However, we suggest that researchers start with 50 samples during the first survey to practice the techniques and to determine the actual number of samples needed to collect the desired percentage of ant species (Chapter 10). Depending on the study objectives, other complementary methods can be added to the standard protocol in order to sample a wider range of ant species (Chapters 1 and 9).

The ALL Protocol

Basic Setup
- 200-m transect (at least one)
- 20 sampling points at 10-m intervals
- 48-hour time period
- 1–2 people (2 people recommended)

Methods Employed at Each Sampling Point
- Standardized, Repeatable Techniques
  - Collect leaf litter, 1 m^2
    - Sift litter
    - Extract ants from litter using mini-Winkler
  - Place 1 pitfall trap
- Optional Techniques to Collect More Species
  - Inspect dead wood
  - Scrape soil (15 × 15-cm area at 1-cm layers down to 10 cm)
  - Direct collecting by hand

Overview of the ALL Protocol

The most important points in implementing the ALL Protocol are outlined in this section, along with references to chapters of this book that contain more information. See Appendixes 1 and 3 for complete lists of equipment needed for the sampling methods and specimen processing.

Transects

Before choosing a particular transect, it is worthwhile to walk through the area to get an impression of the overall environmental variation. Chapters 1 and 9 provide guidance on transect placement.

Ecological Data

In addition to the standard collection information (Chapter 11), ecological data must be recorded. For each transect, the following minimal set of parameters should be described: name of collector, date, choice of transect, locality, habitat, season, soil type, temperature, and microhabitat. See Chapter 9 for a complete list of relevant ecological information and explanations of these parameters.

Labeling Field Samples

It is of the utmost importance to label all samples adequately. Most of the labeling can be done prior to the commencement of field work. Vials used for collecting ants by hand or from logs should preferably be prelabeled as well. See Chapters 9 and 11 for more details.

Pitfall Traps

Pitfall traps should be placed 1 m from the transect line on the opposite side of the transect from where the leaf litter samples were taken. Any plastic drinking cup with smooth sides can be used, but it is best to use cups with openings of the same diameter consistently, in order to standardize the samples. Twenty cups are needed. See Chapter 9 for more information on how to set and collect pitfall traps.

Leaf Litter Samples and Mini-Winkler Extraction

See Chapter 9 for complete instructions on how to collect the 1-m^2 leaf litter samples and extract ants using mini-Winkler sacks. The ALL Protocol requires at least 20 mini-Winkler extractors and one sifter.

Sorting Samples in the Laboratory

Ant specimens and other invertebrates can be separated from debris using the salt water extraction method (Chapter 11). After separation the samples should be washed with ethanol.

Identifying Morphospecies

Ants from each sample should be separated from other invertebrates and housed in a separate vial.