

Northeast Temperate Network And Marsh-Billings-Rockefeller NHP Forest Amphibian Protocol

Version 1.02
June 16, 2006

NOTE: The primary source for these materials is <http://www.pwrc.usgs.gov/Sally/>. This program began on June 30, 1997. Most of the materials provided here were written by Dan Lantz (redleftboy@yahoo.com).

A good ID guide for herps can be found at <http://www.npwrc.usgs.gov/narcam/idguide/>.

Introduction: Salamanders as Unique and Potentially Powerful Indicators of Ecosystem Health

The forests of North America host several genera of small slender salamanders. These salamanders are so adapted to living in woodlands that they have dispensed with the expected amphibian aquatic larval stage and live their entire life cycle within the humus and soil of the forest floor. Here they rely on the moist chambered and channeled soils for retreats, because without direct soil contact, or rain saturated litter, they quickly dry out (desiccate). Their forays to the surface for the necessities of feeding, courting, and mating must coincide with the right conditions of cool, wet, calm weather.

In harmony with a low-input, subterranean life this group of salamanders is the most energetically efficient of the vertebrates (Feder 1983). Energy assimilation rates (conversion of what they eat into mass or energy) varies from 48% to 98% (often in the 90+ % range), contrasting sharply with rates of between 0.5 to 1.22% for vertebrate endotherms (Larsen 1992, Pough 1980, Jaeger pers. comm.).

Other aspects of salamander life histories further underscore their biological stability and tenacity.

- * Extremely long-lived (20+ years) (Hairston 1987)
- * Low reproductive rate (fecundity)
- * Often guard their eggs
- * Take 2 or more years to mature
- * Eliminate the energetic costs of lungs and ventilating
- * Breathe directly through their skin (White 1978)
- * Can go for months, if not years, without eating (Feder 1983)
- * Body temperatures fluctuate with the local substrates (Feder 1982)
- * Store fat in their tails

Energetic efficiency has not been attained at the cost of behavioral simplicity. Plethodontids have complex social systems with territoriality, aggression, site fidelity, and tiny home ranges. Perhaps in part due to their profound efficiency in metabolizing invertebrates, they can, in undisturbed forests, achieve such high densities as to equal or exceed the biomass of other vertebrate groups (Hairston 1987, Burton and Likens 1975). In turn, these salamanders are a source of food for many other vertebrates, transferring the energy of the soil and litter systems up the food chain (Pough 1983).

A consequence of these energetic and life history traits is an inextricable dependence upon moisture. Because this group of salamanders lacks lungs, their skin must remain constantly moist or they lose the ability to transfer oxygen. When moving on the surface they can become dehydrated even when the air is saturated with moisture (Feder 1983). Logging, insect defoliation, development, and canopy fires all can reduce population levels indirectly through loss of soil moisture, exposure to sun, or the compaction and disruption of soils (Welsh and Droege 2001, deMaynadier 1995). In direct contrast,

salamanders achieve their highest densities in ancient or undisturbed forests (Meier et al. 1996, Welsh 1990, Herrington and Larsen 1985).

Despite salamander predominance in some ecosystems and their long co-evolution with forests, we know very little about the population health, cycles, and trends of these woodland endemics (Welsh and Droege 2001). Are populations of these terrestrial species declining, as many have documented for populations of North American frogs and toads (Stebbins and Cohen 1995, Vial and Saylor 1993), or are they comparatively stable? Are currently forestry practices and the recovery of mountain forests from industrial logging at the turn of the century increasing the number of salamanders? Data sets are few and scattered, focused on research rather than monitoring, and largely unpublished. Consequently, existing information provides almost no help in detailing regional status.

Our best knowledge about the population health of terrestrial salamanders comes almost entirely from the repeated field observations of herpetologists. There is now useful information on status, but the information is not nearly sensitive or systematic enough to do more than provide a post-mortem of any population collapse.

What if a monitoring system for terrestrial salamanders existed? What would we gain? How would knowledge of population change in salamanders contribute to our ability to either understand or manage salamander populations or their habitats?

The long-term tenure of salamanders in forested ecosystems (Highton 1995, Welsh 1990) would suggest that their loss or decline may have consequences affecting many other plant and animal species. Numbers and biomass alone dictate that the effects of changes in their populations could cascade throughout the forest as their invertebrate prey and vertebrate predators adjust.

Consequently, the ability to track changes in salamanders, both declines and increases, would provide insights into forest ecosystems and perhaps a more realistic assessment of forest health than those based on the classical forestry measures of stocking and growth rates. Forest health must account for the communities of all plants and animals (such as salamanders) that exist under the defining umbrella of large woody plants. An increasing or stable number of salamanders would indicate that the forest's balance of invertebrates, leaf litter, moisture, pH, debris, burrows, and other habitat features were sufficient, while declines would signal environmental shifts counter to the needs of the species and increasing ecosystem impairment. Detection of salamander declines would hopefully provide adequate forewarning, allowing us to search for the root causes and take action to reverse the damage. Given their longevity and low fecundity, as well as the stability of the forest soil environment, changes in salamander population are much more likely to represent significant environmental changes than changes in other groups of North American amphibians.

A monitoring program's primary functions are to warn of impending population problems, to direct conservation management (activities, monies), and to generate

hypotheses for research. Without such a monitoring network, populations can collapse to the point that conservation and management options become limited and expensive, exactly the sort of situation which led to the endangered listing of several western frogs and toads (Stebbins and Cohen 1995, Vial and Saylor 1993). Salamanders' patterns of population change, when coupled with measurements of their surroundings, would permit the investigation of the effects of geographic location, land management, air quality, vegetation, and associations with other species on salamander population dynamics.

Other less tangible but equally important benefits would also accrue from the long-term monitoring of salamanders. As with bird and forestry trend data, ecological insights into species biology and ecology come to both those who collect the data in the field and those who analyze them in the laboratory. As data and analyses accumulate, so too does the monitoring site's attraction to outside researchers. Monitoring with cover objects is, unlike many other monitoring techniques, attractive and appropriate for citizen participation as well as that of the herpetologist. Because fall and spring are usually the best monitoring periods, this opens up many opportunities for grade school and college student projects. For the reasons outlined above, we propose to contribute to a network of sites (volunteer and professional) in North America that will collect trend information on terrestrial salamanders. The primary technique used in collecting that information will be arrays or transects of cover objects (Sugar et al. 2001, Monti et al. 2000, Bonin and Bachand 1997, Davis 1997, Fellers and Drost 1994, DeGraaf and Yamaski 1992, Grant et al. 1992).

To paraphrase the conclusions of Ted Davis, who evaluated their effectiveness for monitoring salamander populations on Vancouver Island, artificial cover objects are:

- * Non-disruptive to the natural habitat
- * Attract species that are difficult to trap in pitfall traps
- * Providing a standardized sampling unit
- * Minimize observer biases and errors

The rationale for monitoring terrestrial salamanders rests upon:

- * Their key role and high densities in many forests
- * The stability in their counts and populations
- * Their vulnerability to air and water pollution
- * Their sensitivity as a measure of change
- * The threatened and endangered status of several species
- * Their inherent beauty and appeal as a creature to study and conserve

In the remainder of this document we explore the possible sources of bias, provide supporting evidence for these statements, present a series of experiments (Appendix 1) to test for bias and provide validation, and present a conservative protocol for installing a cover object monitoring site.

Before laying out a set of goals and protocols for the program, issues associated with known and potential biases in using cover objects need to be addressed.

Bias: Disguiser of Population Trends

Before laying out a set of goals and protocols for the program, issues associated with known and potential biases in using cover objects need to be addressed.

There are 3 categories of bias in using cover objects to determine salamander population trends.

1. Density and Year Independent
2. Density Dependent
3. Year Dependent

Density and Year Independent Sources of Bias.

These are biases that are acutely felt in the short term but wash out over extended periods of time (10+ years). Time of year, observer effects, rain events, temperature, and soil moisture are all factors that fall within this category.

Time of Year. Salamander captures under cover objects vary greatly with time of year (Taub 1961, Hendrickson 1954, Stebbins 1954). In many regions, the winter and summer months often find cover objects with few or no salamanders under them, with build-ups in the spring and fall. Consequently, each site (or region) needs to define a period of time for salamander counts that will both maximize the number of salamanders found under cover objects, while minimizing the variation in those numbers. The protocol section outlines the basic patterns, and Appendix 1 defines a set of experiments to further define the relationships.

Observer Effects. Unlike many monitoring techniques for amphibians, observer effects are largely confined here to mistakes in identification and not to the skill of the observer in hunting or spotting salamanders. The observer merely has to turn over the objects and accurately report what he or she sees. The level of identification difficulty will vary regionally. Species diversity is high and identification complex in many parts of the Appalachian Mountains, while in the northeast a single species (*Plethodon cinereus*, the Eastern Red-backed Salamander) comprises the vast majority of captures. Both professional and citizen observers will need to go through the quality control procedures outlined later in this document so that their resulting data are deemed credible to outside reviewers and users.

Rain and Temperature. These variables have been shown to have associations with counts under cover objects (Bonin and Bachand 1997, Heatwole 1963, Taub 1961). Over a span of decades these effects act randomly on counts, increasing the variance but not biasing the trends. This is not to say that their impacts should be ignored. Any opportunity to reduce the variability in yearly counts should be pursued. That pursuit should take the

form of a set of rules for the time of year and weather conditions when an observer should check transects or arrays. These rules will vary by location. See Appendix 1 regarding proposed experiments to further explore moisture/count relationships.

Density Dependent Sources of Bias

In these forms of bias, detectability (that is, catchability or observability) of the average salamander changes as the population density varies. The unbiased situation has the proletarian salamander always with the same detectability no matter what the density. Note that this does not mean that all age classes or species are equally detectable (which appears very unlikely to be the case, with both higher and lower detection rates under cover objects reported for juvenile salamanders of different species). As long as the relative rate of detectability remains the same, trends calculated from counts of salamanders under cover objects are a valid and useful measure of change. It is when a species' detectability itself changes (rather than differences among species or age classes) over time that we must be concerned about the meaning of data from our monitoring program.

