

Supplementary material for “Evolution of *Hypolimnas* butterflies (Nymphalidae): Out-of-Africa origin and *Wolbachia*-mediated introgression.”

(Detailed legends are given along with respective table or figure)

Table A1: Distribution, host plant families and putative model taxa of recognized *Hypolimnas* species.

Table A2. Summarizing the divergence times across multiple analyses.

Table A3. Comparison for the fit of different models of diversification.

Table A4. Model selection table comparing the diversification of clades.

Figure A1. Maximum likelihood tree from the mitochondrial dataset.

Figure A2. Maximum likelihood tree from the nuclear dataset.

Figure A3. Lineage Through Time graph for empirical and simulated plots.

Figure A4. Best-tree configuration with diversification rates from BAMM analysis.

Figure A5. Geographical distribution of genera in the tribe Junoniini.

Figure A6. Maximum likelihood tree of mitochondrial COI locus including additional *H. bolina* sequences from GenBank.

Table A1. Geographic distributions, larval host plants, and taxon of putative butterfly models (where applicable) for all recognized *Hypolimnas* species, many of which are Batesian mimics.

Species	Author	Recognized by Lamas (2015)	Sampled	Distribution	Host plant families	Putative model taxa	References
<i>alimena</i>	Linnaeus, 1758	Yes	Yes	Oriental, Australasian	Urticaceae, Acanthaceae, Malvaceae	<i>Euploea</i> spp.	1,3,10,13, 16,18,20
<i>anomala</i>	Wallace, 1869	Yes	Yes	Oriental, Australasian	Urticaceae, Euphorbiaceae	<i>Euploea</i> spp.	1,13,18,20
<i>antevorta</i>	Distant, 1880	Yes	Yes	Afrotropical	Urticaceae	NA	13
<i>anthedon</i>	Doubleday, 1845	No	Yes	Afrotropical	Urticaceae, Asteraceae	<i>Amauris</i> spp.	7,8,13
<i>antilope</i>	Cramer, 1777	Yes	Yes	Oriental, Australasian	Urticaceae, Acanthaceae	<i>Euploea</i> spp.	10,11,13,16, 17,18,20
<i>aubergeri</i>	Hecq, 1987	Yes	No	Afrotropical	NA	<i>Acraea</i> spp.	8,9
<i>bartelotti</i>	Grose-Smith, 1890	Yes	Yes	Afrotropical	NA	NA	4
<i>bolina</i>	Linnaeus 1758	Yes	Yes	Australasian, Oriental, Palearctic, Afrotropical	>10	<i>Euploea</i> spp.	1,3,7,8,10, 11,12,13,16, 17,18,20
<i>chapmani</i>	Hewitson, 1873	Yes	No	Afrotropical	NA	<i>Acraea</i> spp.?	8,20
<i>deceptor</i>	Trimen, 1873	Yes	Yes	Afrotropical	Urticaceae	<i>Amauris</i> spp.	7,13,20
<i>deois</i>	Hewitson, 1858	Yes	Yes	Oriental, Australasian	Urticaceae	<i>Taenaris</i> spp. <i>Euploea</i> spp.	10,20
<i>dexithea</i>	Hewitson, 1863	Yes	Yes	Afrotropical	Urticaceae	NA	4
<i>dimona</i>	Fruhstorfer, 1912	Yes	Yes	Oriental	NA	NA	18
<i>dinarcha</i>	Hewitson, 1863	Yes	Yes	Afrotropical	Urticaceae	<i>Amauris</i> spp.	7,8,13,20
<i>diomea</i>	Hewitson, 1861	Yes	Yes	Oriental	Urticaceae	<i>Euploea</i> spp.	18,20
<i>errabunda</i>	Hopkins, 1927	Yes	Yes	Australasian	Urticaceae, Moraceae	<i>Euploea</i> spp.	11,13,14,20
<i>euploeoides</i>	Rothschild, 1915	Yes	No	Australasian	NA	<i>Euploea</i> spp.	10,20
<i>exiguus</i>	Samson, 1980	Yes	Yes	Australasian	NA	NA	4
<i>inopinata</i>	Waterhouse, 1920	Yes	Yes	Australasian	Urticaceae	NA	11,14,19
<i>mechowi</i>	Dewitz, 1884	Yes	Yes	Afrotropical	NA	<i>Amauris</i> spp.	20
<i>misippus</i>	Linnaeus, 1764	Yes	Yes	Australasian, Oriental, Palearctic, Afrotropical, Neotropical/ Nearctic	>10	<i>Danaus chrysippus</i>	1,7,8,10,13, 16,17,18,20
<i>monteironis</i>	Druce, 1874	Yes	Yes	Afrotropical	Urticaceae	non-mimetic	7,8,13
<i>octocula</i>	Butler, 1869	Yes	Yes	Australasian	Urticaceae, Portulacaceae	NA	2,3,11,14, 15,17
<i>pandarus</i>	Linnaeus, 1758	Yes	Yes	Oriental	Urticaceae	NA	5
<i>pithoeka</i>	Kirsch, 1877	Yes	Yes	Oriental, Australasian	Urticaceae, Malvaceae, Portulacaceae	<i>Euploea</i> spp.	10,16,17,20
<i>salmacis</i>	Drury, 1773	Yes	Yes	Afrotropical	Urticaceae	non-mimetic	7,8,13,20
<i>saundersi</i>	Wallace, 1869	Yes	Yes	Oriental	NA	NA	18

