Protocols for Monitoring Pollinators: Beetles

Objective and Purpose

Many plants are dependent upon animal-aided pollination. Beetles are just one of many pollinating arthropods. Beetles contribute to pollination of flowering species that also host other more commonly identified pollinating species such as bees and butterflies, as well as pollinating vertebrates (Proctor et al., 1996; Bernhardt, 2000). Pollinating beetles either possess unique physical characteristics that allow for consumption of nectar and/or pollen (Bernhardt, 2000) or assist in pollination by transferring pollen on their bodies (Gazit et al., 1982; Krakos et al., 2010). The participation of the beetle in pollination is becoming more crucial due to the fragmentation of the landscape through development. Habitat fragmentation can affect the presence and diversity of pollinating species, including invertebrates (Aizen and Feinsinger, 2003). Assessing the presence of beetles on urban and rural sites may aide in determining the resilience of pollinating species to urbanization and development.

Protocol Consistency

Standardizing sampling protocols for the Urban Biodiversity Inventory Framework is a critical requirement of designing surveys that are replicable and producing results that are spatially and chronologically comparable (Larsen, 2016). This is especially important for long-term monitoring efforts that aim to measure changes over time. While a standardized approach among all cities using the Urban Biodiversity Inventory Framework is preferable, each city may identify a methodology best suited for their species of interest and resources at hand. It is important to record and report the methodology used and remain consistent in protocols unless modifications are essential to its improvement. It is equally important that site conditions and day-of conditions for sampling are kept as similar across sites as possible to reduce the impacts of confounding factors. All methodology will be improved with the use of non-biased approaches to data collection, appropriate sampling efforts and accurate reporting of data. The methodology below follows the assumption that the observers are properly trained, using methods to limit bias, and following designated protocol to ensure consistency among sites and years of sampling efforts.

Protocols for Monitoring Pollinators: Beetles

Invertebrate Pollinators: Beetles
Choosing Appropriate Sampling Methods

Suction Systems: *Ground Beetles, Structure-attached Beetles*

Suction systems are an established method for sampling small invertebrates in grass or low vegetation habitats (Standen, 2000; Brook et al., 2008; Zentane et al., 2016). Multiple suction systems for small invertebrate collection have been modeled after the original D-Vac model. Suction systems for ground application consist of a vacuum apparatus and a wide-mouth suction head attachment with a defined area that can be applied to the ground to pick up lightweight invertebrates and funnel them into a prepared container with a preservative solution. Suction systems can also be used as an 'Absolute' sampling method for calibrating relative methods (Duelli et al., 1999). Using a suction system/aspirator, all arthropods larger than 2 mm are collected in a cubic tent with a ground surface of 2 m2. This allows for an absolute measurement of number of species in a defined area, which can be extrapolated to give abundance estimates for a larger site.

Pitfall Traps: *Ground beetles, beetles with ground movement*

Pitfall traps have been frequently used for sampling epigeal invertebrates. Though established as a qualitative sampling efforts, the use for quantitative sampling was realized. Pitfall traps are buried, nested containers that serve to trap ground-crawling invertebrates. Pitfall traps are inexpensive, transportable, and will capture a variety of species. Pitfalls are efficient for many beetle species across many habitats, especially larger sized beetle species. Researchers should be aware that this method is not a live-trapping method, so it may not be beneficial to use if targeting a limited population. Vegetative structure across comparison sites must remain consistent due to an effect on capture rate. Pitfall traps are sensitive to other biotic and abiotic factors, including species reactions to the attractant liquids, shape and size, and their natural dispersal abilities (Woodcock, 2005; Brown and Matthew, 2016). Thus, the rate of capture may not accurately reflect populations of some species as well as comparison of multiple species' abundance on the same site. When focusing on just one species of interest, consistency and standardization is crucial (Woodcock, 2005; Zhao et al., 2013).