For these salamanders, the only likely mechanism that would change detectability with change in density is territoriality. Strong evidence suggests that some populations of plethodontids may defend natural cover objects from others of their species (Mathis et al. 1995, Jaeger and Forester 1993). A program that uses artificial cover objects to monitor populations may, therefore, underestimate the population size at higher densities as greater proportions of the population would be relegated to the status of non-territorial floaters with limited access to cover objects. At high densities, the population index thus would detect territorial animals and under represent the floating population.

There is some evidence suggesting that artificial cover objects may not be defended in some cases, but are apparently used opportunistically (Mathis et al. 1995). Wells (1980) found that while Slimy Salamanders (*Plethodon glutinosus* complex) occurred together under a log only 16% of the time, the same cover object would be used sequentially by dozens of individuals over the space of 3 years, indicating that cover objects are not under the sole control of single individuals for this species. Despite this pattern of serial occupancy, single males never shared the same log and appear to actively avoid contact with other males. From personal observations, we have found that the use of individual artificial cover objects by Red-backed Salamanders (*Plethodon cinereus*) appears to vary greatly from week to week in the coastal plain of Maryland. When individual Red-backed Salamanders in Maine are captured under both natural and artificial cover objects (wooden shingles) and marked, they rarely are recaptured again under the original cover object, though they remain in the area (L. Monti pers. comm.). This indicates low use of cover objects compared to soil/litter habitats or some negative factor associated with being toe-clipped. While some territorial behavior may be evident in Red-backed Salamanders, occupancy of cover objects is a function of litter and soil moisture. When litter moisture is high, a greater proportion of salamanders are found in the litter, when soil moisture decreases more are found under cover objects, and when soil moisture drops even further they retreat into the soil column (Jaeger 1980a).

In contrast, under arrays of small boards in West Virginia, recapture rates under the same objects are in the 10-30% range for Red-backed Salamanders and Mountain Dusky Salamanders (*Desmognathus ochrophaeus*) (T. Pauley pers. comm.). Slimy Salamanders also have been reported to have high site fidelity to artificial cover objects in South Carolina (Grant et al. 1992) as have Western Red-backed Salamanders (*Plethodon vehiculum*) for both artificial and natural cover objects, with individuals found under larger objects having greater fidelity (T. Davis pers. comm.). California Slender Salamanders (*Batrachoseps attenuatus*) were also found to have long-term tenure under cover objects (Hendrickson 1954).

Observed occupancy rates in the field argue against the notion that territorial animals can saturate a cover object system. We know of no instances where artificial cover arrays have become saturated with individuals, filling all cover objects. Highest single check occupancy rates recorded include: 70% in Southwest Virginia (R. Jaeger pers. comm.), 23% in Quebec (Bonin and Bachand 1997). Average counts from 4 additional surveys were calculated to be 10, 14, 15, 4 percent, assuming only 1 animal per board..

In sum, evidence to date on the effects of density dependent bias is equivocal. There is reason to expect, at least for some populations of Red-backed Salamanders, that territoriality would decrease the detectability of individuals as populations change – though this may break down in northern or consistently moist habitats (R. Jaeger pers. comm.). However, the extent and variability of that impact is unknown, and may be minor if most of the population occurs below the level of cover objects (Taub 1961). Except under the unlikely scenario that detectability decreases so much (per salamander in the population) at higher densities that the index value (count or number of captures) no longer increases, but actually declines, the general effect territoriality has on an index for cover objects will be conservative. That is, both positive and negative trends will be shifted toward zero (no change) from what they should be, but will not cause an estimated trend to change sign (e.g., going from positive to negative). Consequently, any statistically significant changes in the count index would be strong evidence that populations were truly changing. Subtler changes in population size, unfortunately, will be missed if the bias is strong. Alternatively, the index could be viewed as one that tracks changes in the territorial fraction of the salamander population rather than one that tracks the population as a whole.

The proper way to resolve most of these issues, and to correct mathematically for any bias, is through a series of validation experiments. In Appendix 1 we outline a set of experiments to further determine the impact that territorial behavior may have on salamander counts, to either confirm that the index is unbiased or provide a means of correcting the estimates.

Year Dependent Sources of Bias.

In this situation, bias is associated with factors that change over time (multiple years). Here, in the case of plethodontid salamanders, changes in the number and type of

naturally occurring coarse woody debris may bias counts, if their presence, or absence, or changes over time, alters the detection probability of salamanders under cover objects.

Here is an example of how that bias might work to subvert the meaning of the counts. Let's say that an array of artificial cover objects is placed in a wooded area that initially has very few natural cover objects (e.g., downed logs, branches, deep litter). As the forest matures, or an insect outbreak strikes, trees die and the number of natural cover objects increase. During this time the overall salamander population remains stable (rather than responding to the changes themselves, which would be a separate issue). If salamanders prefer to hang out under natural cover objects, the counts under artificial cover objects would drop despite the population's stability, and bias has corrupted our results into falsely signaling a decline when, in fact, there was none.

The literature does indicate that some salamanders have preferences for the location and type of natural cover objects (Mathis 1990, Mathis 1989, Heatwole 1962, Bogert 1952, Test 1948). In a study of Red-backed Salamanders in Maine the size of individuals (size is related to age) under natural cover was nearly identical to that under artificial cover objects (L. Monti unpublished data), but in British Columbia, larger salamanders (e.g., adults) were found more commonly under artificial cover objects for Western Red-backed Salamanders and Clouded Salamanders (*Aneides ferreus*) (Davis 1996). In West Virginia, Red-backed Salamanders and Mountain Dusky Salamanders, and in New Jersey, Red-backed Salamanders, occur more commonly under artificial cover objects than natural (T. Pauley pers. comm., Taub 1961). There is also anecdotal evidence that some species' use of cover objects varies geographically. *Desmognathus* in the coastal ranges occur under cover objects regularly (Stebbins 1954) but in the Sierra Nevada they do so much less commonly (Tim Manolis pers. comm.). So it is reasonable to expect that changes in the type and dispersion of woody debris could affect counts, likely in a way that decreases the detectability of salamanders under a fixed set of artificial cover objects found in the area. However, Monti (M.S. thesis 1996) found that artificial cover objects placed near logs in Maine were no more likely to harbor Red-backed Salamanders than ones placed further away.

The effect of changes in the abundance and type of natural cover objects on counts under artificial cover objects can and needs to be investigated. See Appendix 1 for a simple set of experiments to test for this phenomenon.

Cover objects themselves, in a manner similar to woody debris, may alter the number of salamanders present simply by increasing the capacity of the area to support their populations. Nothing definitive is known about the presence or extent of this phenomenon. However, since soil retreats are the ultimate requirement for almost all species, cover objects – which are used for only a portion of the time (Jaeger 1980a) – may not have a great effect. If such a phenomenon exists, its impacts would stretch over only the first year or two; beyond that time the cover object numbers and placement remains the same.

Validation

One can only go so far in interpreting index data that makes untested assumptions about the relationship between a proposed count index (salamanders under cover boards in this case) and the true population size (Droege 1996). To date, we know of no published validation of the methodology we are proposing. However, a recent study in the southern Appalachians that looked at the relationship between litter searches in plots of salamanders and density estimates found gratifyingly significant and high correlations (J. Petranks pers. comm.). Ted Davis (pers. comm.) on Vancouver Island found that relative abundance was similar between cover board arrays and area searches.

See Appendix 1 for an outline of experimental designs for validating salamander arrays.

National Sampling Frame

Terrestrial plethodontid salamanders are largely restricted to wooded or wet rocky habitats in eastern North America. Densities vary with age of forest and its history of disturbance, grazing, and cutting (deMaynadier and Hunter 1995). In the West, salamanders occur primarily in the forests and Mediterranean scrub of the Coastal Ranges, to a lesser extent in the Sierra Nevada, and in only a few spots along the Rocky Mountain axis. In Mexico less is known about the distribution and life history of plethodontid salamanders, but for several species cover objects may also be an appropriate sampling technique (D. Wake pers. comm.) and testing is greatly encouraged.

Cover objects should not be expected to sample the entire spectrum of herps present in a region or forest. They work best for the most terrestrial of salamanders. While Ambystomas and stream-side salamanders can occur under them, and in some instances their numbers are sufficient to calculate changes, they are not the target group for cover objects.

Developing a random, or even a stratified random, sampling scheme for an artificial cover object network for North America would be difficult. Arrays would fall, in many cases, on private lands and difficult terrain, often far from populated regions. Additionally, the work and expense it takes to initially establish some types of arrays, the access problems, rapidly changing patterns of forest distribution, and a limited pool of local volunteers, further compound the difficulties. This is not to say that small regions, parks, or states could not develop random sampling schemes (and they should be encouraged to do so). It just seems clear that the overall national design can not initially accommodate that type of sampling intensity.

Choosing sites non-randomly calls for a different emphasis in the sampling frame (and yields a different set of outcomes) than a program like the calling frog surveys many states and provinces have begun (Bishop et al. 1996). A collection of non-random sampling sites cannot represent the region as a whole, rather they represent the trends on those sites and those sites alone. This is not to say, however, that regional patterns and inferences cannot be drawn. By graphing the patterns of increases and decrease among forest types, regions, landscape metrics, and other environmental variables, much can be

divined regarding potential causal factors and the overall health of both salamanders and the forests. If interest in the network warrants, it can be expanded into a probability-based system later.

Tracking trends at individual sites, rather than pooling trends among sites, puts the emphasis on detecting individual site trends rather than regional trends. The development of the protocol requires that we first specify the minimum level of precision we would like to achieve; second, collect some pilot data; and third, run a power analysis on the pilot data to see if we can achieve our precision targets.

The overall goal for the national program is to develop a network of sites in North America (Canada, U.S., Mexico) which collect statistically defensible trends in terrestrial salamander populations.

This would not necessarily have to be limited to cover board surveys, but for practical purposes this method would be emphasized as most appropriate for citizen participation. Note too, that once the bugs in the system are worked out there is little reason to limit participation only to North America, as similar issues and problems exist world-wide.

The minimum statistical precision goals for each site are as follows (implicit here is an expectation that most sites would far exceed these targets):

Alpha level = 0.20. Setting the alpha to this level means that we are willing to cry wolf approximately 20% of the time. In other words, we are willing to incorrectly identify a population as one having undergone a significant change, when in reality it had not, 20% of the time.