Species	Author	Recognized by Lamas (2015)	Sampled	Distribution	Host plant families	Putative model taxa	References
<i>sumbawana</i>	Pagenstecher, 1898	No	Yes	Oriental	NA	NA	18
<i>usambara</i>	Ward, 1872	Yes	Yes	Afrotropical	Urticaceae	<i>Amauris</i> spp.	7,13,20

Data References

- Braby, M.F., 2000. Butterflies of Australia: Their Identification, Biology and Distribution. Vol. 2, CSIRO Publishing, Australia.
- Campora, C., Lee, S., 2009. Survey for the Mariana eight spot butterfly, *Hypolimnas octocula marianensis* (Lepidoptera: Nymphalidae), in the Pagat Route 15 area of Yigo Village, Guam. NAVFAC Pacific series. URL
<http://guaminsects.myspecies.info/survey-mariana-eight-spot-butterfly-hypolimna-octocula-marianensis-lepidoptera-nymphalidae-pagat-rou>. Last accessed 20 August 2017.
- Holloway, J.D., Peters, J.V., 1976. The butterflies of New Caledonia and the Loyalty Islands. *J. Nat. Hist.* 10, 273-318. doi: 10.1080/00222937600770211
- Hypolimnas* data base from <http://www.nic.funet.fi>. URL
<http://www.nic.funet.fi/pub/sci/bio/life/insecta/lepidoptera/ditrysia/papilionoidea/nymphalidae/nymphalinae/hypolimnas/index.html> and the references therein. Last accessed 17 February 2017.
- Igarashi, S., Harada, M., 2014. Sequel to “The Life Histories of Asian Butterflies Vols. I and II”. Igarashi & Hirada (Privately published).
- Lamas, G., 2015. Catalog of the butterflies, Available from the author.
- Larsen, T.B., 1996. The Butterflies of Kenya and their Natural History. Oxford University Press, New York.
- Larsen, T.B., 2005. Butterflies of West Africa. Apollo Books, Stenstrup, Denmark.
- Larsen, T.B. 2011. *Hypolimnas aubergeri*. The IUCN Red List of Threatened Species 2011: e.T160287A5360138. URL
<http://dx.doi.org/10.2305/IUCN.UK.2011-2.RLTS.T160287A5360138.en>. Last accessed 13 September 2017.
- Parsons, M., 1998. The Butterflies of Papua New Guinea: Their Systematics and Biology. Academic Press, London.
- Patrick, B., Patrick, H., 2012. Butterflies of the South Pacific. Otago University Press, New Zealand.
- Ramsay, G.W., 1971. The blue moon butterfly (*Hypolimnas bolina nerina*) in New Zealand during autumn. *N.Z. Ent.* 5, 73-75.
- Robinson, G.S., Ackery, P.R., Kitching, I.J., Beccaloni, G.W., Hernández L.M., 2010. HOSTS—a Database of the World’s lepidopteran hostplants. Natural History Museum, London. URL
<http://www.nhm.ac.uk/hosts>. Last accessed 17 February 2017.
- Samon, C., 1986. The *Hypolimnas octocula* complex, with notes on *H. inopinata* (Lepidoptera, Nymphalidae). *Tyô to Ga.* 37, 15-43.
- Schreiner, I.H., Nafus, D.M., 1997. Butterflies of Micronesia. Agricultural Experiment Station, College of Agriculture and Life Sciences, University of Guam, Mangilao.
- Tennent, J., 2002. Butterflies of the Solomon Islands: Systematics and Biogeography. Storm Entomological Publications, United Kingdom.
- Tennent, J., 2009. A Field Guide to the Butterflies of Vanuatu (OI Buttaflae blong Vanuatu). Storm Entomological Publications. United Kingdom.
- Tsukada, E., 1985. Butterflies of the South East Asian Islands (part IV): Nymphalidae (I). Plapac Co. Ltd, Tokyo, Japan.
- Tuiwawa, M.V., Pene, S., Tuiwawa, S.H., (eds.). 2013. A rapid biodiversity assessment and archaeological survey of the Fiji REDD+ plot site: Emalu forest, Viti Levu. Compiled by University of South Pacific for Ministry of Agri. Fishry and Forestry, Republic of Fiji Islands.
<http://www.fiji-reddplus.org/content/rapid-biodiversity-assessment-archaeological-survey-fiji-redd-pilot-site-emalu-forest-viti> Last accessed 10 August 2017.
- Vane-wright, R.I., Ackery, P.R., Smiles, R.L., 1977. The polymorphism, mimicry, and host plant relationships of *Hypolimnas* butterflies. *Biol. J. Linn. Soc.* 9, 285-297.