Additional Methods

There are many other methods that may be used for arthropod collection besides those discussed below. For additional methodology, see the Inventory Methods for Terrestrial Arthropods: Standards for Components of British Columbia's Biodiversity No. 40 publication (1998), which includes sampling methods for a multitude of arthropod species, including beetles. They also provide additional resources, such as references to data collection forms, modifications on the approaches discussed below, and useful charts to help designate a study design (Table 1).

Protocols for Monitoring Pollinators: Beetles

Track 2 & 3
Presence/Absence and Relative Abundance using Vacuum Apparati

Data to be Entered into UBIF Database

- City
- Data Collector(s)
- Date
- Location name
- Ecosystem/habitat of interest
- Taxonomic group
- Species
- GPS coordinates (Lat/Long in decimal degree format)
- Reference or city site
- Target species presence or absence OR relative abundance (%)

Additional Required Information to Record (see Data Collection Sheet)

- Suction level, type of device used, and any modifications made to the device
- Defined amount of time of each touchdown

Optional Information to Record (see Data Collection Sheet)

- Temperature, wind and weather (sunny, partly cloudy, etc.)
- General site conditions
- Vacuum system details
- Dominant vegetation in the area
- Height of the vegetation within each square

Suction Sampling

Vacuum/aspiration devices can be used to collect invertebrates from low vegetation, grass, litter and even high vegetation (with planning) (Duelli et al., 1999; Stewart et al., 2002; Brook et al., 2008, Zentane et al., 2016). While requiring the purchase of a vacuum system and associated materials, this sampling method is an efficient way to sample species on a site in an exhaustive approach. With standardized methods, these vacuum systems can be used to determine presence/absence and species abundance at a site over time. There are two methods of sampling using suction apparati included below which will be discussed in more detail:

1. Suction sampling using vacuum apparati with a defined-circumference nozzle attachment
2. Suction sampling using vacuum apparati in a tented enclosure

Protocols for Monitoring Pollinators: Beetles

Suction Sampling with Nozzle Attachment

Commonly used devices for this sampling method include the Dietrick Vacuum system (D-Vac), Garden-blower Vacuum system (G-Vac) and the Vortis™ system. The D-Vac and G-Vac systems have a mesh screen or bag within the suction hose to collect particles and invertebrates that are vacuumed up, which can later be separated live or preserved using a solution. The D-Vac system is available to purchase, while the G-Vac system is a modified garden vacuum. Modification commonly entails cutting the collection pipe, with a net being inserted to catch particles (Stewart and Wright, 1995; Stewart, 2002; Cherrill, 2015). The Vortis™ system is a suction device that collects the invertebrates in a separate container with no nets or bags. The container can be filled with solution for preservation, or the individuals may be released afterwards. More information about the D-Vac and Vortis™ systems along with product information can be found here: D-Vac; Vortis™.

The above systems all possess or may be modified with a nozzle attachment that allows the system to be placed directly on the ground and vacuum up invertebrates within a defined circumference. This approach can be used with multiple 'touchdown' locations in a quadrat or along a transect. The sampled invertebrates can then be identified to genus or species, and abundance counts for a given area can be calculated through multiple touchdowns.

Suction Sampling Protocols using Nozzle Attachment:

Protocols adapted from those described in Cherrill and Rushton (1993), Cherrill (2015) and Zentane et al. (2016).

Condition Requirements:

» Suction sampling should occur on dry, warm days with little wind. Site features including vegetative structure, light level and weather patterns may influence presence of species. Cities should attempt to keep site features consistent across reference/city sites and sampling periods.

Establishing Survey Grids:

- On a site, randomly select a corner point for a 12 m x 20 m grid, consisting of fifteen 4 m squares

Collecting Data:

» With your suction device assembled, begin collecting within the center of each 4 m x 4 m square. For each device, there are unique methods to ensure that the area of suction is defined and invertebrates around the perimeter do not get collected outside the nozzle area. Zentane et al. (2015) describes appropriate sampling methods for a G-Vac and Vortis™ system using touchdown methods. The appropriate D-Vac system methods can be found in Stewart and Wright (1995) and Stewart (2002).