Why 20% rather than the traditional 5%? The reason lies in our goals. In most experimental settings when we make a statement that there is a "statistically significant difference" between or among things that we have measured, we want to be sure that those differences are real, for if they are not, we look foolish, and people lose respect for us and our interpretation of biology. However, when monitoring populations (as opposed to running experiments on populations), the situation is different. It is imperative that we detect any important changes in populations, because as conservationists we sure would look bad if the population crashed over the span of a few years, and we missed the start of the event (when extinction/extirpation might possibly have been prevented) because we set our standards for detecting trends too high (by setting our alpha level too low). Consequently, the penalty of possibly being wrong 20% of the time in our statements regarding the presence of "significant change" is well worth the aggravation of sometimes crying wolf. The alternative choice is to let a population slip between the cracks (cracks caused by variability in our counting techniques) because we waited too long for the numbers to tell the story.

Minimum Level of Trend Detection = At Least a 50% Decline over 20 Years. The interpretation here is that data from an individual site must have sufficient power to alert us when the population has declined by half over 20 years, with an expectation that it will

be sensitive to much lower levels of decline over longer time periods and that most monitoring programs will be able to detect much smaller changes.. Because of the relative longevity and low fecundity of salamanders, small changes in population size may reflect significant long-term environmental changes. As an aside, by virtue of the variability of natural populations and their counts, you should not expect a monitoring program in its first 10 years to provide a great deal of trend information on populations that are only changing slowly.

Power = 90%. Power is the ability of a monitoring program to detect a trend if one really exists. Here we want to have a monitoring program powerful enough to detect a real population trend 90% of the time it actually is occurring.

Based on compilations in Gibbs' (1996) presentation on the World Wide Web, terrestrial salamanders generally have much lower coefficients of variation than other groups of amphibians (30.3% for terrestrial and stream-side salamanders (8 studies), 53.3% for frogs and toads (6 studies), and 69.5% for pool-breeding salamanders (6 studies), another indication that it is reasonable to expect useful information on trends from individual sites.

Expanding on Gibbs' study, we extracted data from 22 published and unpublished data sets for 11 species of plethodontid salamanders (Table 1). As Table 1 shows, there is a breakpoint at roughly 75% for the Coefficient of Variation (CV) estimates. CV is a standardized measure of the variability of counts. Any CV below 75% will meet our power criteria (i.e., variability is low). In the data presented here, all but 3 studies and 1 borderline case met these criteria. The median CV was 38%. No particular pattern of association with salamander species, geographic regions, or techniques appears evident. These numbers are even more impressive given that the objective of most of the authors was not necessarily to minimize sampling noise and that we made no attempt to remove the effects of any population changes occurring in the longer running studies. However, it should be noted that the 2 longest term studies took their samples at the same time of year every year.

Table 1. Summary of power analyses for 23 studies of plethodontid salamanders. Data include the state, number of years studied, mean count or capture, # of cover boards used, average percentage occurrence under cover boards (assumes only 1 salamander per board), CV, power to detect 3 and 4% per year trends, Type of survey technique (NS = Night Search, NC = Natural Cover Objects, AC Artificial Cover Objects, MR = Mark-recapture, PF = Pitfall Trap), whether the site was mountainous or not and the reference number (Appendix 2).

Species	St	Yr	Mean	#	Ave %	CV	3%	4%	Type	MTN	Ref
Appalachian	NC	19	46.66			20	100	100	NS	X	15
Red-backed	VA	14	35.00			23	96	100	NC	X	3
Eft	CT	4	0.65			23	100	100	NC		14
Mtn Dusky	NC	3	31.39			26	97	99	NS	X	13
Red-backed	CT	4	1.01			26	100	100	NC		2
Red-backed	ME	8	34.95	360	9.7	28	100	100	AC		4
Clouded	BC	3				28	99	100	AC		5
W. Red-	BC	3				30	99	100	AC		5

backed												
Mtn Dusky	NC	7				30	97	99	MR	X	12	
Green	NC	2				31	94	99	NS	X	9	
Red-backed	PA	1	67.29	200	33.5	34	94	100	AC		21	
Appalachian	NC	8	34.08			35	98	100	NS	X	6	
CA Slender	CA	2	26.2			42	92	100	AC/NC		17	
Peaks of	VA	1	0.24			42	94	99	NS	X	7	
Otter												
Mtn. Dusky	WV	6	18.4	135	13.6	52	91	97	AC	X	20	
Ensatina	CA	5				54	90	99	AC/NC		16	
Red-backed	WV	6	20.8	135	15.4	55	90	98	AC	X	20	
Red-backed	MI	1				55	88	95	NC		8	
Slimy	NC	19	14.3			65	86	96	NS	X	15	
Pacific	CA	2	23.4			67	86	96	NC/AC		10	
Slender												
Del Norte	CA	3	3513			74	89	97	MR	X	19	
Red-backed	MA	2				80	84	97	AC		18	
Ensatina	CA	6	26.71			109	79	91	PF	X	11	
Red-backed	NJ	3	2.11	50+	4.2	110	77	91	AC/NC		1	

Marsh-Billings-Rockefeller NHP Sampling Design and Array Setup

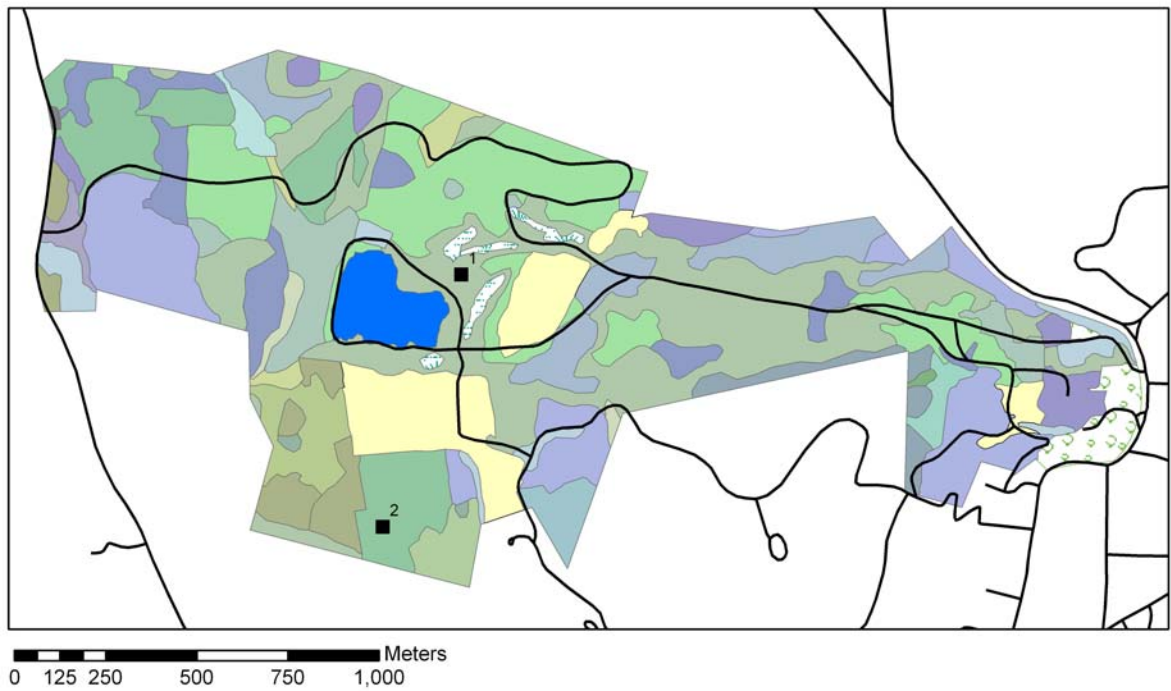
Sampling arrays will be located near long-term forest condition monitoring plots. The arrays will not be at the long-term plots in order to reduce impacts on the forest plots; arrays will be placed 40 m away from the forest plots, and within the same stand whenever possible. NOTE: it is very important that classes avoid disturbing the forest monitoring plots. If you notice that you are in a plot where all of the trees have metal tags, please move your class outside of the area.

This salamander monitoring project will begin with two arrays of 50 stations each (a 5 x 10 array); stations will be 10 m apart, and each will have a pair of cover objects spaced one meter apart. One of the arrays will be in the 1952 red pine plantation, and the other array will be in hemlock-northern hardwood forest. The number of arrays and number of cover objects per array can be adjusted as data is collected and the optimum level of participation is determined. Jim Andrews of Middlebury College has suggested that 15 coverboard stations per array or transect should be adequate (Jim Andrews, pers. comm.), but using 50 stations will provide more of an opportunity for all students to participate in the different aspects of data collection.

Table 1 and Figure 1 display the locations of the two salamander arrays.

Table 1: Corner coordinates for salamander arrays (UTM zone 18N, NAD 1983)

Corner	X coordinate	Y coordinate	Corner	X coordinate	Y coordinate
1A	698401	4834200	2A	698186	4833507
1B	698401	4834240	2B	698186	4833547
1C	698491	4834240	2C	698276	4833547
1D	698491	4834200	2D	698276	4833507



Salamander Arrays at Marsh-Billings-Rockefeller NHP

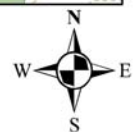
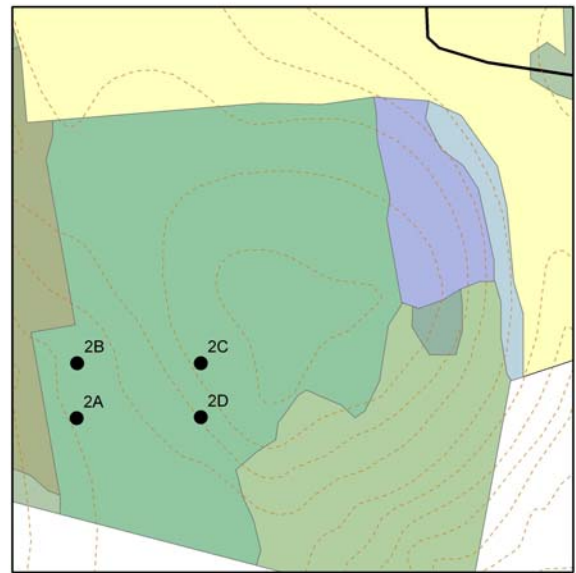
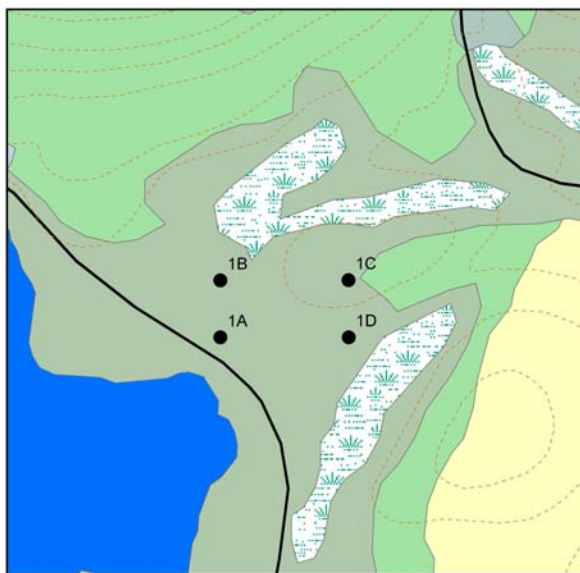


Figure 1: Map of the two salamander arrays.

