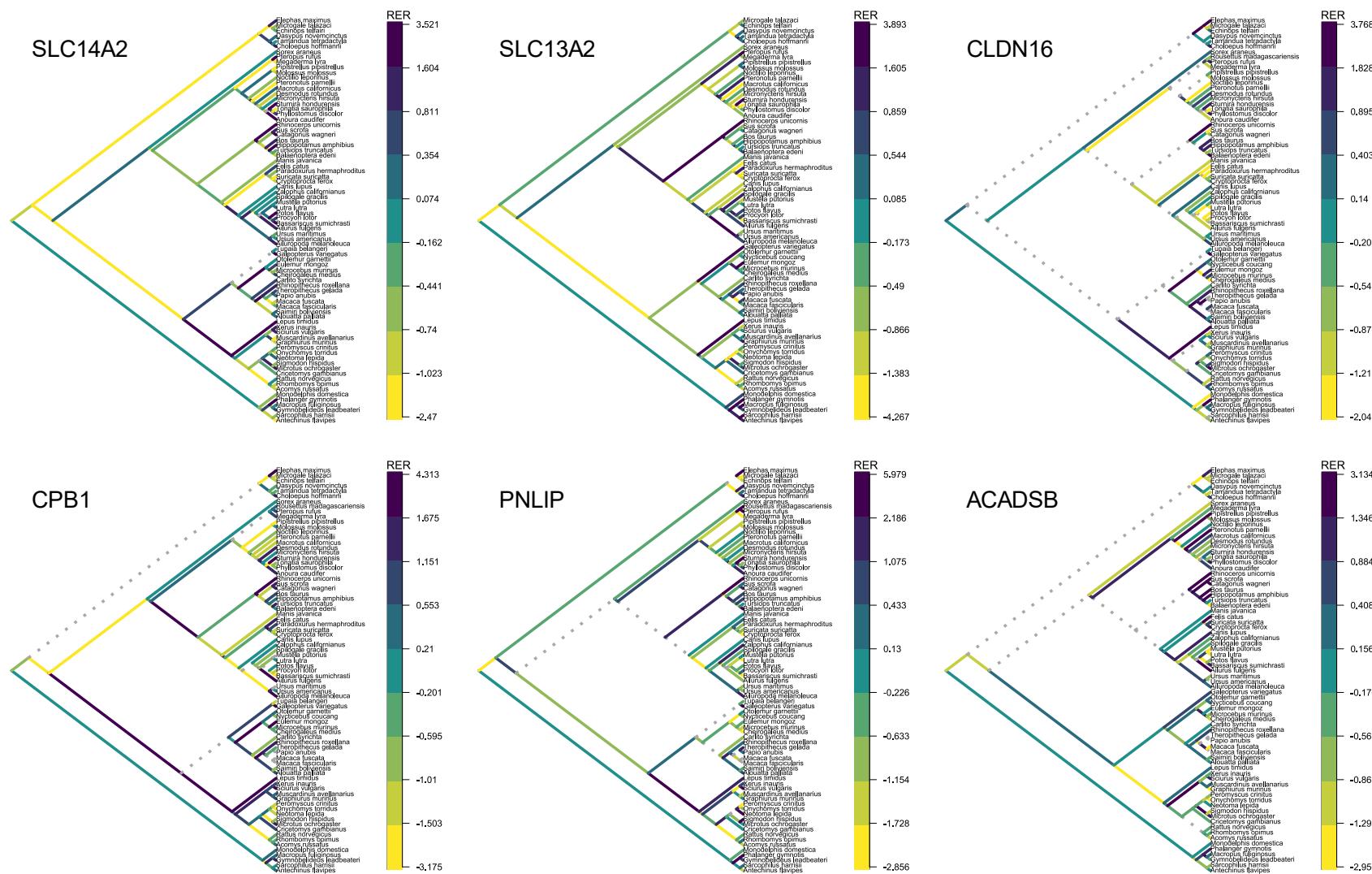
