TOS SCIENCE DESIGN FOR GROUND BEETLE ABUNDANCE AND DIVERSITY

PREPARED BY	ORGANIZATION	DATE
David Hoekman	FSU	12/05/2013

APPROVALS	ORGANIZATION	APPROVAL DATE

RELEASED BY	ORGANIZATION	RELEASE DATE

See configuration management system for approval history.

The National Ecological Observatory Network is a project solely funded by the National Science Foundation and managed under cooperative agreement by NEON, Inc. Any opinions, findings, and conclusions or recommendations expressed in this material are those of the author(s) and do not necessarily reflect the views of the National Science Foundation.

Change Record

REVISION	DATE	ECO #	DESCRIPTION OF CHANGE
A	10/10/2014	ECO-02008	Initial release

TABLE OF CONTENTS

LIST OF TABLES AND FIGURES

1 DESCRIPTION

1.1 Purpose

NEON design documents are required to define the scientific strategy leading to high-level protocols for NEON subsystem components, linking NEON Grand Challenges and science questions to specific measurements. Many NEON *in situ* measurements can be made in specific ways to enable continental-scale science rather than in ways that limit their use to more local or ecosystem-specific questions. NEON strives to make measurements in ways that enable continental-scale science to address the Grand Challenges. Design Documents flow from questions and goals defined in the NEON Science Strategy document, and inform the more detailed procedures described in Level 0 (L0; raw data) protocol and procedure documents, algorithm specifications, and Calibration/Validation (CalVal) and maintenance plans.

1.2 Scope

This document defines the rationale and requirements for ground beetle abundance and diversity sampling in the NEON Science Design.

1.3 Acknowledgments

The design was reviewed by and refined with input from a technical working group consisting of researchers with relevant expertise. The ground beetle technical working group includes George Ball, Robert A. Browne, Robert Davidson, Terry Erwin, James LaBonte, Karen Ober, Barry Knisley, Jonathan Lundgren, David Maddison, Wendy Moore, Jari Niemelä, David Pearson, John Spence, Kip Will and Timothy Work.

2 RELATED DOCUMENTS AND ACRONYMS

2.1 Applicable Documents

Applicable documents contain information that shall be applied in the current document. Examples are higher level requirements documents, standards, rules and regulations.

AD[01]	NEON.DOC.000001	NEON Observatory Design
AD[02]	NEON.DOC.001282	Introduction to the TOS Science Designs
AD[03]	NEON.DOC.000913	TOS Science Design for Spatial Sampling Design
AD[04]	NEON.DOC.004312	NEON Research Coordination Guidelines
AD[05]	NEON.DOC.014015	Fundamental Sentinel Unit Bioarchive Facility Design

2.2 Reference Documents

Reference documents contain information complementing, explaining, detailing, or otherwise supporting the information included in the current document.

RD[01]	NEON.DOC.000008	NEON Acronym List
RD[02]	NEON.DOC.000243	NEON Glossary of Terms
RD[03]	NEON.DOC.005003	NEON Scientific Data Products Catalog
RD[04]	NEON.DOC.014050	TOS Protocol and Procedure: Ground Beetle Sampling
RD[05]	NEON.DOC.001100	Lab Protocol for Beetles and Mosquitoes

2.3 External References

External references contain information pertinent to this document, but are not NEON configuration-controlled. Examples include manuals, brochures, technical notes, and external websites.

ER [01]	
ER [02]	
ER [03]	

2.4 Acronyms

All acronyms used in this document are defined in RD[01].

3 INTRODUCTION

3.1 Overview of the Observatory

The National Ecological Observatory Network (NEON) is a continental-scale ecological observation platform for understanding and forecasting the impacts of climate change, land use change, and invasive species on ecology. NEON is designed to enable users, including scientists, planners and policy makers, educators, and the general public, to address the major areas in environmental sciences, known as the Grand Challenges (Fig. 1). NEON infrastructure and data products are strategically aimed at those aspects of the Grand Challenges for which a coordinated national program of standardized observations and experiments is particularly effective. The open access approach to the Observatory's data and information products will enable users to explore NEON data in order to map, understand, and predict the effects of humans on the earth and understand and effectively address critical ecological questions and issues. Detailed information on the NEON design can be found in AD[01] and AD[02].

