Streamside salamanders are receiving more attention as ecological indicators (Roth et al., 1999; Ohio EPA, 2001). Streamside salamanders in the family Plethodontidae often replace fish as the top vertebrate predators in headwater stream ecosystems. Headwater habitats are the small swales, seeps (where ground water oozes slowly to the surface, usually forming a pool), creeks, and first order streams that form the origins of larger rivers. Streamside salamanders are promising indicators of environmental stressors in small streams due to their longevity, relatively stable populations, small home ranges, abundance, and ubiquity (Rocco and Brooks, 2000; Welsh and Ollivier, 1998). Studies have found reduced salamander species richness or abundance at streams with higher impervious surface area in the basin (Boward et al., 1999), increased urbanization (Orser and Shure, 1972) and acid mine drainage (Middlekoop et al., 1999; Rocco and Brooks, 2000), and with nearby road construction (Welsh and Ollivier, 1998) and logging (Bury and Corn, 1988; Corn and Bury, 1989). To monitor changes in populations of streamside salamanders in relation to environmental variables, efficient and effective standardized sampling techniques that detect and accurately characterize presence and abundance of all species and age classes are essential.

Streamside salamanders are most active at night, avoiding predation by diurnal vertebrate predators. During the day, they hide under or in different types of microhabitat cover including rocks, logs, leaves, moss, bark, burrows, and overhanging banks. Streamside salamanders are often difficult to survey because they can escape into crevices and interstices among rocks along the stream and stream bank (Pauley and Little, 1998). Survey techniques do not sample all species with equal efficiency (Fellers, 1997) and almost certainly differ in their ability to detect larvae and adults. Estimating attributes of larval populations is important because larvae may be more sensitive than adults to environmental stressors (e.g., stream acidification, Kucken et al., 1994; Middlekoop et al., 1999; Rocco and Brooks, 2000). The presence of larval salamanders indicates the population is reproducing and resident in the stream on an annual basis. Comparisons of streamside amphibian sampling methods have been conducted to identify effective monitoring techniques (Fellers and Freel, 1995; Jung et al., 2000; Mitchell, 1998a,b, 1999; Pauley and Little, 1998; Welsh, 1987). From these studies, quadrat and transect methods appear to be efficient in capturing adults and larvae and promising for long-term monitoring.

This project seeks to answer the following questions: 1) How are streamside salamanders doing on protected lands in the Northeast? and 2) Are they effective indicators of ecological condition in small streams undergoing some form of degradation or disturbance? Examples of disturbance could include logging or clear cutting in the adjacent riparian zone, the presence of horse or cow pastures along the stream, a recent fire, storm water runoff, urbanization, point sources (e.g., maintenance yard drainage), acid mine drainage, and road construction. The objectives of this project are to: (1) conduct transect and quadrat sampling for streamside
salamanders, (2) determine detection rates and population estimates along transects, (3) obtain data from a range of degraded and non-degraded sites, and (4) establish a long-term streamside salamander monitoring program on Federal lands within the Northeast. Only first- and second-order streams (those likely draining less than 1000-acre catchments) will be surveyed. Predominant land use will be used to designate streams as developed (>25% urban or 50% agriculture) or undeveloped (>50% forested). Using GIS, actual upstream catchment land use will be calculated and used in analyses. Classification of sites as to degree of degradation will be based on the surveyor’s assessment of disturbance or degradation, land use, and physical habitat.

Biologists with the National Park Service, U.S. Geological Survey, and National Wildlife Refuges will participate in the streamside salamander surveys. Our goal is to survey a minimum of eight stream sections at each of 5 Refuges and three National Parks, using quadrat and transect methods. Typically this will consist of picking 4 streams and conducting two transects and two quadrats per stream. A stream section consists of one transect and one quadrant (a transect-quadrat pair). We will estimate streamside salamander populations at all transects using removal sampling based on two or three removal passes (Bruce, 1995; Rexstad and Burnham, 1991; Salvidio, 1998).

MATERIALS AND METHODS

Stream and Site Selection

A stream is defined as a “surface watercourse having a channel with well defined bed and banks, either natural or artificial, which confines and conducts continuous or periodical flowing water” (Ohio EPA, 2001). You can locate streams using either 7.5-minute series USGS topographic maps or NRCS county soil maps. Sometimes small headwater streams will not be identified at the USGS 1:24,000 mapping scale and you will have to scout these by foot. Choose headwater, first- or second-order stream sections that have a lot of cobble or cover objects to turn over. Avoid stretches that have primarily sandy substrates, boulders, or waterfalls.