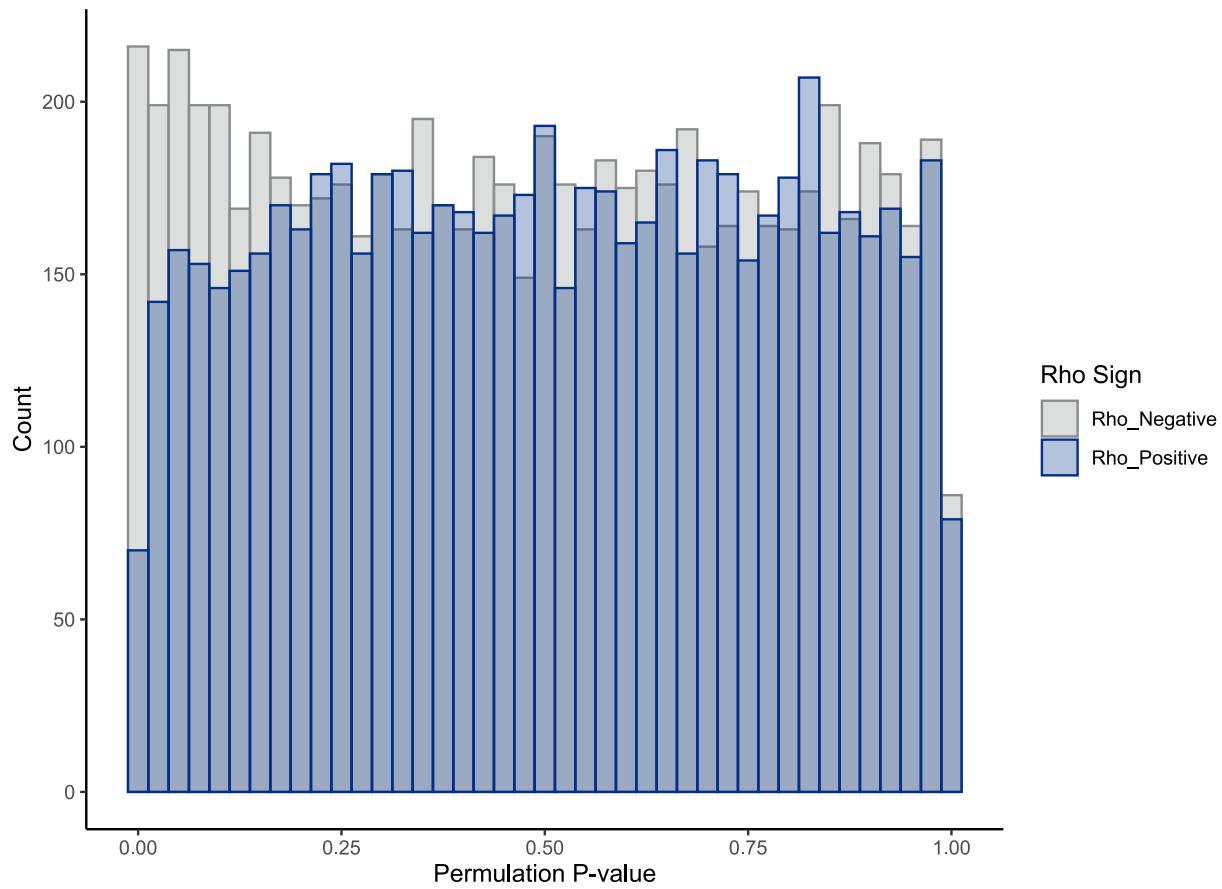


