them safe and together. (See Section 4. Choose, Measure, & Bag)
  11. Gently return extra site water and invertebrates to the sample site.
  12. Pack out all trash, disinfect gear.
3. Identify

Please identify dragonfly larvae to family if you are comfortable doing so. There are only 6 major families in most places (see following pages). Most identifications can be accomplished in the field with a hand lens and guide book, or you can bag samples (Section 4) and bring them to a clean indoor workspace to look more closely.

The online dragonfly family identification key is available to assist efforts at: http://go.nps.gov/dragonflylarvaeflashcards

The online video “Identifying Maine Dragonfly Larvae to Family” (http://vimeo.com/76713446) will also assist with identification because similar larvae families are found outside of Maine.

Identifying to family requires careful inspection. The ID will be checked once the laboratory receives samples, and some samples will be further identified to species through genetic barcoding or a taxonomist, but identifying samples when they are fresh is helpful in the event of damage during shipping.

A. It is helpful to distinguish between the two Suborder levels: Dragonflies (Anisoptera) vs. Damselflies (Zygoptera). Although both are predators, the Anisoptera bodies are more robust and may therefore consume and store more mercury. Anisoptera also tend to have longer aquatic lifespans (up to 5 years) than Zygoptera (usually 1 year).

B. It takes specialized training to classify dragonfly larvae to the species level; we will do this for some individuals in the lab and provide the identification information to you along with the mercury and other data. This information is especially useful for biodiversity discovery activities!

4. Choose, Measure, & Bag

- Look at the samples you’ve collected. If you have identified to family already, please choose at least 3 individuals in each of the families you found. If you only found one individual in a certain family, please don’t submit that one. Ideally, at least 10 and up to 15 individual larvae will be selected per site. We would like to analyze samples from across the range of sizes present at a site, however, larvae larger than 15 mm are easiest to measure mercury in—please choose medium to larger samples over very small ones when possible.
- Given the risk of contamination, it is very important to follow the Keep it Clean! Protocol (above) while choosing, measuring, & bagging dragonfly larvae samples, and through the whole sampling outing.
- In sum, once samples have been collected, identified, and selected, each dragonfly larvae is double-bagged; placed first in an inner zipper bag, then with label placed in an outer bag, and stored on wet ice.
- Use iNaturalist to record each specimen collected. (See Appendix D for instructions.) If you are taking a photo of the specimen, consider using an iPhone/Android macro lens, if available, and doing so just before the larva is bagged to obtain the clearest picture possible!
Photos of exuviae: A. Anderson, Kingfisher Photography, Old Town, ME
Diagram: Ken Soltesz

Note: The illustration shows a typical Gomphid, with paddle-shaped antennae that have fewer segments; most larvae have long, filamentous antennae with more segments.
5. Store & Send

- In the field, put dragonfly sample bags into a cooler with ice packs or wet ice, being careful not to squish dragonfly larvae.
- Upon return to the laboratory or office, store dragonfly larvae in a freezer until shipping.
- **Do not open the inner dragonfly bags!** If you want to check your identification, do so quickly, before freezing, and through the inner bag. Do not allow dragonfly samples to thaw and refreeze; it is important that they remain frozen once placed in the freezer.
- Dragonfly samples can be stored until all sites are sampled, then **must be shipped in a cooler on DRY ICE.** Include a copy of your field sheet in the shipment, and keep a copy for yourself. Alternatively, the datasheets can be scanned and emailed to the coordinating lab with the originals included in the shipment. Contact us if you have trouble finding dry ice, or have questions about it.
- Note: When shipping samples on dry ice, you must **check the dry ice box on the FedEx® label.** Additionally, the cooler must have the Dry Ice label (included in sampling kit) affixed with waterproof tape.
- **Contact the coordinator when a shipment is ready** to verify that someone is available to accept the shipment. Shipment of coolers should be sent FedEx overnight (with pre-paid return label included in kit).

**Coordinator for Shipment of Samples:**

James Willacker (jwillacker@usgs.gov); 541-750-0957 (office) or 541-243-3606 (cell)

**Shipping Address:**

USGS Forest and Rangeland Ecosystem Science Center  
Attn: James Willacker  
3200 SW Jefferson Way  
Corvallis, OR 97331

6. Use the data!

We aim to make preliminary park-specific data available ~1 year after the samples have been collected. Work with your citizen scientists to describe the findings at your park. Submit entries to the local science fair. Feel free to contact us if you want to discuss your park’s data and how to incorporate it into park programs. UMaine and USGS will interpret the results for the national parks each project year, through webinars or other outlets.
Appendix A. Nets

Participating parks must provide their own sampling nets. There are many options for nets, ranging from expensive D-nets to modified inexpensive baitwell nets. See a few options below. None is necessarily better than the other, so choose according to substrate (if known), resource availability, and group size.