The ideal situation would be to choose two streams that are not degraded and two streams that are degraded, conducting two sections (i.e., two transect-quadrat pairs) per stream. Alternatively, streams could represent both conditions if an undegraded stretch was surveyed directly above a source of degradation (e.g., cow pasture, point source) and a degraded stretch was surveyed directly below the source of degradation. However the stream sections are picked, we would like each Refuge or Park to survey a total of 4 transect-quadrat pairs representing undegraded streams and 4 transect-quadrat pairs representing degraded streams. If there are no obvious sources of degradation in the Refuge impacting streams, then streams representing different habitats (e.g., streams through woods versus grasslands), histories (e.g., burned, unburned), or covering the geographical extent within the Refuge or Park could be chosen.

The first step will be to scout the length of the stream within the Refuge. A good place to start would be at the headwaters (spring, seep) if that occurs within the Refuge boundaries. You can turn rocks and logs along the stream as you go, recording what you find. This will serve as an inventory of the amphibians along the stream and will also allow you to identify good stretches (rocky areas) where the transect-quadrat pairs can be conducted.

In general, the first transect-quadrat pair should be conducted at or near the headwaters of the stream (the spring or seep source) or directly above a source of degradation (e.g., stormwater pipe). The other transect-quadrat pair should be conducted at a lower elevation, at least 50 m
distant if possible, or directly below a source of degradation (if this applies) from the first transect-quadrat pair.

**Time Frame for Surveys**

The full suite of transect-quadrat surveys should be conducted **once in June-August**.

**Transects**

Transects (15 x 2 m) are conducted along either the right or left side of the stream, searching 1 m from the water’s edge along the bank and 1 m from the water’s edge within the stream channel. The salamander biologists will carefully survey transects by turning over the surface layer of cover objects (i.e., rocks and logs). Try to turn over as many cover objects in the transect as you can, excluding, of course, those that are too heavy or embedded to lift. Make sure to replace the cover objects that are lifted to their original position to minimize habitat disturbance.

As you go along, use the clicker counter to tally the number of rocks (cobble size or greater) or logs turned over that exceed 6.4 cm (2.5 inches) in width or length. Be sure to record the total number of cover objects turned over as well as all the other information on the data sheet: observer names, time begin and end of each pass (the actual time spent searching for amphibians), air and water temperature, stream water depth and width at the beginning, middle, and end of the transect, etc.

Start at the downstream end. If you have two people, one person can start at the downstream end, and a second person can start at the mid-point at 7.5 m. Alternatively, one person can conduct the transect, while the other conducts the quadrat (see below). As you move upstream, first place the net firmly against the bottom substrate just below (downstream of) the cover object. Next, lift the cover object in front of the net. Sometimes salamanders immediately (and quickly) swim away, but often larvae stay in the area where the rock had been. If you don’t see movement, wait for the sediment to settle and look carefully in the area. Then, to capture larval salamanders, position the net in front of the salamander’s head and gently touch the tail; more often than not they will move forward into the net. Sometimes larvae are swimming around in the open and you can direct them into the net and then transfer them into the zip-lock bag from there.

For adults, you can use the same tactic, or you may have to go after them using your hand or dip net if they try to escape. On land, have your net ready to catch amphibians from under overturned rocks. The salamanders can be quite fast! Once you have turned over the rock or log, you might see movement right away, in which case you need to catch it quickly! If a salamander escapes, write down the information about the species (put a question mark by the species if you are uncertain) and estimate the total length. In this case, make sure to write, “ESCAPE” in the Notes section of the data sheet. Once you have the salamander in your hand or net, transfer the salamander to a zip-lock bag (see “Amphibian Capturing…” section).

After the first pass, measure all the amphibians caught (snout-vent length, total length), in the meantime keeping them in the shade in their plastic bags. You can even place the bags at the edge of the stream in a pool (so they don’t float away!) to keep them cooler. Do not return these salamanders yet to the stream. Once they are measured, you have the option to transfer these first pass amphibians to larger containers (e.g., plastic tubs with water for larvae, spackle buckets
with lids and a little water for adults) so that they have more room to move around. Then, conduct the second pass, turning over approximately the same number of rocks or logs as you did during the first pass. If the number of salamanders you catch during the second pass is smaller than the number you caught the first pass, two passes is enough. If, however, you catch more or the same number of salamanders the second pass compared to the first pass, follow the same procedures as above, but continue on to conduct a third pass. Two or three passes allow us to calculate salamander detection rates and to estimate population sizes. However, statistically, three passes is better than two passes!

**Quadrats**

The salamander biologists will also sample a 4 m² quadrat (a square formed by 2 m² on the bank and 2 m² in the water) near the transect (Rocco and Brooks, 2000). Mark out the quadrat using meter sticks, PVC pipe, or the 50 m tape, and/or marking the four corners with wire flags. Quadrats will be searched intensively, removing all cover objects and substrate as practicable. Quadrats represent destructive sampling, such that all rocks and gravel and debris within the quadrat are temporarily removed and only the underlying sand or bedrock is left. The goal is to ensure that no salamanders escape detection, providing a complete census of the quadrat area. Use the same amphibian catching techniques as described above. For quadrats, count and record the number of all large surface rocks overturned, but do not count underlying rocks or gravel.

