SUPPLEMENTARY MATERIAL

Reinforcement alone does not explain increased reproductive isolation in sympatry

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FIGURE S1. 4PL regression parameters for *Drosophila*. The density plots correspond to *a* (left), *b* (middle), and *d* (right) for premating isolation (top) and postzygotic isolation (bottom). Blue: sympatric; red: allopatric. The values of *c*, the fourth parameter of the regression, are shown in Figure 1.

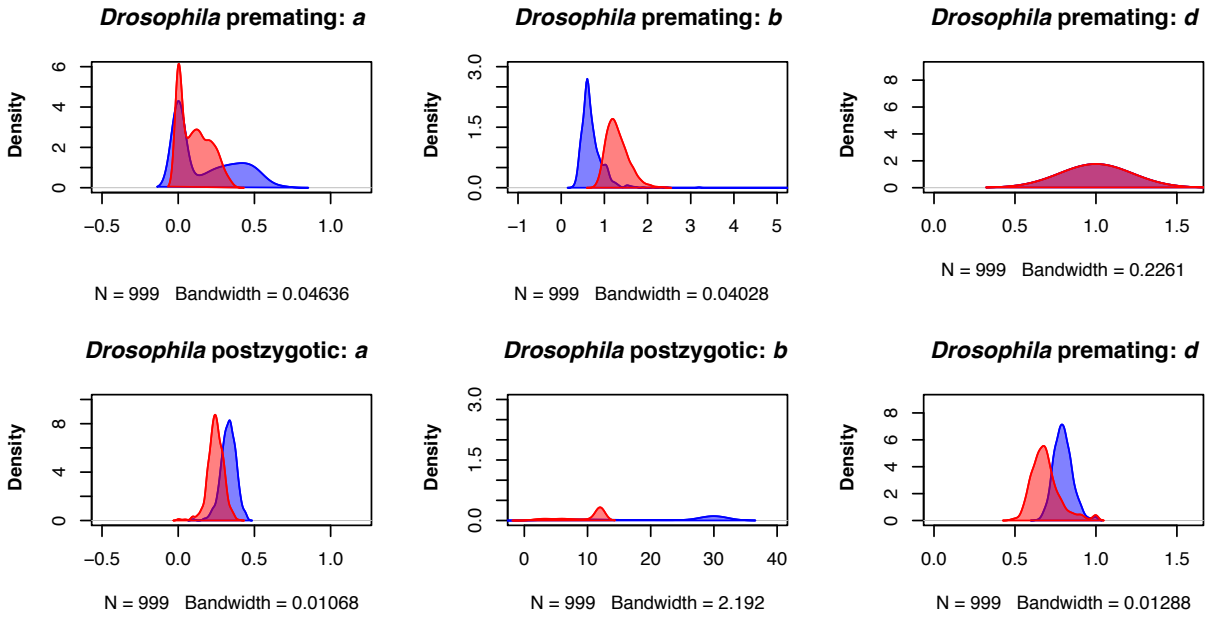


FIGURE S2. *Drosophila* 4PL regression parameters after phylogenetic subsamplings at the species-group level. The density plots correspond to *a* (left), *b* (middle left), *c* (middle right), and *d* (right) for premating isolation (top) and postzygotic isolation (bottom). Blue: sympatric; red: allopatric.