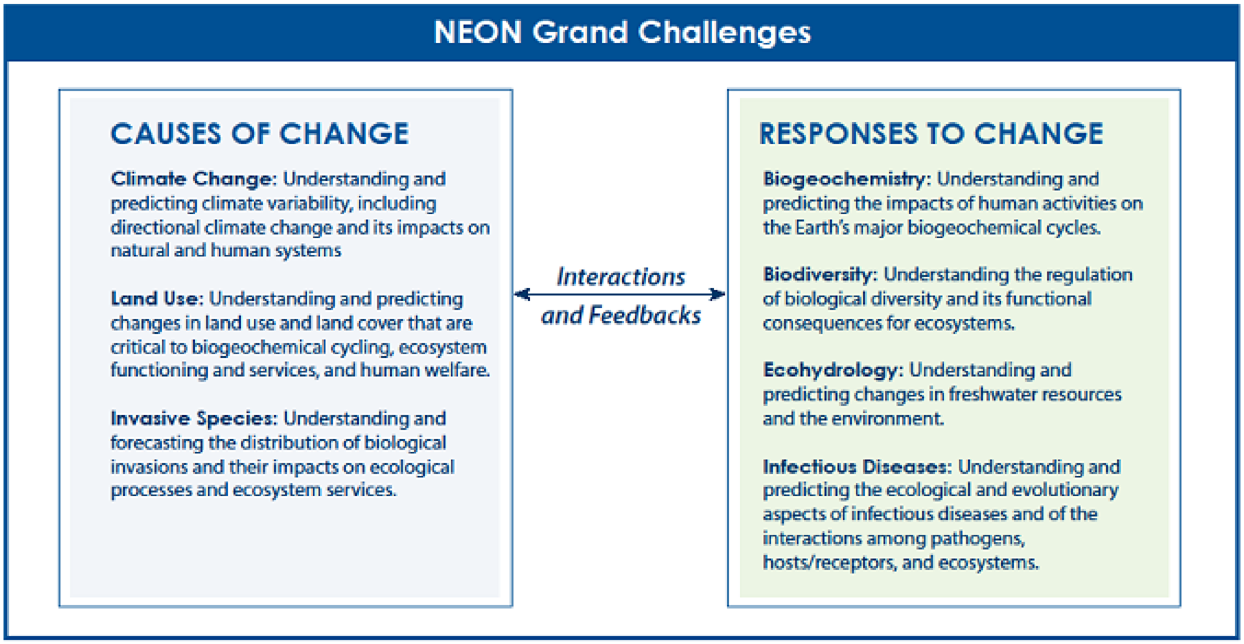

Figure 1. The seven Grand Challenges defined by the National Research Council (Committee on Grand Challenges in Environmental Sciences 2001)

3.2 Components of the Observatory

There are five components of the Observatory: the Airborne Observation Platform (AOP), Terrestrial Instrument System (TIS), Aquatic Observation System (AOS), Aquatic Instrument System (AIS), and Terrestrial Observation System (TOS). Collocation of measurements associated with each of these components will allow for linkage and comparison of data products. For example, remote sensing data provided by the Airborne Observation Platform (AOP) will link diversity and productivity data collected on individual plants and stands by the Terrestrial Observation System (TOS) and flux data captured by instruments on the tower (TIS) to that of satellite-based remote sensing. For additional information on these systems, see Keller et al. (2008) and Schimel et al. (2011).

3.3 The Terrestrial Observation System (TOS)

The NEON TOS will quantify the impacts of climate change, land use, and biological invasions on terrestrial populations and processes by sampling key groups of organisms (sentinel taxa), infectious disease, soil, and nutrient fluxes across system interfaces (air, land, and water) (AD[01], AD[02]). The sentinel taxa were selected to include organisms with varying life spans and generation times, and wide geographic distributions to allow for standardized comparisons across the continent. Many of the biological measurements will enable inference at regional and continental scales using statistical or process-based modeling approaches. The TOS sampling design captures heterogeneity representative of each site to facilitate this inference when possible. Plot and organism-scale measurements will also be coordinated with the larger-scale airborne measurements, which provide a set of synergistic biological data products at the regional scale. Details of these design elements and algorithms can be found in individual design documents available through the NEON website (www.NEONinc.org).

The standardization of protocols across all sites is key to the success of NEON (and its novelty) and must be maintained at all sites through time. Thus, although specific techniques may be required at some sites (e.g., due to different vegetation types), protocols have been developed to ensure data comparability. These details can also be found in individual design documents available through the NEON website (www.NEONinc.org).

The TOS Science Designs define the scientific strategies leading to high-level sampling designs for NEON sentinel taxa, terrestrial biogeochemistry, and infectious disease, linking NEON Grand Challenges and science questions to specific measurements (AD[02]). The TOS Spatial Sampling Design document describes the sampling design that collocates observations of the components of the TOS (AD[03]). TOS Science Design documents were developed following input from the scientific community, including module-specific Technical Working Groups, and the National Science Foundation (AD[02]). Science Designs will be reviewed periodically to ensure that the data collected by NEON are those best suited to meet the requirements of the observatory (AD[01]), are (to the extent possible) consistent with standards used by the scientific community, and fit within the scope of NEON. Additional information on the development and review process can be found in AD[02].

4 INTRODUCTION TO THE GROUND BEETLE ABUNDANCE AND DIVERSITY SAMPLING DESIGN

4.1 Background

4.1.1 Ground Beetles as a Sentinel Taxon

The Terrestrial Observation System (TOS) at NEON is charged with monitoring the responses of biodiversity and ecosystems to environmental change. While several different invertebrate groups were considered, a NEON design committee (AIBSnews 2007) selected ground beetles (Coleoptera: Carabidae) as a focal taxon for measurement.

