Supplementary Information for
Layered evolution of gene expression in “superfast” muscles for courtship

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Mitochondrial Assembly and Read Filtering. We detected a high proportion of mitochondrial or mitochondrial-like transcripts in our sequenced read set, so each individual's reads sets were de novo assembled using Trinity under default settings (version 2.9.1; 1). Fragments were aligned using BLASTn to the *Mionectes oleagineus* reference mitochondrial genome (NC_024682), a partial *L. coronata* genome, and a library of other species-specific available for the target species. High-scoring hits were then aligned to the *Mi*. oleagineus reference using MUSCLE (2) and draft consensus sequences for each individual were generated by consensus overlap using SeaView (3). Finally, trimmed reads from each individual were mapped to their individual-specific mitochondrial draft consensus genome using BWA-mem (v. 0.7.17-r1188; 4). Read alignments were analyzed using SAMtools mpileup, and a final consensus sequence was determined from the pileup by majority allele using a custom Python script. Read pairs from each of the 42 individual samples were mapped using BWA mem using default settings. Of trimmed read pairs in each sample 16.3–44.9% (mean 35.5%) mapped to their individual mitochondrial genomes. These mitochondrial-mapped reads were filtered out of the main reads set used in the later quantification and sequence analyses.

Transcriptome Mapping and Quantification. Non-mitochondrial reads for each sample were mapped using STAR (version 2.7.3a; 5) to the *Pipra filicauda* reference genome (version 1; GenBank: GCA_003945595.1) using single-pass mode with the reference GTF and all other parameters default. *M. vitellinus* and *L. coronata* genomes were also available, but we determined they were less complete than the *P. filicauda* genome and we obtained higher mapping rates using *P. filicauda* as a single standard reference genome space for all samples. The unmapped read pairs from STAR were then aligned using BWA-mem (v. 0.7.17-r1188; 4) using a slightly relaxed mismatch parameter (–B 2), split hits as secondary (–M), and all other command flags default. The reference *P. pipra* GTF file was filtered to remove snoRNA, miRNA, snRNA, guide RNAs, and pseudogenes. Read pairs were counted per gene using full gene coordinate boundaries using featureCounts (version 2.0.1; 6). An average of 95% of uniquely mapped read pairs were assigned uniquely to a gene. Quantification was repeated allowing fractional counting of multi-mapped reads and reads mapping to multiple features. However, the similarity of counts for all samples was high ($r^2 \approx 0.98$) and so we used the uniquely mapped reads table only. On average, 13.9 million read pairs were mapped per sample (range 11.8–19.1 million) with final read counts proportionate to starting library size and showing no apparent sign of increase in quantification rates with genetic distance.

In addition to the final workflow above, we tried two other methods for completeness that did not improve data quality and so were not used for final analyses. A small number of remaining reads were de novo assembled using Trinity (1) and quantified but represented largely micro-contaminants or low-expression lineage-specific paralogs and were not included in final read count tables. We also annotated each sample's read alignment de novo by Cufflinks (version 2.2.1; 7) requiring paired sequences and with all other parameters default. Comparison of the individual annotations to the reference indicated no consistent new features among species and so only *P. filicauda* reference genes were used for the final quantification step. Quantification for featureCounts was also repeated allowing fractional counting of multi-mapped reads and reads mapping to multiple features. However, the similarity of counts for all samples was high ($r^2 \geq 0.98$) and so we used the uniquely mapped reads table only.

Phylogenetic Differential Gene Expression Tool (PhyDGET). Whole-transcriptome expression profiling more directly examines gene-by-gene activity within a specific tissue, but models for studying the evolution of transcriptome-wide expression profiles are still nascent and phase and face substantial challenges (8, 9). Previous approaches to modeling transcriptome evolution have focused on shifts in gene or gene family expression levels (10, 11, 12), changes in expression variance under selection (13, 14, 15, 16), and tissue-specificity or profile divergence (17, 18, 19). Despite the technical challenges inherent in RNA-Seq, Differential Gene Expression (DGE) analysis, and Phylogenetic Comparative Methods (PCM), an approach combining these techniques is the most direct approach to studying a tissue-specific molecular shifts in situ for complex traits.
The Phylogenetic Differential Gene Expression Tool (PhyDGET) is based primarily on Phylogenetic Comparative Methods, specifically modeling shifts in the levels of a quantitative trait that have been established previously for physiological traits. PhyDGET treats each gene’s mRNA expression level as a quantitative trait and applies a model likelihood comparison strategy to determine the fit of the data to various models of accelerated change in expression level compared to the stochastic changes under a Brownian Motion null model. PhyDGET is a Python3-based script engine that performs data transformation and parallelizes calls to a Bayesian PCM engine (in this case BayesTrait3; 20). PhyDGET is available open-source and free at http://www.github.com/peaselab/phydget.

The first step in PhyDGET is to normalize and adapt RNA-seq expression data to be usable under the conditions of Comparative Methods analyses. First, the raw count tables are normalized to counts-per-million reads (cpm) in each sample. Next, these values are log\(_2\)-transformed into the log\(_2\)(cpm) values used in the actual comparative analyses. The values are calculated as follows (to accommodate zeros):

\[
\log_2(\text{cpm}) = \log_2\left(\frac{\text{raw count} + 0.5}{\text{total sample library size}/10^6}\right)
\]

(1)

The normalization in a log\(_2\)(cpm) values (1) normalizes different library sizes in the case of different sequencing yields, (2) makes each integer unit of log\(_2\)(cpm) values correspond to a doubling of expression, and (3) brings the raw values of RNA-seq counts that can regularly span a range from 0 to > 500,000 into a more unified quantitative space where log\(_2\)(cpm) values are in the range from \(-5\) to \(\approx 16\), and (4) accommodates log-of-zero problem. This normalization scheme makes genes at various scales of expression levels more comparable when placed in the next phase of the comparative analyses.

The second step in PhyDGET is to feed the log\(_2\)(cpm) normalized data one gene at a time into BayesTrait3 (20) for evaluating the likelihood of the gene’s values evolving under a variety of user-specified models (Fig. S1) compared to each other and a null model. The script automatically translates single- or multi-branch models into the correct syntax for the BayesTrait3 control file and then uses a Python3 framework to parallelize the running of the \((n \times m)\) runs for \(n\) genes and \(m\) models. The marginal likelihood scores and transformation values are extracted from the outputs and then can be compared among models for each gene.

**Example BayesTrait3 Alternative Model Specification:**

```
7
2
Burnin 1000000
Iterations 10000000
PriorAll uniform -10 30
DistData $TEMPDATAFILE
Stones 200 20000
AddTag Target0 MAVI
LocalTransform TransBranch0 Target0
```

**Simulations to determine significance cutoffs for PhyDGET.** While the likelihood ratio test associated with BayesTrait models has rough significance cutoffs that have become convention, we hypothesized that the log\(_2\)-transformation and normalization of the data would make the Bayes Factor (BF) differences smaller and thus the standard cutoff of BF > 2 might be overly conservative for this application. Therefore, we wanted to assess the false discovery rate (FDR) directly for this phylogeny under a range of expression values to empirically determine an appropriate cutoff values for this method.

We used the *phangorn* package (21) to generate an ultrametric tree from the consensus nuclear phylogeny. To simulate values, we wrote a custom R-script with the *ape* (21), *evd*, and *phytools* (22) packages. The *fastBM* function from *phytools* was used to simulate five sets of 10000 genes using starting \(\alpha\) values of 10, 100, 1000, 10000, 100000. In all six treatments, \(\sigma^2\) values was calculated as 0.22\(\alpha^2\) as determined by calculating the median of the variance of across all genes in the empirical dataset. This follows the biological pattern of a positive mean-variance relationship for gene expression values observed in nearly all datasets.
These six Brownian Motion simulated datasets of 2000 genes were transformed into log_{2}(cpm) values using approximated library sizes to achieve a similar log_{2}(cpm) values to genes at those raw count values in the empirical file. These data were tested in PhyDGET under three models: (1) the M4 ancestor branch model with a single target branch, (2) the Parallel single-rate model (MC1) where the Manacus vitellinus and Ceratopipra ancestor branches were both targets under a single alternative rate parameter, (3) the Reversion two-rate model (R42) with the M4 ancestor branch and the Pseudopipra pipra branch both target but under two separate rate parameters.