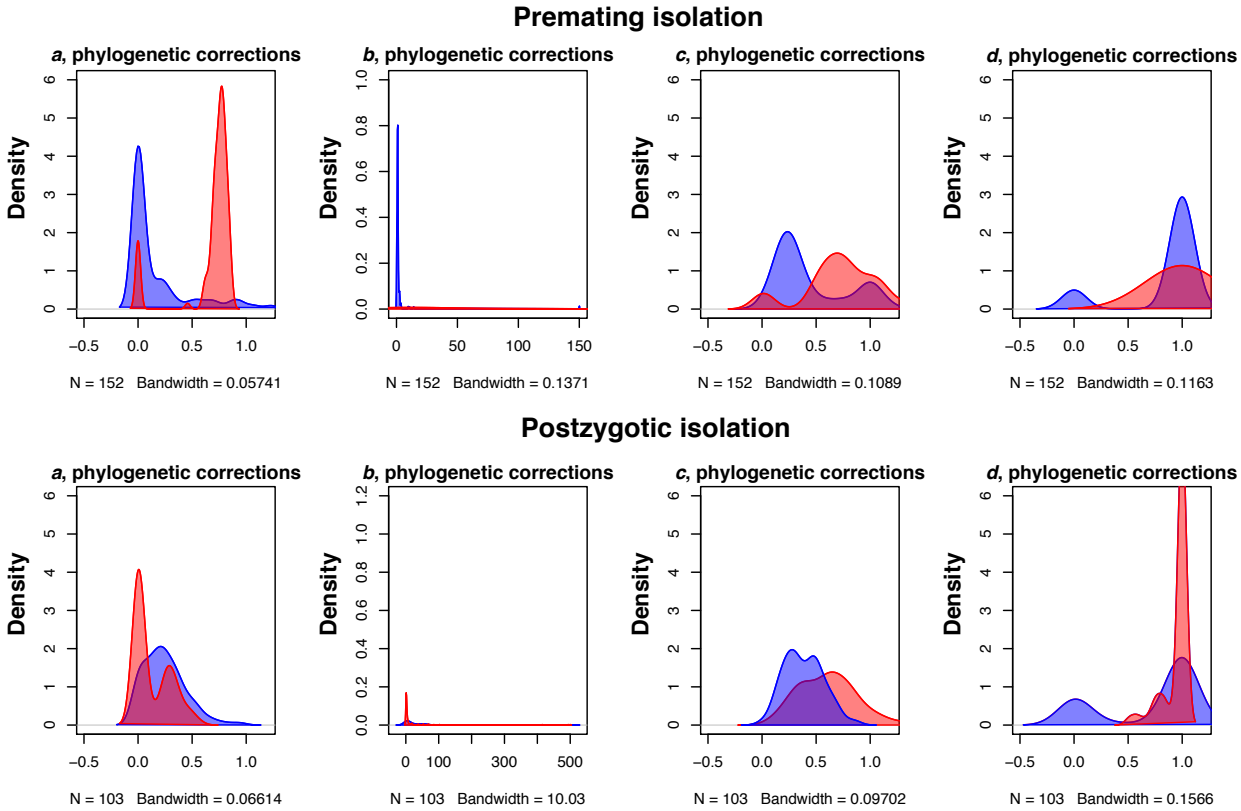


FIGURE S3. 4PL regression parameters for Lepidopterans and *Bufo* frogs. The density plots correspond to *a* (left), *b* (middle), and *d* (right). Blue: sympatric; red: allopatric. The values of *c*, the fourth parameter of the regression, are shown in Figure 2.

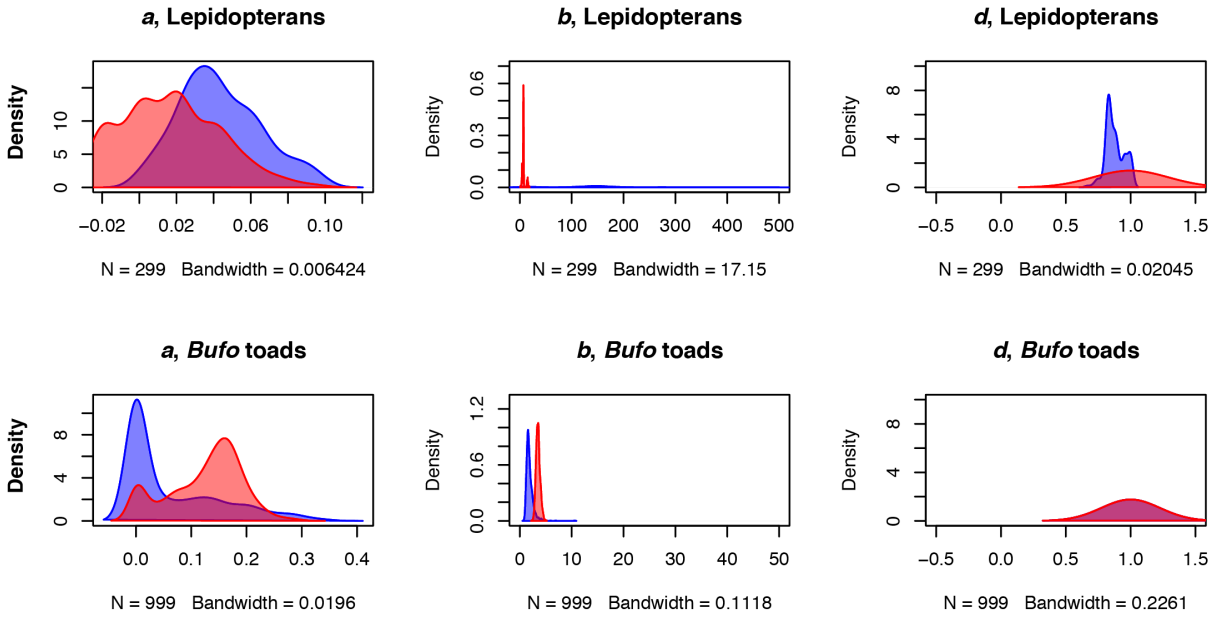


FIGURE S4. Comparisons of RI depend greatly on divergence time. Wilcoxon tests comparing the strength of premating (black) and postzygotic (purple) RI in sympatry and allopatry when using different thresholds to define young species. The solid horizontal line indicates the threshold of significance defined by Coyne and Orr (1, 2). Shaded intervals represent 95% confidence intervals.

