The Science Approach to the Smokies ATBI

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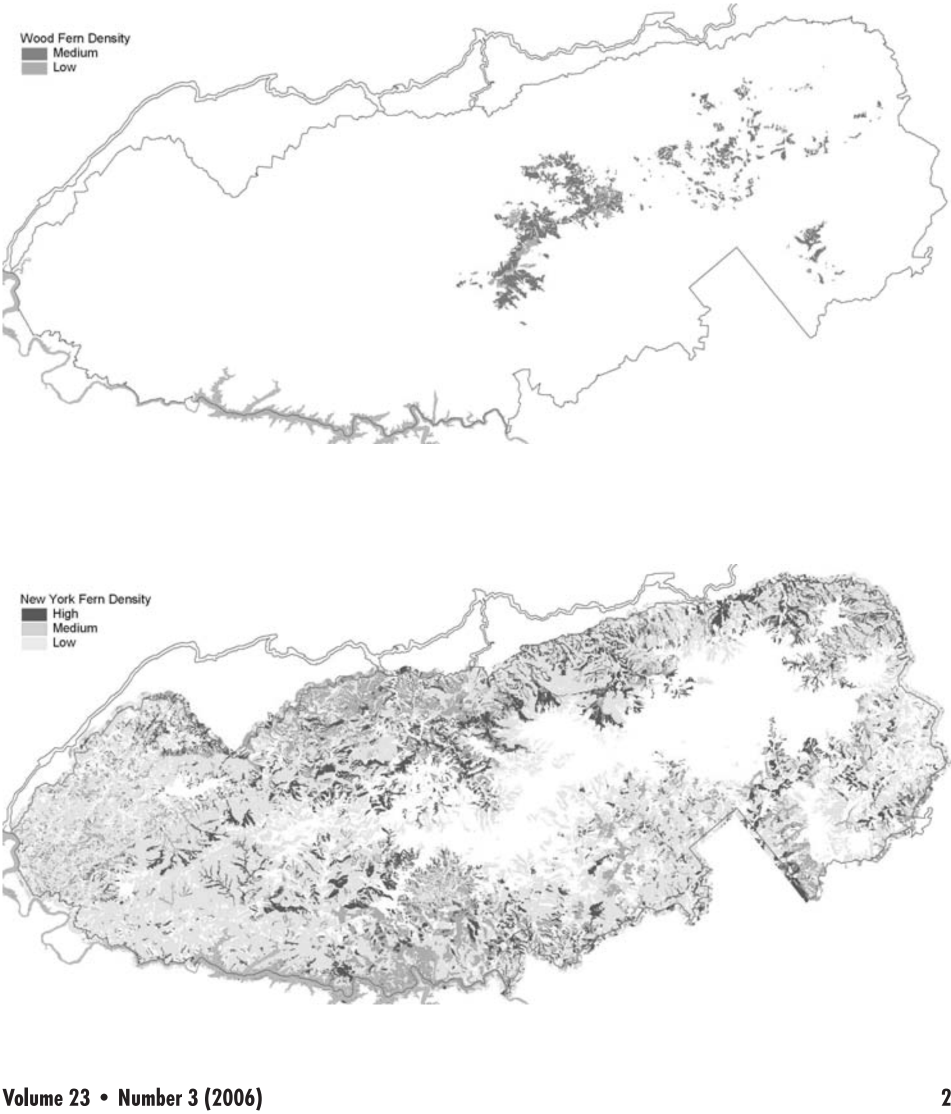
When the Smokies ATBI officially began on Earth Day, 1998, procedures for conducting a comprehensive inventory of life in a diverse natural landscape were not available. A "generic protocol" is contained in the report on a workshop held in 1993 to consider conducting an ATBI in Costa Rica (Janzen and Hallwachs 1994), and methods are available for selected groups of organisms (e.g., soil organisms—Hall 1996; fungi—Rossman et al. 1998; ants—Agosti et al. 2000). The science committee of the ATBI, therefore, developed a Science Plan to guide our initial efforts, relying on the needs for information as expressed by park resource specialists and on the knowledge and experiences of the scientists interested in participating. The Smokies ATBI Science Plan calls for a traditional sampling approach to operate in parallel with a structured sampling approach (see White and Langdon, this volume), and relies on taxonomic authorities organized into Taxonomic Working Groups (TWIGs) for the critical tasks of identifying specimens, describing species, developing species lists, and training students. Here we describe the traditional and structured sampling approaches, giving examples of the results from each approach, and how the TWIGs function to meet the goals of the ATBI.