Table A2. The mean crown age (in millions of years) estimates along with the 95% highest posterior density (HPD), in parentheses, for three specific clades (Root, Junoniini and *Hypolimnas*) across multiple analyses. Detail of the calibrations from Wahlberg et al. (2009) are mentioned in the Methods section. Regarding calibrations from Su et al. (2017): we assigned 42.3 Mya (95% HPD: 35.3 - 49.8) to the crown age of Junoniini and 56.2 Mya (95% HPD: 47.3 - 64.5) to the root (Junoniini+Victorinini).

		Root (Victorinini+Junoniini)	Junoniini	<i>Hypolimnas</i>
Calibrations from Wahlberg et al. (2009)	BEAST analysis (birth-death tree prior)	39.5 (33.3 - 45.6)	29.5 (24.6 - 34.6)	13.4 (9.3 - 18.1)
	BEAST analysis (Yule-model tree prior)	37.9 (32.0 - 43.9)	30.9 (26.1 - 35.8)	16.8 (12.1 - 21.6)
	PATHd8	41.1	34.0	15.1
Calibrations from Su et al. (2017)	BEAST analysis (birth-death tree prior)	54.6 (45.6 - 64.4)	41.9 (34.5 - 49.5)	18.7 (12.8 - 25.3)
	BEAST analysis (Yule-model tree prior)	52.2 (43.0 - 61.2)	43.9 (36.5 - 51.3)	24.4 (17.4 - 31.9)
	PATHd8	56.2	46.8	20.7

Table A3. Comparison of different models of diversification fitted to the diversification pattern in the time tree. Pure Birth is a pure birth or Yule model of diversification, Birth Death is a constant birth-death model of diversification, SPVAR considers a variable speciation rate and constant extinction rate, EXVAR considers a variable extinction rate and constant speciation rate, BOTHVAR considers variability in both speciation and extinction rates. AIC = Akaike information criteria; logLik = log-likelihood; λ , μ are speciation and extinction rates, respectively.

	Pure Birth	Birth Death	SPVAR	EXVAR	BOTHVAR
logLik	-4.426	-4.426	-4.383	-4.433	-4.378
AIC	10.852	12.852	14.766	14.866	16.756
λ	0.202	0.202	0.244	0.202	0.243
μ	-	0	0.001	0.001	0.001

Table A4. Model selection table comparing the diversification of African and non-African clades. The wide spread species *H. bolina* and *H. misippus* are within the non-African clade. The African clade based almost exclusively on Urticaceae, while non-African species often feed on several other families. K = number of parameters; logLik = log-likelihood; AIC = Akaike information criteria; λ , μ are speciation, extinction rates for Africa (0) or non-Africa (1) clade. The sampling frequency for Africa clade was 10/12 and that for non-African clade was 16/17.

