DNA Barcodes of Caterpillars (Lepidoptera) from Papua New Guinea

Author(s): Scott E. Miller, Jan Hrcek, Vojtech Novotny, George D. Weiblen and Paul D.N. Hebert


Published By: Entomological Society of Washington

DOI: http://dx.doi.org/10.4289/0013-8797.115.1.107

URL: http://www.bioone.org/doi/full/10.4289/0013-8797.115.1.107
DNA barcodes of caterpillars (Lepidoptera) from Papua New Guinea

DOI: 10.4289/0013-8797.115.1.107

This paper provides metadata for DNA barcode (COI) data in GenBank for a collection of caterpillar vouchers from ecological studies in Wanang, Papua New Guinea, sampled as part of long-term research on the ecology and taxonomy of herbivorous insects there (Miller et al. 2003, Craft et al. 2010, Novotny et al. 2010, Whitfeld et al. 2012). This paper aims to make DNA barcode data available to document ongoing research, to contribute to the International Barcode of Life (iBOL; www.ibol.org) project, and to encourage enhancement in identifications, in line with the concept of DNA barcode data release papers and the Fort Lauderdale principles for genetic data (Schindel et al. 2011). Data released on Genbank (accession numbers HM906186-HM906529, HQ558240-HQ558310, KC158225-KC158271) includes the standard fields for the BARCODE data standard (Benson et al. 2012) and more data, including images and host plant codes, are available on BOLD (www.boldsystems.org; Ratnasingham and Hebert 2007), accessible from the project CATS using a DOI (dx.doi.org/10.5883/DS-CATS1).

Field methods and host plants: Trees and shrubs with stems greater or equal to 5 cm in diameter at breast height (dbh) in two 100 x 100 m plots near Wanang (145.182° E, 5.231° S), Madang Province, Papua New Guinea were destructively sampled (see Whitfeld et al. 2012 for details). The two plots were 800 m apart in a mosaic of primary and secondary rain forest vegetation at 100–200 m above sea level in an extensive mixed evergreen forest on latosols in the Ramu River basin (Paijmans 1976). The climate is generally humid and relatively aseasonal. Historical readings from Madang (70 km east, 1956–1970; McAlpine et al. 1983) indicate a mean annual rainfall of 3500 mm and mean monthly temperature between 26.2 °C and 26.7 °C. Local landowners practice subsistence agriculture in 0.25–1.0 ha gardens planted after felling and burning of primary forest, and we coordinated our sampling with local landowners who were planning to clear the sites for subsistence gardens. After mapping and clearing adjacent vegetation, trees were individually felled and immediately inspected for the presence of caterpillars and leaf miners by a team of eight field workers. Live caterpillars were hand-collected and placed in plastic vials for processing. The live caterpillars were assigned to morphospecies (numbered in the CATX series), documented with photographs, and, in most cases, with preserved vouchers. Host plant data are included in the BOLD records via the plant number in the form WS2B2315. All plants were vouchedered with full data available as part of the Digital Flora of New Guinea in the Atrium database (http://ng.atrium-biodiversity.org/atrium). DNA data (rbcL) for representatives of most plant species are in GenBank (accession numbers JF738369–JF739166).

Lepidoptera methods: General field and laboratory methods for Lepidoptera are described in Miller et al. (2003) and Novotny et al. (2010). DNA sequencing (COI barcode) followed standard methods at the Biodiversity Institute of Ontario,
University of Guelph (Craft et al. 2010, Wilson 2012), using tissue from the anal end of the caterpillars. 475 vouchers were sampled for DNA, resulting in 462 successful sequences (including one Coleoptera larva and three larvae of Hymenoptera parasitoids accidently sampled with the caterpillar tissue and preferentially amplified from the samples). Caterpillar vouchers are deposited in the Smithsonian National Museum of Natural History. The images in the BOLD records were taken from live caterpillars to characterize the morphospecies as they were classified in the field. In many cases, these images represent the same specimens that were sequenced, but not always, and we have noticed (and removed) several cases of confused labeling of images, so the images should be used with care.

Identifications: The 462 successful sequences group into 351 barcode clusters. These were identified using the BOLD identification tool, primarily against some 18,000 sequences that we have generated for adult New Guinea Lepidoptera, and a larger library for adult Australian Lepidoptera, including more than 30,000 specimens from the Australian National Insect Collection. Identifications were made either based on very close matches at the species level (less than one percent difference) or using NJ tree topology at the generic level or above. Some species remain unidentified, and we welcome comments on all identifications.

We are currently preparing ecological analyses of the caterpillar communities, their host plants, and their parasites (e.g., Hrcek et al. 2011) and continuing taxonomic analyses of the adult Lepidoptera that can be linked to these larvae by their DNA barcodes.

ACKNOWLEDGMENTS

We thank the staff at the New Guinea Binatang Research Center for field assistance, Wanang landowners for access to field sites and assistance, Kipiro Damas and PNG Forest Research Institute for plant species identification. Karolyn Darrow, Lauren Helgen, Margaret Rosati, and many taxonomists have assisted in building the DNA barcode library used for Lepidoptera identifications. Field work was supported by the U.S. National Science Foundation grant DEB-0515678, Czech Science Foundation grant 206/09/0115, and Czech Ministry of Education & European Union grant CZ.1.07/2.3.00/20.0064. DNA sequencing was supported by a grant from the Government of Canada through Genome Canada and the Ontario Genomics Institute in support of the iBOL project.

LITERATURE CITED


Scott E. Miller, National Museum of Natural History, Smithsonian Institution, P.O. Box 37012, MRC 105, Washington, D.C. 20013-7012, U.S.A. (e-mail: millers@si.edu)

Jan Hrcek, Faculty of Science, University of South Bohemia and Biology Center, Czech Academy of Sciences, Branisovska 31, 37005 Ceske Budejovice, Czech Republic

Vojtech Novotny, Czech Academy of Sciences, Biology Center and University of South Bohemia, Faculty of Science, Branisovska 31, 37005 Ceske Budejovice, Czech Republic

George D. Weiblen, University of Minnesota, Bell Museum and Department of Plant Biology, 250 Biological Sciences Center, 1445 Gortner Avenue, St. Paul, Minnesota 55108-1095, U.S.A.

Paul D.N. Hebert, Department of Integrative Biology and the Biodiversity Institute of Ontario, University of Guelph, Guelph, Ontario N1G 2W1, Canada