The distributions of the BF values from these 15 sets of 10000 tests showed generally low rates of false positives (Fig. S9). A slight increase in the rate of false positives was observed as the raw value increases from 10 to 100000. Given the scaling of the variance in expression in the BM simulations, this is expected and the increase in false positive rate is modest. For genes in the range of raw expression values from 10 to 1,000 (most genes detected in the empirical set), the FDR rate for BF $\geq$ 1.5 was 0.65 to 4.5%. Based on these results we adopted a BF cutoff of 1.5 as an appropriate standard cutoff for all tests, though these results show that especially for genes at the lower levels of expression (and thus lower levels of potential variance) that this cutoff may be conservative.

Considerations of functional phylotranscriptomic expression modeling. In considering the application of this analysis framework to other traits, systems, and questions, many other factors should be considered. We benefited here from past empirical results that identified tissues of interest, a muscle contraction trait with established molecular systems and phenotypic diversity among close relatives, and high similarity of expression profiles within species, individuals, and tissues. The high molecular concordance of the species phylogeny and low genetic distances among species also largely precluded confounding effects from hemiplasy (23). All of these factors would have to be weighed carefully in applying our framework in other systems as the expression landscape, trait complexity, and time scale and phylogenetic discordance increase. We also recognize that these expression measurements are a time slice in the moment of adult tissue activity, and therefore more work may be necessary to further enrich the functional developmental context of these physiological adaptations. With these considerations in mind, we conclude that this approach could be applied readily to many complex traits appearing among closely related species where comparison of tissue-specific expression levels might offer more direct evidence of evolutionary changes than sequence variations. As we further explore the diverse evolutionary histories of genomes and their circuitous connections to traits, an evolutionary view of the intermediate transcriptional layer is a vital window for disentangling the layered molecular bases of traits.

Pairwise Group Approximations of Phylogenetic Expression Tests. We conducted the following pairwise tests using species groupings where individuals were encoded two states in a linear model and analyzed using limma+voom using the same general parameters at the pairwise SH-PEC tests. (1) RWM species (C. cornuta, C. mentalis, M. vitellinus) versus all others, (2) Mi. oleagineus and X. atronitens versus all other species to approximate the M5 ancestor model, (3) Mi. oleagineus, X. atronitens, and L. coronata versus all other species to approximate the M4 ancestor model, and (4) both Ceratopipra species versus all other species to approximate the Ceratopipra ancestor model (CE).

Gene Ontology Annotation and Analysis. We first created a Pipra filicauda reference by BLAST search for a strong match to the NCBI database of amniote sequences. We used a consensus of these BLAST hits to assign each gene to a human ortholog. The GO terms were assigned to each P. filicauda reference gene symbol based on the human GO terms for that gene. If a specific ortholog could not be identified confidently but the gene was clearly in a specific gene family, we assigned the gene the intersection of common GO terms from that genes in that gene family. We assigned GO terms to 14,752 out of 18,065 P. filicauda reference gene symbols.

Focal study gene sets (target group) were determined for intraspecific pairwise SH-versus-PEC tests, we set the focal study set as genes with Adjusted $P$-value $\leq$ 0.05 from limma analysis. Focal study sets were determined for each PhyDGET model test using a cutoff of BF $\geq$ 1.5 (for the CE, LC, MV, PP, XA, and M6 terminal branch/outgroup branch models) or BF $\geq$ 1.0 (for the M5, M4, R41, and MC1 ancestral and double-branch models). The background list of 14,777 genes were those with at least 10 counts-per-million in at least three samples.
We conducted Gene Ontology analyses using Ontologizer (v2.1; 24) using the Parent-Child Union method (25) and the Benjamini-Hochberg multiple testing correction (26). The top SH-vs.-PEC differentially expressed genes for each species are shown in Table S9, and the top GO terms are shown in Table S10 (no GO results were available for X. atronitens). The lists of PhyDGET-identified genes for each model are shown in Table S14 and the top GO terms associated are shown in Table S12.

**Phylogenetic Tests of Selection.** Starting from the transcriptome sequence alignment used for the phylogenetic inference, we used MVTools to merge alleles ("FilterMVF") from the two tissues sampled from each individual (27). We then used MVTools (MVF2FastaGene) to extract coding sequences using the P. filicauda reference annotation GTF file (V1) selecting the option to choose a random allele at each heterozygous site. We then used a custom Python3 script to filter out 1,015 genes with missing species data premature stop codons in the alignment. These were mostly due to lower coverage and, in a few cases, likely paralog collapse. None of these genes were implicated as important in any of the differential gene expression tests (phylogenetic or pairwise).

Once the alignment of each gene's coding sequence was extracted, we tested for evidence of positive selection in each gene in the sequenced transcriptomes using the branch-test in PAML (28). Using a custom Python3 script, we prepared sequence alignment files containing a random individual's sequence from each species for each gene. We ran four PAML tests on these focal branches: (1) M5 ancestor branch, (2) the M4 ancestor branch, (3) the Ceratopipra ancestor branch, and (4) the terminal branch for M. vitellinus (see Fig. 4). To identify genes experiencing positive selection in each foreground branch, estimated branch-specific rates of non-synonymous substitution per non-synonymous site (\(d_N\)) and the ratio of \(d_N\) to synonymous substitutions per synonymous site (\(d_N/d_S\)) values in the focal test branches.

We ran `codeml` from PAML (28) using a control files with the following parameters for both the null and alternative models:

```
runmode = 0
method = 0
seqtype = 1
CodonFreq = 2
clock = 0
NSSites = 0
fix_kappa = 0
kappa = 2
fix_omega = 0
omega = 1
fix_alpha = 1
alpha = 0
Malph = 0
ncatG = 4
cleandata = 1
```

The `cleandata = 1` ensures all gap and ambiguous positions in the alignment are ignored, so that the only positions analyzed were those shared between all taxa. For the null model control file, we set `model = 0`, specifying `codeml` to calculate a single \(\omega_0\) (\(d_N/d_S\)) value for the whole alignment. This alignment-wide \(\omega_0\) serves as the null hypothesis to test against branch-specific omega values to detect a significant difference. To run a test model and estimate different omega values among lineages, we specified `model = 2` in the test model control file to estimate the focal branch \(\omega_1\) rate parameter separately from the background \(\omega_0\). Top gene candidates from the M5, M4, MV, and CE branch tests are shown in Table S15. We performed multiple runs to check for effects of the species and allele randomization procedures did not and found any substantive changes to the results.