**Amphibian Capturing, Handling, and Photodocumentation of Species and Streams**

All captured amphibians (frogs, toads, salamanders) will be placed into sealable zip-lock plastic bags for identification and measuring. Larvae (with gills) must be kept in plenty of stream water such that their entire body is covered with water. Adults (without gills) should have access to a little water (enough to keep moist, but not to drown). Plethodontid salamanders are lungless and respire through their skin. Puff the zip-lock bag a little so that adults have air and are not stuck between sheets of plastic in the zip-lock bag. Amphibians should be kept in the shade at all times to avoid overheating. At the end of sampling (i.e., after the quadrat or final pass of the transect), all amphibians must be returned to the quadrat or transect. To return salamanders to the exact point of capture, you can use numbered wire flags to mark the capture point, writing the same number of the wire flag onto the zip lock bag in which the amphibian is placed.

It is highly recommended that you take a camera with you into the field (digital cameras preferred) so that you can take representative pictures of the stream sections and species and age classes you encounter, malformed amphibians, or any other unusual sightings.

**Field Work Code of Practice**

Biologists can spread various diseases among sites that can impact amphibians (e.g., fungi and viruses such as chytrid fungus, iridovirus, ichthyophonus, etc.). This can be avoided by 1) designating specific dip nets for exclusive use at each stream (tie flagging tape to each dip net and write the stream name on the tape), and 2) cleaning and bleaching boots or other
equipment thoroughly between sites. Below are highly recommended procedures to follow during all amphibian survey work:

1) Take a stiff scrub brush, a spackle bucket half-filled with water (covered by a lid), and a 50% solution of bleach:water in a squirt bottle with you into the field.
2) Label dip nets for each stream with flagging tape (e.g., 2 per stream for 2 people) and use only those dip nets for those streams.
3) Clean boots of all wet or dried mud using a stiff scrub brush and the bucket of water.
4) After boots are cleaned of mud, spray the boots with a 50% solution of bleach:water.
5) Rinse the boots after 5 minutes, or once you arrive at the next site, by dipping them into the spackle bucket with water.

Description of Data Fields

Unit Name: Record the name of the Refuge or Park (e.g., Canaan Valley NWR) and Subunit if applicable (e.g., Great Meadows NWR-Oxbow subunit)
Stream Name: Record the name of the stream (e.g., Cow Knob Creek)
Date: Record Month, Day, Year (e.g., June 20, 2002)
Observer Name(s): Record the name(s) of the person(s) conducting the amphibian survey
Recorder Name: Record the name of the person recording the data on the data sheet
Transect: Record whether it is transect 1 (upper, headwater) or 2 (lower) for that stream
Distance from Transect to Quadrat (m): Record the distance in meters from the beginning of the transect to the quadrat
Date of Last Precipitation: Record (as best you can remember) the last time it rained
Air Temperature (ºC): Record 1 meter above the ground in the shade
Water Temperature (ºC): Record about 1/3 meter out from shore 2 cm below surface
Other Water Quality Variables (Optional):
   Water pH, Acid-Neutralizing Capacity (ANC), Conductivity, Dissolved Oxygen, Heavy Metals, Ammonia-N, Nitrate/nitrite-N, Chlorides, Total Phosphorus, Fecal Coliform Bacteria, Acid Mine Drainage (Iron, Manganese, Sulfate)
Turbidity: Record whether water is clear or turbid (e.g., cloudy with algae, muck or precipitate)
Sky Code: Use the following codes:
   0 = Clear or few clouds (< 20% of sky covered with clouds)
   1 = Partly cloudy or variable (20-50% of sky covered with clouds)
   2 = Cloudy or overcast (> 50% of sky covered with clouds)
   3 = Fog
   4 = Mist or drizzle
   5 = Showers or light rain
   6 = Heavy rain (don’t do survey!)
   7 = Sleet or hail (don’t do survey!)
   8 = Snow (don’t do survey!)
Wind Code: Use the Beaufort wind scale codes
   0 = < 1 mph, calm, smoke rises vertically
   1 = 2-3 mph, light air movement, smoke drifts
   2 = 4-7 mph, light breeze, wind felt on face, leaves rustle
   3 = 8-12 mph, gentle breeze, leaves in constant motion, raises dust
4 = 13-18 mph, moderate breeze, small branches move
5 = 19-24 mph, fresh breeze, small trees begin to sway
6 = 25-31 mph, strong breeze, large branches move (go home!)
7 = 32-38 mph, near gale, large trees begin to sway, difficult to walk (go home!)