Ground beetles have been extensively used as indicator species of arthropod biodiversity, environmental change (Rainio and Niemela 2003, Koivula 2011), land use (Vanbergen et al. 2005a) and land management (Work et al. 2008). The ground beetle family is large, widespread, and associated with the ground layer, rather than any particular plant community, ensuring that some ground beetles are present in virtually all terrestrial habitats over the entire extent of the observatory (Lövei and Sunderland 1996). The family is rich in species (over 40,000 species described globally, circa 3,000 species in NEON's spatial extent), abundant in individuals and well known taxonomically (Larochelle and Larivière 2003). Though ground beetles range in body size from less than a millimeter to several centimeters, they are straightforward to sample and identify (Kotze et al. 2011). For example, they have distinctive features, such as a 5-5-5 tarsal formula and a first abdominal segment completely separated by their rear legs, that facilitate their identification by non-experts (Forsythe 2000).

In addition to being widespread and well-known biologically, ground beetles are sensitive to environmental conditions and form well-defined richness gradients. Consequently, ground beetles are well suited as "sentinels" of arthropod biodiversity (Rainio and Niemela 2003, Koivula 2011). Their value to environmental science is evident in their status as model organisms for population biology, landscape ecology and conservation biology (Kotze et al. 2011). Ground beetles form an important component of terrestrial food webs and can influence terrestrial trophic structure. They are omnivorous, and most species are predators as both adults and larvae. In addition, they are common prey for small mammals, birds, reptiles, amphibians and other larger arthropods (Larochelle and Larivière 2003). Their role in food webs is recognized both in wildland and agricultural settings, where they contribute to biological control of pests (Kromp 1999). Ground beetles are prevalent in agricultural systems, where their community composition is affected by land management (Purvis and Fadl 2002, Legrand et al. 2011) and their diversity can influence weed seed predation (Gaines and Gratton 2010). In addition ground beetles have been used to assess ecological effects of urbanization (Niemelä and Kotze 2009).

4.2 NEON's Contribution

Earth's environment is changing rapidly, and data collected at broad temporal and spatial scales will increase our ability to understand, forecast and manage our changing biosphere (Keller et al. 2008). NEON's ground beetle sampling will provide a cost effective and informative measure of biodiversity, environmental and land-use change. This work will add to the considerable base of ground beetle studies and enable us to characterize their response to ecosystem drivers quantified across the observatory (Vanbergen et al. 2005b, Work et al. 2008).

The need for and value of long-term and large scale research to understand our changing environment is well recognized and best practices include high species resolution taxonomy and well-characterized variability in the local environment (Work et al. 2008). NEON has been designed to meet these research needs and expectations. NEON will collect data for 30 years, much longer than most sampling efforts and the spatial extent of NEON includes Alaska, Hawaii, the continental US, and Puerto Rico. Taxonomic resolution will be to the species level and will be temporally stabilized using DNA barcoding and expert taxonomists. In addition, the sampling environment will be thoroughly characterized by a broad suite of biotic and abiotic measurements (Schimel et al. 2011).

NEON ground beetle sampling will improve our understanding of the distribution, ecology and evolution of ground beetles in both wildland and human-dominated landscapes at a very wide scale over several decades. NEON ground beetle data will have added value through active collocation at the plot and site level with other NEON data (e.g., plant productivity and diversity, rodent density, temperature, precipitation and hundreds of other NEON data products). NEON ground beetle data will be complementary to other ongoing projects to monitor particular groups of invertebrates (e.g., The Xerces Society for Invertebrate Conservation). NEON's archived samples will be available for loan, for example for gut analyses, isotopic measurements, symbiont surveys and genomic analyses of rapid evolution in response to environmental change. In addition, bycatch from ground beetle sampling will be available for loan and represents a valuable resource for the ecological community (Buchholz et al. 2011).

4.3 Purpose and Scope

This document defines the rationale and requirements for ground beetle (Coleoptera: Carabidae) abundance and diversity in the NEON Science Design. Details about protocols including educational materials to conduct protocols, quality assurance and quality control or calibration and validation procedures are addressed in protocol specific documents.

5 SAMPLING FRAMEWORK

5.1 Science Requirements

This science design is based on Observatory science requirements that reside in NEON's Dynamic Object-Oriented Requirements System (DOORS). Copies of approved science requirements have been exported from DOORS and are available in NEON's document repository, or upon request.

5.2 Data Products

Execution of the protocols that stem from this science design procures samples and/or generates raw data satisfying NEON Observatory scientific requirements. These data and samples are used to create NEON data products, and are documented in the NEON Scientific Data Products Catalog (RD[03]).