The traditional sampling approach

The traditional approach is defined as the types of activities employed by taxonomic authorities to collect the species of their expertise, normally accomplished by these authorities visiting habitats favored by the organisms under investigation, and using collecting techniques most likely to result in specimens. These techniques may involve, for example, turning over rocks and inspecting them individually for minute pauropod specimens, sweeping with an insect net in vegetation for planthoppers, or spraying a mixture of cola and honey on bushes where certain kinds of parasitic flies aggregate. Other approaches include using a light to attract flying insects after dark, collecting leaf litter for processing in a Tüllgren funnel to sample arthropods, and examining individual flowers and mushrooms for thrips. Because of the specialized nature of many of these approaches, they are best accomplished by experienced authorities, usually working on their own or with a trained technician. A variation on the traditional approach is a foray, or blitz, in which groups of experts and dedicated volunteers conduct intense, short-term efforts focused on particular taxonomic groups or habitat types. Since the Smokies ATBI began, we have held 26 forays, 23 of them focused on taxonomic groups such as moths and butterflies, beetles, snails, flies, ants, slime molds, bats, and millipedes. The other three have focused on particular ecosystems: a leaf-litter quest, a high-country quest, and a karst quest. Finally, we have had highly successful fern forays for several years running, in which groups of scientists and volunteers hike designated trails in the park and map the occurrences of fern species following a specific protocol. To date, fern forays have covered more than 250 miles of trails, and the results have been used to develop GIS models of probability distribution maps of fern species throughout the park (Figure 1).

Traditional approaches are excellent for rapidly developing lists of species, and for finding unusual species that are restrict-

Figure 1. Probability distributions of two species of ferns in Great Smoky Mountains National Park. Probabilities were determined from the results of fern forays along 250 miles of park trails.



ed to unique habitats likely to be overlooked by collectors with less experience. Traditional methods of sampling can result in the collection of any type of organism, and in some cases do not actually require the collection of specimens. For example, observations by qualified ornithologists listening for bird songs can suffice as a reliable record of a species occurrence at a specified location at a particular point in time, without the need for a specimen to be collected. The U.S. Geological Survey conducted more than 4,000 such observation sessions of breeding birds in the park over a period of three years, resulting in nearly 75,000 observations of 115 species (Susan Shriner and Ted Simons, personal communication).

Of the more than 600 species new to science and the more than 4,400 new park records discovered since the beginning of the ATBI (see Langdon et al., this volume), more than two-thirds of each category resulted from traditional sampling. However, traditional approaches are less successful at providing the type of data needed to evaluate the completeness of an inventory for a group of taxa, and for quantifying relationships among taxa and community types. These types of data are more accessible using the structured approach.

The structured sampling approach

The structured approach is based on biodiversity reference areas and uses various types of standard traps that operate for long periods of time in every "ecological zip code" in the park (see White and Langdon, this volume). Structured sampling is a quantifiable approach that allows us to develop estimates of species–effort relationships for multiple taxa per habitat type simultaneously, and to discover biotic relationships at a scale that ultimately will permit modeling of the occurrences of numerous species across the park landscape. Of course, structured sampling is not appropriate for organisms that cannot be captured in a trap. Even for those organisms that can be trapped by some device, sampling is biased by the types of traps used. Malaise traps (Figure 2), a type of trap favored by entomologists, predominantly sample insects flying within 1–2 m of the ground, and more specifically, those insects that fly upwards when they encounter an obstacle. Malaise traps are less successful at sampling insects that drop to the ground

Figure 2. A Malaise trap in an acid bog on Andrews Bald, with an electric fence to keep bears and other wildlife away.

and fly away in the opposite direction when they encounter an obstacle. Some groups of flying insects are rarely captured in Malaise traps under any circumstances. Pitfall traps are designed to sample the leaf litter community of the forest floor, but collections are biased by the activity levels of the individuals in the community; for instance, springtails are more active than slugs. The type of preservative used in the collection cup, as well as seasonality, temperature, and moisture, are other qualifying considerations. Thus, several methods must be used simultaneously in order to sample different segments of the communities present. In order to overcome seasonality-, temperature-, and moisture-related variations, sampling should be extended over multiple seasons, preferably over several years. No consensus exists on how best to sample multiple communities that exist at one location. Therefore, a pilot study was designed to address this all-important question.