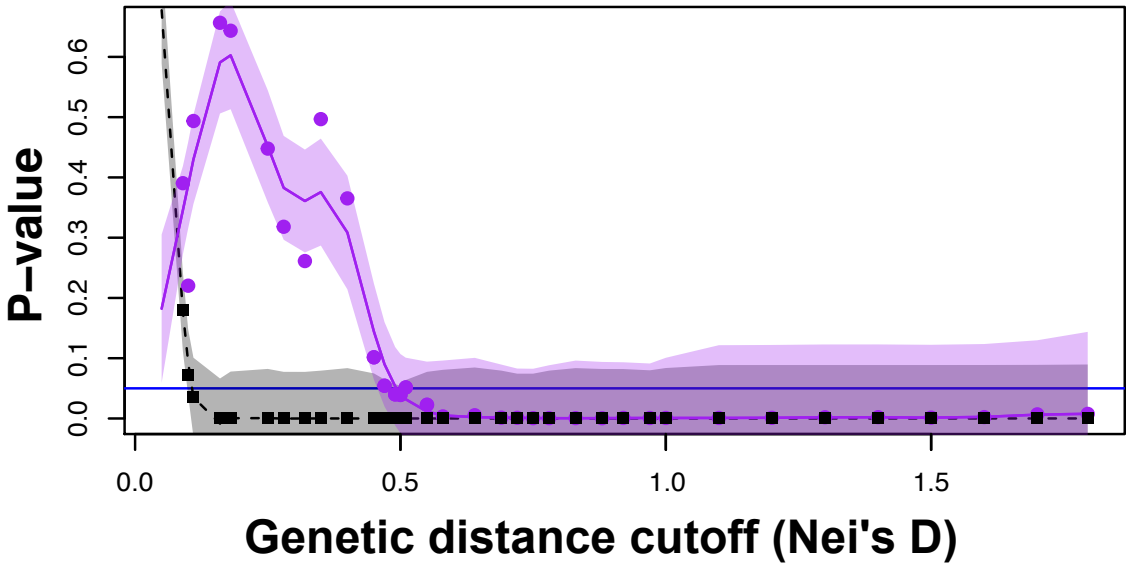


FIGURE S5. Postzygotic isolation evolves faster in sympatric species than in allopatric species when the data set is restricted to 'young' species. For these analyses we defined young as the species with the third lowest divergence time (i.e., Nei's $D < 0.5$ for *Drosophila* and NJ distance < 0.05 for *Bufo)*. 4PL regressions in the youngest *Drosophila* (Nei's $D < 0.51$) and youngest *Bufo* species (Nei's $D < 0.51$) show similar results to the full range of divergence. In both cases (young species and full divergence), sympatric species pairs show an earlier inflection point. Note that we did not do the same analyses for Lepidopterans as the 4PL regressions did not converge in datasets that did not include all the data.