5.3 Priorities and Challenges for Ground Beetle Abundance and Diversity Sampling

In keeping with NEON's broad mission, this design must be able to be deployed across a wide range of sites in a standardized way (e.g., methods, sampling frequencies) such that data are comparable across time and space. The design must be relatively uncomplicated so that it can be deployed consistently by disparate field crews over multiple years with minimal chance of alteration. An additional sampling priority to promote comparability and the characterization of relationships between disparate measurements is collocation with other measurements across the observatory. To this end, ground beetle sampling will occur at distributed plots within NEON sites, where other organismal and abiotic measurements are taken.

To promote utility to the scientific community, ground beetles will be sampled using pitfall traps, a standard and effective sampling method (Kromp 1999, Rainio and Niemela 2003, Kotze et al. 2011). This standardized, well established and widely used sampling method was selected to maximize comparability across time and between domains within the observatory and also to be comparable to external data sets. Pitfall trapping has been used for more than a century and is still the most commonly used method for ground beetle collection.

6 SAMPLING DESIGN FOR GROUND BEETLE ABUNDANCE AND DIVERSITY

NEON's ground beetle abundance and diversity sampling will target all members of the family Carabidae (ground beetles). Ground beetles are ubiquitous and can be found at all NEON sites.

Pitfall traps are a passive collection technique for estimating terrestrial invertebrate species richness and relative abundance and therefore provide a reliable measure of ground beetle abundance and diversity (Baars 1979). Pitfall traps collect a wide range of arthropod taxa, and are particularly effective in sampling mobile, surface-active taxa like ground beetles. Specifically, pitfall traps measure the density of activity in the area of a trap, or more simply "activity density," a measure that combines both arthropod abundance and movement.

Pitfalls will be deployed continuously during the entire growing season (length varies among sites) to encompass the activity of all ground beetle species. Pitfalls at both core and relocatable sites will be sampled (emptied and re-set) every 2 weeks. Across the observatory and within each domain, core and relocatable sites will be sampled with equal intensity. This strategy prioritizes even sampling across sites to maximize comparability of ground beetle data among all NEON sites.

Ground beetles in pitfall samples will be separated from bycatch and identified to species, thus providing an estimate of ground beetle abundance and biodiversity in each domain each year. NEON pitfall traps will collect large numbers of many common ground-dwelling arthropods in addition to ground beetles. The non-ground beetle arthropods are termed "bycatch" and although the identification of these additional taxa would be advantageous, as their inclusion would encompass additional taxonomic diversity and feeding guilds (e.g. herbivores, detritivores), their inclusion would considerably increase sampling costs in terms of processing time, analysis and storage space relative to the advantages of focusing on ground beetles. The bycatch will be archived and available from the NEON collections to other researchers for additional analyses.

6.1 Sampling Methods

6.1.1 Field Sampling

NEON's pitfall traps are cup traps, 11 cm in diameter by 7 cm deep, a medium-sized trap that has been shown to perform well in comparison to other sizes (Work et al. 2002). Propylene glycol (non-toxic antifreeze) is used as the preserving fluid.

Pitfall trapping is the most common method for sampling ground-level arthropods (Kotze et al. 2011). Alternative methods exist for capturing ground beetles, including sticky traps, malaise traps, ultraviolet light traps, flight intercept traps, sweep netting, hand picking, point counts, fogging, quadrat sampling and litter washing (Rainio and Niemela 2003). However, pitfall trapping is overwhelmingly the most used method in a wide variety of environments as it is the easiest, cheapest and most reliable method to use over the long term.

The primary bias of pitfall trapping is that it confounds abundance and activity. Activity varies among species and can be affected by environmental or physiological factors that influence activity, e.g., temperature, humidity, seasonality and food availability (Purvis and Fadl 2002). Thus, it can not necessarily be inferred from pitfall data that species A is more abundant than species B in the environment simply because it is more abundant in pitfall samples. It may be that species A is more active and therefore more likely to be trapped. Because of this activity bias, larger beetles are more likely to be trapped than smaller beetles because they cover greater distances (Spence and Niemela 1994). Despite these known biases, pitfall trapping is the standard method for characterizing ground beetle communities (Rainio and Niemela 2003). Other potential disadvantages of pitfall trapping can be neutralized using specific techniques. For example, the NEON sampling design includes an odorless preservative to avoid attracting arthropods; pitfalls covers will be installed to prevent both flooding and desiccation; and rather than solitary pitfall traps, sets of 4 well-spaced traps will be deployed to overcome occasional trap losses or other problems with individual traps.

While pitfall traps may require slight modifications for deployment in specific locations, the sampling technique is versatile and can be utilized at all sites. Site-specific considerations include substrate, temperature, humidity and seasonal water cover. Substrate is important because sand content, soil moisture and vegetation cover all influence the relative ease of digging and servicing pitfall traps. For example, different vegetation types, such as sphagnum moss, grasses, or dense woody roots may require different tools and/or techniques for pitfall deployment and ensuring durability of pitfall holes. Temperature and humidity will affect how quickly water in the trap evaporates, effectively concentrating the preservative. Finally, seasonal ponds or stream flooding may limit the specific placement of pitfall trap locations within a site.