Pilot study design

The pilot study was designed to test techniques for adoption in the full-scale structured sampling program. Funding was obtained from the U.S. Geological Survey for a three-year study with the objectives of (1) determining how to efficiently sample and process many thousands of specimens using a variety of collecting methods in a variety of habitats, (2) estimating species accumulation curves and stopping rules for different taxa and methods of sampling, and (3) developing reliable approximations of the time, effort, and costs of doing the full-scale ATBI in Great Smoky Mountains National Park.

The first 19 ATBI plots were set up by the park's forest ecologist, Mike Jenkins, using the North Carolina Vegetation Survey methodology (Peet et al. 1998). We selected 11 of these plots for the pilot study, ensuring a range of habitat types from low to high elevation, including old-growth and second-growth forest, and grassy balds and heath balds (Table 1). The specifics of all 19 plots, including additional details about the 11 used in the pilot study, are found in Jenkins (in press). The initial invertebrate sampling design used in the plots included aspects of the efforts then being employed by a University of Georgia researcher to sample ichneumonoid wasps in Panama, Costa Rica, Georgia, and the Smokies. His design used paired Malaise traps in each plot. To this we added paired funnel traps (Lindgren traps, Figure 3) to sample the canopy fauna, and 10 pitfall traps to sample the litter fauna. Thus, we arrayed 14 traps on each of 11 plots (Figure 4). Sampling began in October 2000 and traps were left operating continuously until June 2003. Weather and other circumstances often prevented us from reaching every plot on an exact 2-week schedule, especially in the winter months, and occasionally, traps were damaged or destroyed by wildlife, tree-fall, or prescribed burning. Ultimately, we had 6,812 sampling events, totaling 129,380 trap-days.

Samples from each 2-week interval were sorted to TWIG level, generally consisting of an order of arthropods (i.e., flies, beetles, spiders). Selected taxa were segregated to finer levels, and ultimately sorted to the species level. These taxa were chosen because we or our cooperators have the taxonomic expertise to identify specimens of these groups to the species level. This is a relatively short list, which highlights a general problem facing not just the Smokies ATBI, but all similar comprehensive inventory efforts. That problem is the "taxonom-

Plot	Elevation (m)	Vegetation Classification†	Temperature (Average °C)
Albright Grove	1,033	Southern Appalachian Acid Cove Forest (Silverbell Type)	9.4
Andrews Bald	1,757	Grassy Bald (Southern Grass Type)	7.5
Brushy Mountain	1,468	Southern Appalachian Mountain Laurel Bald	7.4
Cades Cove	522	Pasture	11.6
Cataloochee	1,385	High Elevation Red Oak Forest (Deciduous Shrub Type)	8.3
Clingmans Dome	1,956	Fir Forest (Deciduous Shrub Type)	4.1
Goshen Prong	917	Southern Appalachian Cove Forest (Rich Montane Type)	7.5
Indian Gap	1,634	Southern Appalachian Beech Gap	5.9
Purchase Knob	1,324	Southern Appalachian Cove Forest (Typic Montane Type)	8.4
Snake Den Ridge	932	Southern Appalachian Acid Cove Forest (Silverbell Type)	10.4
Twin Creeks	594	Southern Appalachian Cove Forest (Typic Montane Type)	11.6

†From the NatureServe vegetation classification of Great Smoky Mountains National Park (White et al. 2003)