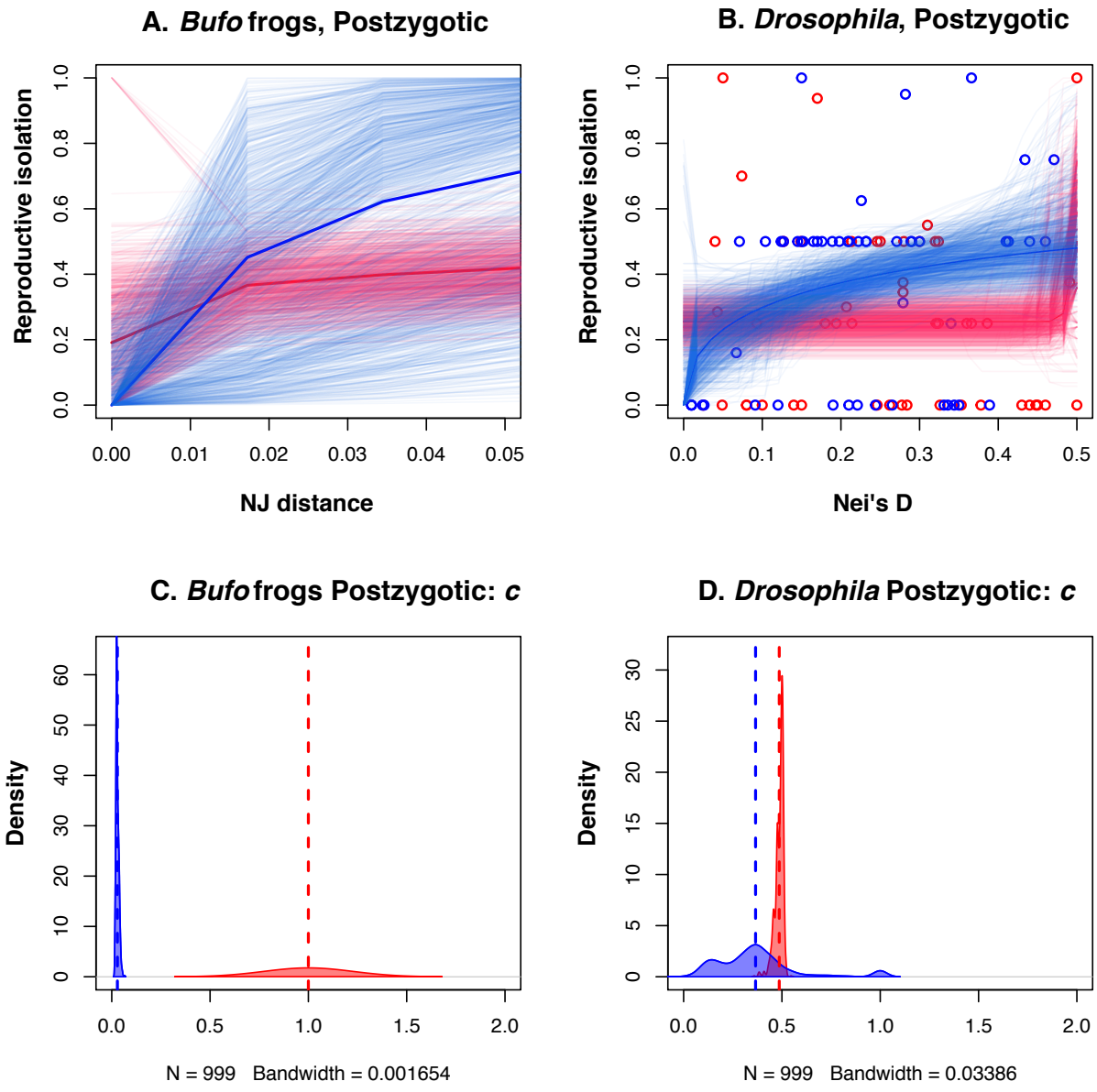


FIGURE S6. **The association between premating and postzygotic asymmetries is contingent on the way that data are filtered in *Drosophila*.** The *y*-axis shows the significance (*P*-value) of a Wilcoxon rank sum test with continuity correction. **A.** Correlation between premating and postzygotic isolation when we used a lower threshold (i.e., including data from the lower threshold to the maximum of the range) for allopatric (red) and sympatric (blue) species. **B.** Correlation between premating and postzygotic isolation when we used a upper threshold (i.e., including data from 0 to the upper threshold) for allopatric (red) and sympatric (blue) species.

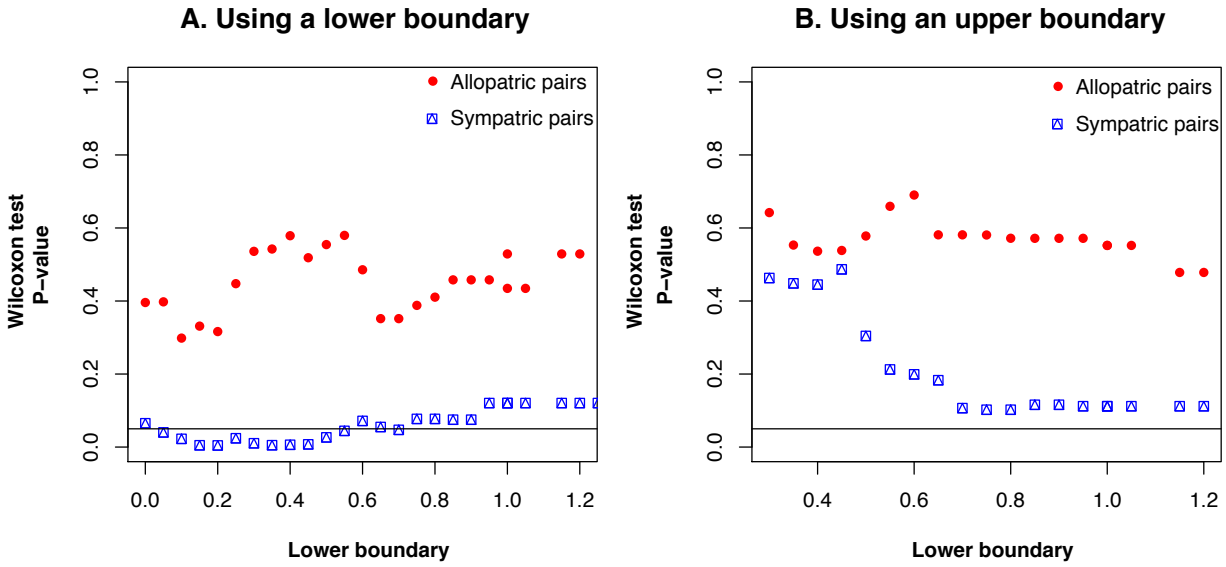


FIGURE S7. Mean premating and postzygotic RI are higher in sympatry in *Drosophila* but the ranges are overlapping. Premating (**A**) and postzygotic (**B**) RI in sympatry (S) is a subset of the range in allopatry (A). This result for premating (**C**) and postzygotic (**D**) RI is independent of the arbitrary threshold chosen to define young species. Similar plots for postzygotic RI in Lepidopterans and *Bufo* are shown in Figure S8.