**Parvalbumin phylogeny.** The parvalbumin family includes four primary vetebrate genes known as: *parvalbumin* (*PVALB*), *oncomodulin* (*OCM*), *oncomodulin 2* (*OCM2*), and *oncomodulin 3* (*OCM3*; only found in non-placental tetrapods). We used NCBI BLAST to find sequences related to the manakin,
chicken, and human sequences of these genes among diverse vertebrate lineages. A set of 158 parvalbumin family sequences was trimmed to their coding sequences and aligned using MUSCLE (2)). We inferred a phylogeny from these genes using RAxML-ng (29) using the GTR+Γ4.
Fig. S1. **Distribution map of the six manakin study species.** Ranged estimated using data from www.birdlife.org and www.gbif.org. Abbreviations: Cc = *Ceratopipra cornuta*, Cm = *Ceratopipra mentalis*, Lc = *Lepidothrix coronata*, Mv = *Manacus vitellinus*, Pp = *Pseudopipra pipra*, Xa = *Xenopipo atronitens*. 
Fig. S2. Phylogenomic diversity by QS and sCF scores. For the 7-taxon tree (A) and 8-taxon tree (B; including the *P. filicauda* reference), the proportion of concordant gene trees (fC), quartet concordance score (QC), and site concordance factors (sCF) show generally strong support for the consensus tree. The high quartest differential (QD) scores indicate no evidence of biased discordance that might indicate introgression. The Quartet Informativeness (QI) scores are relatively low indicating that most individual gene alignments do not contain sufficient phylogenetic signal to be measurably concordant or discordant. Collectively, this indicates low phylogenetic signal (rather than a biological ILS or introgression mechanism) is the primary contributor to lowering our concordance scores.
Fig. S3. Correlation coefficients of gene expression counts-per-million. Samples are labeled as: first two letters of genus, first two letters of species, individual number, P(EC) or (S)H muscle. Values above the diagonal are rounded linear regression $r^2 \times 100$, and values below the diagonal are rounded Pearson's $\rho \times 100$. 

| Sample | Value | Sample | Value | Sample | Value | Sample | Value | Sample | Value | Sample | Value |
|--------|-------|--------|-------|--------|-------|--------|-------|--------|-------|--------|-------|
| **CEN** | **19** | **31** | **53** | **75** | **97** | **119** | **41** | **63** | **85** | **107** | **129** |
| **CEN** | **01** | **23** | **45** | **67** | **89** | **111** | **33** | **55** | **77** | **99** | **121** |
| **CEN** | **12** | **34** | **56** | **78** | **100** | **122** | **42** | **64** | **86** | **108** | **130** |
| **CEN** | **24** | **46** | **68** | **90** | **112** | **132** | **53** | **75** | **97** | **119** | **141** |
| **CEN** | **36** | **58** | **80** | **102** | **124** | **143** | **65** | **87** | **109** | **131** | **153** |
| **CEN** | **48** | **70** | **92** | **114** | **136** | **155** | **89** | **111** | **133** | **156** | **177** |
| **CEN** | **60** | **82** | **104** | **126** | **148** | **169** | **91** | **113** | **135** | **157** | **179** |
| **CEN** | **72** | **94** | **106** | **128** | **140** | **162** | **105** | **127** | **149** | **171** | **193** |
| **CEN** | **84** | **96** | **108** | **130** | **152** | **174** | **117** | **139** | **161** | **183** | **205** |
| **CEN** | **96** | **100** | **112** | **132** | **154** | **176** | **129** | **151** | **173** | **195** | **217** |

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**Fig. S4. Diagrams of PhyDGET Branch Models and Trait Hypotheses**

Several alternative models with alternative rates parameters of expression value change were tested, including models with a single alternative rate on a single branch (M6, M5, M4, XA, LC, MV, PP, CE) and models with two branches tested either under a single alternative rate parameter (R51, R41, and MC1) or two alternative rate parameters on the two branches (R52, R42, MC2). Sunbursts at the bottom indicate presence (filled) or absence (empty) of the rapid wing movement (RWM). Colors on the branch marker dots correspond to the colors in Figure 3, or are highlighted in black otherwise. **b,c,d**, Three models of the evolution of the rapid wing movement trait.
Fig. S5. Comparison of PhyDGET to pairwise differential gene expression. Comparison of PhyDGET results from each model (Bayes Factor on x-axis) compared to grouped pairwise differential gene expression approximation of the same comparisons using \textit{limma}+\textit{voom} (\log_{10} P on the y-axis). Grey points are genes non-significant under both tests, green points showing genes significant (BF geq 1.5) under PhyDGET only, blue points significant (P < 10^{-4}) in \textit{limma} only, and magenta points significant under both. The number grid above each graph shows the number of genes highlighted in each quadrant of the graph, with the upper right value being the number significant under both analyses.
Fig. S6. Comparison of pairs of model results from PhyDGET for the SH tissue. Bayes Factor (BF) values for pairs of models are shown, highlighting the cases where models with overlapping branches (e.g., MC1/MC2, MC1/MV, MC1/CE, R41/PP, R41/M4) show strong or moderate correlation. Genes significant in one of the models are highlighted in blue or green. Models without overlapping target branches do not show correlation and have few (if any) genes significant in both tests (magenta points).
Fig. S7. (continued on next page)
**Fig. S7.** (continued on next page)
Fig. S7. (continued on next page)
Fig. S7. (continued on next page)
Fig. S7. Expression profiles for PhyDGET-detected genes Genes highlighted in Figure 3 showing the mean counts-per-million per species (horizontal line) with standard error (shaded bars) and three individual values (open circles) for both tissues. Color-coding for the models matches Fig. 3 and Fig. S4. The color-highlighted species are expected to have different expression compared to grey-shaded species in each model’s representation (when the test has higher BF values).
Fig. S8. Phylogeny and protein alignment of parvalbumin family genes. (caption on next page)
Fig. S8. Phylogeny and protein alignment of parvalbumin family genes. (A) A phylogeny of the parvalbumin-oncomodulin gene family showing our PVALB-like-1 genes that show strong increase in expression in Ceratopipra are from the “oncomodulin 3” subfamily defined by the G. gallus reference OCM3 gene. This subfamily is a related but distinct paralog from the PVALB-like-2 gene. These two paralogs are ancient duplicates of parvalbumin beta (PVALB), oncomodulin (OCM), oncomodulin 2 (OCM2) in the human genome and CPV3 in the chicken genome. Sequences from our RNA-Seq data are highlighted in yellow and with labels preceded by an asterisk. Reference human and chicken genes are highlighted in red. (B) An amino acid alignment of parvalbumin/oncomodulin proteins showing the focal OCM3/PVALB-L1 genes at the top with amino acids differing from our Ceratopipra mentalis sequence highlighted (identical residues are shown as dots). High conservation is observed among Passerine birds and the chicken reference sequence (directly above the horizontal divider) is only two residues different from the manakin conserved sequence. The PVALB-L2 from C. mentalis, and PVALB, OCM, and OCM2 from chicken and human are all highly divergent in sequence from OCM3/PVALB-L1, as shown below the divider.
Fig. S9. PhyDGET Results from Simulated Values. Histograms show the distribution of Bayes Factor (BF) scores between the null and alternative model of a shift on the M4 ancestor branch (left), two-branch Parallel model (center), and two-branch two-rate Reversion model (R42; right) using a variety of starting \( \alpha \) values. Dashed lines and inset text show the 95th, 99th percentiles (green) and the False Discovery Rate (FDR) of values with BF \( \geq 1.0, 1.5, 2.0, \) and 5.0 (black). The rate of false positives increases slightly with the raw value, as expected given the increased variance scaling with the starting value in the simulations and reflecting the biological pattern of mean-variance relationships. Slightly higher FDR rates were observed in the 2-branch models than the 1-branch model.
Fig. S10. (continued on next page)
Fig. S10. Expression profiles for other genes of interest. Genes with high expression, from gene families of interest, or known candidate functional loci. The mean counts-per-million per species (horizontal line) with standard error (shaded bars) and three individual values (open circles) are shown for both tissues: (P)EC and (S)H.
Supplementary Tables

| Location                  | Species Sampled                  | Approximate Latitude | Approximate Longitude |
|---------------------------|----------------------------------|----------------------|-----------------------|
| Gamboa, Panama            | *Ceratopipra mentalis*           | 9°07'N               | 9°41'W                |
|                           | (Red-capped manakin)             |                      |                       |
|                           | *Lepidothrix coronata*           |                      |                       |
|                           | (Blue-crowned manakin)           |                      |                       |
|                           | *Manacus vitellinus*             |                      |                       |
|                           | (Golden-collared manakin)        |                      |                       |
|                           | *Mionectes oleagineus*           |                      |                       |
|                           | (Ochre-bellied flycatcher)       |                      |                       |
| Pakaraima Mountains, Guyana | *Ceratopipra cornuta*           | 4°56'N               | 59°53'W               |
|                           | (Scarlet-horned manakin)         |                      |                       |
| Boro Boro River, Guyana   | *Pseudopipra pipra*              | 4°8'N                | 59°4'W                |
|                           | (White-collared manakin)         |                      |                       |
| coastal savannah, Guyana  | *Xenopipo atronitens*            | 6°22'N               | 58°7'W                |
|                           | (Black manakin)                  |                      |                       |