Establishing each array will require 100 labeled white or red oak boards measuring 20 x 30 x 2.5 cm (approximately 8 x 12 x 1 inches), 4 rebars or other posts for marking corners, a GPS for locating plot boundaries, a compass (with declination set correctly), and a 100 m measuring tape. Locate and mark corners A and D, and run the tape between these corners as straight as possible. Place a cover board 0.25 m north and 0.25 m south of each corner, and 0.5 m north and south every 10 meters along the measuring tape (each pair of boards is one station). Make sure to remove all litter and level the mineral soil before placing each cover object. If a tree or other object is in the way, place the board as close as possible to the desired location.

After establishing the first row of stations, use the GPS and locate and mark corners B and C. Run the measuring tape between A and B, and establish stations at the corner and every 10 meters between the corners (at 10 m, 20 m, and 30 m). The cover boards should always be placed 0.25 m north and 0.25 m south of the station point. Repeat this process with corners C and D.

Next, run the tape between the stations that have been established 10 m north of corners A and D, and establish the stations between them. Continue with the stations 20 m north of the line between A and D, the 30 m stations, and then the stations between corners B and C (Figure 2)

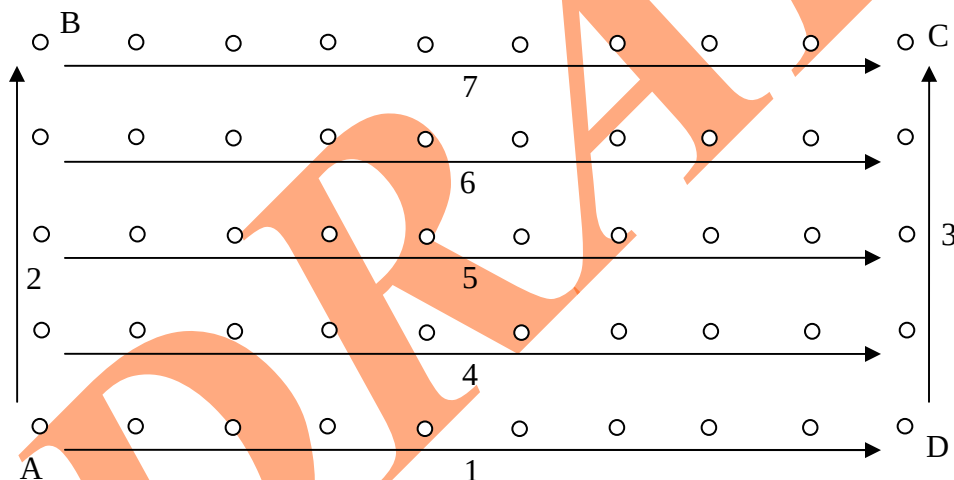


Figure 2: Diagram of stations in an array, with corners (letters) and suggested order of station establishment (numbers). Each station consists of two boards, 0.25 m north and south of the point indicated.

Additional Details About Cover Object Arrays

This section includes the rationale for the array design above. Note that there are other valid ways to monitor terrestrial salamander populations, in particular those using mark-recapture estimators. However, those techniques are most appropriate for academic and agency use since they require more time, special techniques, and can result in the injury of the salamanders.

Transects or arrays of cover objects can be installed anywhere there are terrestrial salamanders to crawl under them. The objective of the monitoring program is to look at patterns of changes of salamanders in relationship to local and regional habitat, climate, location, and landscape factors. Monitoring at old-growth and disturbed sites is particularly important; data from these types of sites may have particular relevance for understanding forest herps and their habitat relationships.

Ancient or old-growth sites are those sites that have had little disturbance due to cutting, grazing, plowing, or other major anthropogenic disturbance. These sites act as reservoirs of plant and animal populations, and, while all are inescapably affected by the air, water, and landscape changes that occur around them, they represent something of a natural control for comparison with other sites. Also, if widespread declines are detected in these, the most optimal of salamander sites, then we have grounds for increased worry regarding global changes.

Disturbed sites are sites where salamander populations are or are likely to become stressed. Such sites could include fragmented woodlands, urban/suburban sites, forest management sites, grazed woodlands. Sites could be established prior to an impact, during the impact, or in the aftermath of these changes. As a general rule, the situation with the most potential is one where the monitoring can begin well before a disturbance takes place.

Scouting. Potential monitoring sites should be scouted to determine if populations of any of the target monitoring species are present.

Target Species. Species known to occur regularly and consistently under artificial cover objects are Red-backed Salamander, Slimy Salamander (not under small wood boards, however, in WV (T. Pauley pers. comm)), Western Red-backed Salamander, Del Norte Salamander (*Plethodon elongatus*), *Ensatina* (*Ensatina eschscholtzii*), Mountain Dusky Salamander, California Slender Salamander, Pacific Slender Salamander (*Batrachoseps pacificus*), Dwarf Salamander (*Eurecea quadridigitata*), Clouded Salamander, Rough-skinned Newt (*Taricha granulosa*), Eastern Newt (*Notophthalmus viridescens*). In the northeastern United States, the most likely species are the Red-backed Salamander and efts (juveniles) of the Eastern Newt. Any species that is found regularly under natural cover objects should be an appropriate target species. Ambystomas and stream-side salamanders also occur periodically under terrestrial cover objects, but appear to rarely occur with sufficient numbers to warrant anything but the evaluation of very long-term changes.

Tension Zones. Tension zones between closely related (and potentially competing) species should be avoided, until the effects of interspecific competition on the results are determined.

Distance from Edge. Cover objects should be placed so they are more than 50 m from the edge of the forest. Most species are affected by forest edges only to a distance of 25-35m (deMaynadier and Hunter In press).

Habitat. The transect/array should traverse only one habitat, if at all possible.

Habitat Discontinuity. Transects can be divided into several subsections to avoid inappropriate habitat.

Pairing Boards. To minimize the potential effects of territoriality, cover objects (if using single boards) should be laid out in pairs, 0.5 m apart.

Distance Apart. Based on recapture distances for several species, distance between sets of cover objects should be a minimum of 6m (Mathis et al. 1995).

Numbers of Cover Objects in a Transect. The number of cover objects (or clusters of objects in some cases) in a transect should be enough so that salamander counts average 10 or more per check (during the season of peak abundance of salamanders). A rough estimate of this number can be made by scouting a prospective area during the appropriate season and counting the proportion of natural cover objects with salamanders under them. Double this proportion and use that number to estimate the number of cover objects to put in place. For example, if you found that 1 out of 10 natural cover objects had a salamander under it in your scouting runs, then you would want 50 cover objects in your array. Another way to approach the problem is to put 100 objects out in a suspected low salamander density area, 50 at moderate densities, and 25 at high densities.

Numbering Cover Objects. Each object in the array should be assigned an individual number, starting with the number 1.

First Year Effects. Data from the first year an array is established should not be used to calculate long-term trends. Newly-placed cover objects (as opposed to new objects that replace already existing cover objects) appear to have depressed capture rates in relation to ones that have been there for a long period of time (Grant et al. 1992, L. Monti unpublished data, Gary Schneider pers. comm.)

Transect vs. Array. The objects are best placed in a transect (to cover the greatest area) but can be curved to fit topography or put in a matrix to fit the limitations of the site. We have elected to use an array because of logistical constraints (i.e., need to keep a school group close together), and in order to keep the array close to permanent forest monitoring sites.

Installing a Cover Object. When installing a cover object the leaf litter should be removed, the mineral soil leveled, and the board placed so its entire surface is in contact with the earth. Boards that are not in contact with the earth are not attractive to most of the *Plethodon* genus (Bonin and Bachand 1997, Gary Schneider pers. comm.). The bark-

hiding species find 2-layered boards more attractive (Davis 1997), but it is not worth the extra effort if only Plethodons are present (Bonin and Bachand 1997).

Changes in the Cover Object Type - Once Established. Experiments to date (Monti 1996) indicate that wood boards/shingles work the best. Sets of bricks also work satisfactorily for eastern salamanders, plastic bags may work for western species. Once a cover object type and dimensions are decided upon for a site, it must be maintained, ad infinitum, for the sake of repeatability. Because cover objects appear to have different rates of attractability, they cannot be interchanged without biasing the resulting trends.

Advantages and disadvantages by type of cover object:

Wood. Wood appears to often be the most preferred substrate in preference tests (Monti 1996, Grant et al. 1992) and in comparisons of rock vs. log natural cover objects (Bogert 1952). Most researchers in the past have used wooden cover objects. Rot eventually destroys their usefulness (2-10 years). Type, dimensions, and species of board must remain the same over time.

Wood Species Characteristics. No studies have been published that have documented a preference among salamanders for one species type of board vs. another. However, given our understanding of the needs of salamanders for moist environments and the moisture retention characteristics of wood, some general principles emerge and should serve well until direct experimental evidence comes in.

Theoretically, the best cover object is one that retains as much moisture as possible for the longest period of time. Thus green (fresh-cut) lumber is preferable to dried (particularly kiln-dried). Green lumber can be obtained directly from a local sawmill.