	df	AIC	logLik	λ_0	λ_1	μ_0	μ_1
Full model	6	141.858	-64.929	0.167	0.266	0	0
Equal speciation	5	140.678	-65.339	0.240	-	0.103	0
Equal extinction	5	139.857	-64.929	0.167	0.266	0	-
Equal diversification	4	139.174	-65.587	0.215	-	0	-
Null	3	138.819	-66.409	0.215	-	0	-

mtDNA

- H. alimena* ●
- H. anomala* ●
- H. antevorta* ●
- H. anthedon* ●
- H. antilope* ●
- H. bartelotti* ●
- H. bolina* ●
- H. deceptor* ●
- H. deois* ●
- H. dexithea* ●
- H. dimona* ●
- H. dinarcha* ●
- H. diomea* ●
- H. errabunda* ●
- H. exiguus* ●
- H. inopinata* ●
- H. mechowi* ●
- H. misippus* ●
- H. monteironis* ●
- H. octocula* ●
- H. pandarus* ●
- H. pithoeka* ●
- H. salmacis* ●
- H. saundersi* ●
- H. sumbawana* ●
- H. usambara* ●

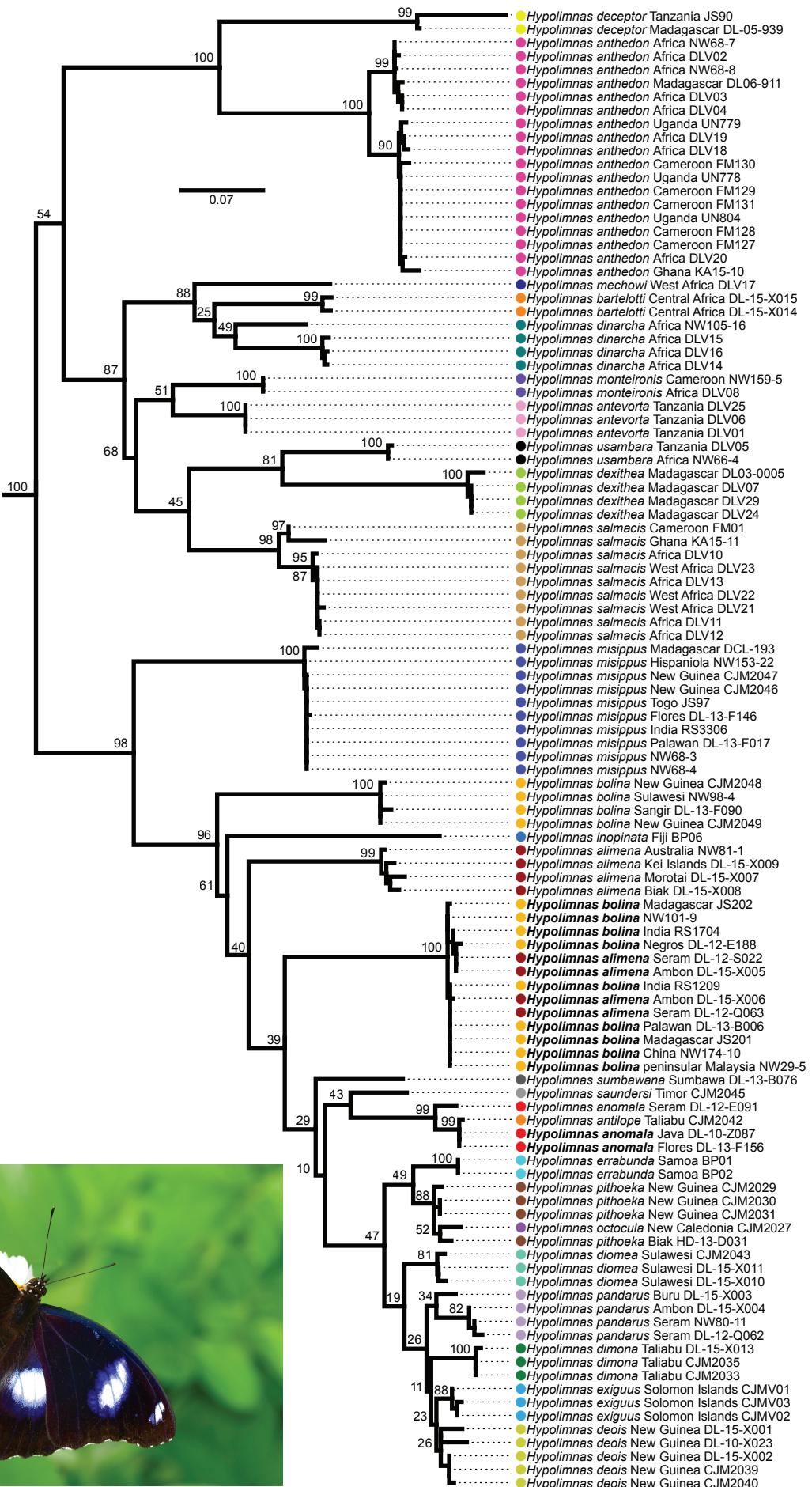


Figure A1. Maximum likelihood tree based on the mitochondrial COI locus (1475 bp). Colors indicate species and values at select nodes indicate bootstrap support. Samples in bold were infected with *Wolbachia*. Inset: *Hypolimnas bolina* male in Singapore (photo by Khew Sin Khoon and used with permission).

nDNA

- H. alimena* ●
- H. anomala* ●
- H. antevorta* ●
- H. anthedon* ●
- H. antilope* ●
- H. bartelotti* ●
- H. bolina* ●
- H. deceptor* ●
- H. deois* ●
- H. dexithea* ●
- H. dimona* ●
- H. dinarcha* ●
- H. diomea* ●
- H. errabunda* ●
- H. exiguum* ●
- H. inopinata* ●
- H. mechowi* ●
- H. misippus* ●
- H. monteironis* ●
- H. octocula* ●
- H. pandarus* ●
- H. pithoeka* ●
- H. salmacis* ●
- H. saundersi* ●
- H. sumbawana* ●
- H. usambara* ●