In an effort to test the broad applicability of pitfall trapping, NEON prototyped pitfall traps at a variety of different sites from 2009-2013. While testing different trap designs and components, ground beetles were collected at 16 sites with broad geographical coverage, including sites in Alabama, Colorado, Florida, Massachusetts, Michigan, New Hampshire, North Dakota, Tennessee, Utah and Virginia.

Several specific deployment and equipment issues were resolved during prototyping, including the critical importance of burying the cup flush with the ground and the tools necessary to achieve this in different soil types. Different cover designs were tested and it was determined that plywood is susceptible to rotting and splintering, so it has been replaced with composite plastic. The height of the cover was modified to reduce vertebrate bycatch. The propensity of covers to slide down nails and close the trap was overcome by adding plastic spacers to the trap design. The tendency for nails to rust in some environments was fixed by switching to plastic stakes. A variety of cup sizes were tested and smaller cups were deemed problematic because spiders built webs across the cup opening.

Prototyping efforts culminated in an abbreviated field season of ground beetle sampling in the summer of 2012 at three sites in Domain 03 (Jones Ecological Research Center, Ordway-Swisher Biological Station and Disney Wilderness Preserve). At each site, up to 40 pitfall traps were deployed for a total of

5 weeks from the beginning of August through the first week of September (Table 1). On average between 2 and 5 ground beetles were captured per plot in a week of pitfall trapping. The abbreviated trapping season yielded a total of 412 ground beetles, representing 26 unique species (Table 1) and demonstrated the feasibility of the proposed methods to be deployed at the domain scale (Hoekman et al. 2013).

Table 1. Summary of ground beetle data from Domain 03 prototype, summer 2012

	Total number of ground beetles collected	Number of plots sampled (1 week deployment)	Average number of ground beetles collected per plot per week	Average number of ground beetles collected per site per week	Number of ground beetle species recorded
Jones	91	52	4.1	15	11
Ordway	46	22	1.9	8	5
Disney	275	24	5.3	46	17
Domain 3 total	**412**	**98**			**26**

NEON plan: Each sampling bout will consist of 14 days of continuous pitfall trapping. Every two weeks, specimens trapped in pitfall cups will be collected and processed in the domain lab. Specifications for beetle trap construction and deployment are described in the TOS Protocol and Procedure: Ground Beetle Sampling (RD[04]).

6.1.2 Vertebrate Bycatch

Animals collected in pitfall traps other than ground beetles are termed bycatch. Bycatch is primarily composed of spiders and insects. Depending on their size, pitfall traps can be used to capture a wide range of ground-dwelling animals; e.g., larger pitfalls are used to survey amphibians, reptiles and small mammals (Bury and Corn 1987, Hobbs and James 1999). Small traps designed to capture invertebrates do occasionally capture small vertebrates, including frogs, salamanders, and shrews.

NEON will take various steps to reduce vertebrate bycatch in our pitfall traps. First, the pitfalls are relatively small (11 cm diameter) and shallow (7 cm depth), thus only vertebrates about the size of large ground beetles are vulnerable to being captured. Recent studies suggest that shallower (7 cm) pitfalls are less likely to capture vertebrate bycatch than deeper (15 cm) pitfalls (Pendola and New 2007 and references therein). Larger vertebrates could easily exit the cup if they happened to fall in (but see cover below). The preservation fluid in the pitfalls (propylene glycol) is colorless and unscented, so the traps are not baited and do not attract ground beetles or bycatch. Each pitfall trap will be covered with a 20x20 cm hard plate, 1.5 cm above the lip of the trap. The cover will deny access to all but the smallest vertebrates.

6.1.3 Sample Processing in the Lab

Specimens collected in pitfall traps will be sieved out of the propylene glycol and transferred into 95% ethanol. After 24 hours, the ethanol will be replaced to ensure low water content in the ethanol used for storage of the samples. Pitfall samples may remain in 95% ethanol for several months after collection until the end of the field season, when the technicians have time to process them.

The effects of time in propylene glycol and storage temperature on DNA degradation were tested in a laboratory experiment in the fall of 2012. Ladybird beetles were preserved in propylene glycol at 5, 20 and 35 degrees C. Twenty beetles were collected and processed for DNA barcoding from each temperature treatment after 1, 2, and 3 weeks. We found propylene glycol to be an excellent DNA preservative for up to three weeks. Barcoding success rates were high (>95%) and statistically indistinguishable for all temperature treatments and did not differ between weekly collections.