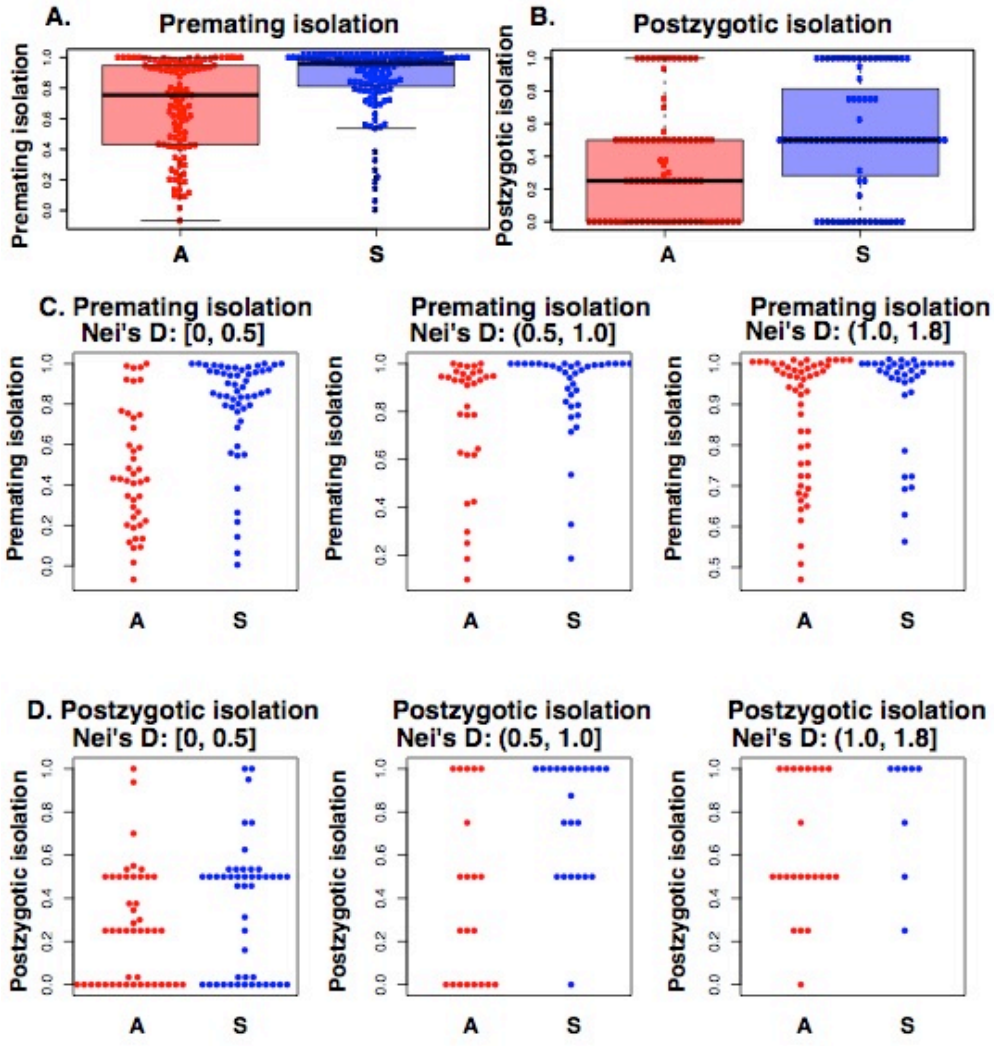


FIGURE S8. The range of postzygotic isolation in sympatry is similar to that in allopatry in Lepidopterans and in *Bufo* toads. Regardless of the cutoff of genetic distance, the range of postzygotic isolation usually overlaps with the range of RI in allopatry (highly diverged Lepidopterans being an exception). Top panels: Lepidopterans; lower panels: *Bufo* frogs.