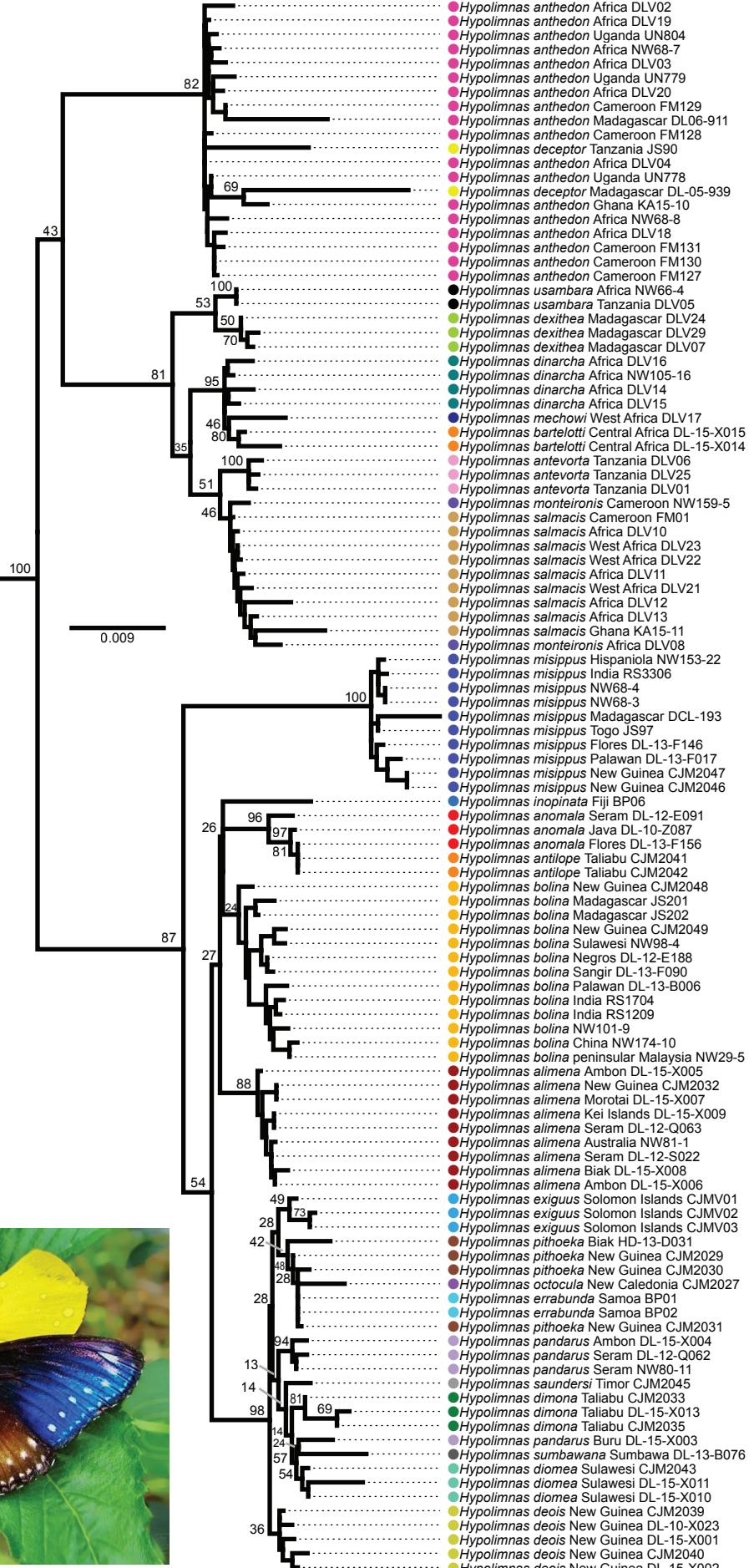


Figure A2. Maximum likelihood tree based on the nuclear dataset (5538 bp). Colors indicate species and values at select nodes indicate bootstrap support. Inset: *Hypolimnas anomala* female in Panay, Philippines (photo by Shekainah D. Alaban and used with permission).

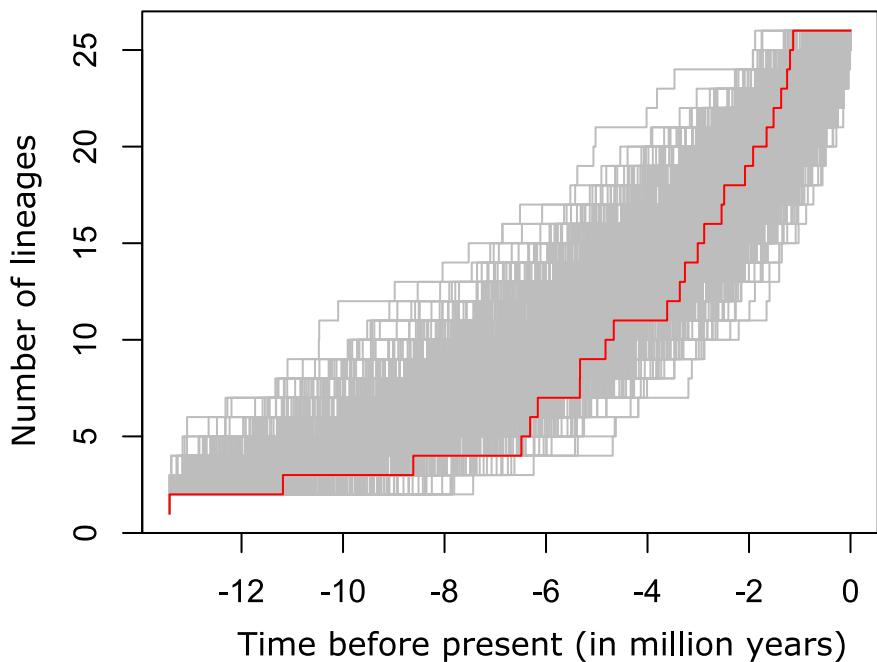