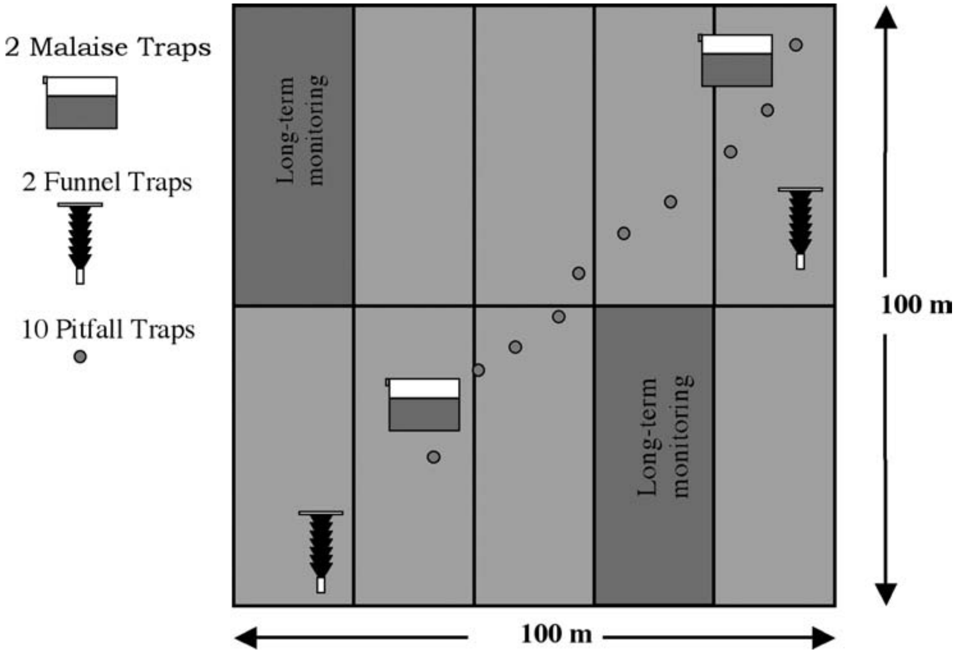
Table 1. ATBI plots used in the pilot study.

ic impediment," a critical shortage of taxonomic authorities available and willing to identify samples from such undertakings. This will be discussed further below.

Pilot study data: crane flies. Crane flies are collected in Malaise traps in large numbers, and an extensive list of species known from Great Smoky Mountains National Park was published decades ago (Alexander 1940, 1941). Thus, this group was selected for study by Matthew Petersen, then a graduate student at the University of Tennessee. The results were astounding; 176 species in 52 genera and 6 families were identified among over 9,000 specimens, bringing the total number of crane flies known from the park to 250 species (Petersen et al. 2005). Seventy species were recorded for the first time, including two species new to science (Petersen et al. 2004). The data also were analyzed for seasonal occurrence of species (Figure 5). The species shown in the plot at the top of Figure 5 occurs in the spring and early summer and at all elevations, but appears at low and mid elevations five weeks earlier than at the highest elevations. The species in the middle graph occurs in the fall and early winter at all elevations, but this species appears first at the higher elevations. The

Figure 3. A Lindgren funnel trap hung in the forest canopy adjacent to Andrews Bald.

Figure 4. The 1-ha monitoring plots used in the structured sampling pilot study, showing a typical layout of bulk sampling devices on the plot. The rectangles labeled "Long-term monitoring" represent areas intensively sampled for vegetation characteristics. See Jenkins (in press) for details on the vegetation measures recorded and the methods used.



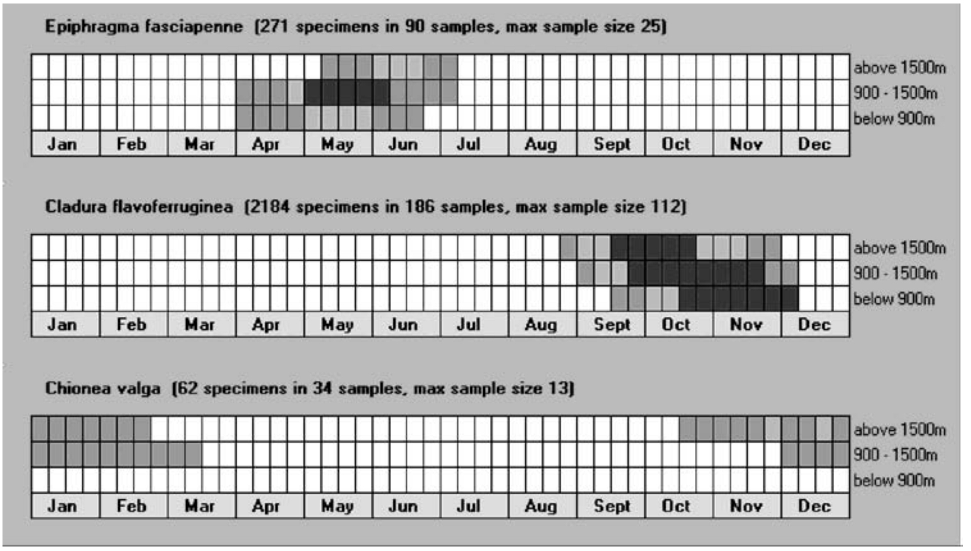


Figure 5. Seasonal occurrence of three crane fly species as revealed by the structured sampling pilot study of the Smokies ATBI. In each plot, the three rows of white boxes represent high, middle, and low elevations and approximate weekly intervals of time. The shades of gray in the boxes indicate relative numbers of specimens captured, with medium gray < light gray < dark gray.

species in the bottom plot is a winter-emerging species that does not occur at all in the lowest elevations of the park.