1 Supplemental Figures



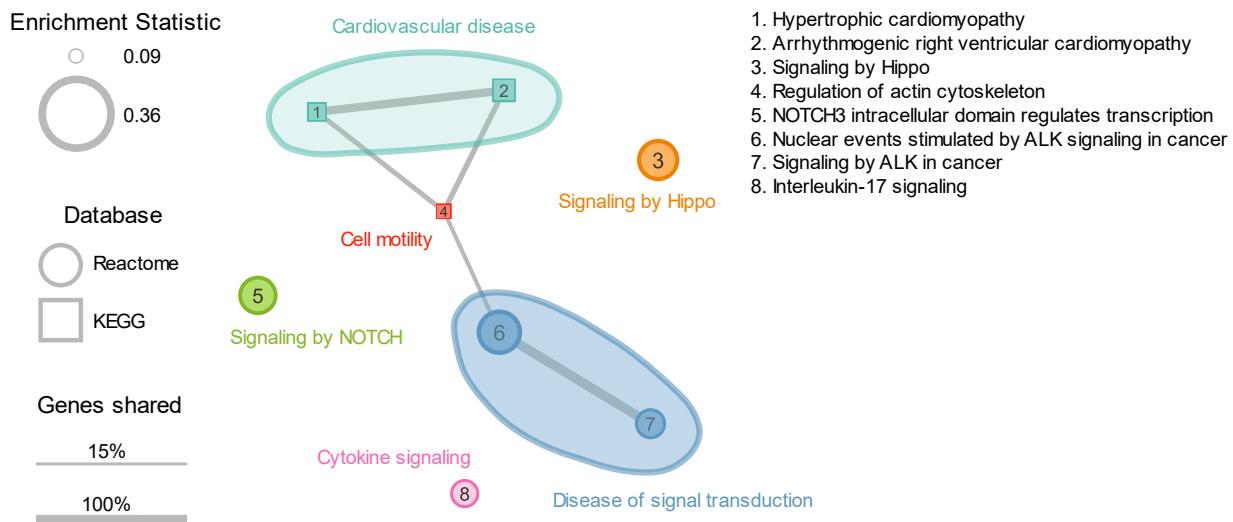
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Figure S1. Relative evolutionary rates (RERs) across the mammalian phylogeny for genes significantly associated with change in carnivory score. Branch colors represent the RER for a given gene in each part of the phylogeny. Purple and yellow branches indicate positive and negative RERs, respectively. Gray dashed lines represent branches that were excluded from our analyses due to default branch length filtering in RERconverge, which set the shortest 5% of branches across all gene trees to N/A. In each gene, positive and negative RERs are distributed throughout the phylogeny.



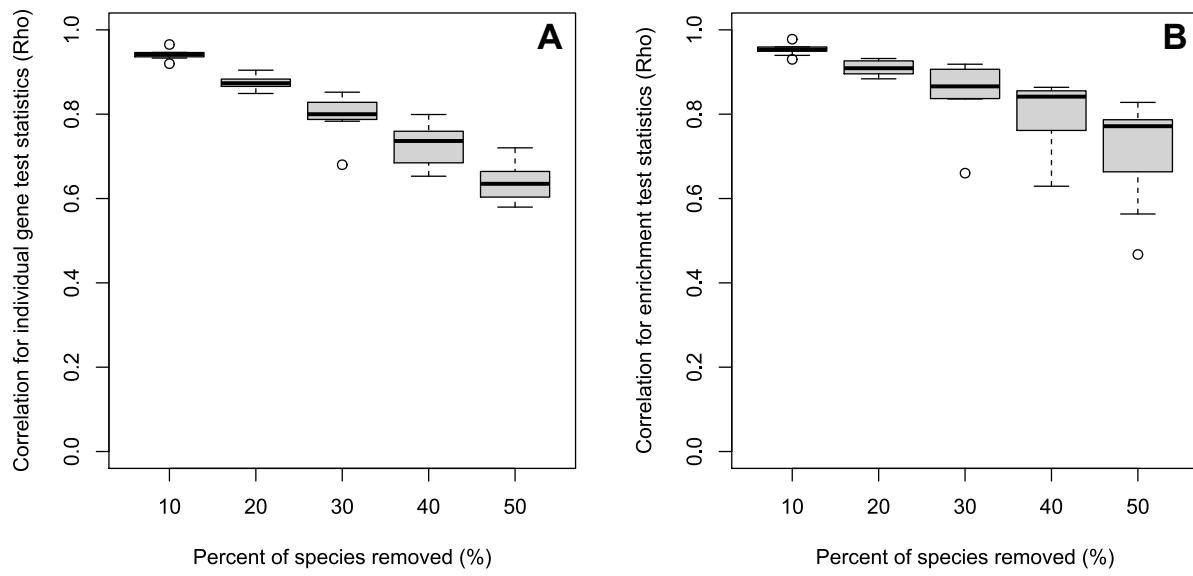
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9 **Figure S2.** Significant differences in the distribution of empirical, permutation-based *P*-values for
10 positive and negative associations between gene relative evolutionary rates (RERs) and carnivority
11 score (Mann-Whitney U test, AUC=0.523, $P=2.16 \times 10^{-6}$). Our analysis showed reduced power to
12 detect positive associations between RER and change in carnivority, as indicated by fewer low *P*-
13 values being generated during permutation for genes with a positive correlation.