Figure A3. Lineage Through Time (LTT) graph showing the empirical LTT (in red) plot superimposed on 1000 LTT plots (in grey) simulated under a pure birth model.

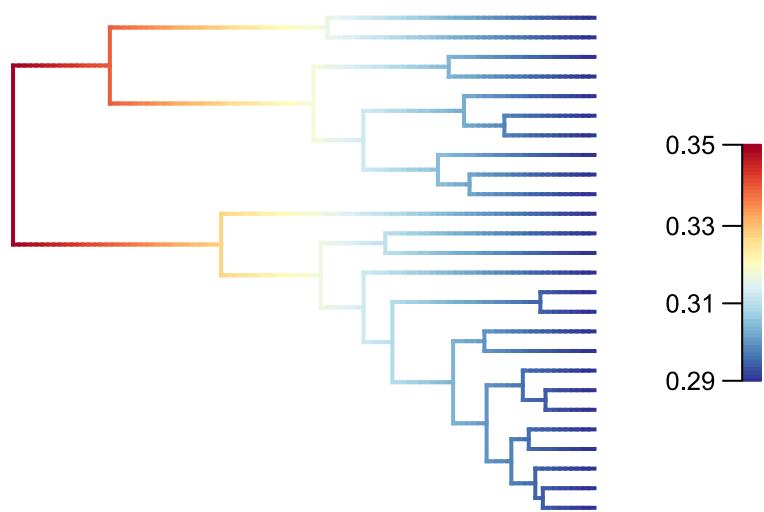


Figure A4. The best tree configuration from BAMM analysis indicating diversification rate (colour coded) across lineages.