A major strength of data from a structured sampling program is its quantitative nature, which permits researchers to use statistical methods to investigate relationships among species and the environment. One of the most pressing questions in conducting an inventory is, "Can we stop yet?" To obtain the answer to this question we need to determine where we stand in terms of the number of species known to occur in an area versus the number of species believed to live in an area but not yet confirmed to occur there. The most reliable method for determining the answer is to develop species accumulation curves (Figure 6). These curves represent the rate at which new finds are added to the existing body of data based on some measure of the effort required to find them. At first, the curve of new discoveries is very steep as it is initially very easy to find new records with little effort (Figure 6). As efforts continue, the rate of discovery slows even if the level of effort stays the same. Eventually the curve will level to an asymptote that represents the maximum number of species that can be found. In practice, the asymptote is likely to never be reached, because resources (and patience) are limited. Therefore, statistical estimators of the limit can be used to determine what percentage of the theoretical maximum we have achieved, and how much more effort is required to achieve any desired level of completion. Using these estimators, Petersen (2002) estimated that actual richness in the 11 plots was 228 species, and that sampling had achieved 77% of the estimated total. To completely census the crane fly populations of the 11 plots used in the pilot study, without a change in the level of effort, would require an additional eight years of continuous sampling.

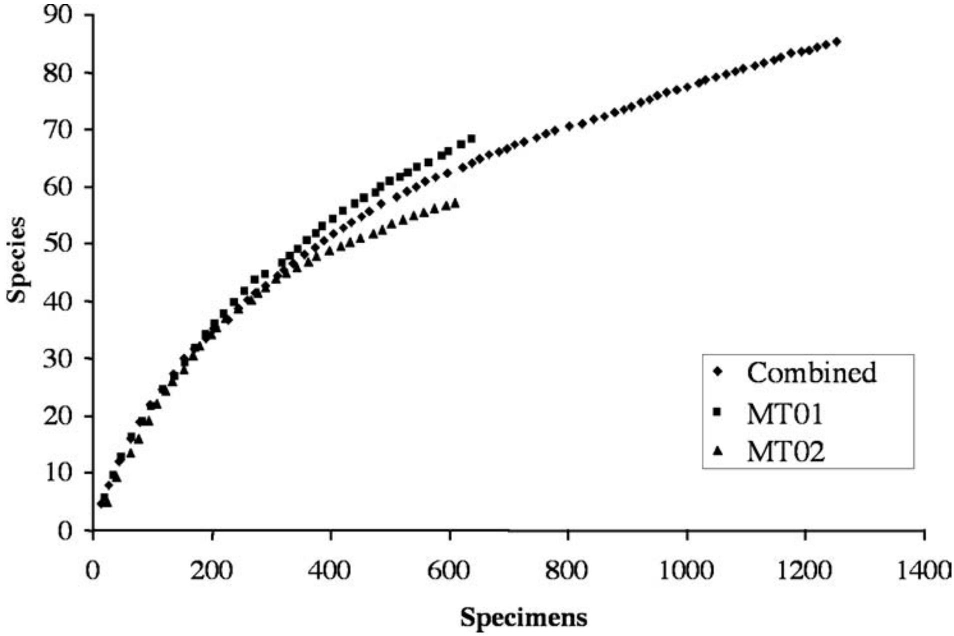


Figure 6. Species accumulation curve for crane flies from the two Malaise traps operated in the Twin Creeks structured sampling plot. The number of species found is plotted against the number of specimens examined, which is a measure of the amount of effort expended. The curves show no sign of leveling off, indicating that more sampling is required to reach an asymptote. MT01 and MT02 are the identifiers of the Malaise traps deployed on the Twin Creeks plot.