» Be sure to keep suction level, type of device used, the operator name, and any modifications to the device consistent across sites and plots.

» For any system, a defined amount of time must be determined for each touchdown.

- A total of 90 seconds may be used, with three rounds of 25 second touches following a 5 second hover. Some systems may have smaller increments, such as nine 10 second touches (Vortis™).
- These increments may be chosen by the city, but the methods should remain constant for each square in the grid and each year of sampling.

» The catch of collected invertebrates should be removed and placed in a larger container whenever the vacuum is not actively intaking air as to prevent loss of individuals.

» After completion of the grid, the sampled individuals can be separated into larger containers for preservation and identified later, or identified to species or genus in the field if resources are available. The benefit of in-field identification is the ability to live-release, however, this may not be an option for more cryptic species.

Determining Presence/Absence

» Presence/absence of the target species will be determined for each survey grid

Calculating Relative Abundance

» All individuals should be speciated and relative abundance will be calculated as the proportion of target beetle species to total collected beetle species

$$\text{Relative Abundance (\%)} = \frac{\text{Number of target species}}{\text{Number of target species + Number of non-target}} \times 100$$

Suction Sampling within a Tented Enclosure

This method uses the same vacuum devices (D-Vac, G-Vac, Vortis™) to collect all invertebrate species within a tented enclosure. This method may be better suited for higher vegetation, or species that cling to vegetation where a touchdown method is not suitable. In a tented enclosure method, a defined area (e.g. 2 m x 2 m) is exhaustively aspirated until all species have been caught in the area. This approach can be combined with a beating method where a white tray is placed under a plant which is then hit, causing the invertebrates to fall onto the tray. Description of beating and sweeping can be found in the Redmon et al. (2000) article.

Protocols for Monitoring Pollinators: Beetles

Suction Sampling Protocols within Tented Enclosure:

Protocol adapted from those described in Duelli et al. (1999).

Condition Requirements

» Sampling should occur on dry, warm days with little moisture.

Establishing Tent Locations

» Obtain a 2 m x 2 m mesh cubic tent that is completely enclosed with a metal frame around the bottom that can extend approximately 6 inches into the soil.
» Using stratified random sampling, define four points in each homogenous habitat to place a 2 m x 2 m tent.
» To place the tent, use an overarm approach and swiftly place the net down against the wind on the randomly selected point. This will limit escape of flying invertebrates due to disturbance of the net placement.

Collecting Data

» Immediately after placing the net down, hammer the metal frame into the soil to prevent terrestrial beetles from fleeing the tented area.
» Using a vacuum system (a compact, more portable model may be preferable due to the restricted space) systematically collect all of the species in the tent. Take care to collect all vegetation dwelling species which may not be apparent. A combined beating approach may be beneficial to ensuring all species are removed from the vegetation.
» After collecting all of the species in the area, immediately repeat the collection process two more times. This will serve as a calibration exercise to see the exhaustiveness of the first collection.
» Once calibration data is collected, repeat the tenting and one-time exhaustive collection method at all four points, recording site data at each point.
» Species identification can occur in lab or in field, with either live or preserved samples. This is up to the cities discretion, though the approach should be consistent.

Determining Presence/Absence

» Presence/absence of the target species will be determined for each tent

Calculating Relative Abundance

» All collected individuals should be speciated and relative abundance will be calculated as the proportion of target beetle species to total collected beetle species

$$\text{Relative Abundance (\%)} = \frac{\text{Number of target species}}{\text{Number of target species + Number of non-target}} \times 100$$

Protocols for Monitoring Pollinators: Beetles

Track 2 and 3
Presence/Absence and Relative Abundance using Pitfall Traps

Note: For presence/absence and relative abundance, there are multiple modifications that can be made to pitfall traps to increase efficiency, including small barriers to direct movement into the trap. Depending on the species of interest, each city should use their best judgement to decide on modification, but should remain consistent in their protocol. Brown and Matthew (2016) suggest a standardized approach for pitfall traps that is used below.