Stream Width (cm): Record stream width at the beginning, middle and end of the transect
Maximum Pool Depth (cm): Record maximum pool depths near the beginning, middle and end of the transect

Begin Time: Record the hour and minute survey begins (use 24 hour clock) for each pass.
End Time: Record the hour and minute survey ends (use 24 hour clock) for each pass.
Number Objects: Record the number of overturned rocks and logs for each pass (transect) or quadrat
Species: Record the species observed using codes on the data sheet or writing the full common or scientific name
Age Class: Record whether the species is a larva (gills present) or an adult (no gills present)
Snout-Vent Length (mm): Measure the snout-vent length (snout to posterior end of the cloaca).
For very small larvae, sometimes the cloaca is not very visible. In these cases, just measure to behind the hind limbs.
Total length (mm): Measure the total length (snout to end of tail)

Notes: Record whether amphibian is an escape, whether it is nesting (provide details about the number of eggs) or whether there is anything else unusual about it (e.g., malformed – describe malformation in detail, missing tail, etc.)
Fish? Record whether present (Yes) or absent (No)
Crayfish? Record whether present (Yes) or absent (No)
Aquatic Invertebrates? Record whether present (Yes) or absent (No); Describe

Data Fields only on the Stream Transect Habitat Data Sheet

For Coordinates, record using either UTM E and N OR Latitude and Longitude
(you don’t have to record both)
UTM N: Record the upstream coordinate for the transect
UTM E: Record the upstream coordinate for the transect
Latitude: Record the upstream coordinate for the transect
Longitude: Record the upstream coordinate for the transect
Stream Channel Modification: Select one of the following:
1 = Stream channel is natural without modification
2 = Stream channel is modified (e.g., with cement, pipe, dredged, etc.); describe
Stream Order: Select the best description of the stream order category:
1 = Starting from the headwaters; headwater area
2 = Stream segment from unbranched tributary further downstream from headwater area; first order
3 = Stream segment resulting from the joining of 2 or more unbranched tributaries; second order
Slope: Record the slope of transect from highest to lowest point (between 0º and 90º)
Flow Regime: Record % of the transect that is covered by each of the categories below:
% Dry: No visible moisture or water
% Moist: No flow, but moist soil
% Seep: Slow flow, trickle or drip
% Pool: Standing/Stagnant water
% Riffle: Riffles/small waves, not caused by obstruction
% Run: Swiftly-moving, smooth surface current

% Substrate Embeddedness: Record the percent (0-100%) of visible vertical surfaces (rock) that are surrounded by line sediment or flocculent material (buried or embedded in either silt, fine sediments or sand) (Lowe and Bolger 2002, Welsch et al. 1997).

% Substrate Type: Record the percent substrate covering the transect. Should sum to 100%.
% Sand (< 2 mm, gritty texture)
% Gravel (2-32 mm)
% Pebble (33-64 mm)
% Cobble (65-256 mm)
% Boulder/Boulder slabs (> 256 mm)
% Bedrock
% Silt (particles < 2 mm, greasy texture when rubbed with fingers; clay and fine organic)
% Detritus (partially or undecayed sticks, wood, leaves or other plant material)
% Clay/Hardpan (hard and gummy clay, hard to penetrate)
% Muck (decayed organic matter with little or no clay content)
% Artifical (cement, pipe, etc.)

Riparian Width: Record the appropriate width of the adjacent forested buffer area along both the right and left banks of the stream.

Land Use: Percent of land use adjacent to the site (within 50 m both sides). Should sum to 100%.
% Agriculture/Field
% Pasture
% Industrial/Urban
% Residential/Suburban/Park
% Mature Forest
% Immature Forest/Shrub
% Meadow/Marsh
% Mining/Construction
% Road
% Other: Describe

Disturbance or Habitat Type Represented: Describe

EQUIPMENT LIST

Dip Nets (fine mesh – 6” x 8” or 8” x 10”)
Tally Counters
Plastic Tubs with Lids and/or Spackle Buckets
Rubber Boots (knee-high) or Hip Waders (for deeper streams)
Meter Stick
Water Quality Equipment (if available)
Zip-Lock Bags (Sandwich size with triple color) – Box of 100
Rulers (30 cm)
Meter stick
50 m fiberglass tape
Data Sheets
Clipboard
Pencils
Bug Repellent (if necessary, though do not apply to hands or other parts of body that may come in contact with amphibians or the stream itself)
Bleach solution in Spray bottle
Scrub brush
GPS Unit
Camera (with film or card)
Wire Flags
Sharpie

LITERATURE CITED


Ohio EPA. 2001. Field evaluation manual for Ohio’s primary headwater habitat streams. Ohio EPA Division of Surface Water, P.O. Box 1049, Columbus, OH.


REFERENCE MATERIALS