Different species of wood retain water differently. That ability is reflected in their relative drying time rates. We will use white oak cover boards because white oak is a native species with wood that is highly resistant to rot (USFS 1977, Jim Andrews pers. comm.).

Small vs. Large Boards. In the Northeast and Appalachian Plateau, small boards or shingles appear to be more attractive to most species than large boards, apparently due to the increased moisture under the boards (T. Pauley, L. Monti pers. comm.). Results from tests of preferences for large (24 X 48 X 3/8 in.) vs. small (7 X 42 X 0.5 in.) plywood cover objects indicated that significantly more salamanders were found under the small boards (both per object and even more so per unit area) and that larger boards were significantly drier and warmer (K. Heister pers. comm.). In the drier parts of the West, and perhaps in dry portions of the East too, larger boards or even plywood may be more appropriate in the drier/scrub sections - it is not at all clear. A drawback of larger board sizes is that in regions where the soil surface is rocky or uneven it is difficult to get soil contact along the entire length of the wood, resulting in decreased attractiveness and warping (G. Schneider pers. comm.). In the rainforests of British Columbia Ted Davis has had good success using built-up boards with Western Red-backed Salamanders occurring between the board and the soil and Clouded Salamanders within the built up

sections of the board (Davis 1996, Fellers and Drost 1994). Slabwood from sawmills should not be used because of the variation in thickness, size, and species. We recommend, that until more information comes in that most groups consider installing arrays of approximately 12" X 12" X 1" to 8" X 12" X 1" natural wood boards.

Brick (Clusters of 6 or more common bricks). Closely-spaced bricks permit moisture to seep into the interior of the cluster as well as providing additional hiding places in the cracks. Bricks are heavy to haul into the site, initial costs are usually higher than wood, and checking the arrays can take longer. However, they are permanent and may be more difficult for predators to disturb. J. Gibbs has found bricks to work well with Mountain Dusky and Red-backed salamanders as well as Eastern Newts in the Adirondack Mountains of New York.

Plastic Bags (roughly 1m X 1m black plastic). Experience seems to indicate that plastic dries out the ground too much for eastern woodland salamanders. Bags probably need to be replaced every year. The edges need to be anchored to prevent them from blowing away, making checking more tedious than boards. Finally, the esthetics of plastic are less than desirable for many land managers.

Other Materials. Cardboard, plywood, and masonry block all have been found with salamanders under them, but have various disadvantages of weight, expense, cost, or environmental problems in addition to much lower attractiveness than wood. Other objects should continue to be tested and evaluated. Galvanized metal, when tested against wood, has been found to be very inferior to wood in the coastal plain of South Carolina (Grant et al. 1992). Dinner plates have been tested and in most regions, were not very attractive to salamanders (Monti 1996) (but then, who would want to live under a dinner plate?).

Rules for Moving Cover Objects Already in Place. Rules for deciding what situations will require moving or replacing individual cover objects will need to be developed.

Permanent Marking of Transect Locations. Some permanent means of marking the arrays should be established, both visible and concealed, in case of vandalism or disuse. Two foot pieces of rebar pounded into the ground at every 5th cover object can be relocated with a metal detector many years later. Small flags are useful for refinding boards that are covered by leaves.

Data Collection

Time of Year. High air moisture, calm wind conditions, high soil and litter moisture, and low (but above freezing) temperatures are the physiologically optimal conditions that prompt salamanders to move out of their soil retreats and under cover objects (Feder 1983). As many studies have shown early spring and very late fall (especially pronounced in Eastern populations) are when salamander numbers reach their highest surface densities, particularly the younger age classes which are the most vulnerable group to dessication. The Fall peaks in counts (in the East) tend to be compressed into the last few

weeks prior to the first hard freeze, and often average higher than spring counts (Bonin and Bachand 1997), probably due to courting activities. In fact, numbers can drop precipitously (even after it warms back up) following the first freeze (Hairston 1983, Hendrickson 1954). In cool moist northern and eastern mountain areas salamander populations may occur throughout the summer; however, all data sets (most unpublished) show that the summer numbers are low (on average) compared to those of the spring or fall. Inclusion of mid-summer data almost always leads to increased sampling variability and bias (see Appendix 1) and therefore decreased ability to detect trends.

General recommendations for the timing of taking counts (assuming no prior knowledge about the surface abundance patterns at a site) are as follows:

Western. In the West, the fall sampling period should begin after the first soaking set of rains, and cease when the first hard freeze occurs. The spring sampling period following the mid-winter thaw and ends when temperatures move above the 50s (10oC+) or the rains stop.

Eastern. In the East, the fall sampling period should begin 1 month prior to the average hard freeze data in the fall and ends with the first hard freeze. The spring sampling period starts following the complete thawing of the soil column, roughly when temperatures move into the upper 30s and low 40s. Sampling should stop about 2 months after that date or if daytime temperatures begin to regularly exceed the 50s.

Northern. In the Northern interior latitudes more work needs to be done to determine the best sampling strategy, for now, it would be best to follow the general Eastern recommendations.

Further Work. See Appendix 1 for experiments that will help further quantify moisture and temperature relationships.

When to Check Cover Objects. Arrays should not be checked if it is misty or rainy, times when salamanders may be out foraging in the litter. Taub (1961) showed no strong relationship between rainfall and captures under cover objects. However, in the Mediterranean zones of California Fellers and Drost (1994) report that captures appear to be higher after winter storms, and Jaeger (1979, 1980a) found presence of Red-backed Salamanders under cover objects in western Virginia to be directly but negatively related to rain in the previous 24h. Consequently, until we know more about the relationship between rainfall and counts, we recommend that checks for salamanders should not occur within 24 hours of a rain.

Number of Times Arrays Should be Checked Within a Year. Computer models of sampling power have shown that arrays should be checked at least 3 times during a spring or fall sampling season.

Check Intervals. Nothing is known about what the disturbance factor that checking cover objects may have on the probability of subsequent counts of salamanders. Weekly

intervals between checks do not appear to be harmful and are the current recommended minimal interval. See Appendix 1 for experiments to determine the effect that board checking may have on salamander occurrence under boards.

Temperature. Taub (1961) found that captures of Red-backed Salamanders in the Piedmont of New Jersey were highest when temperatures just above the litter were from 7.5-15C. Finer temperature differences were not found to have a relationship (at least a strong one) with presence of Red-backed Salamanders under cover objects over an 11 day period in the mountains of western Virginia (Jaeger et al. 1995). As long as the temperature is above freezing there are currently no temperature recommendations for checking boards.

Data to Record During a Check of Cover Objects. Data collected on the day of the count should include the date, time, temperature, wind speed, cloud cover, amount of rain within 24 hours, amount of rain within the week, and observer(s) name. Core data collected for each species should include species, relative body and tail length measured using the grid method outlined below (see Measurements section below), and the object number. Ancillary data would include sex, the presence of worms, ants, non-target amphibians and reptiles, or ant nests under the cover objects and would be up to the interest of the observer to record.

Measurements. We feel that direct handling of salamanders to get precise measurements is too stressful, which, being small and energy minimizers, burns up an exorbitant amount of energy in their struggles. However, sizes of salamanders are useful for determining rough age classes. Ratios of large to small salamanders are an indication of breeding success and cohort size. While sizes can be estimated by eye, there is great potential for error and bias in such a process. Three general techniques can be used to get salamander measurements.

Box. A small clear plastic box can be made with a 5 mm grid measured out in indelible pen on the bottom. By putting the salamander in the box and waiting for it to remain in a relatively straight position, the number of 5 mm boxes and partial boxes its body lies can be counted. Keep a bit of moisture in the bottom for the salamanders benefit and to keep them from moving around as much.

Mander Masher. Wise and Buchanan (1992) have designed a sandwich device composed of a piece of plexiglass on one side and a small sponge attached to a piece of sheet aluminum or board on the other. Those 2 pieces are hinged together with metal hinges or duct tape. A set of rigid stops made of wood, bolts, etc. are placed at the corners to prevent too much compression of the salamander into the sponge. Salamanders are placed on the wetted glass side and when they are roughly straight the top of the masher (i.e., with the sponge) is closed and the salamander can be measured through the glass.

Plastic Baggie. The salamander to be measured is placed in a plastic ziplock baggie. The bag is put on a hard surface (e.g., a clipboard) and a thin swizzle stick sized rod is used to press them against the side of the baggie where they can be measured.

Measurements should be made (in millimeters) from the tip of the snout to the posterior end of the cloaca and then from the posterior end of the cloacae to the end of the tail using a ruler or calipers. If you are using the salamander in a box technique, then those two measurements should also be estimated, as many salamanders lose portions of their tails.

Crushing Salamanders. After an animal is found under a cover object, it should be captured and released immediately after measuring next to the object so that it can crawl back under without being crushed.

Predator Proofing. In a few cases large mammals have been known to flip cover objects while searching for food (R. Jaeger pers. comm.). No effective predator proofing has yet been devised, but it may become more of an issue once permanent systems of cover arrays become established. Rocks placed on top of the boards can be used in regions where they are available. See Appendix 1 for some ideas that need be testing.

Data Entry. All data and information are managed on the World Wide Web. Dan Lantz (dlantz@minn.net) can also mail out information packages by email or snail mail. Dan's registration and data entry site is now ready for your information. [MAY GO WITH A SPREADSHEET OR DATABASE INSTEAD]

Data Forms. Field forms can be developed by the participant. If you develop your own field sheets be sure to include the main fields listed in the protocol. Dan Lantz has some examples of field forms. [NEED TO GET EXAMPLE FIELD FORMS]

Literature Cited

Bishop, C. 1996. Calling amphibian surveys. NAAMP Protocol Document. <http://www.im.nbs.gov/amphib/naampcall.html>.

Bogert, C.M. 1952. Relative abundance, habitats, and normal thermal levels of some Virginian salamanders. *Ecology* 33:16-30.

Bonin, J. and Y. Bachand. 1997. The use of artificial covers to survey terrestrial salamanders in Québec. *Herpetological Conservation* 1:175-179.

Burton, T.M. and G.E. Likens. 1975. Salamander populations and biomass in the Hubbard Brook Experimental Forest, New Hampshire. *Copeia* 1975:541-546.