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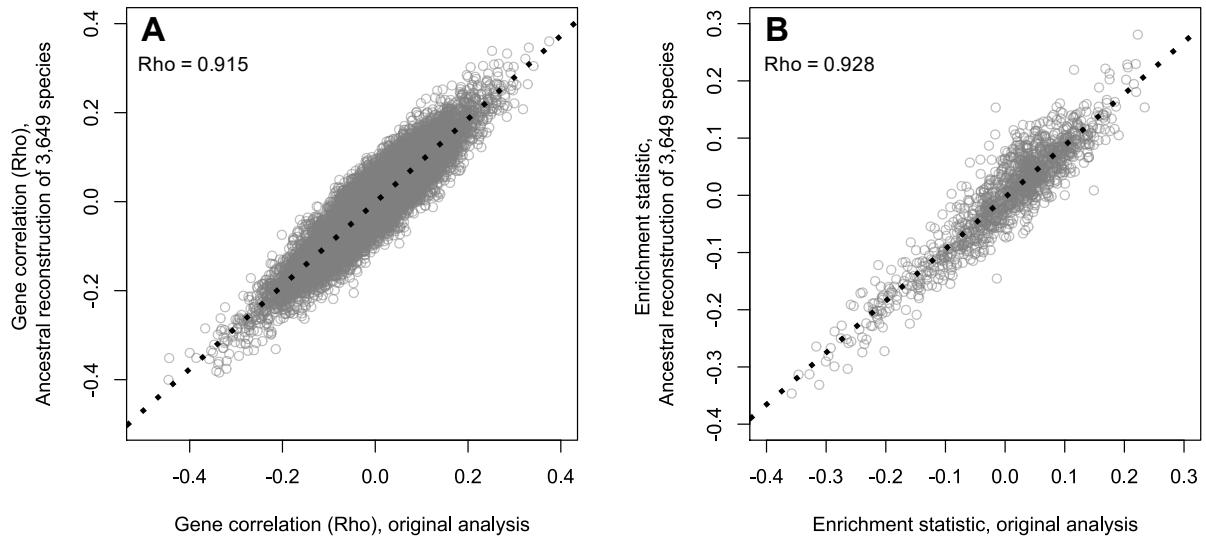
15 **Figure S3.** Significantly enriched gene pathways (n=8) for genes whose evolutionary rates are
 16 positively correlated with carnivoriness. Each circle or square represents a gene pathway. Circles
 17 and squares represent pathways from the Reactome and KEGG databases, respectively. The size
 18 of the shape represents the magnitude of the difference between the distribution of test statistics
 19 for genes in that pathway and the distribution for all other genes. Larger shapes, representing
 20 pathways with larger positive correlation statistics, indicate greater reductions in evolutionary
 21 constraint as carnivoriness score increases. The width of lines connecting pathways represents the
 22 proportion of shared genes in the smaller gene set. Colors represent the broad functional categories
 23 that the pathways occupy.