1. The D-Net: LaMotte D-Net, $70, item #138658 at www.benmeadows.com

2. For smaller mountain streams – or to facilitate group management – aquarium-sized nets may be more useful than a large D-Net. These nets can be purchased for as low as $4 (see examples at www.carolina.com). A helpful technique may be to allow each citizen scientist or small group one net. Each group or individual then searches a designated section of the stream/shore, bringing potential positive samples back to a central location (i.e., bucket) where the larger group can collectively observe, identify, and select.

3. A good, inexpensive net (specific to dragonfly larvae and not good for quantitative biomonitoring) can be made by modifying a baitwell net – stretch the net across like a pool skimmer and attach with plastic zip ties or small nylon strings. This works well because it won’t plug up with sediment (drains quickly) and retains large invertebrates like dragonfly larvae. Cost is $20-30 and they are available online (http://www.forestry-suppliers.com/product_pages/View_Catalog_Page.asp?mi=5061) or at local fishing shops. These nets are lightweight as well.
Appendix B. How clean is “clean”? 

Mercury is all around us: in our hair, in soil, leaves, etc. We need to keep everything clean so to not contaminate our samples. Read on for recommendations on getting gear ready....

How we are using terms in this project:
• **Clean**: Materials and supplies prepared in such a way that they would not have contamination that could affect our analyses and interpretation of mercury.
• **New**: We use this term when referring to items like plastic spoons and dishpans. By “new”, we mean that the materials are either brand new from the store, or you know they have only been used to hold natural water and invertebrates, similar to what is done in this study. For instance, dishpans might have been used before in your field sampling for macroinvertebrates, which is fine. What we mean by “new” is that this is a designated field supply, not an old dishpan or spoon that was used with food, or materials that you really don’t know about.

Level of cleaning needed for each field item:
Many supplies are “**disposable**”: plastic spoons, gloves, and extra bags. They come to you pre-cleaned and ready to take into the field to use just as they are. These should be marked as “used” when you are done at one site and heading to the next. Each site has its own sampling kit and you should start fresh at each new site. A good practice is to draw an “X” on the trash bag with any supplies that have been used at a site, to keep them from being confused with fresh, un-used supplies.

Before you go in the field:
• For new supplies (e.g., if you bought a dishpan), rinse it out three times with hot tap water before taking it out in the field. Then rinse it in site water as with items you are re-using (see “New”).
• You can use new garbage bags to transport large items into the field (to minimize road/trail dirt), and to store equipment between sampling days. Just be sure everything is dry first to avoid mildew.

Before your first site or between sites: (even with new items) If you are heading from Site A to Site B, you have some field gear that will contact the samples that needs to be **re-used**: nets, and dishpans. What do you do to ensure you are not introducing Site A contamination at Site B? And for new items, how do you ensure that there’s limited contamination from the store or from transit?
• First, you will probably have disinfected your nets, boots/waders, and dishpans after you finished work at Site A. Many parks require disinfection to reduce the spread of amphibian disease or other aquatic-borne pests from site to site. Each park has its own policy and requirements. Often (but not always) this means soaking or spraying nets, boots/waders, and dishpans with a bleach-water solution. Check your permit for procedures.
• Then, you will probably toss your wet, disinfected gear in the back of a vehicle and maybe drive down dusty roads or hike through the woods to get to Site B.
• When you get to Site B (or your first site, for new gear), locate an area of the pond away from that target sampling location, or downstream of it in the case of a river/stream, and rinse your waders.
• Have the rest of the crew rinse their boots, waders, and nets in pond/stream water away from the probable sampling site or downstream. Dunk gear in the water three times to rinse. Then proceed with sampling.