Table S1. Wing beat frequencies of birds with similar size to manakins.
| species                      | muscle action      | wingbeat frequency (Hz) | body weight (g) |
|------------------------------|--------------------|-------------------------|-----------------|
| *Ceratopipra mentalis*      | SH “claps” display | ≈50 (30)                | 13.0–15.5 (31)  |
| (red-capped manakin)         |                    |                         |                 |
| *Ceratopipra cornuta*        |                    |                         | 21.9–23.75 (31) |
| (scarlet-horned manakin)     |                    |                         |                 |
| *Manacus vitellinus*         | SH “roll-snap” display | 60–70 (32)         | 16.95–18.7 (31) |
| (golden-collared manakin)    | PEC wing flap      | 17.6 (33)               |                 |
| *Taeniopygia guttata*        | PEC wing flap      | 26.7–29.3 (34)         | 10–16 (35)      |
| (zebra finch)                |                    |                         |                 |
| *Fringilla coelebs*          | PEC wing flap      | 18.2 ± 1.74 S.E. (36)  | 17–30 (37)      |
| (common chaffinch)           |                    |                         |                 |
| *Catharus ustulatus*         | PEC wing flap      | 11.9–19.6 (38)         | 23–45 (37)      |
| (Swainson's thrush)          |                    |                         |                 |
| *Hylocichla mustelinae*      | PEC wing flap      | 10.1–18.6 (38)         | 40–50 (37)      |
| (wood thrush)                |                    |                         |                 |
| *Delichon urbica*            | PEC wing flap      | 2.5–8.4 (39)           | 15–21 (37)      |
| (house martin)               |                    |                         |                 |
| *Hirundo rustica*            | PEC wing flap      | 3.0–8.1 (39)           | 16–25 (37)      |
| (barn swallow)               |                    |                         |                 |

**Table S2.** Wing beat frequencies and body weight ranges of birds with similar size to manakins. Data compiled from the cited sources.
| #  | Sample   | Species     | Date extracted | Amount of RNA on qubit (ng/ul) | Bioanalyzer concentration (pg/ul) |
|----|----------|-------------|----------------|--------------------------------|----------------------------------|
| 1  | LECO1-SH | *L. coronata* | 05/02/16 | 149                         | 745                              |
| 2  | LECO2-SH | *L. coronata* | 05/02/16 | 362                         | 1810                             |
| 3  | LECO3-SH | *L. coronata* | 05/02/16 | 436                         | 2180                             |
| 4  | LECO1-PEC | *L. coronata* | 5/7/2016 | 266                         | 1330                             |
| 5  | LECO2-PEC | *L. coronata* | 5/7/2016 | 210                         | 1050                             |
| 6  | LECO3-PEC | *L. coronata* | 5/7/2016 | 236                         | 1180                             |
| 7  | MAVI1-SH | *M. vitellinus* | 5/6/2016 | 572                         | 2860                             |
| 8  | MAVI2-SH | *M. vitellinus* | 5/6/2016 | 308                         | 1540                             |
| 9  | MAVI3-SH | *M. vitellinus* | 5/6/2016 | 240                         | 1200                             |
| 10 | MAVI1-PEC | *M. vitellinus* | 5/6/2016 | 1200                        | 3000                             |
| 11 | MAVI2-PEC | *M. vitellinus* | 5/6/2016 | 1140                        | 2850                             |
| 12 | MAVI3-PEC | *M. vitellinus* | 5/6/2016 | 1200                        | 3000                             |
| 13 | CECO1-SH | *C. cornuta* | 5/7/2016 | 400                         | 2000                             |
| 14 | CECO2-SH | *C. cornuta* | 5/7/2016 | 242                         | 1210                             |
| 15 | CECO3-SH | *C. cornuta* | 5/7/2016 | 106                         | 530                              |
| 16 | CECO1-PEC | *C. cornuta* | 5/3/2016 | 442                         | 2210                             |
| 17 | CECO2-PEC | *C. cornuta* | 5/3/2016 | 400                         | 2000                             |
| 18 | CECO3-PEC | *C. cornuta* | 5/3/2016 | 388                         | 1945                             |
| 19 | XEAT1-SH | *X. atronitrens* | 5/7/2016 | 398                         | 1990                             |
| 20 | XEAT2-SH | *X. atronitrens* | 5/7/2016 | 226                         | 1130                             |
| 21 | XEAT3-SH | *X. atronitrens* | 5/7/2016 | 155                         | 775                              |
| 22 | XEAT1-PEC | *X. atronitrens* | 5/6/2016 | 137.2                       | 860                              |
| 23 | XEAT2-PEC | *X. atronitrens* | 5/6/2016 | 86                          | 3220                             |
| 24 | XEAT3-PEC | *X. atronitrens* | 5/6/2016 | 322                         | 1600                             |
| 25 | PSPI1-SH | *P. pipra* | 5/6/2016 | 160                         | 670                              |
| 26 | PSPI2-SH | *P. pipra* | 5/6/2016 | 67.2                        | 1480                             |
| 27 | PSPI3-SH | *P. pipra* | 5/6/2016 | 148                         | 1480                             |
| 28 | PSPI1-PEC | *P. pipra* | 5/3/2016 | 622                         | 3110                             |
| 29 | PSPI2-PEC | *P. pipra* | 5/3/2016 | 590                         | 2950                             |
| 30 | PSPI3-PEC | *P. pipra* | 5/3/2016 | 136                         | 1360                             |
| 31 | CEME1-SH | *C. mentalis* | 5/7/2016 | 330                         | 1580                             |
| 32 | CEME2-SH | *C. mentalis* | 5/7/2016 | 316                         | 2490                             |
| 33 | CEME3-SH | *C. mentalis* | 5/7/2016 | 498                         | 3335                             |
| 34 | CEME1-PEC | *C. mentalis* | 5/7/2016 | 667                         | 1550                             |
| 35 | CEME2-PEC | *C. mentalis* | 5/7/2016 | 310                         | 2150                             |
| 36 | CEME3-PEC | *C. mentalis* | 5/7/2016 | 430                         | 0                                |
| 37 | MIOL1-SH | *Mi. oleagineus* | 5/6/2016 | 396                         | 1980                             |
| 38 | MIOL2-SH | *Mi. oleagineus* | 5/6/2016 | 428                         | 2140                             |
| 39 | MIOL3-SH | *Mi. oleagineus* | 5/6/2016 | 308                         | 1540                             |
| 40 | MIOL1-PEC | *Mi. oleagineus* | 5/3/2016 | 696                         | 3480                             |
| 41 | MIOL2-PEC | *Mi. oleagineus* | 5/3/2016 | 750                         | 3750                             |
| 42 | MIOL3-PEC | *Mi. oleagineus* | 5/3/2016 | 1100                        | 3667                             |