24

25 **Figure S4.** Correlations between RERconverge results using our full species list and using species
 26 subsets where 10, 20, 30, 40, or 50 percent of species have been randomly removed. For each
 27 percent of species removed, we created 10 random subsets. Boxes represent the distribution of
 28 correlation values (Pearson's correlation coefficient, rho) between the original results and the
 29 results with a given percent of species removed. (A) Correlations between gene correlation
 30 statistics. (B) Correlations between gene set enrichment statistics.

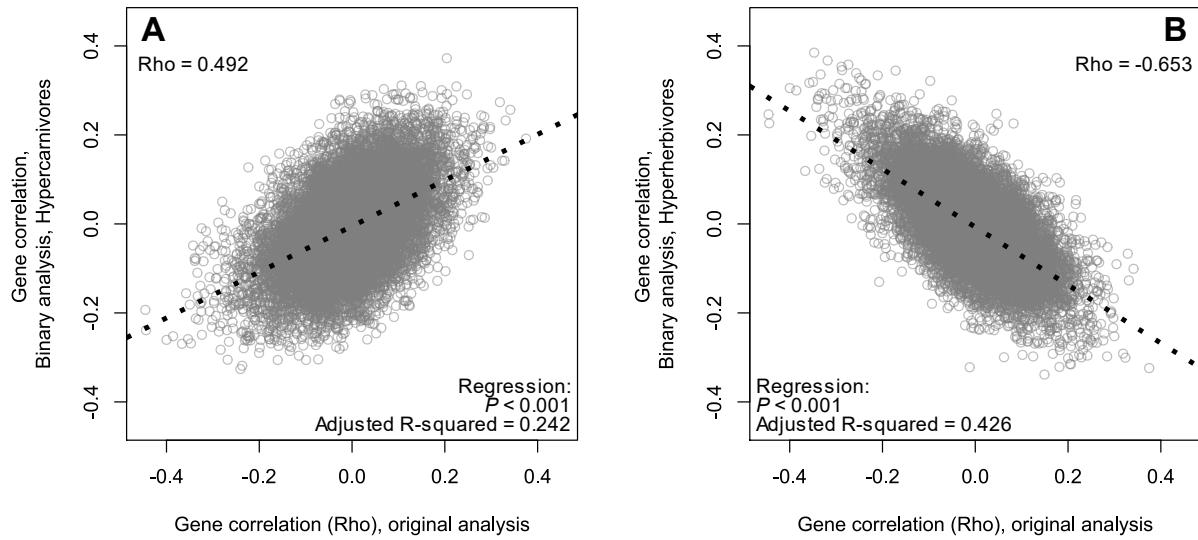
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33 **Figure S5.** Correlations between RERconverge results using the original ancestral state
 34 reconstruction method built into RERconverge and a modified reconstruction method. For the
 35 modified method, we performed fast estimation of maximum likelihood ancestral states (Revell
 36 2012) on a larger tree of 3,649 species for which we could obtain diet data from EltonTraits
 37 (Wilman et al. 2014). We then pruned this reconstruction to our study species and used it as input
 38 in the RERconverge analysis. Correlations were calculated using Pearson's correlation coefficient
 39 (rho). Dashed lines indicate the regression line. (A) Correlation between gene correlation statistics.
 40 (B) Correlation between gene set enrichment statistics.