Within a site: If you are at Site A, and you are bagging all of your dragonfly larvae from Site A, you can rinse your plastic spoon in lake/streamwater between samples. The same is true for your Nitrile (purple) gloves. But don’t use those Site A spoons or gloves at Site B – they are contaminated with Site A materials.
Appendix C. Research Permit Application – example RPRS text

Project title: The Dragonfly Mercury Project – engaging citizen scientists in monitoring mercury contamination in National Parks

Purpose: Mercury, in its toxic methylated form, is a potent neurotoxin that is delivered to ecosystems via deposition from a global atmospheric pool, and ultimately bioaccumulates in aquatic and terrestrial foodwebs. In the northeastern U.S., research sites in ‘pristine’ areas have fish and other biota that exceed thresholds considered safe for human consumption or wildlife protection. All New England states, and many other states, have statewide fish consumption advisories with respect to mercury because, in large part, of the difficulty in predicting which systems are likely to be most affected. This study will be part of ongoing citizen science research that is evaluating the utility of dragonfly larvae (Odonata: Anisoptera) as indicators of mercury status. Our early data are being used to develop hypotheses regarding whether mercury varies with odonate larval body size or by family; or whether a site’s landscape setting drives variability in mercury in odonate larvae. To date, research has been primarily carried out by citizen scientists in national parks in the pilot program; this permit request is in support of an effort to expand the work beyond the pilot parks to identify broader-scale spatial patterns and better understand the utility of this bio-sentinel.

Summary of proposed field methods and activities: Aquatic macroinvertebrates are typically collected using standard D-nets with 500 micron mesh or dip nets with larger mesh (if dragonfly larvae only are targeted), and/or by inspection of cobbles, submerged or emergent vegetation, and woody material. Individual dragonfly larvae are picked (with gloves or pre-cleaned forceps or spoons) from nets and double bagged in zipper seal bags. Individuals are frozen until shipment and analysis. Body length of individuals and identification to taxonomic family may be done at the field site or after received in the lab, by prior arrangement. At the field site, individuals not selected for analysis are immediately returned to the waterbody of origin.

Repository Type: Will be destroyed through analysis or discarded after analysis

Objects Collected:

- Odonata: anisoptera (dragonfly) nymphs; 45 per park per sampling outing. (15 per site, 2–3 sites).

Location Information:

- List sites: *(Specific to each park; to be determined.)*
- Access: *(Specific to each park; access by foot on roads or trails.)*

Where will data, maps, photos, etc. (not specimens) reside upon completion of this project?
Data are stored in original laboratory files, on the project PIs computer (and associated backups), and will be uploaded to IRMA after final quality assurance and publication of results. Data collected by students/teachers from sites in the pilot studies are already available on IRMA.
Appendix D. How to use iNaturalist

iNaturalist.org is a place where you can record what you see in nature, meet other nature lovers, and learn about the natural world.

From hikers to hunters, birders to beach-combers, the world is filled with naturalists, and many of us record what we find. What if all those observations could be shared online? You might discover someone who finds beautiful wildflowers at your favorite birding spot, or learn about the birds you see on the way to work. If enough people recorded their observations, it would be like a living record of life on Earth that scientists and land managers could use to monitor changes in biodiversity, and that anyone could use to learn more about nature.

That’s the vision behind iNaturalist.org. So if you like recording your findings from the outdoors, or if you just like learning about life, join us!

What Are Observations?
An observation records an encounter with an individual organism at a particular time and location. This includes encounters with signs of organisms like tracks, nests, or things that just died. When you make an observation, you’ll record:

Who you are
You’ll need to make an iNaturalist account and please only post your own personal observations

Where you saw it
Record both the coordinates of the encounter as well as their accuracy. You can obscure the location from the public

What you saw
Choose a group of organisms like butterflies or better yet a specific organism like the Monarch butterfly. If you provide evidence you can leave this blank and the community can help

When you saw it
Record the date of your encounter, not the date you post it to iNaturalist

Evidence of what you saw
By including evidence like a photo or sound, the community can help add, improve, or confirm the identification of the organism you encountered. Help the community by taking clear well framed photos, by including multiple photos from different angles
Make an observation from the Web

1. Start by tapping **Add** from the dropdown in the top menu.

2. Type a name for **what** you saw and choose from one of the choices. If you can't find what you're looking for, leave it blank or use some placeholder text. If you entered a scientific name that's not recognized, click 'Search external name providers' to first import the organism from elsewhere. Let us know if the organism you observed was captive or cultivated.

3. Type in an address and click 'Search' in order to calculate coordinates and an accuracy circle describing **where** you were. You'll probably also need to zoom into the map and manually adjust the marker position and accuracy circle size.

4. Adding **evidence** for what you saw in the form of photos or sounds provides. You can upload photos from your hard drive or import photos or sound from your other social networks by linking them.

5. Use the calendar to enter **when** you observed the organism.

6. **Save** your observation.
How to Make an Observation with your iPhone

1. Tap **observe**.

2. Add one or more photos as **evidence**.

3. Enter **what** you saw or leave this blank.

4. **When** you saw it should be added automatically.

5. **Where** you saw it should be added automatically, if not check Privacy in the Settings app.

6. **Save** your observation.