Cunningham, J.D. 1960. Aspects of the ecology of the Pacific Slender Salamander *Batrachoseps pacificus*, in southern California. *Ecology* 41:88-99.

Davis, T. M. 1996. Distribution, abundance, microhabitat use and interspecific relationships among terrestrial salamanders on Vancouver Island, British Columbia, University of Victoria. Doctoral Dissertation.

---. 1997. Nondisruptive monitoring of terrestrial salamanders with artificial cover objects on southern Vancouver Island, British Columbia. *Herpetological Conservation* 1:161-174.

DeGraaf, R.M. and M. Yamasaki. 1992. A nondestructive technique to monitor the relative abundance of terrestrial salamanders. *Wild. Soc. Bull.* 20:260-264.

deMaynadier, P.G. and M.L. Hunter Jr. 1995. The relationship between forest management and amphibian ecology: a review of the North American literature. *Environmental Review* 3:230-261.

---. In press. Effects of silvicultural edges on the distribution and abundance of amphibians in Maine. *Conservation Biology*.

Droege, S. 1996. A heuristic approach to validating monitoring programs based on count indices (damn the statisticians, full speed ahead). <http://www.im.nbs.gov/naamp3/papers/44nn.html>

Feder, M.E. 1983. Integrating the ecology and physiology of plethodontid salamanders. *Herpetologica* 39:291-310.

---. 1982. Thermal ecology of neotropical lungless salamanders (Amphibia: Plethodontidae): environmental temperatures and behavioral responses. *Ecology* 63:1665-1674.

Fellers, G.M. and C.A. Drost. 1994. Sampling with artificial cover. In Heyer, W.R., M.A. Donnelly, R.W. McDiarmid, and others. Editors. *Measuring and monitoring biological diversity: Standard methods for amphibians*. Washington, Smithsonian Institution Press. p. 146-150.

Gordon, R.E., J.A. MacMahon, and D.B. Wake. 1962. Relative abundance, microhabitat and behavior of some southern Appalachian salamanders. *Zoologica* 47:9-14.

Grant, B. W., A.D. Tucker, J.E. Lovich and others. 1992. The use of coverboards in estimating patterns of reptile and amphibian biodiversity. In McCullough, D. R., and Barrett, R. H. Editors. *Wildlife 2001*. London, England, Elsevier Science Publ. p. 379-403.

Hairston, N.G. 1983. Growth, survival and reproduction of *Plethodon jordani*. Trade-offs between selective pressures: *Copeia* 1983:1024-1035.

---. 1987. *Community ecology and salamander guilds*: Cambridge, Cambridge University Press.

---. 1986. Species packing in *Desmognathus* salamanders: experimental demonstration of predation and competition. *American Naturalist* 127:266-291.

Hairston, N.G. and R.H. Wiley. 1993. No decline in salamander (Amphibia: Caudata) populations: a twenty-year study in the southern Appalachians. *Brimleyana* 18:59-64.

Heatwole, H. 1963. Environmental factors influencing local distribution and activity of the salamander, *Plethodon cinereus*. *Ecology* 43:460-472.

Hendrickson, J.R. 1954. Ecology and systematics of salamanders of the genus *Batrachoseps*. *Univ. Calif. Publ. Zool.* 54:1-46.

Herrington, R.E. and J.H. Larsen. 1985. Current status, habitat requirements and management of the Larch Mountain salamander *Plethodon larselli* (Burns). *Biological Conservation* 34:169-179.

Highton, R. 1995. Speciation in eastern North American salamanders of the genus *Plethodon*: *Ann. Rev. Ecol. Syst.* 26:579-600.

Jaeger, R.G. 1979. Seasonal spatial distributions of the terrestrial salamander *Plethodon cinereus*. *Herpetologica* 35:90-93.

---. 1980a. Microhabitats of a terrestrial forest salamander. *Copeia* 1980:265-268.

---. 1980b. Density-dependent and density-independent causes of extinction of a salamander population. *Evolution* 34:617-621.

Kramer, P., Reichenbach, N., Hayslett, M., and others. 1993. Population dynamics and conservation of the Peaks of Otter Salamander, *Plethodon hubrichti*. *J. Herpetology* 27:431-435.

Nishikawa, K.C. and P.M. Service. 1988. A fluorescent marking technique for individual recognition of terrestrial salamanders. *J. Herpetology* 2:351-353.

Larsen, L. O. 1992. Feeding and Digestion. In Feder, M.E. and W.W. Burggren. Editors. *Environmental physiology of the amphibians*. p.378-394.

Mathis, A. 1989. Do seasonal spatial distributions in a terrestrial salamander reflect reproductive behavior or territoriality? *Copeia* 1989:788-791.

---. 1990. Territoriality in a terrestrial salamander: The influence of resource quality and body size. *Behaviour* 112:162-175.

Mathis, A., R.G. Jaeger, W.H. Keen and others. 1995. Aggression and territoriality by salamanders and a comparison with the territorial behaviour of frogs. In. Heatwole, H.

and B.K. Sullivan, Editors. Amphibian Biology, Vol. 2. Social Behaviour: Chipping Norton, NSW, Australia, Surrey Beatty and Sons, p. 633-676.

Meier, A. J., S.P. Bratton, and D.C. Duff. 1996. Biodiversity in the herbaceous layer and salamanders in Appalachian primary forests, In Davis, M. B., Editor. Eastern old-growth forests: Washington D.C., Island Press, p. 49-64.

Monti, L. In prep. The Redback Salamanders (*Plethodon cinereus*) Population in a Maine Oak- Pine Forest. Master's Thesis, due for submission at U. Maine. December 1997.

Monti, L., M. Hunter Jr., J. Witham. 2000. An evaluation of the artificial cover object (ACO) method for monitoring populations of the redback salamander *Plethodon cinereus*. *Journal of Herpetology* 34:624-629.

Pough, F.H. 1980. The advantages of ectothermy for tetrapods. *Am. Nat.* 115:92-112.

---. 1983. Amphibians and reptiles as low-energy systems. In Aspey, W.P. and S.I. Lustick, Editors. Behavior energetics: the cost of survival in vertebrates. Ohio State University Press, Columbus, Ohio. p. 141-188.

Stebbins, R.C. 1954. Natural history of the salamanders of the plethodontid genus *Desmognathus*. U. Calif. Publ. Zool. 54:47-124.

Stebbins, R.C. and N.W. Cohen. 1995. A natural history of amphibians. Princeton University Press, Princeton, NJ.

Sugar, A., T. Bellhouse, D. Phoenix, N. Dawson, and G. Holborn. 2001. A sampling protocol for red-backed salamander (*Plethodon cinereus*) populations in Ontario: the 2nd pilot study. Wildlife Assessment Program, Ontario Ministry of Natural Resources WAP-2001-01.

Taub, F.B. 1961. The distribution of the red-backed salamander, *Plethodon c. cinereus*, within the soil. *Ecology* 42:681-698.

Test, F.H. 1948. Census of a population of the red-backed salamander (*Plethodon cinereus*). *Am. Midl. Nat.* 39:362-372.

Tilley, S.G. 1980. Life histories and comparative demography of two salamander populations. *Copeia* 1980:806-821.

Vial, J.L. and L. Saylor. 1993. The status of amphibian populations: A compilation and analysis. International Union for Conservation of Nature and Natural Resources. Species Survival Commission. Declining Amphibian Populations Task Force. Corvallis, OR, Center for Analysis of Environmental Change.

Wells, K.D. 1980. Spatial associations among individuals in a population of slimy salamanders (*Plethodon glutinosus*). *Herpetologica* 36:271-275.

Welsh Jr., H.H. 1990. Relictual amphibians and old-growth forests. *Conservation Biology* 4:309-319.

Welsh Jr., H.H. and S. Droege. 2001. A case for using Plethodontid Salamanders for monitoring biodiversity and ecosystem integrity of North American Forests. *Conservation Biology* 15:558-569.

Welsh Jr., H.H. and A.J. Lind. 1991. Population ecology of two relictual salamanders from the Klamath Mountains of northwestern California. In McCullough, D. R. and Barrett, R. H., Editors. *Wildlife 2001: Populations*. New York, Elsevier Applied Science, p. 419-437.

White, F.N. 1978. Comparative aspects of vertebrate cardiorespiratory physiology. *Ann. Rev. Physiol.* 40:471-499.

Wise, S.E. and B.W. Buchanan. 1992. An efficient method for measuring salamanders. *Herpetological Review* 23:56-57.

Appendix 1: Salamander Experiments

The text of the main salamander monitoring document summarizes the many studies that have used, as part of their investigations, some aspect of locating salamanders under natural or artificial cover objects. Those reports include both population and ecological investigations. Enough can be gleaned from these papers to confidently propose the creation of a long-term monitoring program for terrestrial salamanders. However, it is also clear that few studies to date have focused on optimizing these methodologies for the purpose of long-term monitoring. We know that the relationships between occurrence of salamanders under cover objects and time of year, weather, and the total population appear to vary to some degree with species and location. But the controlling factors are not completely clear, though they probably revolve around the relationship between soil moisture and freezing temperatures.

We outline here a number of experiments that we hope will interest the greater scientific, education, and salamanderphilic community. Some experiments are quite simple, for example, investigating the preference of salamanders for different types of cover objects. While others should be attempted only by those experienced with marking salamanders and have the time to complete the project.

The creators of the national salamander protocol will, for those interested and willing to share their information, document those groups who are running these tests and will post their results. The results from these experiments will greatly help the Terrestrial Salamander Monitoring Network develop into a useful, precise, and statistically sound

conservation and monitoring program. A program that is rigorously designed is one that will be powerful in both the statistical and conservation sense. If we expect our data to be used by policy makers and scientists to direct conservation activities, money, and priorities our network cannot rely upon untested assumptions. If we look at the contentious world of environmental contaminants studies, undocumented work is quickly shredded by the opponents' scientific experts, often rightly so. We can do better. Please consider both establishing a coverboard monitoring array and running some of the experiments below.

For questions, or to register your intentions please contact Sam Droege (sam_droege@usgs.gov, 301-497-5840).

Salamander Experiment 1. Investigating the Distribution of Counts of Salamanders Over Time.