**Table S3.** **Sequencing quality metrics.** Id number (for sequencing), sample and species names, date of extraction of the RNA, and concentrations on Qubit and Bioanalyzer.
| Sample   | Raw reads  | % After Quality Filtering | % Raw Map to Mitochondria | % Nuclear Reads Mapped to Genome | Total Counted of Nuclear Mapped Reads | % Counted of Nuclear Mapped Reads |
|----------|------------|---------------------------|---------------------------|----------------------------------|--------------------------------------|-----------------------------------|
| LECO1-SH | 24,240,465 | 99.87%                    | 16.32%                    | 91.79%                           | 17,440,240                           | 86.24%                            |
| LECO2-SH | 28,245,653 | 99.86%                    | 20.11%                    | 90.18%                           | 19,066,123                           | 84.76%                            |
| LECO3-SH | 27,791,546 | 99.89%                    | 22.30%                    | 89.96%                           | 18,248,487                           | 84.77%                            |
| LECO1-PEC| 25,866,131 | 99.89%                    | 31.73%                    | 86.62%                           | 14,620,430                           | 83.19%                            |
| LECO2-PEC| 25,399,293 | 99.89%                    | 32.97%                    | 88.30%                           | 14,288,649                           | 84.34%                            |
| LECO3-PEC| 26,386,738 | 99.90%                    | 36.12%                    | 88.68%                           | 14,192,607                           | 84.55%                            |
| MAVI1-SH | 23,867,517 | 99.92%                    | 33.03%                    | 85.36%                           | 12,958,295                           | 81.41%                            |
| MAVI2-SH | 24,439,612 | 99.92%                    | 31.86%                    | 86.04%                           | 13,585,673                           | 81.79%                            |
| MAVI3-SH | 25,318,789 | 99.90%                    | 30.20%                    | 85.36%                           | 13,323,028                           | 83.11%                            |
| MAVI1-PEC| 30,105,559 | 99.92%                    | 44.77%                    | 87.21%                           | 13,817,608                           | 83.45%                            |
| MAVI2-PEC| 26,055,131 | 99.91%                    | 38.07%                    | 86.26%                           | 13,323,028                           | 83.11%                            |
| MAVI3-PEC| 26,303,029 | 99.91%                    | 39.23%                    | 85.88%                           | 13,114,388                           | 82.73%                            |
| CECO1-SH | 25,521,654 | 99.91%                    | 30.81%                    | 87.18%                           | 14,517,277                           | 82.60%                            |
| CECO2-SH | 25,885,362 | 99.89%                    | 30.98%                    | 86.77%                           | 14,633,982                           | 82.32%                            |
| CECO3-SH | 26,035,482 | 99.88%                    | 32.94%                    | 85.36%                           | 13,253,687                           | 82.84%                            |
| XEAT1-SH | 27,144,487 | 99.91%                    | 40.83%                    | 86.52%                           | 13,293,623                           | 83.95%                            |
| XEAT2-SH | 24,964,724 | 99.89%                    | 37.39%                    | 87.29%                           | 12,798,538                           | 83.28%                            |
| XEAT3-SH | 26,650,402 | 99.89%                    | 33.77%                    | 86.88%                           | 14,406,384                           | 83.01%                            |
| XEAT1-PEC| 24,341,291 | 99.91%                    | 42.39%                    | 87.35%                           | 13,263,626                           | 83.82%                            |
| XEAT2-PEC| 26,055,131 | 99.91%                    | 38.07%                    | 86.26%                           | 13,323,028                           | 83.11%                            |
| XEAT3-PEC| 28,933,085 | 99.91%                    | 39.23%                    | 85.88%                           | 13,114,388                           | 82.73%                            |
| PSPI1-SH | 24,095,715 | 99.90%                    | 38.81%                    | 85.92%                           | 11,873,860                           | 81.97%                            |
| PSPI2-SH | 26,498,928 | 99.91%                    | 43.19%                    | 87.14%                           | 12,245,928                           | 82.86%                            |
| PSPI3-SH | 25,333,635 | 99.90%                    | 42.42%                    | 87.62%                           | 11,941,114                           | 83.42%                            |
| PSPI1-PEC| 26,469,426 | 99.90%                    | 41.46%                    | 86.22%                           | 12,575,976                           | 82.49%                            |
| PSPI2-PEC| 26,434,185 | 99.90%                    | 40.58%                    | 86.73%                           | 12,282,222                           | 82.51%                            |
| PSPI3-PEC| 25,441,092 | 99.88%                    | 39.80%                    | 87.69%                           | 12,387,728                           | 83.95%                            |
| CEME1-SH | 25,095,251 | 99.86%                    | 29.70%                    | 88.95%                           | 14,721,145                           | 84.02%                            |
| CEME2-SH | 24,995,771 | 99.87%                    | 28.64%                    | 86.93%                           | 14,526,463                           | 81.77%                            |
| CEME3-SH | 26,029,065 | 99.90%                    | 25.19%                    | 89.78%                           | 16,428,178                           | 84.87%                            |
| CEME1-PEC| 26,258,958 | 99.90%                    | 44.21%                    | 87.59%                           | 12,108,735                           | 83.18%                            |
| CEME2-PEC| 28,288,172 | 99.90%                    | 39.35%                    | 86.31%                           | 13,936,682                           | 81.70%                            |
| CEME3-PEC| 26,832,237 | 99.89%                    | 44.86%                    | 87.03%                           | 12,106,808                           | 82.68%                            |
| MIOL1-SH | 27,595,869 | 99.88%                    | 29.70%                    | 86.95%                           | 16,364,458                           | 84.94%                            |
| MIOL2-SH | 25,912,309 | 99.88%                    | 38.94%                    | 87.51%                           | 13,439,728                           | 85.49%                            |
| MIOL3-SH | 22,627,541 | 99.85%                    | 35.07%                    | 86.47%                           | 12,361,971                           | 84.68%                            |
| MIOL1-PEC| 24,249,406 | 99.88%                    | 35.57%                    | 86.47%                           | 13,154,542                           | 85.02%                            |
| MIOL2-PEC| 25,968,132 | 99.87%                    | 33.11%                    | 86.33%                           | 14,694,711                           | 85.00%                            |
| MIOL3-PEC| 28,943,901 | 99.88%                    | 39.99%                    | 85.68%                           | 14,583,879                           | 84.70%                            |

Table S4. Sequence Mapping Metrics. Raw number of reads sequenced, percent of raw retained after quality control trimming and filtering (see Supplementary Methods), percent mapped to the mitochondrial genomes, percent of the remainder mapped to the nuclear genome, total number of mapped reads, and total percent of nuclear genome-mapped that were quantified uniquely to a gene feature.
| Individual | Total Bases Covered at Depth ≥ 3 | % Percent Coverage |
|------------|----------------------------------|--------------------|
| P. filicauda Reference | 187,053,847 | |
| LECO1 | 94,139,392 | 50.33% |
| LECO2 | 85,198,563 | 45.55% |
| LECO3 | 80,225,667 | 42.89% |
| MAVI1 | 55,198,388 | 29.51% |
| MAVI2 | 58,390,019 | 31.22% |
| MAVI3 | 60,060,711 | 32.11% |
| CECO1 | 55,838,882 | 29.85% |
| CECO2 | 56,078,814 | 29.98% |
| CECO3 | 53,009,227 | 28.34% |
| XEAT1 | 56,074,253 | 29.98% |
| XEAT2 | 56,182,688 | 30.04% |
| XEAT3 | 57,496,392 | 30.74% |
| PSPI1 | 52,119,833 | 27.86% |
| PSPI2 | 52,334,977 | 27.98% |
| PSPI3 | 50,689,389 | 27.10% |
| CEME1 | 62,573,398 | 33.45% |
| CEME2 | 60,058,501 | 32.11% |
| CEME3 | 70,416,863 | 37.65% |
| MIOL1 | 50,470,135 | 26.98% |
| MIOL2 | 50,173,041 | 26.82% |
| MIOL3 | 49,068,149 | 26.23% |