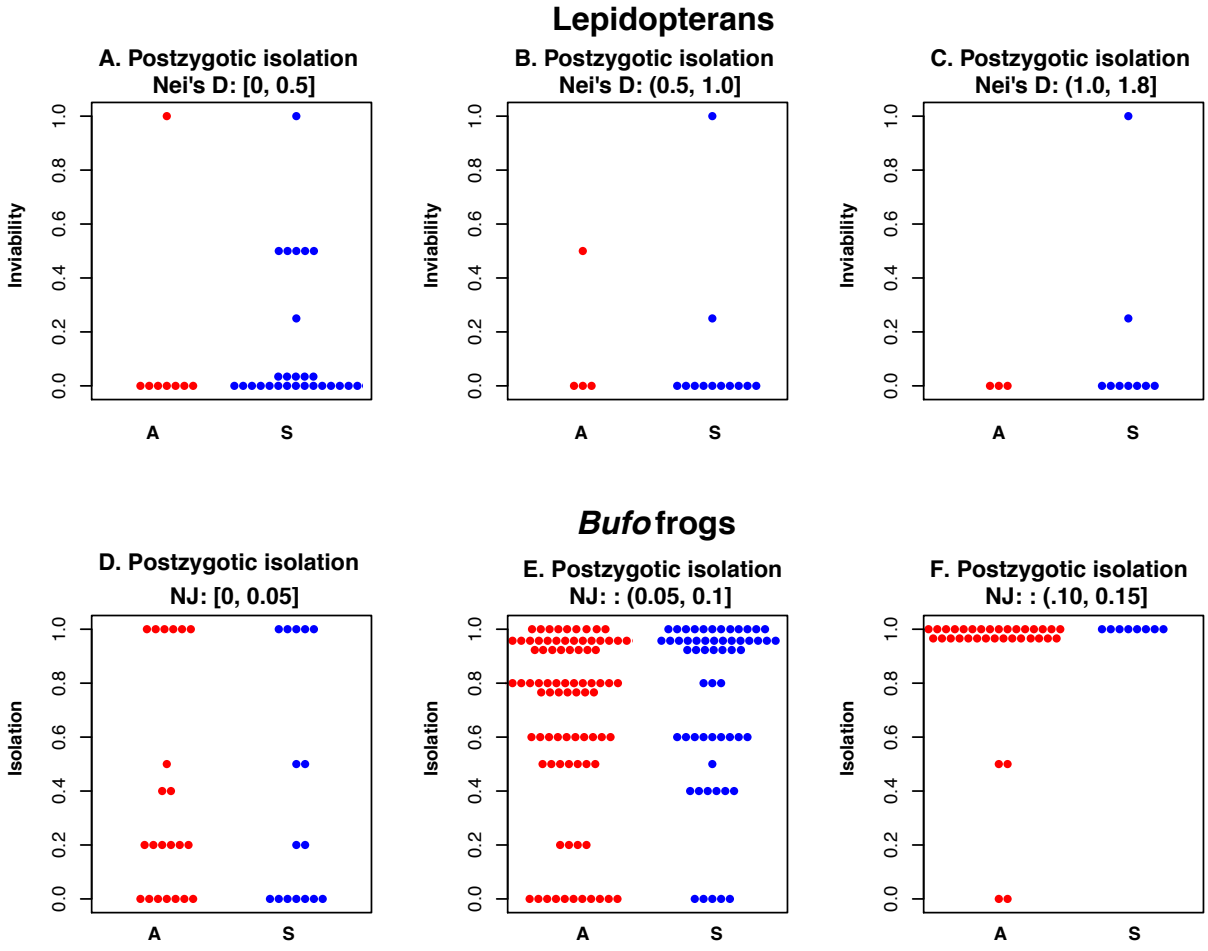
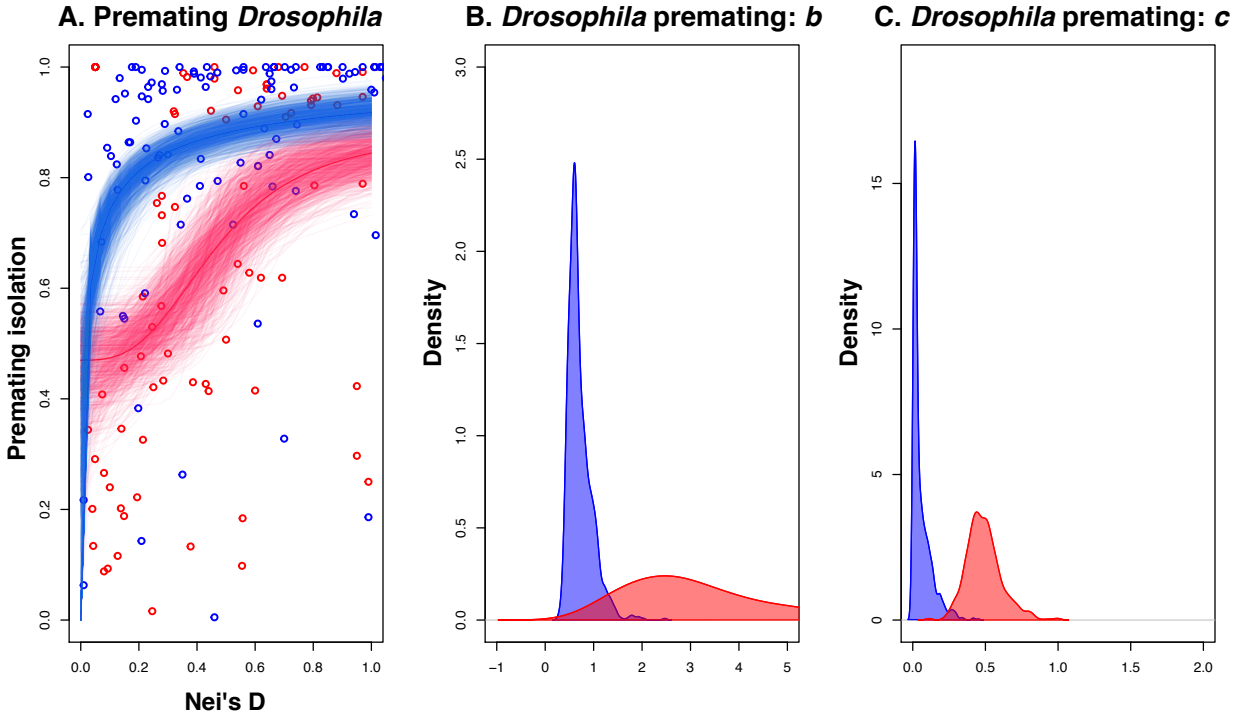


FIGURE S9. Premating isolation accumulates faster in sympatric than in allopatric *Drosophila* species, even after including previously estimated metrics of premating isolation in allopatric population pairs of the same species. These estimates are similar to the ones shown in Figures 1 and S1 but include metrics of isolation provided in (3–5).



SUPPLEMENTARY TABLES

TABLE S1. 4PL models fit the data better than linear regressions. Akaike information criterion for each fitted model.

