



DNA LEARNING CENTER

Barcode Long Island DNA Barcoding and Biodiversity Campaigns and Independent Projects



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DNA Barcoding

• DNA barcoding allows for identification of organisms using their DNA.



DNA barcoding lends itself to studies involving

biodiversity and species identification

Biodiversity

• The variety of living things in a given area or environment





Biodiversity

• What is species diversity?

• Describes the number of different kinds of organisms within a community or ecosystem.

How many species are there?

- Currently 1.7 million species identified
- Estimates range between 3-50 million
- Recent data support an estimate of 4 to 6 million insect species alone.
- Invertebrates make up 65% of all known species, and probably most of yet to be discovered species.

Benefits of Biodiversity

- Ecological Benefits
 - Soil formation
 - Waste disposal
 - Air and water purification
 - Nutrient cycling
 - Solar energy absorption
 - We do not fully understand biological communities. Loss of a seemingly insignificant species can be damaging.

Benefits of Biodiversity

• Food

• Wild plants could provide new sources of food or more genetic diversity for existing crops

Drugs and Medicines

- More than half of all modern medicines contain some natural product from a wild species
- Pharmaceutical companies actively prospect tropical countries for products
 - \$30 billion/year!

Accelerating Extinction Rates

- We are currently in the 6th extinction
 - Worst species die-off since the dinosaurs 65 million years ago
 - 99% of currently threatened species are at risk from human activities

TABLE 5.1	Estimated Number of Species		
CLASS	NUMBER DESCRIBED	NOT YET EVALUATED ¹	PERCENTAGE THREATENED ²
Mammals	5,491	0%	21%
Birds	9,998	0%	12%
Reptiles	9,084	82%	28%
Amphibians	6,433	2%	30%
Fishes	31,300	86%	32%
Insects	1,000,000	100%	27%
Mollusks	85,000	97%	45%
Crustaceans	47,000	96%	35%
Other invertebrates	173,250	99%	30%
Mosses	16,236	99%	86%
Ferns and Allies	12,000	98%	66%
Gymnosperms	1,021	11%	35%
Flowering Plants	281,821	96%	73%
Fungi, Lichens, Protists	51,563	100%	50%

¹Evaluated by IUCN for threatened status.

²Number of species as a percentage of those evaluated. Includes IUCN categories critically endangered, endangered, or vulnerable.

SOURCE: IUCN Red List, 2012

Insects as Bioindicators



Bioindicators – organisms whose presence or absence can inform us about ecosystem health

Chowdhury S, Dubey VK, Choudhury S, Das A, Jeengar D, Sujatha B, Kumar A, Kumar N, Semwal A and Kumar V (2023) Insects as bioindicator: A hidden gem for environmental monitoring. *Front. Environ. Sci.* 11:1146052. doi: 10.3389/fenvs.2023.1146052

Species Identification

Identification of living things is not trivial...





Figure 2. Morphological differences between F. btophtlica and F. incerta: a) F. btophtlica (closed circles) generally has a longer mesosoma (WL) relative to head proportions than F. incerta (open diamonds). b) Whereas F. btophtlica rarely has more macrochaetae on the propodeum than the pronotum (12 of 32 specimens), this is more often the case for the F. incerta (22 of 31). The line indicates the relationship PnM = Ppm.



Adding to the complexity: *immature, damaged, or incomplete specimen may make identification impossible.*

Trager, J. C., MacGown, J. A., Trager, M. D. 2007. Revision of the Nearctic endemic Formica pallidefulva group, pp. 610-636. In Snelling, R. R. B. L. Euber, and P. S. Ward (eds). Advances in an systematics (Hymenoptera: Formicidae): homoge to E. O. Wilson – 30 years of contributions. Memory of the American Entomological Institute, 80.

Species Identification

Complex terminology!

Take this family diagnosis:



The body form ranges from <u>hemispherical</u> (e.g., *Cleidostethus*) to <u>elongate</u> oval (e.g., *Clypastraea*) to latridiid-like (e.g., *Foadia*). Corylophids are typically dull brown, but some species have contrasting yellowish-brown patches on the <u>pronotum</u> or <u>elytra</u>. The <u>integument</u> is often densely punctured and may be <u>glabrous</u> or bear short, fine <u>recumbent setae</u>. Most corylophid adults can be diagnosed using the following morphological features: <u>Maxilla</u> with single <u>apical lobe</u>; <u>Mesotrochanter</u> short and strongly oblique; Head usually covered by <u>pronotum</u>; <u>Frontoclypeal suture</u> absent; Antennae elongate with <u>3-segmented club</u>; <u>Procoxal</u> cavities closed externally; <u>Tarsal formula 4-4-4</u>; <u>Pygidium</u> exposed

Diagnosis: an articulated list of characters that defines a particular taxonomic group

Leaves <u>alternate proximally</u>, opposite and ultimately <u>decussate distally</u>, 6–16 × 4–13 cm; <u>petiole</u> ca. as long as blade, <u>winged</u>, base clasping, <u>basal lobes stipulate</u>, growing as extensions of wings, less than 1 mm wide; blade 5– 7-veined, <u>ovate</u>, <u>glabrous</u>, base typically <u>sagittate</u>, margins entire, <u>apex acute</u> to <u>acuminate</u>. <u>Staminate</u> <u>inflorescences axillary</u>, 1–2 per <u>axil</u>, <u>paniculate</u>, <u>fasciculate</u>; <u>panicles</u> bearing flowers <u>singly</u>, <u>bracteolate</u>, in a zigzag pattern along <u>rachis</u>, <u>internodes</u> less than 2 mm; <u>rachis</u> to 25 cm, secondary axes 1–3(–6), <u>fasciculate</u>, less than 3 cm, each subtended by <u>deltate-ovate</u> <u>bracteole</u> shorter than 1 mm. *Pistillate* <u>inflorescences</u> solitary, 4–8(–20)flowered, 6–35 cm, <u>internodes</u> ca. 1 cm

Complex and somewhat objective



>Dioscorea alata (matK) gene, partial

Simple (A,T,G, or C) and more objective

DNA Barcoding Workflow



Organism is sampled

DNA is extracted



"Barcode" amplified

ACGAGTCGGTAGCTGCCCTCTGACTGCAT CGAATTGCTCCCCTACTACGTGCTATATGC GCTTACGATCGTACGAAGATTTATAGAAT GCTGCTAGCTGCTCCCTTATTCGATAACTA G C T C G A T T A T A G C T A C G A T G



Sequenced DNA is compared with a barcode database

Genes with the right number of differences



Animals: Mitochondrion



Fungi, Bacteria: Nucleus





Species Identification

DNA Barcoding is a way quicker way to identify the species living in an area and just about anyone can do it, almost anywhere!

Species Diversity vs. Genetic Diversity

While we are ultimately using variation in DNA to ID a species, BLI projects should focus on **species diversity**

Linking an environmental mutagen to mutation (leading to variation in DNA) is difficult and should not be the focus of BLI studies

AKA: avoid genetic diversity studies...

Species Abundance vs. Species Diversity

Species abundance – the population size of a species in an area

Out of 20 samples, finding that half of the barcodes match one species *is not* a strong measure of *abundance*

Focus on **species diversity** – abundance can be measured through observation or other means