**Table S5. Sequencing Quality Metrics.** Short-read pairs mapped at depth of 3 or greater to the *P. filicauda* reference genome and their coverage as approximated by the total size of the annotated *P. filicauda* reference transcriptome.
Table S6. Pairwise Nucleotide Sequence Distances. Pairwise nucleotide distances for the complete alignment of all nucleotides at depth of coverage ≥ 3.
| Rank | Tree Topology | Count | Type | Rank | Tree Topology | Count | Type |
|------|--------------|-------|------|------|--------------|-------|------|
| 1    | ((Cc,Cm,L,M,P,X),O); | 517   | c    | 34   | ((L,(X,(Cc,Cm,M,P)),O); | 38    | d    |
| 2    | ((X,(Cc,Cm,L,M,P)),O); | 497   | c    | 35   | ((L,(X,(Cc,Cm,M,P)),O); | 38    | d    |
| 3    | ((X,(L,(M,(P,(Cc,Cm)))),O); | 476   | CON  | 36   | ((X,(L,(M,(Cc,Cm,M,P)),O); | 37    | DIS  |
| 4    | ((X,(L,M,P,(Cc,Cm)),O); | 257   | c    | 37   | ((L,(X,(Cc,Cm,M,P)),O); | 37    | d    |
| 5    | ((X,(L,M,P,(Cc,Cm)),O); | 245   | c    | 38   | ((L,(X,(Cc,Cm,M,P)),O); | 36    | DIS* |
| 6    | ((X,(L,M,(Cc,Cm,P)),O); | 223   | c    | 39   | ((L,(X,(Cc,Cm,M,P)),O); | 36    | d    |
| 7    | ((X,(L,(Cc,Cm,M,P)),O); | 210   | c    | 40   | ((L,(X,(Cc,Cm,M,P)),O); | 36    | d    |
| 8    | ((X,(L,(Cc,Cm,M,P)),O); | 208   | c    | 41   | ((L,(X,(Cc,Cm,M,P)),O); | 36    | d    |
| 9    | ((X,(L,(M,P,(Cc,Cm)),O); | 153   | d    | 42   | ((L,(M,(Cc,Cm,M,P)),O); | 35    | DIS  |
| 10   | ((X,((L,M),(P,(Cc,Cm)),O); | 137   | d    | 43   | ((L,(X,(Cc,Cm,M,P)),O); | 35    | d    |
| 11   | ((X,(L,M,(P,(Cc,Cm)),O); | 130   | DIS  | 44   | ((L,(X,(Cc,Cm,M,P)),O); | 32    | c    |
| 12   | ((L,M,X,(Cc,Cm,P)),O); | 130   | c    | 45   | ((L,(X,(M,P,(Cc,Cm)),O); | 32    | d    |
| 13   | ((L,M,P,X,(Cc,Cm)),O); | 130   | c    | 46   | ((L,(X,(Cc,Cm,M,P)),O); | 31    | d    |
| 14   | ((L,X,(Cc,Cm,M,P)),O); | 123   | c    | 47   | ((L,(X,(Cc,Cm,M,P)),O); | 30    | d    |
| 15   | ((M,(Cc,Cm,L,P,X)),O); | 102   | d    | 48   | ((L,(Cc,Cm,M,P,X)),O); | 30    | d    |
| 16   | ((L,Cc,Cm,M,P,X),O); | 95    | d    | 49   | ((L,(Cc,Cm,L,P,X)),O); | 30    | d    |
| 17   | ((L,(Cc,Cm,L,P,X)),O); | 87    | d    | 50   | ((L,(Cc,Cm,L,P,X)),O); | 29    | d    |
| 18   | ((L,(X,(Cc,Cm,M,P,X)),O); | 79    | DIS  | 51   | ((L,(X,(Cc,Cm,M,P,X)),O); | 29    | d    |
| 19   | ((L,(X,(Cc,Cm,M,P,X)),O); | 62    | d    | 52   | ((L,(X,(Cc,Cm,M,P,X)),O); | 29    | d    |
| 20   | ((L,(Cc,Cm,L,P,X)),O); | 60    | d    | 53   | ((L,(X,(Cc,Cm,M,P,X)),O); | 29    | d    |
| 21   | ((L,X,(Cc,Cm,M,P)),O); | 59    | d    | 54   | ((L,X,(Cc,Cm,M,P,X)),O); | 28    | d    |
| 22   | ((L,M,X,(Cc,Cm)),O); | 59    | c    | 55   | ((L,M,X,(Cc,Cm,L,P)),O); | 27    | d    |
| 23   | ((Cc,Cm,P,(L,M,X)),O); | 57    | d    | 56   | ((L,(X,(Cc,Cm,M,P,X)),O); | 26    | d    |
| 24   | ((L,(Cc,Cm,L,P,X)),O); | 55    | d    | 57   | ((L,(Cc,Cm,L,P,X)),O); | 26    | d    |
| 25   | ((L,(X,(Cc,Cm,M,P)),O); | 55    | c    | 58   | ((L,(X,(Cc,Cm,M,P)),O); | 25    | d    |
| 26   | ((L,(M,X,(Cc,Cm,P)),O); | 50    | d    | 59   | ((L,(M,X,(Cc,Cm,P)),O); | 25    | d    |
| 27   | ((L,(X,(M,P,(Cc,Cm)),O); | 49    | DIS  | 60   | ((L,(X,(Cc,Cm,L,M,X),O); | 23    | d    |
| 28   | ((L,(X,(M,(Cc,Cm)),O); | 48    | d    | 61   | ((L,(X,(Cc,Cm,L,M,X),O); | 23    | d    |
| 29   | ((X,(L,(Cc,Cm,M,P)),O); | 46    | DIS  | 62   | ((L,(X,(Cc,Cm,M,P,X)),O); | 21    | d    |
| 30   | ((X,(P,(Cc,Cm),(L,M)),O); | 44    | DIS  | 63   | ((L,(X,(Cc,Cm,L,M,X)),O); | 21    | d    |
| 31   | ((L,(X,(Cc,Cm,M,P)),O); | 42    | d    | 64   | ((L,(X,(Cc,Cm,M,P,X)),O); | 20    | d*   |
| 32   | ((X,(L,(Cc,Cm,M,P)),O); | 41    | d    | 65   | ((L,(X,(Cc,Cm,M,P,X)),O); | 20    | d*   |
| 33   | ((L,(X,(Cc,Cm,L,M,X)),O); | 39    | d    | 66   | ((L,(X,(Cc,Cm,M,P,X)),O); | 18    | d*   |