During the field season or subsequent off season, pitfall samples will be sorted, separating ground beetles from invertebrate bycatch. Ground beetles will be geo-referenced, pinned and labeled, and sorted to morphospecies, morphologically distinct types, by technicians at the domain lab. The number of pinned individuals of the most common species, defined and updated in site-specific protocols, will be limited to 20 per site. The technicians will use a voucher collection, specific to each domain, to identify ground beetle morphospecies to species. Before operations begin at each site the voucher collection will be assembled and this collection will be supplemented over time with additional collections.

From each domain each year, some ground beetles will be sent to external facilities for taxonomic verification. This subset will be selected in order to represent some individuals from every morphospecies; individuals that are rare or difficult to identify will be prioritized. Some beetles will be sent to expert taxonomists for identification and some ground beetles will be photographed and submitted for DNA barcoding, which has been shown to be effective for distinguishing ground beetle species (Raupach et al. 2010). These subsets may overlap and exact numbers will depend on the abundance and diversity of collected ground beetles and annual funding.

6.1.4 DNA Barcoding

Identifying specimens using DNA barcoding requires the building of a reference barcode library. The specimens for the NEON reference library are collected during field prototype campaigns or obtained from museum archives (Gibson et al. 2012). All the assembled resources for each specimen - sequence data, photos, and other ecological information - can be publicly accessed online from the Barcode of Life Datasystem (BOLD, http://www.boldsystems.org/index.php/databases). Before sites enter the operations phase, ground beetles from each site will be collected during site characterization efforts to build the domain-specific voucher collection and DNA barcode library. In every subsequent sampling year, a subset of ground beetles will be selected for barcoding. Beetles that are rare, particularly difficult to identify or poorly represented in the archive will be prioritized for DNA barcoding. Specimens will be selected to ensure species coverage at the domain level first, then the site level. DNA barcoding will be used to confirm species identifications and maintain continuity of identifications through time. The exact number submitted for barcoding will vary among sites (approximately 95 per site per year). Both the voucher collection and the barcode library are useful tools for identifying ground beetles and will become more complete as specimens are added over time.

6.1.5 Archiving

All specimens (including unsorted "bycatch"), and any extracted DNA will be archived in existing collections. Archiving plans, including guidance on how specimens can be requested for loan are detailed in the Fundamental Sentinel Unit Bioarchive Facility Design AD[05].

6.2 Spatial Distribution of Sampling

Each domain includes three sites, a single "core" site in a wildland location where sampling will occur for the entire 30-year lifespan of the observatory, and two "relocatable" sites that may periodically be reassigned (i.e., moved within the domain, estimated 7-10 years per location). At each NEON site, 10 distributed plots will be selected for pitfall sampling with 4 pitfall traps deployed per plot (40 traps per site). Plots will be distributed across up to three dominant vegetation types to best represent the different habitats present at each site while maintaining sufficient replication within each vegetation type. The number of plots per vegetation type will be proportional to the percent cover of that type at the site. This stratified approach will benefit the ground beetle sampling because vegetation cover has been shown to be an important predictor of ground beetle community composition (Dufrene and Legendre 1997, Work et al. 2008). Therefore the important ground beetle species present at a site are more likely to be encountered and recorded if sampling effort is spread across different plant communities.

At each distributed plot a pitfall trap will be placed in each cardinal direction 20 meters from the center of the plot. This level of replication will provide a sufficiently large sample to characterize the ground beetle community, including rare species, and is a greater or comparable sampling effort to other large-scale pitfall trapping schemes (Dufrene and Legendre 1997, Vanbergen et al. 2005b, Work et al. 2008, Brooks et al. 2012). Collectively, the 4 pitfall traps will represent the ground beetle community at the plot level, but traps will be far enough apart (30-40m) to represent independent samples of the beetle community at a site level (Digweed et al. 1995) to maximize site-level replication (N=40).

6.3 Temporal Distribution of Sampling

Sampling will take place every 14 days throughout the growing season, when ground beetles are active and likely to be caught in pitfall traps. Every two weeks, specimens collected in pitfall cups will be sieved out of the propylene glycol and transferred into 95% ethanol. After 24 hours, the ethanol will be replaced to ensure low water content in the ethanol used for storage of the sample. Pitfall samples may remain in 95% ethanol for several months after collection until the end of the field season, when the technicians have time to process them. During the field season or subsequent off season, pitfall samples will be sorted, separating ground beetles and bycatch.

Ground beetles display seasonal abundance and diversity patterns. The exact sampling dates will vary among domains based on the length of the growing season. The growing season may vary between years and change over longer time scales. A temperature threshold will be used to determine the start and end of the field season for NEON sampling. Pitfall trapping at a site will begin when the 10-day running average low temperature is >4°C and end when it is <4°C. NEON staff scientists may align this threshold with sampling of other NEON modules based on other biologically relevant thresholds (e.g., degree days, vegetation index, phenological observation) and the season may be truncated when necessary based on logistical and budgetary constraints.