Objective. The objective of this experiment is to determine the optimal sampling time for salamanders using artificial cover objects. This is a useful exercise for any group establishing a cover object monitoring station. Unlike the other experiments, it does not require the creation of a special array, your monitoring array may be used.

Location. See the main document for information about how to choose a place to sample and how to estimate the number of cover objects you need.

Test. Salamander cover objects are checked weekly using the standard handling and measurement procedures. Checks should begin whenever it is remotely possible to find salamanders under the boards and end once salamanders again disappear from under the boards.

Suggested presentation of data. A simple x-y chart of time on the x-axis and count on the y can be used to graph out the relationship between time of year and count. By inspecting the graph you should be able to detect periods when salamander counts are high and relatively stable. Stability can be measured by calculating the mean and standard deviation (specifically the sample standard deviation not the population standard deviation) of what appears to be the best time period. The coefficient of variation (CV) provides a standardized measurement of variability and is calculated by dividing the standard deviation by the mean and multiplying by 100. The CV is useful when compare data with different means (from either different periods or other sites and species). Any period with a coefficient of variation of less than 50% is a good indication of an appropriate time period for counting salamanders.

Salamander Experiment 2. Cover Object Comparisons.

Objective. The objective of this class of experiments is to determine preferences of salamander species for different configurations and types of cover objects.

Location. See the main document for information about how to choose a place to sample and how to estimate the number of cover objects you need.

General Test Procedure. Pairs of cover objects, 0.1 m apart, are placed on site 6m apart. The number of pairs should be sufficient to average about 20 salamanders under them in the right situation (see the primary document for how to estimate that). For those of you who relish complex ANOVA analyses, these tests can be generalized beyond the paired comparisons listed in the Test Group section below.

Test Groups.

1ft. X 1ft. natural board vs. 6in. X 6in. natural board
1ft. X 1ft. natural board vs. 1ft. X 1ft. ½in. thick plywood
1ft. X 1ft. plywood vs. 2ft. X 2ft. plywood
1ft. X 1ft. natural wood vs. black plastic garbage bag (plastic bags appear to be primarily relevant in the dry West, but may be appropriate in the drier sections of the East)
1ft. X 1ft. natural wood vs. (4) 6in. X 6in. natural wood boards spaced 1/2in. apart
(4) 6in. X 6in. natural wood boards vs. (8) 6in. X 6in. natural wood boards

Other testing pairs that individuals feel are appropriate are also welcome.

Checking the test pairs. Test cover objects should be established for at least 3 months before starting the test and it would be wise to continue the test over several sampling seasons to make sure the initial results stay the same in case the establishment of the array has any effect.

Suggested presentation of data. A table showing the numbers of each species under each cover type broken down by season, if appropriate. Paired t-test should be used to see if the numbers of salamanders differ.

Salamander Experiment 3. The Effects of Checking Cover Objects on Counts of Salamanders

Objective. The objective of these experiments is to determine if counts of salamanders are affected by previous checks of cover objects for salamanders.

Location. See the main document for information about how to choose a place to sample and how to estimate the number of cover objects you need. In this case you will want to have enough boards so that the number of salamanders per check is roughly 20.

Test 1. Pairs of cover objects, 0.1 m apart, are placed on the site 6m apart. A board from each pair is randomly chosen to be checked (flipping a coin is fine). Handling and measurements on any salamanders are made as in the general protocol. The chosen board is recorded. The next day both the chosen board and its mate are checked and data recorded as before.

Test 2. The procedure is the same as in Test 1, however, the interval between checks is not one day, but one week.

Test 3. The procedure is again the same as in Test 1, however, this time only single boards (not pairs) are placed 6m apart. One half the boards are again randomly chosen and the procedure in Test 1 used for checking.

Test 4. This variant uses the spacing of boards as outlined in Test 3, but the timing of checks as outlined in Test 2.

Checking the test pairs. Test cover objects should be established for at least 3 months before starting the test and the test should occur during the time when salamanders are most abundant under cover objects. If the same array of boards is used repeatedly for tests then a 2-week rest period with no checks should occur between test runs.

Suggested presentation of data. For each test, a table showing the numbers of each species found under checked boards (for both periods) and unchecked boards (for the second sampling period only, obviously). A 2-sample Chi-squared test can be used to check for significant differences in counts among checked and unchecked boards. A test for differences in the means in the 2 counts taken from checked boards can also be made, however, if significant differences exist, it will not be clear whether those differences were caused by differences due to being checked or differences due to different environmental conditions across the 2 days.

Salamander Experiment 4. What is the Relationship Between Numbers of Salamanders under Artificial Cover Objects and Numbers of Salamanders Under Natural Cover Objects?

Objective. The objective of these experiments is to determine if counts of salamanders are affected by changes in the number of natural cover objects in the same area.

Location. See the main document for information about how to choose a place to sample and how to estimate the number of cover objects you need. In this case you will want to have enough boards so that the number of salamanders per check is roughly 10-20.

Test. This experiment actually combines a validation of the technique with a test for the effects of natural cover objects impacts. Cover boards are placed in a grid 6m apart. Those boards are allowed to weather-in for 3 months. A metal or silt barrier fence is built around the grid, going down at least 1 foot into the ground to prevent salamanders from crossing under. The top of the fence needs to have a lip barrier on both sides to prevent salamanders from climbing out or climbing in (they readily climb trees and other plants). See validation experiment (not written, as of yet) for the procedure to use for removing salamanders from the plot and using that to estimate their total populations.

Because natural changes in the number of salamanders under boards could confound the results of this experiment a control site should also be monitored to correct for natural

changes. The control plot need not have the removal or fencing. The control sites should be checked on the same days as the experimental plot(s). While a treatment plot and a control plot may be sufficient to look at effects a more robust design would have 2 controls and 4 treatment plots. All plots should be in the same woodland and soil type.

After salamanders are removed a known number of salamanders can be returned to the plot, this should be done during the time of year in which salamander are at the surface. Let the salamanders acclimatize for 2 weeks and then do two checks of the cover board array (following standard procedures) spaced one week apart. Following the last check randomly remove 25% of the natural cover objects (calculate that percentage as the percentage of linear area in contact with the soil). Record both the surface area of natural cover removed, the number of objects, and the length of those objects. Wait a week and do 2 more sets of counts spaced a week apart, as before. Repeat this procedure while removing 50%, 75%, and 100% of the natural cover.

After the completion of the initial experiment, logs can be added back (in increments of 25%) to the treatment plots to see if the same results occur (standardizing the counts using the controls).

Suggested presentation of data. A table (or set of tables) showing the numbers of each species under the boards, arranged by plot and each weekly check. Data from the control boards should be used to standardize data for the experimental plot(s). Counts should be transformed to account for non-normal distributions. Split-plot in time ANOVA (this is repeated measures situation) can be used to look for differences in mean count across sampling periods. Linear regression can be used to look for trends in the counts with increased removal of natural cover objects. A bar chart can be used to graphically present the differences among treatments and plots.

Validation Experiments. These are fairly complex undertakings, we haven't finished writing up this section. Stayed tuned.

Other Experiments. Those of you who would like to propose additional experiments are encouraged to do so. Please contact Sam Droege.

Contacts. To register or for further information please contact:

Sam Droege
Patuxent Wildlife Research Center
12100 Beech Forest Rd.
Laurel, MD 20708
sdroege@usgs.gov
w - 301-497-5840
h- 301-390-7759

Appendix 2: Power Analysis Details

Below is a more detailed listing of information summarized in Table 1. Each listing starts with its Reference Number, and then goes into species, research group, publication, and the specifics of the technique and field site, depending upon what information was available from the publication or the authors.

The data summary at the bottom of each account lists the Mean, Standard Deviation, and Coefficient of Variation. The line below these estimates includes the power estimates to detect a positive population change at levels that start with 1% per year and increase from there by steps of 1% (i.e., 1%, 2%, 3%, 4%, etc) levels. Levels that have 100% power are not listed. The next line presents the same power information, but for negative trends.

No attempt was made to detrend (correct for actual population changes) long-term data sets. Consequently, the CV's for the longer term counts are inflated (but not usually to a very great extent).

Note too that most of these studies have lumped all age classes together. Analyzing data for adults alone should further stabilize the numbers and increase the CV.

Reference 1

Data from Taub 1961.

Red-backed Salamander

Hutcheson Memorial Forest, eastern edge of East Millstone, Somerset Co., NJ. Mature Oak-Hickory. Piedmont.

70 X 90 foot plot, 39 pine boards (1 foot by 10 in. X 1 in.), one piece of tar paper, and natural cover. All of which were turned for every census. Data were used from mid-September to Mid-November, parts of 3 years. Included day, night, cold, hot, no attempt to standardize.

Mean 2.11, SD = 2.33, CV = 110.4

+34 57 86 95

-30 55 77 91 97 99

Reference 2

Red-backed Salamander

Data and site description were kindly provided by James Gibbs from Columbia University. *Plethodon cinereus* were counted by college students overturning all natural cover objects on 6x10 m plots (sample sizes range from 100 to 24) randomly located within the a 60 year-old oak forest during August over 4 consecutive years at the Yale Forest in NE Connecticut . 1993 was an unusually dry year, so this time series, though short, captures some important inter-annual variation.

Mean 1.01, SD 0.26, CV 26

+73

-71 98

Reference 3

Jaeger 1980b

1966-1979 Hawksbill Mountain, Blue Ridge Mountains, VA. 20 Spring and Autumn surveys during or just after a rain, under natural cover objects. 100 m² quadrat in deep soil. No trend line was apparent and no detrending of the data was attempted.

Mean 35, SD 7.99, CV 23

+73 97 99

-55 86 96

Reference 4

Red-backed Salamander

Data were kindly provided by Jack Witham, Laura Monti, and Mac Hunter from the University of Maine, Orono. Sixty stations composed of 6, 10x25 cm cedar shingles with an area of 1 meter squared were placed non-randomly throughout a 40 ha oak-pine study plot at the Holt Research Forest on the midcoastal island of Arrosic, Maine. Half of the cedar shingles were placed on the surface and the other half 2.5 cm below the soil surface. Data from 1990-1994 were used. No attempt was made to detrend the results nor correct for known seasonal differences in capture rates. From 5-7 checks of arrays were made each year.

Mean 34.95,SD 9.67,CV 28

+78

-68 94

Reference 5.