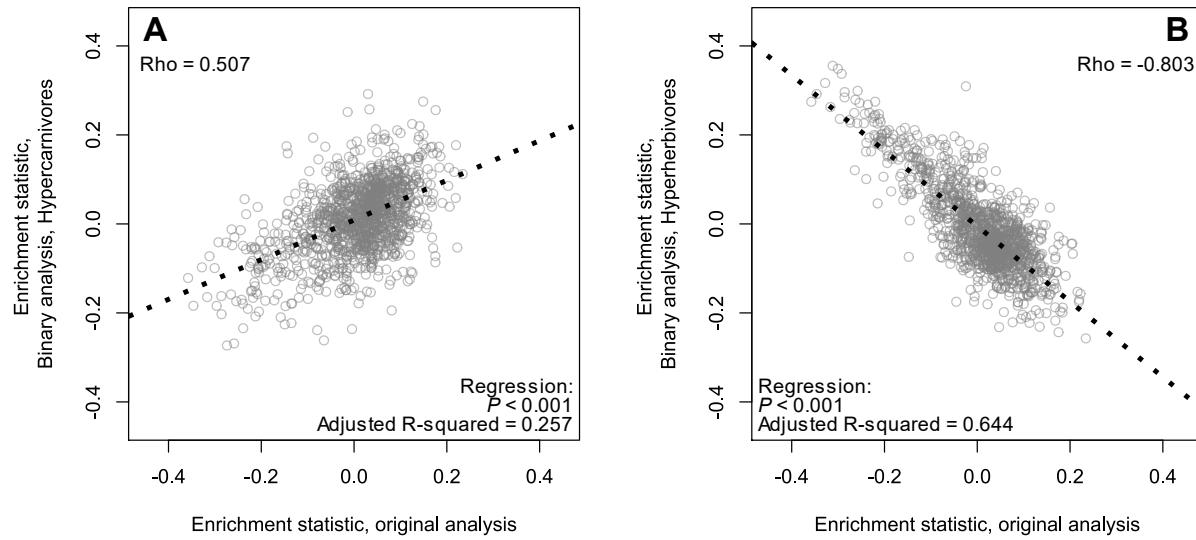
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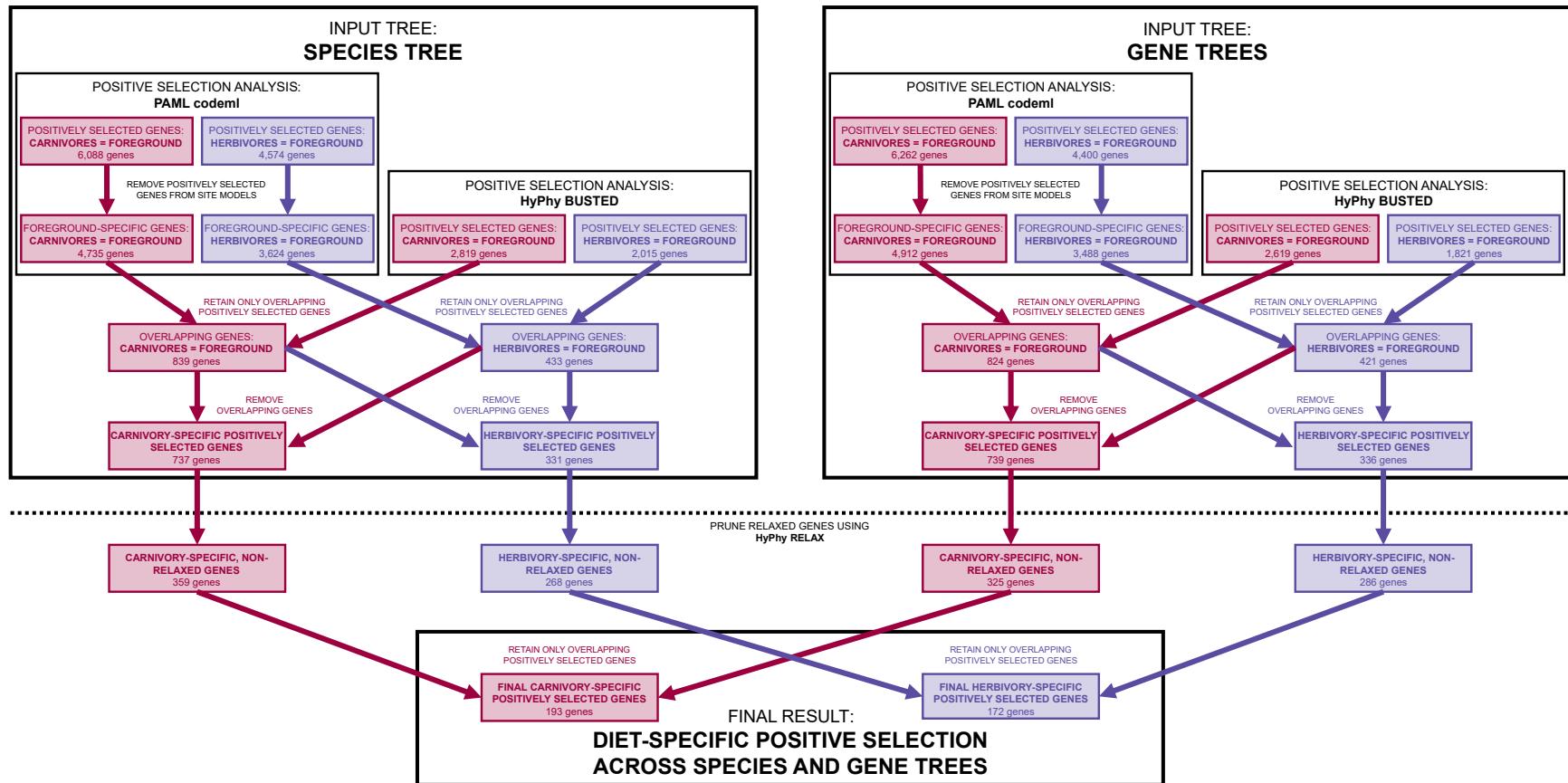
43 **Figure S6.** Correlations between the test statistics for the continuous and binary RER converge
 44 analyses. Inset rho values represent the Pearson correlation coefficient between the two analyses.
 45 Dashed lines indicate the regression line. (A) Correlation between the individual gene test statistics
 46 from the original continuous analysis and the binary analysis with hypercarnivores (carnivory
 47 scores ≥ 90) as foreground. (B) Correlation between the individual gene test statistics from the
 48 original continuous analysis and the binary analysis with hyperherbivores (carnivory scores ≤ 10)
 49 as foreground.