Taxon	Barrier	Linear	Logistic	4PL
Drosophila	**Premating. Allopatric**	3.111	7.167	-19.557
Drosophila	**Premating. Sympatric**	-44.350	-45.059	-53.562
Drosophila	**Postzygotic. Allopatric**	53.975	53.383	50.630
Drosophila	**Postzygotic. Sympatric**	36.043	138.178	30.949
Lepidopterans	**Postzygotic. Allopatric**	-13.427	-16.391	-19.366
Lepidopterans	**Postzygotic. Sympatric**	-11.07777	5.3061	-44.391

TABLE S2. Pairwise comparisons between coefficients of the four-parameter logistic model between sympatric and allopatric species. To assess the significance of the difference between coefficients we compared bootstrapped distributions using a Wilcoxon test.

	Coefficient	$Coef_{sympatric}$	$Coef_{allopatric}$	Wilcoxon test
Premating, *Drosophila*	*a*	0.184 (0.211)	0.115 (0.102)	*W* = 468,480; *P* = 0.015
Premating, *Drosophila*	*b*	0.740 (0.517)	1.316 (0.270)	*W* = 949,860; *P* < 0.0001
Premating, *Drosophila*	*c*	0.059 (0.083)	0.318 (0.085)	*W* = 978,080; P < 0.0001
Premating, *Drosophila*	*d*	1 (0)	1 (0)	*W* = 499,000; *P* = NA
Postzygotic, *Drosophila*	*a*	0.329 (0.049)	0.242 (0.050)	*W* = 93,010; *P* < 0.0001
Postzygotic, *Drosophila*	*b*	22.843 (9.801)	9.402 (3.532)	*W* = 164,870; *P* < 0.0001
Postzygotic, *Drosophila*	*c*	0.439 (0.044)	0.751 (0.114)	*W* = 993,050; *P* < 0.0001
Postzygotic, *Drosophila*	*d*	0.799(0.061)	0.682 (0.091)	*W* = 126,760; *P* < 0.0001
Postzygotic, Lepidopterans	*a*	0.015 (0.025)	0.043 (0.021)	*W* = 17155; *P* < 0.0001
Postzygotic, Lepidopterans	*b*	7.252 (2.955)	222.696 (426.678)	*W* = 1,189; *P* < 0.0001
Postzygotic, Lepidopterans	*c*	1 (0)	0.686 (0.012)	*W* = 89,401; *P* < 0.0001
Postzygotic, Lepidopterans	*d*	1(0)	0.872 (0.069)	*W* = 85,364; *P* < 0.0001
Postzygotic,	*a*	0.062 (0.087)	0.124 (0.065)	*W* = 739,185;

***Bufo* toads**				$P < 0.0001$
Postzygotic, *Bufo* toads	*b*	1.854(0.699)	3.520 (0.414)	$W = 971{,}916$; $P < 0.0001$
Postzygotic, *Bufo* toads	*c*	0.028 (0.008)	0.038 (0.003)	$W = 877{,}636$; $P < 0.0001$
Postzygotic, *Bufo* toads	*d*	1 (0)	1 (0)	$W = 499{,}000$, P = NA

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TABLE S3. Coefficients for a phylogenetically informed linear regression for premating and postzygotic isolation between *Drosophila* species pairs. The confidence intervals are based in 5 different chains.

Isolation	Coefficient	Isolation mean	95% CI	Effective sample size	pMCMC
Premating	(Intercept)	0.417	[0.253, 0.616]	888.760	<0.001
Premating	Genetic Distance	0.297	[0.179, 0.414]	11.710	<0.001
Premating	Sympatry (Sympatric-Allopatric)	0.364	[0.245, 0.464]	575.470	<0.001
Premating	Genetic Distance × Sympatry (Sympatric-Allopatric)	-0.184	[-0.323, -0.058]	1,000.00	0.014
Postzygotic	Intercept	0.214	[-0.018. 0.446]	910.500	0.084
Postzygotic	Genetic Distance	0.306	[0.087, 0.530]	704.400	0.004
Postzygotic	Sympatry (Sympatric-Allopatric)	0.034	[-0.121, 0.198]	1,000.00	0.660
Postzygotic	Genetic Distance × Sympatry (Sympatric-Allopatric)	0.388	[0.138, 0.699]	984.50	0.002

TABLE S4. Regression coefficients from a phylogenetically informed regression for postzygotic isolation between Lepidopteran and *Bufo* species pairs. The confidence intervals are based in 5 different chains.