Western Red-backed Salamander

Data were kindly provided by Ted Davis from his Doctoral Dissertation (Davis 1996). Cover objects were composed of 2-layered 180x30cm boards designed to provide hiding spaces for salamanders that mimic loose bark (Davis, in press). Boards were placed in a range for conifer forest stands on Vancouver Island British Columbia (clearcuts to mature second growth). The number and arrangement of cover objects varied with each site ranging from 24-144. Detection rates per cover object for Western Red-backed Salamander were around 0.2. Because counts were depressed in the summer as

salamanders left the litter zone, only counts taken in the spring were used. Data were not combined across years.

Eight year and site combination analyses were performed on data for Western Red-backed Salamanders. In all cases power to detect trends exceeded the minimum goals, at times dramatically. Note that Davis found significant differences between counts taken in 1993 and 1994 for some populations. Because inter-year sampling variability was not included in the calculations, actual power would be less, but given the high power the current results provide, overall power should remain reasonable.

Median CV was 30
+56 94 99
-44 77 99

Clouded Salamander

Power for Clouded Salamander counts on one plot were also calculated.

Mean 6.17, SD 1.72, CV 27.9
+55 79 92
-57 90 99

Reference 6

Appalachian Salamander

Hairston, N. G. 1983

Pisgah National Forest, 14km NW of Rosman Transylvania County, Courthouse Creek. Forest type not reported. Data came from 2 24.83meter in diameter octagonal plots. 3 counts taken each year. Each count is the mean of 2 counts on each of the 2 plots taken within 2 weeks of each other , 1974-1981 Night search. Cold weather accounted for population lows.

Mean 34.08, SD 11.96,CV 35
+59 98
-50 90 98

Reference 7

Kramer et al. 1993

P. hubrichti

NPS near blue ridge parkway steep slope se aspect, mixed oaks, understory with logs and rocks in VA. 10mx10m area 121 subplots of 1m2 only surface active salamanders were

counted nothing was turned, night surveys were conducted during or shortly after a rain. Approximately 10 surveys were made, across all months.

Mean 0.24,SD 0.10,CV 42

+52 94

-42 81 94 99

Reference 8

Heatwole, H. 1962

Red-backed Salamander

Western edge of the village Carp Lake, Emmet Co. MI, beech-maple forest, 1958

searches taken by turning over same objects in July and first part of August

Under logs - 8 counts, Measured as: Mean number/m² area of log

Mean 0.67, SD 0.27,CV 40

+54 96

-47 84 95 98

Under small objects (mean number/small object)

Mean 0.056,SD .024,CV 42

+45 92

-46 76 93 97 99

Inside chunky logs (number of salamanders per cubic meter of wood)

Mean 28.17,SD 13.93,CV 49

+49 90

-34 80 95 99

Inside crumbly wood (number of salamanders per cubic meter of wood)

Mean 21.6,SD 19.27,CV 89

+30 74 87 99

-38 56 84 93

Reference 9

Gordon 1962

Green Salamander

Highlands, NC (1949-50) 2 sites. Rock face and south slope of Satulah Mountain and cliff and rock face area near highway 28.

Highlands site: (number of salamanders seen in crevices - during breeding season April-August)

Mean 6.94, SD 1.85, CV 27
+45 63 94
-52 91 98

Satulah Mountain

Mean 7.33, SD 2.57, CV 35
+38 71 97
-42 83 97

Reference 10

Cunningham 1960

Pacific Slender Salamander

Surface counts in a suburban backyard December 1955 - May 1956

Santa Monica, CA, Upper Sonoran life zone lawn and garden areas.

Mean 23.44, SD 15.82, CV 67
+35 66 94 99
-34 63 86 96 99 99

Reference 11

Ensatina

***Need more information

Mean 26.71, SD 29.13, CV 109
+33 57 79 98
-26 56 79 91 98 98

Reference 12

Tilley 1980 as written in Hairston 1987

Mountain Dusky Salamander

Mark-recapture of Mountain Dusky Salamanders captured at night on two vertical rock faces near Highlands, NC (1970-1976).

Whiteside

Mean 218.3,SD 45.25,CV 27
+88
-76 99

Dry Falls

Mean 71.8,SD 22.84,CV 32
+62
-54 94 98

Reference 13

Hairston 1986

Mountain Dusky Salamander

3 plots, 30m x 7m, night searches were made from spring to fall, all data were used, Nantahala Mountains, NC (1982-1984)

Mean 31.39,SD 8.12,CV 26
+95 98
-55 84 97 99

Reference 14

James Gibbs unpublished data

Eft

Same procedures as for Reference 2.

Mean 0.645,SD 0.151,CV 23
+80
-72 99

Reference 15

Hairston, data from Great Smokies files, but reported in Hairston and Wiley, 1993

Appalachian Salamander

Slimy Salamander

Data taken from September only 1973-1991 in the NC side of the Great Smoky Mountains. Groups of 15 students were told to go out at night and collect salamanders. Time constrained search on the ground at night and under cover. Numbers taken from data files not from the graphs

Appalachian Salamander

Mean 46.66, SD 9.29, CV 20
+68 87 92
-64 98

Slimy Salamander

Mean 14.29, SD 9.36, CV 65
+33 70 92
-36 59 86 96 99

Reference 16

Stebbins 1954

Ensatina

Contra Costa County, CA, Upper Sonoran Zone/Transition Zone along the Coast Range near San Francisco.

Searched weekly under boards and debris found on the site. Always searched under the same set of boards. 3 years of November data and 5 years of March data. No attempt was made to correct for weather, despite known effect of temperature and rainfall/soil moisture.

March data

Mean 19.59, SD 11.22, CV 57.3
+36 80 92 99
-35 66 90 98

November data

Mean 13.92, SD 7.23, CV 51.9
+40 80 98
-29 74 90 99

Reference 17

Hendrickson 1954

California Slender Salamander

Contra Costa County, CA, Upper Sonoran Zone/Transition Zone along the Coast Range near San Francisco.

Searched weekly under boards and debris found on the site. Always searched under the same set of boards. 2 years of data taken from November to March of each year. Data were not used on the two occasions when the surface was frozen and no salamanders were found, otherwise no attempt was made to include or exclude data from dry spells.

Mean 26.2, SD 11.1, CV 42

+41 86

-37 75 90

Reference 18

Red-backed Salamander

Southern New England forest type: upland oak, pine, mixed hardwoods

Data kindly supplied by Bob Brooks and come from 16 plots of 60 cover boards (1m X 25cm) each near the Quabbin Reservoir in central Massachusetts. Cover objects checked once a month from May to October in all types of weather and moisture conditions over 2 years.

Median CV for all 16 plots all months included in the analysis: 98.0

+26 61 87 99

-34 54 75 93 98

Median CV for all 16 plots with the dryish months of July, August, September removed: 79.5

+27 69 94 98

-32 63 84 97 98

Reference 19

Welsh and Lind 1991

Del Norte Salamander

Density estimates came from old-growth Douglas Fir (*Pseudotsuga menziesii*)/hardwood in the Klamath Mountains of Humboldt County, CA. Densities were among the highest

recorded for any salamander ($0.9/\text{m}^2 \pm 0.3$). Standard errors were high due to low capture probabilities. Estimates were made using mark-recapture methodology for 5 sampling periods over 3 years on a 4500 m² plot. The initial estimate used a simple Lincoln-Peterson estimator while subsequent ones Model A in program JOLLY.

Mean 3513.6, SD 2606.7, CV 74.2
+37 77 95
-36 64 89 97 99

Excluding the first estimate

Mean 4034.5, SD 2692.7, CV 66.7
+48 84
-32 68 88 98

Reference 20

Red-backed Salamander

Mountain Dusky Salamander

Data were kindly provided by Tom Pauley from his work at Fernow Experiment Station, Tucker County, West Virginia. Data were collected (1989-1994) from 27 sets of 5 pine boards (20 X 10 X 1.3-2.5 cm) placed in a domino pattern of 4 boards placed 60cm from the center board. Boards were checked periodically throughout the spring, summer, and fall.

When all data are used:

Red-backed Salamander

Mean 8.13, SD 8.85, CV 109
+29 60 85 94
-32 52 80 91 96 99

Mountain Dusky Salamander

Mean 17.29, SD 14.17, CV 82
+31 66 90 99
-36 61 81 92 98

When May data only are used:

Red-backed Salamander

Mean 13.8, SD 12.0, CV 86.8

+34 64 91 99
-33 56 82 94 98

Mountain Dusky Salamander

Mean 18.4, SD 9.6, CV 51.9
+34 79 92
-40 67 91 97 99

When Data from 9/20 to 10/30 only are used:

Red-backed Salamander

Mean 20.8, SD 11.5, CV 55.3
+38 72 93
-32 62 90 98 99

Mountain Dusky Salamander Mean 5.9, SD 5.2, CV 88
+29 66 90 97
-32 60 77 95 98

Reference 21

Red-backed Salamander

Preliminary Results of *Plethodon cinereus* Survey
in Valley Forge National Historical Park, Pennsylvania
by

Harry M. Tiebout III

Kristina Heister

Fredric C. Lucher

10 June 1997

NAME AND LOCATION OF SITE

The study is being conducted at Valley Forge National Historical Park, which is 20 km northwest of Philadelphia, Pennsylvania.

HABITAT

All sampling is being conducted in mature mixed hardwood forest dominated by northern red oak, sassafras, yellow poplar, red maple, and chestnut oak.

TECHNIQUE

Five transects were laid out on Mount Misery between 73 and 134 meters elevation. Each transect has 10 sampling stations evenly spaced, with 4 coverboards per station, for a total of 200 coverboards. The boards at each station consist of four different designs: large (4' x 2') and small (42" x 7") uncleated (boards laid flat on the substrate) and cleated (boards were supported 1.5" above the substrate by wooden cleats attached at one end).

TIME OF YEAR DATA WERE COLLECTED AND NUMBER OF SAMPLES

We flipped boards twice per week during periods of salamander activity between July 3, 1996 and May 26, 1997, for a total of 50 censuses. We are continuing to collect data as funding allows. To date, abundance has peaked in mid-October and in mid-to-late-March.

Mean 67.29, SD 22.52, CV 34

36 89

43 80 94