7. **Upload** to share with the community. This should happen automatically, if not tap the button. You can turn off autoupload from the app settings from the Me tab.

8. Check back for **activity** on your observation from the community.
Make an observation from your Android

1. Tap the **observe** button from the My Observations tab.

2. Add one or more photos as **evidence**.

3. Enter **what** you saw or leave this blank.

4. **When** you saw it should be added automatically.

5. **Where** you saw it should be added automatically, if not check Privacy in the Settings app.

6. Save your observation.

7. **Sync** to share with the community. This should happen automatically, if not tap the button. You can turn off autoupload from the Settings tab.

8. Check back for **activity** on your observation from the community.
Appendix E. Sacrificing Dragonfly Larvae for Science: What is the Impact?

Dragonflies are typically found in very high densities, and our project samples a relatively tiny number per waterbody.

Evidence:
- Range is 20-200 individuals/m² depending on time of year and species\(^1,2\)

Dragonflies are prolific reproducers (r strategists): they produce a lot of offspring because it is expected that most won’t survive to adulthood.

Evidence:
- Rearing studies in Maine: a single female lays 400-1600 eggs and can do this more than once per season.
- Other studies show that females can lay from 800-2600 eggs over a 9 day period.\(^3\)
- In nature, >95% of larvae don’t survive to adulthood.\(^4,5\)

Other threats such as vehicle traffic can pose a significant threat to dragonflies:
- To a dragonfly, roads are warm, sunny clearings with ample flying insect prey; thus, dragonflies often use roads as resting and foraging sites.
- In one study at Acadia National Park, researchers found that vehicle collisions with flying adult dragonflies were common near roadways: at least 3.2 Odonata per kilometer of road were struck each day during the breeding season, along high-traffic roadways near freshwaters.\(^6\) Further, these mortality rates were likely underestimates because researchers were unable to count insects that remained stuck to vehicles, and weather was unusually rainy during the study.

The impact of this project on dragonflies is expected to be minimal.
- We sample the smallest number of individuals that are statistically meaningful.
- Power analyses are used to optimize the number of samples.
- Other factors are responsible for even higher rates of mortality (cannibalism, predation, automobile collisions (see above))
- Dragonflies are ubiquitous and found in so many habitats around your region – even urban sites, irrigation ditches, cattle ponds, cold regions, and nutrient-enriched sites.

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Appendix F. Dragonfly Ecology 101: How do I find them?

Dragonflies inhabit a wide range of microhabitats so it may not be completely intuitive where to start sampling. If you are having trouble finding dragonflies one of two factors may play a part:

1. Habitat selection
2. Timing of sampling

Finding buggy habitats:
Dragonfly guilds are separated by their behavior, morphology and microhabitat in which they reside.

- **Claspers: Cling to structure in the water column**
  - Family*: Aeshnidae
    - Habitat: Aquatic vegetation or snags
  - Collection: Claspers typically climb up onto vegetation, so sweeping macrophytes or grass along the banks is effective for collection of this family
    * Nymphs in the genera *Epitheca* (Corduliidae) and *Leucorrhinia* (Libellulidae) are also claspers

- **Burrowers: morphology to burrow and resides under sandy substrates**
  - Family: Cordulegastridae – shallow burrower
    - Habitat: lotic systems within depositional zones containing fine sediments
  - Family: Gomphidae – deep burrower
    - Habitat: lentic or lotic systems containing fine sandy sediment
  - Collection: Skimming the top inch or so of sandy substrates is effective for collection of this family.
    - You can also look for lines in the sand. Both of these families will keep their posterior end out of sediments to be able to respire, this leads to a line that is drawn out in the sand as they move within substrates. Sweeping your net along these lines can yield specimens.

- **Swarplers: longer legs to stabilize on top littoral sediments or vegetation**
  - Families: Libellulidae, Corduliidae, Macromiidae
    - Habitat: Lentic-littoral on top of sediments or aquatic vegetation
  - Collection: variable

- **Hiders: conceal their bodies under leaf litter and detritus**
  - Species*: Hagenius brevistylus (Gomphidae) - Abdomen strongly flattened dorsoventrally
  - Collection: Sweep your net into depositional zones containing high detritus
  *A few Cordulid nymphs are hiders

*This general guidance is simplified, there are exceptions for each and it’s not restricted to these families.*

Timing of sampling:

- Depending on your climate, you may be between populations that are emerging and those that are laying new eggs – making the individuals harder to find or even non-existent in that microhabitat
- If you are looking for 20 minutes and find nothing, try another spot along the shore to explore these different microhabitats

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