Pilot study data: Collembola. The only comprehensive list of Collembola (minute arthropods; springtails) from the Smokies prior to the ATBI contained 55 species (Wray et al. 1963). In the structured study, more than 150,000 Collembola were collected in pitfall traps and more than 20,000 in Malaise traps; together they comprise about 14,000 park records. Approximately 112,000 of these specimens (10,000 records) have been identified to the species level, and the discussion below refers to these identifications. All four orders of Collembola, 11 families, and about 120 species were collected in pitfall traps. Three orders, six families, and 21 species were collected in Malaise traps. The total number of species collected during the structured study is 127. Many of the Malaise trap taxa were never or rarely collected in pitfall traps, and would have been missed in a typical unstructured litter sampling effort. At least 25 of the taxa collected in the study are new to science, and descriptions are being published (e.g., Bernard 2006). The Collembola sampling effort with pitfall and Malaise traps appears to have been efficient at collecting most of the active or climbing species that can be obtained by these methods, since species-accumulation curves are near asymptote for most of the 11 sites. These kinds of traps are poor for collecting the many less-active species of springtails, which are better obtained with Tüllgren funnels.

Taxonomists and structured sampling

The linchpin of taxonomic inventories

is the taxonomic authority who identifies specimens, describes new species, and develops tools for non-experts to identify and understand the diversity of life. Unfortunately, there is a severe shortage of taxonomists, especially those who work on the most diverse groups of organisms, such as arthropods, fungi, and bacteria. This shortage has been termed the "taxonomic impediment" (Taylor 1983), and has serious consequences for biodiversity studies and conservation (Mikkelsen and Cracraft 2001; Hopkins and Freckleton 2002; O'Connell and Yallop 2002; Giangrande 2003; Terlizzi et al. 2003). The numbers of specialists who have the time and inclination to identify specimens for ambitious projects such as all taxa inventories seem to be in steady decline, with fewer young scientists going into taxonomy to replace those who retire or die. Those who do work in taxonomy often are so busy that they have little time to devote to identifications of large mixed samples of organisms to find the few gems of interesting specimens that may represent rare, unusual, or undescribed species. In the Smokies ATBI for example, we have been unable to find taxonomists with expertise in Hymenoptera willing to identify material from the park, with the notable exceptions of ants, mutillid wasps, sawflies, and bees. The majority of parasitic wasps, which number in the thousands of species, thus are being stored on shelves in the hope that some day authorities will be found to identify the samples. This limitation is true for other groups as well. Even when authorities are willing to identify Smokies material, they often have only limited time to devote to it, which results in a further difficulty. The bulk sampling methods used in structured sampling were operated continuously for several days or weeks, which resulted in enormous numbers of specimens of common species, as well as small numbers of rare or otherwise interesting species. In a sense, the original design of the pilot study was too successful for its own good. Since the end of the pilot study, the park has developed funding that has allowed us to procure the services of specialists in various groups to begin processing this backlog. Thus, some of the hyper-diverse groups, such as Diptera and Lepidoptera, are finally receiving attention where previously they had not.

A modified approach to structured sampling

Because of the problems mentioned above, and because of the difficulty of operating the plots on a continuous basis, we have modified our approach to the structured sampling program. The park has provided funds for a test of the revised protocols that is currently underway (Becky Nichols, personal communication). In the revised protocol, the structured sampling plots consist of points established in "ecological zip codes" by a GIS algorithm (see White and Langdon, this volume). At each point, a 6-m Malaise trap and a canopy trap are deployed. No pitfall traps are used. These Malaise traps are three times the size of the ones used in the original pilot study, and the canopy traps are larger than the Lindgren funnel traps. However, the traps are operated for just 48 hours every two weeks, rather than continuously. The shorter time frame allows us to collect the specimens dry, resulting in higher-quality specimens that we can pin, making the specimens more attractive to cooperating specialists. In addition, since the traps are operated for shorter time periods, they will not trap as many specimens, thus reducing the "fatigue of the commons" that the original samples produced. By using the larger traps for shorter periods of time, we hope that we will improve the quality of specimens, reduce the number of individuals of common species, and still maintain a high rate of new species recovery. In order to sample the litter fauna that the pitfall traps collected in the original pilot study, we will take litter samples periodically and process them in Tüllgren funnels. We anticipate that this approach will reduce the biases discussed above (more active species predominating) that pitfall traps are known to present.

Conclusions

The parallel operation of traditional and structured sampling approaches is highly productive. We believe it represents the most comprehensive and feasible way in which to inventory the biodiversity of complex terrestrial natural areas. The design of biodiversity reference areas and structured sampling plots can change from natural area to natural area, depending on the ecosystems represented. However, the inclusion of georeferenced plots at which specific protocols are followed strengthens the scientific credibility of the inventory program, and, for the Smokies, ensures that we will be able to achieve the management-driven goals of the effort.

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