Data to be Entered into UBIF Database

- City
- Data Collector(s)
- Date
- Location name
- Ecosystem/habitat of interest
- Taxonomic group
- Species
- GPS coordinates (Lat/Long in decimal degree format)
- Reference or city site
- Target species presence or absence OR relative abundance (%)

Additional Required Information to Record (see Data Collection Sheet)

- Transect or grid information (number, size/length, etc.)
- Counts of target and non-target individuals (for Track 3 data only)

Optional Information to Record (see Data Collection Sheet)

- Site and weather conditions
- Non-target species information (for Track 3 data only)

Protocols for Monitoring Pollinators: Beetles

Pitfall Sampling Protocol:

Protocols adapted from those described in Garvey (2012) and Yekwayo et al. (2006).

Establishing Transects or Grids

» Randomly select start points and direction of transects (if assessing multiple habitat types on one site, random stratified sampling is recommended). Systematic sampling through a grid approach may also be used.

» If using transects, mark a point with an identifying marker every 20 m (smaller distances will work, but no less than 10 m).

- At each point, place four (4) pitfall traps in a square arrangement spaced at least 5 m apart. Greater than 5 m distancing has higher yield in invertebrate captures (Ward et al., 2001).

» For the grid approach, randomly select a starting point in a homogenous habitat type, and mark points in a 5 m x 5 m arrangement spaced 10 m apart. At each point, place two pitfall traps.

Collecting Data

» Each pitfall trap should consist of nested containers (Figure 1) buried in the ground with lip flush to surface. Typical pitfall trap includes inner cup for easy removal (90-110 mm width and 90-110 mm depth), transparent cup, transparent funnel and transparent rain guard. If rain is expected, use soil to raise the ground level up an inch. The inner container should have a preservative solution of a of propylene glycol and ethyl alcohol or water. Brown and Matthew (2016) recommend 100 ml of a suitable transparent, nontoxic killing preservative such as propylene glycol, with concentration clearly reported. When determining the amount of solution to use, the researcher should take into account temperature and evaporation rates.

» A transparent funnel should be placed on top of the inner container to limit escape of fallen invertebrates as well as to lessen the chance of accidental small mammal trapping.

» The assembled pitfall trap should have a transparent cover on top (slightly lifted) to prevent excess precipitation from entering the solution.

» The pitfall traps should be checked once weekly for the active season of the species of interest. To check, lift the cover, and pull out the inner cup containing the solution and the invertebrates. Pour the solution with the invertebrates over a wire mesh to collect, and place in a 70% alcohol solution to preserve for genus identification.

Determining Presence/Absence

» Presence/absence of the target species will be determined for each transect or grid

Calculating Relative Abundance

» All collected individuals should be speciated and relative abundance will be calculated as the proportion of target beetle species to total collected beetle species

$$\text{Relative Abundance (\%)} = \frac{\text{Number of target species}}{\text{Number of target species + Number of non-target}} \times 100$$

To improve efficiency of trapping, an attractant solution or a lighted pitfall trap may be used (Figure 2). Pit-light traps have better efficacy in catching beetles in forests, both in number and in diversity (Hébert et al., 2000). This trap type is particularly suited for nocturnal light-attracted species. Lighted pitfalls may artificially increase abundance calculations, so they are best used for presence/absence community composition studies.

Lighted pitfall trap: The trap is 38 cm in height and is made of two parts: a 1-L collection container inserted into the ground which has a diameter of 10 cm at its base and 13 cm at its rim, and an upper container which houses a 6-V lantern alkaline battery and a circuit for electronic control of a 1.8-W miniature blue fluorescent tube. Pit-light traps should be switched on at nightfall by a photoelectric cell and remained in operation until dawn. A cover should be used to prevent precipitation from entering (Hébert et al., 2000). A more complete description of the Luminoc trap and of its components can be found in Jobin and Coulombe (1994).