6.4 Logistics and Adaptability

6.4.1 Potential for Changing Physical Locations of Sampling Plots

Spatial patterns associated with ground beetle-related phenomena of interest are likely to change over the course of NEON's lifespan. These changes could include local patterns of distribution, abundance and diversity. A fully fixed plot design in which plot locations within sites do not change has a number of logistic and statistical conveniences but does not allow for spatial flexibility to accommodate/track such changes. One way to capture this anticipated variability is to sample at different distributed plots in different years.

After three years of data collection, we will evaluate the effectiveness of our sampling design in meeting the requirements for ground beetle sampling. At this time, we will consider a sampling with a partial replacement design. Following this design, the locations of a sub-set of ground beetle sampling plots would be fixed while the location of another subset of plots would be moved to other distributed plots. The number of plots to remain fixed vs. moved would be based on the covariance in ground beetle abundance and diversity between years (Skalski 1990). Higher inter-annual variability would result in a higher proportion of fixed plots. In contrast, lower inter-annual variability would result in a higher proportion of re-assigned plots; in effect, maximizing the amount of information at the site scale by redistributing sampling effort. Fixed plots serve to monitor trends while moved plots gather data from new locations within the area being sampled. Decisions about if and when to change the sampling design will be made in consultation with an expert review committee, the Assistant Director of the Fundamental Sentinel Unit, and the Observatory Director.

6.4.2 Supplemental Trapping Methods

While pitfall traps are the most efficient, versatile and widespread method for collecting ground beetles, their effectiveness at sampling the entire ground beetle community varies between different habitat types (e.g., pitfalls are particularly effective in grassland and boreal forest sites). For this reason, it may be prudent to supplement pitfall trapping with other collection methods at a sub-set of NEON sites. NEON encourages researchers to conduct supplemental sampling of ground beetles utilizing alternative methods; details on working with NEON can be found in RD[05].

7 REFERENCES

AIBSnews. 2007. NEON design 2007. BioScience 57:198–200.

Baars, M. A. 1979. Catches in pitfall traps in relation to mean densities of carabids. Oecologia 46:25–46.

Brooks, D. R., J. E. Bater, S. J. Clark, D. T. Monteith, S. J. Corbett, D. A. Beaumont, and J. W. Chapman. 2012. Large carabid beetle declines in a United Kingdom monitoring network increases evidence for a widespread loss in insect biodiversity. Journal of Applied Ecology. doi: 10.1111/j.1365-2664.2012.02194.x.

Buchholz, S., M. Kreuels, A. Kronshage, H. Terlutter, and O.-D. Finch. 2011. Bycatches of ecological field studies: bothersome or valuable? Methods in Ecology and Evolution 2:99–102. doi: 10.1111/j.2041-210X.2010.00051.x.

Bury, R. B., and P. S. Corn. 1987. Evaluation of Pitfall Trapping in Northwestern Forests: Trap Arrays with Drift Fences. Journal of Wildlife Management 51:112–119.

Committee on Grand Challenges in Environmental Sciences, O. C. for the C. on G. C. in E. S. 2001. Grand Challenges in Environmental Sciences. Page 106. The National Academies Press, Washington, D.C. Retrieved September 21, 2012, from http://www.nap.edu/catalog.php?record_id=9975.

Digweed, S. C., C. R. Currie, H. A. Carcamo, and J. R. Spence. 1995. Digging out the digging-in effect'' of pitfall traps: Influences depletion and disturbance on catches of ground beetles (Coleoptera: Carabidae). Pedobiologia 39:561–576. Gustav Fischer Verlag Jena. Retrieved from http://www.mendeley.com/research/digging-diggingin-effect-pitfall-traps-influences-depletion-disturbance-catches-ground-beetles-coleoptera-carabidae/.

Dufrene, M., and P. Legendre. 1997. Species Assemblages and Indicator Species: The Need for a Flexible Asymmetrical Approach. Ecological Monographs 67:345–366. Eco Soc America. doi: 10.2307/2963459.

Forsythe, T. G. 2000. Ground Beetles. Page 96 (S. A. Corbet and R. H. L. Disney, Eds.)2nd editio. The Richmond Publishing Co. Ltd, Slough.

Gaines, H. R., and C. Gratton. 2010. Seed predation increases with ground beetle diversity in a Wisconsin (USA) potato agroecosystem. Agriculture, Ecosystems & Environment 137:329–336. Elsevier B.V. doi: 10.1016/j.agee.2010.03.003.

Gibson, C. M., R. H. Kao, K. K. Blevins, and P. D. Travers. 2012. Integrative taxonomy for continental-scale terrestrial insect observations. PLoS ONE 7:e37528. doi: 10.1371/journal.pone.0037528.

Hobbs, T. J., and C. D. James. 1999. Influence of shade covers on pitfall trap temperatures and capture success of reptiles and small mammals in arid Australia. Wildlife Research 26:341–349.