Group		Postzygotic mean	95% CI	Effective sample size	pMCMC
Lepidopterans	(Intercept)	-0.100	[-0.392, 0. 273]	1,000.0	0.488
Lepidopterans	Genetic Distance	0.906	[0.670, 1.125]	892.2	<0.001
Lepidopterans	Sympatry (Sympatric-Allopatric)	0.123	[-0.018, 0.257]	1,000.0	0.076
Lepidopterans	Genetic Distance × Sympatry (Sympatric-Allopatric)	-0.382	[-0.674, -0.093]	1,000.0	0.014
***Bufo* frogs**	(Intercept)	0.502	[0.294, 0.691]	1,000.0	<0.001
***Bufo* frogs**	Genetic Distance	4.849	[3.668, 5.950]	1,000.0	<0.001
***Bufo* frogs**	Sympatry (Sympatric-Allopatric)	-0.161	[-0.272, -0.032]	1,000.0	0.006
***Bufo* frogs**	Genetic Distance × Sympatry (Sympatric-Allopatric)	1.938	[0.486, 3.637]	1,000.0	0.012

TABLE S5. Recently diverged species show faster accumulation of postzygotic isolation when they are sympatric than when they are allopatric in *Bufo* frogs. Wilcoxon tests comparing the strength of postzygotic isolation in sympatry and allopatry when using different thresholds to define young species in lepidopterans and *Bufo* frogs. NC: regression did not converged and the analysis was not possible. Figure 3 shows similar results for premating and postzygotic isolation in *Drosophila.*

Group	Cutoff	c_{symp}	c_{allop}	*W*	*P*-value
Lepidopterans	0.5	NC	NC	—	—
Lepidopterans	1	NC	NC	—	—
***Bufo* frogs**	0.05	0.028	>0.05	995,004	<0.0001
***Bufo* frogs**	0.06	0.028	>0.06	946,053	<0.0001
***Bufo* frogs**	0.07	0.028	0.037	816,440	<0.0001
***Bufo* frogs**	0.08	0.029	>0.08	971,028	<0.0001
***Bufo* frogs**	0.09	0.028	0.037	839,586	<0.0001
***Bufo* frogs**	0.10	0.029	0.038	868,986	<0.0001
***Bufo* frogs**	0.11	0.029	0.038	869,901	<0.0001
***Bufo* frogs**	0.12	0.028	0.038	871,175	<0.0001

TABLE S6. Linear models show that the overlap of geographic range significantly affects the magnitude of premating and postzygotic isolation between *Drosophila* species pairs.

Response (type of barrier)	Model	Effect	F-value	P-value
Premating	Factorial	Genetic distance (Nei's D)	57.187	7.171×10^{-13}
Premating	Factorial	% Sympatry	50.809	1.044×10^{-11}
Premating	Factorial	Genetic distance × % Sympatry	3.982	0.047
Premating	No-interaction	Genetic distance (Nei's D)	56.528	9.338×10^{-13}
Premating	No-interaction	% Sympatry	50.224	1.329×10^{-11}
Postzygotic	Factorial	Genetic distance (Nei's D)	28.259	6.097×10^{-7}
Postzygotic	Factorial	% Sympatry	14.283	2.625×10^{-4}
Postzygotic	Factorial	Genetic distance × % Sympatry	2.380	0.126
Postzygotic	No-interaction	Genetic distance (Nei's D)	27.892	6.98×10^{-7}
Postzygotic	No-interaction	% Sympatry	14.098	2.85×10^{-4}

TABLE S7. The effect of geographic overlap on the effect of the magnitude of premating and postzygotic RI is mostly driven by species with disjoint geographic ranges. For each overlap threshold, we fit a linear model in which the strength of isolation, either premating or postzygotic, depends on genetic distance and the percentage of overlap (% sympatry).

		Nei's *D*		% sympatry	
RI type	**Lower threshold (not included)**	***F*-value**	***P*-value**	***F*-value**	***P*-value**
Premating	All data	56.528	9.338×10^{-13}	50.22	1.329×10^{-11}
Premating	0.00	10.873	1.268×10^{-3}	7.022	9.082×10^{-3}
Premating	0.01	10.873	1.268×10^{-3}	7.022	9.082×10^{-3}
Premating	0.02	10.450	1.569×10^{-3}	7.487	7.121×10^{-3}
Premating	0.03	8.946	3.36×10^{-3}	6.704	0.011
Premating	0.04	9.158	3.030×10^{-3}	8.601	4.026×10^{-3}
Premating	0.05	9.443	2.637×10^{-3}	3.242	0.074
Premating	0.10	9.920	2.101×10^{-3}	0.095	0.758
Postzygotic	All data	27.892	6.98×10^{-7}	14.098	2.85×10^{-3}
Postzygotic	0.00	16.626	1.632×10^{-4}	1.520	0.223
Postzygotic	0.01	16.626	1.632×10^{-4}	1.520	0.223
Postzygotic	0.02	16.626	1.632×10^{-4}	1.520	0.223
Postzygotic	0.03	14.006	4.788×10^{-4}	1.459	0.233
Postzygotic	0.04	14.274	4.443×10^{-4}	2.355	0.132
Postzygotic	0.05	14.263	4.549×10^{-4}	2.841	0.090
Postzygotic	0.10	12.629	9.371×10^{-4}	3.641	0.063

TABLE S8. Datasets that have compiled data on the magnitude of reproductive isolation in multiple species pairs of animals.

	N	Premating isolation	Postzygotic isolation	Distance	Geographic status	Reference
Drosophila	630	X	X	X	X	(6)
Toads	669		X	X	X	(7, 8)
Lepidoptera	212		X	X	X	(9)
Heliconius	12	X	X	X		(10)
Birds	407		X	X		(11)
Sticklebacks	5	X	X	X		(12)
Centrarchid fish	130		X	X		(13)