Table S7. Gene trees with 200+ bp of fully occupied alignment. The tree concordant with the concatenated species tree is bolded. "c"=concordant with the concatenated species tree, but contains polytomies. "DIS"=discordant bifurcating trees. "d"=discordant trees with polytomies. "*"=discordance tree with Manacus and Ceratopipra monophyletic.
Table S8. Gene trees with 1000+ bp of fully occupied alignment. The tree concordant with the concatenated species tree is bolded. “c”=concordant with the concatenated species tree, but contains polytomies. “DIS”=discordant bifurcating trees. “d”=discordant trees with polytomies. “*”=discordance tree with *Manacus* and *Ceratopipra* monophyletic.
| gene             | log₂ fold change | log₂ average expression | t-statistic | P-value     | Adjusted P-value |
|------------------|------------------|-------------------------|-------------|-------------|------------------|
| **Mionectes oleagineus** |                  |                         |             |             |                  |
| TNNC1            | −4.94            | 9.87                    | −26.56      | 1.5 × 10⁻⁸  | 0.0002           |
| MYLK2            | 8.46             | 4.81                    | 13.88       | 1.6 × 10⁻⁶  | 0.0097           |
| ABHD2            | −1.67            | 9.83                    | −13.21      | 2.3 × 10⁻⁶  | 0.0097           |
| FANCI            | −2.87            | 3.91                    | −9.94       | 1.7 × 10⁻⁵  | 0.0526           |
| PITX1            | 1.95             | 2.96                    | 8.87        | 3.6 × 10⁻⁵  | 0.0824           |
| MYBPC1           | 6.92             | 5.36                    | 8.31        | 5.6 × 10⁻⁵  | 0.0824           |
| PITX3            | 2.89             | 4.45                    | 8.23        | 6.0 × 10⁻⁵  | 0.0824           |
| MFGE8            | −1.66            | 5.91                    | −8.1        | 6.6 × 10⁻⁵  | 0.0824           |
| FHL1             | 5.46             | 5.23                    | 8.05        | 6.9 × 10⁻⁵  | 0.0824           |
| EML4             | −1.39            | 4.7                     | −7.97       | 7.4 × 10⁻⁵  | 0.0824           |
| LOC113996532     | 5.19             | 0.91                    | 7.66        | 9.6 × 10⁻⁵  | 0.0824           |
| MYBPH            | −1.64            | 8.51                    | −7.62       | 9.9 × 10⁻⁵  | 0.0824           |
| ENAH             | 2.98             | 7.17                    | 7.54        | 1.0 × 10⁻⁴  | 0.0824           |
| HSPB1            | 1.6              | 9.22                    | 7.49        | 1.1 × 10⁻⁴  | 0.0824           |
| UCGG             | 1.33             | 5.63                    | 7.39        | 1.2 × 10⁻⁴  | 0.0824           |
| LOC113998929     | −1.81            | 4.36                    | −7.39       | 1.2 × 10⁻⁴  | 0.0824           |
| CUNH1orf21       | 1.22             | 5.97                    | 7.34        | 1.3 × 10⁻⁴  | 0.0824           |
| RAPGEF2          | 1.2              | 4.65                    | 7.28        | 1.3 × 10⁻⁴  | 0.0824           |
| LOC113990814     | 3.4              | 6.87                    | 7.28        | 1.3 × 10⁻⁴  | 0.0824           |
| SLC02B1          | 1.28             | 7.31                    | 7.27        | 1.3 × 10⁻⁴  | 0.0824           |
| HOMER2           | −1.48            | 6.06                    | −7.26       | 1.4 × 10⁻⁴  | 0.0824           |
| **Xenopipo atronitens** |                  |                         |             |             |                  |
| LOC113994916     | 5.35             | −1.8                    | 10.63       | 1.7 × 10⁻⁵  | 0.21             |
| LOC114000920     | 1.73             | 3.38                    | 9.39        | 3.8 × 10⁻⁵  | 0.24             |
| GJD2             | −2.42            | 2.16                    | −8.11       | 9.7 × 10⁻⁵  | 0.4              |
| ERICH3           | 2.72             | 0.09                    | 6.97        | 0.0002      | 0.66             |
| LOC114002263     | −1.48            | 3.3                     | −6.89       | 0.0003      | 0.66             |
| LOC113990672     | 2.18             | 3.21                    | 6.43        | 0.0004      | 0.74             |
| ST8SIA2          | −2.21            | 1.35                    | −6.39       | 0.0004      | 0.74             |
| UNC45B           | −0.61            | 8.62                    | −6.18       | 0.0005      | 0.74             |
| PRKCA            | 1.23             | 6.63                    | 6.06        | 0.0006      | 0.74             |
| TIGD4            | −2.39            | 2.58                    | −5.94       | 0.0006      | 0.74             |
| LOC114004148     | 2.71             | 0.32                    | 5.69        | 0.0008      | 0.74             |
| LOC113993176     | −1.54            | 3.68                    | −5.65       | 0.0009      | 0.74             |

*Table S9.* (continued on next page)
| gene            | log₂ fold change | log₂ average expression | t-statistic | P-value | Adjusted P-value |
|-----------------|------------------|-------------------------|-------------|---------|-----------------|
| **Lepidothrix coronata** |                  |                         |             |         |                 |
| TPM2            | 4.88             | 11.13                   | 21.31       | 2.1 x 10⁻⁷ | 1.5 x 10⁻³     |
| MYL2            | 4.79             | 5.44                    | 21.04       | 2.3 x 10⁻⁷ | 1.5 x 10⁻³     |
| LOC113983529    | 5.96             | 5.37                    | 20.15       | 3.1 x 10⁻⁷ | 1.5 x 10⁻³     |
| LOC113989700    | -3.32            | 5.87                    | -18.19      | 6.0 x 10⁻⁷ | 2.2 x 10⁻³     |
| LOC113999653    | 5.44             | 3.87                    | 15.47       | 1.7 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC113998630    | -4.71            | 3.24                    | -15.21      | 1.9 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC114002580    | -2.06            | 12.29                   | -14.62      | 2.5 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC113989355    | 3.12             | 4.36                    | 14.5        | 2.7 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC113983388    | 3.16             | 4.43                    | 14.44       | 2.7 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC113999652    | 7.28             | 7.67                    | 14.31       | 2.9 x 10⁻⁶ | 3.9 x 10⁻³     |
| KIAA2026        | -2.01            | 6.75                    | -14.27      | 2.9 x 10⁻⁶ | 3.9 x 10⁻³     |
| TNNT1           | 4.19             | 5.15                    | 13.06       | 5.2 x 10⁻⁶ | 5.7 x 10⁻³     |
| **Manacus vitellinus** |            |                         |             |         |                 |
| TPM2            | 4.88             | 11.13                   | 21.31       | 2.1 x 10⁻⁷ | 1.5 x 10⁻³     |
| MYL2            | 4.79             | 5.44                    | 21.04       | 2.3 x 10⁻⁷ | 1.5 x 10⁻³     |
| LOC113983529    | 5.96             | 5.37                    | 20.15       | 3.1 x 10⁻⁷ | 1.5 x 10⁻³     |
| LOC113989700    | -3.32            | 5.87                    | -18.19      | 6.0 x 10⁻⁷ | 2.2 x 10⁻³     |
| LOC113999653    | 5.44             | 3.87                    | 15.47       | 1.7 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC113998630    | -4.71            | 3.24                    | -15.21      | 1.9 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC114002580    | -2.06            | 12.29                   | -14.62      | 2.5 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC113989355    | 3.12             | 4.36                    | 14.5        | 2.7 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC113983388    | 3.16             | 4.43                    | 14.44       | 2.7 x 10⁻⁶ | 3.9 x 10⁻³     |
| LOC113999652    | 7.28             | 7.67                    | 14.31       | 2.9 x 10⁻⁶ | 3.9 x 10⁻³     |
| KIAA2026        | -2.01            | 6.75                    | -14.27      | 2.9 x 10⁻⁶ | 3.9 x 10⁻³     |
| TNNT1           | 4.19             | 5.15                    | 13.06       | 5.2 x 10⁻⁶ | 5.7 x 10⁻³     |
| **Pseudopipra pipra** |            |                         |             |         |                 |
| MYLK2           | 8.5              | 4.87                    | 35.75       | 2.5 x 10⁻⁹ | 3.07 x 10⁻⁵    |
| LOC114002581    | 4.23             | 10.75                   | 31.17       | 6.6 x 10⁻⁹ | 4.07 x 10⁻⁵    |
| MYBPC1          | 7.36             | 5.66                    | 28.01       | 1.4 x 10⁻⁸ | 5.79 x 10⁻⁵    |
| TPM2            | 6.8              | 8.65                    | 26.23       | 2.2 x 10⁻⁸ | 6.93 x 10⁻⁵    |
| TNNC1           | -5.5             | 9.78                    | -24.73      | 3.4 x 10⁻⁸ | 8.41 x 10⁻⁵    |
| FHL1            | 4.75             | 6.11                    | 18.87       | 2.3 x 10⁻⁷ | 0.00047        |
| NOS1            | 2.8              | 4.37                    | 17.46       | 4.0 x 10⁻⁷ | 0.0007         |
| TMOD1           | 5.47             | 5.67                    | 15.72       | 8.3 x 10⁻⁷ | 0.00128        |
| TSTD2           | 3.08             | 6.34                    | 14.22       | 1.7 x 10⁻⁶ | 0.00227        |
| AGBL1           | 5.97             | 1.38                    | 13.66       | 2.2 x 10⁻⁶ | 0.00271        |
| SMPX            | 4.33             | 4.01                    | 13.16       | 2.8 x 10⁻⁶ | 0.00317        |
| CNTNAP2         | 3.09             | 3.14                    | 11.86       | 5.8 x 10⁻⁶ | 0.00597        |