Hoekman, D., Y. Springer, and K. K. Blevins. 2013. Abundance and diversity of ground beetles and mosquitoes at three southeastern US sites: NEON data in action. Poster presentation: the 61st Annual Meeting of the Entomological Society of America. Austin, TX.

Keller, M., D. S. Schimel, W. W. Hargrove, and F. M. Hoffman. 2008. A continental strategy for the National Ecological Observatory Network. Frontiers in Ecology and the Environment 6:282–284. Eco Soc America. doi: 10.1890/1540-9295(2008)6[282:ACSFTN]2.0.CO;2.

Koivula, M. J. 2011. Useful model organisms, indicators, or both? Ground beetles (Coleoptera, Carabidae) reflecting environmental conditions. ZooKeys:287–317.

Kotze, D. J., P. Brandmayr, A. Casale, E. Dauffy-Richard, W. Dekoninck, M. J. Koivula, G. L. Lövei, D. Mossakowski, J. Noordijk, W. Paarmann, R. Pizzolotto, P. Saska, A. Schwerk, J. Serrano, J. Szyszko, A. Taboada, H. Turin, S. Venn, R. Vermeulen, and T. Zetto. 2011. Forty years of carabid beetle research in Europe - from taxonomy, biology, ecology and population studies to bioindication, habitat assessment and conservation. ZooKeys:55–148.

Kromp, B. 1999. Carabid beetles in sustainable agriculture : a review on pest control efficacy , cultivation impacts and enhancement. Agriculture Ecosystems & Environment 74:187–228.

Larochelle, A., and M. C. Larivière. 2003. A Natural History of the Ground-Beetles (Coleoptera : Carabidae) of America North of Mexico. Page 583 Faunistica No 27. Pensoft.

Legrand, A., C. Gaucherel, J. Baudry, and J.-M. Meynard. 2011. Long-term effects of organic, conventional, and integrated crop systems on Carabids. Agronomy for Sustainable Development 31:515–524. Springer Paris. doi: 10.1007/s13593-011-0007-3.

Lövei, G. L., and K. D. Sunderland. 1996. Ecology and behavior of ground beetles (Coleoptera: Carabidae). Annual review of entomology 41:231–56. doi: 10.1146/annurev.en.41.010196.001311.

Niemelä, J., and D. J. Kotze. 2009. Carabid beetle assemblages along urban to rural gradients: A review. Landscape and Urban Planning 92:65–71.

Pendola, A., and T. R. New. 2007. Depth of pitfall traps—does it affect interpretation of ant (Hymenoptera: Formicidae) assemblages? Journal of Insect Conservation 11:199–201. doi: 10.1007/s10841-006-9028-7.

Purvis, G., and A. Fadl. 2002. The influence of cropping rotations and soil cultivation practice on the population ecology of carabids (Coleoptera : Carabidae) in arable land. Pedobiologia 46:452–474.

Rainio, J., and J. Niemela. 2003. Ground beetles (Coleoptera: Carabidae) as bioindicators. Biodiversity and Conservation 12:487–506.

Raupach, M. J., J. J. Astrin, K. Hannig, M. K. Peters, M. Y. Stoeckle, and J.-W. Wägele. 2010. Molecular species identification of Central European ground beetles (Coleoptera: Carabidae) using nuclear rDNA expansion segments and DNA barcodes. Frontiers in Zoology 7:26. BioMed Central.

Schimel, D., M. Keller, S. Berukoff, R. Kao, H. Loescher, H. Powell, T. Kampe, D. Moore, W. Gram, D. Barnett, R. Gallery, C. Gibson, K. Goodman, C. Meier, S. Parker, L. Pitelka, Y. Springer, and R. U. K Thibault. 2011. NEON SCIENCE STRATEGY: Enabling Continental-Scale Ecological Forecasting. Page 56.

Skalski, J. R. 1990. A design for long-term status and trends monitoring. Journal of Environmental Management 30:139–144. doi: 10.1016/0301-4797(90)90011-K.

Vanbergen, A. J., B. A. Woodcook, A. D. Watt, and J. Niemelä. 2005a. Effect of land-use heterogeneity on carabid communities at the landscape scale. Ecography 28:3–16.

Vanbergen, A. J., B. A. Woodcook, A. D. Watt, and J. Niemelä. 2005b. Effect of land-use heterogeneity on carabid communities at the landscape scale. Ecography 28:3–16.

Work, T. T., C. M. Buddle, L. M. Korinus, and J. R. Spence. 2002. Pitfall Trap Size and Capture of Three Taxa of Litter-Dwelling Arthropods : Implications for Biodiversity Studies. Environmental Entomology 31:438–448. Entomological Society of America. doi: 10.1603/0046-225X-31.3.438.

Work, T. T., M. J. Koivula, J. Klimaszewski, D. Langor, J. Spence, J. Sweeney, and C. Hebert. 2008. Evaluation of carabid beetles as indicators of forest change in Canada. Canadian Entomologist 140:393.