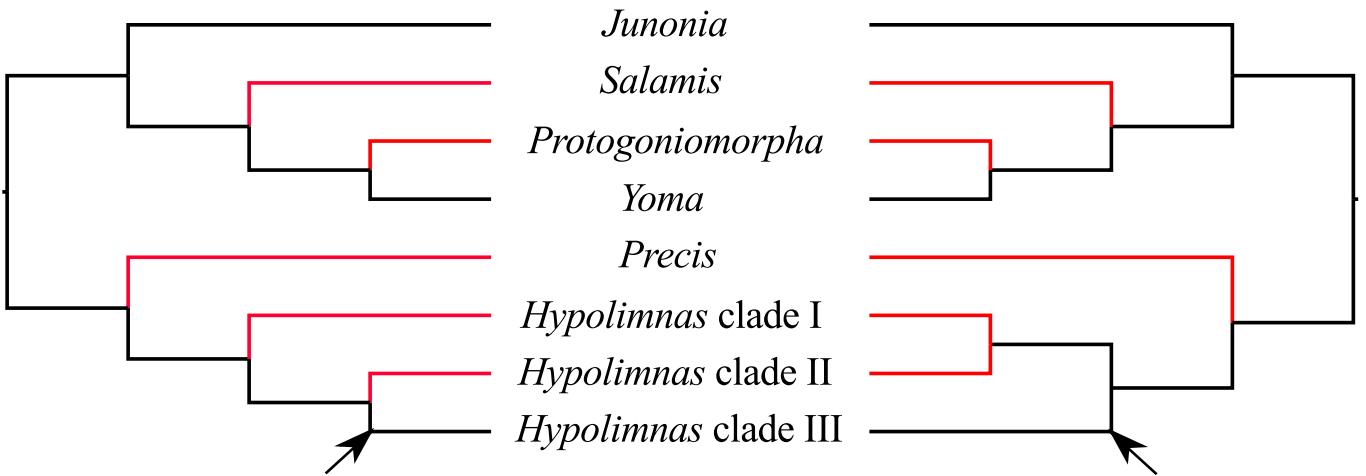


Figure A5. Topological uncertainty within *Hypolimnas* does not affect inference that the genus originated and diversified in Africa before one lineage dispersed east and radiated. Relationships among genera in the tribe Junoniini were consistent between the studies of Wahlberg et al. (2009) and Su et al. (2017). However, there is not strong support for relationships among the three earliest diverging *Hypolimnas* clades recovered by our analyses. The genera *Salamis*, *Protagoniomorpha*, *Precis*, and *Hypolimnas* clades I and II are restricted to the Afrotropics (marked in red). *Junonia* likely originated in Afrotropics and dispersed to all other tropical areas. *Yoma* is restricted to Southeast Asia and Australasia. Species in *Hypolimnas* clade III are mostly restricted to the Oriental and Australasian regions, but two species in this clade are found across multiple geographical areas. The arrow indicates possible dispersal of *Hypolimnas* from Africa to Asia.

mtDNA

- H. alimena* ●
- H. anomala* ●
- H. antevorta* ●
- H. anthedon* ●
- H. antilope* ●
- H. bartelotti* ●
- H. bolina* ●
- H. deceptor* ●
- H. deois* ●
- H. dexithea* ●
- H. dimona* ●
- H. dinarcha* ●
- H. diomea* ●
- H. errabunda* ●
- H. exiguis* ●
- H. inopinata* ●
- H. mechowi* ●
- H. misippus* ●
- H. monteironis* ●
- H. octocula* ●
- H. pandarus* ●
- H. pitheoka* ●
- H. salmacis* ●
- H. saundersi* ●
- H. sumbawana* ●
- H. usambara* ●



Figure A6. Maximum likelihood tree of mitochondrial COI locus including additional sequences of *H. bolina* from Genbank; accession numbers are indicated at the terminal branches along with species name and collection locality. Values at nodes indicate bootstrap support. *Hypolimnas misippus* female from Negros, Philippines (photo by Romana Pahilanga Delos Reyes and used with permission).