Figures

TAXA	Pan/Pitfall	Window/Malaise	Berlese	Beating	Sweeping	Searching	Sifting	Netting & Chasing	Dipnet - sweeping	Dipnet - shuffling	Light trap
MOLLUSCA (fresh water)						X			X	X	
ANNELIDA (terrestrial)	X		X								
CRUSTACEA AMPHIPOD						X			X	X	
ISOPODA						X			X	X	
ARACHNIDA SOLPUGIDA	X										
SCORPIONIDA	X					X					
ARANEAE	X		X	X	X	X	X		X		
ACARIFORMES			X						X	X	
DIPLODA	X		X			X	X				
DIPLURA			X								
COLLEMBOLA	X		X								
INSECTA MICROCORYPHIA	X		X			X					
ODONATA								X	X	X	
EPHEMEROPTERA				X	X	X		X	X	X	X
PLECOPTERA	X			X	X	X		X	X	X	
NOTOPTERA						X					
DICTUOPTERA	X					X					
GRYLLOPTERA	X			X	X	X		X			X
ORTHOPTERA	X	X		X	X	X		X			X
HETEROPTERA	X	X	X	X	X	X	X	X	X		X
HOMOPTERA (Auchenoprrhyncha)	X	X		X	X	X		X			X
MEGALOPTERA		X		X	X	X		X	X	X	X
RAPHIDIOPTERA		X		X	X	X		X			X
NEUROPTERA		X		X	X	X		X			X
COLEOPTERA (Carabidae, etc.)	X	X	X	X	X	X	X	X	X		X
MECOPTERA		X				X		X			
DIPTERA (some)	X	X		X	X	X		X			
LEPIDOPTERA (Macrolepidoptera)								X			X
TRICHOPTERA		X		X	X	X			X	X	X
HYMENOPTERA (Aculeates)	X	X	X	X	X	X		X			

Table 1. Taxa Collected by Various Sampling Techniques. Image from Inventory Methods for Terrestrial Arthropods (1998).

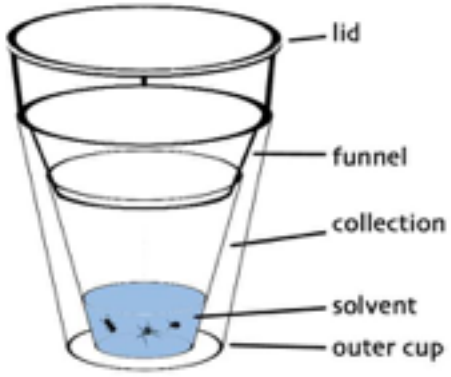

Figure 1. Typical pitfall setup includes an inner cup for easy removal (90-110 mm width, 90-110 mm depth), transparent cup, transparent funnel and transparent rain guard. Image from University of St. Andrews (n.d.).

Figure 2. Lighted Pitfall Trap. Image from Hébert et al. (2000).

Citations and Additional Resources

Aizen, M.A. and P. Feinsinger. 2003. Bees not to be? Responses of insect pollinator faunas and flower pollination to habitat fragmentation. How landscapes change: 111-129. Springer Berlin Heidelberg.

Bernhardt, P. 2000. Convergent evolution and adaptive radiation of beetle-pollinated angiosperms. Plant Systematics and Evolution 222: 293-320.

Brook, A.J., B.A. Woodcock, M. Sinka, A.J. Vanbergen. 2008. Experimental verification of suction sampler capture efficiency in grasslands of differing vegetation height and structure. Journal of Applied Ecology 45(5): 1357-1363.

Brown, G.R. and I.M. Matthews. 2016. A review of extensive variation in the design of pitfall traps and a proposal for a standard pitfall trap design for monitoring ground□active arthropod biodiversity. Ecology and evolution.

Cherrill, A. 2015. Suction sampling of grassland invertebrates using the G-vac: Quantifying and avoiding peripheral suction effects. European Journal of Entomology 112(3): 520.