50



51

52 **Figure S7.** Correlations between the gene pathway enrichment statistics for the continuous and
 53 binary RERconverge analyses. Inset rho values represent the Pearson correlation coefficient
 54 between the two analyses. (A) Correlation between the enrichment statistics from the original
 55 continuous analysis and the binary analysis with hypercarnivores (carnivory scores ≥ 90) as
 56 foreground. (B) Correlation between the enrichment statistics from the original continuous analysis
 57 and the binary analysis with hyperherbivores (carnivory scores ≤ 10) as foreground.



60 **Figure S8.** Conservative strategy for generating final lists of positively selected genes associated with carnivorism and herbivorism in
61 mammals. We tested 15,117 genes for positive selection in carnivores (species with carnivorism scores ≥ 90) and herbivores (species with
62 carnivorism scores ≤ 10) separately, using both codeml from PAML (Yang 2007) and BUSTED from HyPhy (Murrell et al. 2015; Pond et
63 al. 2020). To account for the impact of gene tree discordance, we performed our positive selection analyses using both the species tree
64 (Upham et al. 2019) and gene trees inferred using RAxML (Stamatakis 2014). We only considered a gene to have experienced diet-
65 associated positive selection when it produced a significant result in both our codeml and BUSTED analyses, and if we got this result
66 using both the species and gene trees. As an additional filtering step, we tested the genes returned by codeml and BUSTED for relaxed
67 selection using RELAX (Wertheim et al. 2015) and excluded any genes showing significant relaxation in the foreground species from
68 our final gene lists. The final products of this process were two conservative lists representing positively selected genes in the most
69 carnivorous and herbivorous mammals, respectively (Tables S7, S8).

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