Cherrill, A.J. and S.P. Rushton. 1993. The Auchenorrhyncha of an unimproved moorland in northern England. Ecological Entomology (18): 95–103.

Duelli, P., M.K. Obrist, D.R. Schmatz. 1999. Biodiversity evaluation in agricultural landscapes: above-ground insects. Agriculture, Ecosystems & Environment 74(1): 33-64.

Hébert, C., L. Jobin, M. Fréchette, G. Pelletier, C. Coulombe, C. Germain, M. Auger. 2000. An efficient pit-light trap to study beetle diversity. Journal of Insect Conservation 4(3): 189-200.

Garvey, L. 2012. A Survey of Beetles (Coleoptera) for Greenham Common and Bowdown Woods during 2012.

Gazit, S., I. Galon, H. Podoler. 1982. The role of nitidulid beetles in natural pollination of annona in Israel. Journal of the American Society for Horticultural Science 107(5): 849-852.

Inventory Methods for Terrestrial Arthropods: Standards for Components of British Columbia's Biodiversity (Version 2.0). 1998. Canadian Ministry of Environment, Lands and Parks: Resources Inventory Branch. Resources Inventory Committee, The Province of British Columbia, Canada.

Jobin, L. and C. Coulombe. 1994. Canada Minister Of Forestry, Portable luminous insect trap. U.S. Patent 5,301,456.

Krakos, K.N., G.M. Booth, P. Bernhardt. 2010. Mechanical vs. beetle-mediated self-pollination in Gossypium tomentosum (Malvaceae), an endangered shrub. International Journal of Insect Science 2: 35-49.

Larsen, T.H. 2016. Core standardized methods for rapid biological field assessment. Conservation International, Arlington, VA. 207 p.

Redmon, S.G., T.G. Forrest, G.P. Markin. 2000. Biology of Bruchidius villosus (Coleoptera: Bruchidae) on scotch broom in North Carolina. Florida Entomologist 83(3): 242-253.

Proctor, M., P. Yeo, A. Lack. 1996. The natural history of pollination. HarperCollins Publishers.

Standen, V. 2000: The adequacy of collecting techniques for estimating species richness of grassland invertebrates. J. Appl. Ecol. 37: 884–893.

Stewart, A.J. and A.F. Wright. 1995. A new inexpensive suction apparatus for sampling arthropods in grassland. Ecological Entomology 20(1): 98-102.

Stewart, A.J. 2002. Techniques for sampling Auchenorrhyncha in grasslands. Na.

University of St. Andrews. n.d. GardenLife: a study of gardens as a reservoir for arthropod biodiversity. Sampling. University of St. Andrews and Stellenbosch University, Fife, Scotland. <http://biology.st-andrews.ac.uk/gardenlife/sampling.html>.

Woodcock, B.A. (2005) Pitfall Trapping in Ecological Studies, in Insect Sampling in Forest Ecosystems (ed S. R. Leather), Blackwell Science Ltd, Oxford, UK.

Ward, D.F., T.R. New, A.L. Yen. 2001. Effects of pitfall trap spacing on the abundance, richness and composition of invertebrate catches. Journal of Insect Conservation 5(1): 47-53.

Yekwayo, I., J.S. Pryke, F. Roets, M.J. Samways. 2016. Surrounding vegetation matters for arthropods of small, natural patches of indigenous forest. Insect Conservation and Diversity 9(3): 224-235.

Zentane, E., H. Quenu, R.I. Graham, A. Cherrill. 2016. Suction samplers for grassland invertebrates: comparison of numbers caught using Vortis™ and G□vac devices. Insect Conservation and Diversity 9(5): 470-474.

Zhao, Z.H., P.J. Shi, C. Hui, F. Ouyang, F. Ge, B.L. Li. 2013. Solving the pitfalls of pitfall trapping: a two□circle method for density estimation of ground□dwelling arthropods. Methods in Ecology and Evolution 4(9): 865-871.