Table S9. (continued on next page)
| gene             | log2 fold change | log2 average expression | t-statistic | P-value     | Adjusted P-value |
|------------------|------------------|-------------------------|-------------|-------------|------------------|
| Ceratopipra cornuta |                 |                         |             |             |                  |
| TNNC1            | −8.16            | 8.36                    | −49.29      | 4.1 × 10⁻¹¹ | 3.91 × 10⁻⁷      |
| LOC114002581     | 5.4              | 11.14                   | 46.64       | 6.3 × 10⁻¹¹ | 3.91 × 10⁻⁷      |
| TPM2             | 7.54             | 8.62                    | 37.81       | 3.3 × 10⁻¹⁰ | 1.36 × 10⁻⁶      |
| TMOD1            | 6.21             | 6.01                    | 35.94       | 4.9 × 10⁻¹⁰ | 1.52 × 10⁻⁶      |
| TSTD2            | 3.36             | 6.37                    | 29.79       | 2.1 × 10⁻⁹  | 5.29 × 10⁻⁶      |
| MYBPC1           | 7.92             | 5.68                    | 26.8        | 4.9 × 10⁻⁹  | 1.01 × 10⁻⁵      |
| LOC113998858     | −2.38            | 6.87                    | −22.9       | 1.7 × 10⁻⁹  | 2.81 × 10⁻⁵      |
| LOC113990140     | 3.08             | 5.43                    | 22.32       | 2.0 × 10⁻⁸  | 2.81 × 10⁻⁵      |
| CDC177           | 2.63             | 5.31                    | 22.31       | 2.0 × 10⁻⁸  | 2.81 × 10⁻⁵      |
| FHL1             | 4.38             | 5.9                     | 21.02       | 3.3 × 10⁻⁸  | 4.01 × 10⁻⁵      |
| MB               | −1.66            | 12.55                   | −18.57      | 8.5 × 10⁻⁸  | 9.53 × 10⁻⁵      |
| SMPX             | 5.18             | 3.39                    | 18.23       | 9.8 × 10⁻⁸  | 1.01 × 10⁻⁴      |
| Ceratopipra mentalis |            |                         |             |             |                  |
| MYLK2            | 6.28             | 6.03                    | 29.49       | 9.4 × 10⁻⁹  | 1.3 × 10⁻⁴       |
| MYBPC1           | 7.53             | 7.37                    | 21.27       | 9.6 × 10⁻⁸  | 0.00064          |
| TPM2             | 7.73             | 9.54                    | 16.48       | 5.8 × 10⁻⁷  | 0.00256          |
| LOC113990140     | 2.62             | 6.27                    | 13.94       | 1.9 × 10⁻⁶  | 0.00469          |
| CLDN1            | 3.31             | 4.09                    | 13.85       | 1.9 × 10⁻⁶  | 0.00469          |
| CRIK1            | 2.45             | 4.63                    | 13.49       | 2.3 × 10⁻⁶  | 0.00469          |
| ZIC1             | 6.57             | −0.71                   | 13.26       | 2.6 × 10⁻⁶  | 0.00469          |
| TMOD1            | 4.57             | 7.17                    | 13.13       | 2.8 × 10⁻⁶  | 0.00469          |
| LOC114000838     | −3.57            | 2.73                    | −12.68      | 3.6 × 10⁻⁶  | 0.00514          |
| PPP3CA           | 2.33             | 5.02                    | 12.54       | 3.9 × 10⁻⁶  | 0.00514          |
| SV2B             | 3.15             | 4.98                    | 12.05       | 5.1 × 10⁻⁶  | 0.00617          |
| SIM2             | −3.72            | 3.42                    | −11.73      | 6.1 × 10⁻⁶  | 0.00678          |

Table S9. Top differentially expressed genes in an SH-PEC contrast for each species. List of top genes (by P-value) identified by limma as differentially expressed for a comparison of the SH and PEC tissues (three replicates of each).
| GO Term                  | Definition                                      | P-value          | Adj. P-value | Study | Family | Population |
|-------------------------|-------------------------------------------------|------------------|--------------|-------|--------|------------|
| **Mionectes oleagineus**|                                                 |                  |              |       |        |            |
| GO:0032971              | regulation of muscle filament sliding           | $3.00 \times 10^{-5}$ | 0.01         | 2 / 4 | 2 / 451 | 3 / 14776  |
| GO:0032972              | regulation of muscle filament sliding speed      | $6.30 \times 10^{-4}$ | 0.08         | 1 / 4 | 2 / 3190 | 1 / 14776  |
| GO:1903115              | regulation of actin filament-based movement      | 0.003            | 0.23         | 2 / 4 | 3 / 1100 | 36 / 14776 |
| GO:0051270              | regulation of cellular component movement        | 0.004            | 0.23         | 3 / 4 | 4 / 7906 | 836 / 14776|
| GO:0048644              | muscle organ morphogenesis                       | 0.004            | 0.23         | 2 / 4 | 2 / 1004 | 67 / 14776 |
| GO:0048240              | sperm capacitation                               | 0.008            | 0.26         | 1 / 4 | 1 / 727  | 6 / 14776  |
| GO:0007517              | muscle organ development                         | 0.011            | 0.26         | 2 / 4 | 2 / 2684 | 276 / 14776|
| GO:0060415              | muscle tissue morphogenesis                       | 0.011            | 0.26         | 2 / 4 | 2 / 597  | 63 / 14776 |
| GO:0030048              | actin filament-based movement                    | 0.011            | 0.26         | 2 / 4 | 3 / 2182 | 137 / 14776|
| GO:0032970              | regulation of actin filament-based process       | 0.011            | 0.26         | 2 / 4 | 4 / 7724 | 347 / 14776|
| GO:0030049              | muscle filament sliding                           | 0.011            | 0.26         | 2 / 4 | 2 / 304  | 33 / 14776 |
| GO:0032386              | regulation of intracellular transport            | 0.012            | 0.26         | 2 / 4 | 2 / 2799 | 309 / 14776|
| **Xenopipo atronitensis**|                                                 |                  |              |       |        |            |
|                         | N/A                                             |                  |              |       |        |            |
| **Lepidothrix coronata**|                                                 |                  |              |       |        |            |
| GO:0032971              | regulation of muscle filament sliding           | $3.00 \times 10^{-5}$ | 0.01         | 2 / 4 | 2 / 451 | 3 / 14776  |
| GO:0032972              | regulation of muscle filament sliding speed      | $6.30 \times 10^{-4}$ | 0.08         | 1 / 4 | 2 / 3190 | 1 / 14776  |
| GO:1903115              | regulation of actin filament-based movement      | 0.003            | 0.23         | 2 / 4 | 3 / 1100 | 36 / 14776 |
| GO:0051270              | regulation of cellular component movement        | 0.004            | 0.23         | 3 / 4 | 4 / 7906 | 836 / 14776|
| GO:0048644              | muscle organ morphogenesis                       | 0.004            | 0.23         | 2 / 4 | 2 / 1004 | 67 / 14776 |
| GO:0048240              | sperm capacitation                               | 0.008            | 0.26         | 1 / 4 | 1 / 727  | 6 / 14776  |
| GO:0007517              | muscle organ development                         | 0.011            | 0.26         | 2 / 4 | 2 / 2684 | 276 / 14776|
| GO:0060415              | muscle tissue morphogenesis                       | 0.011            | 0.26         | 2 / 4 | 2 / 597  | 63 / 14776 |
| GO:0030048              | actin filament-based movement                    | 0.011            | 0.26         | 2 / 4 | 3 / 2182 | 137 / 14776|
| GO:0032970              | regulation of actin filament-based process       | 0.011            | 0.26         | 2 / 4 | 4 / 7724 | 347 / 14776|
| GO:0030049              | muscle filament sliding                           | 0.011            | 0.26         | 2 / 4 | 2 / 304  | 33 / 14776 |
| GO:0032386              | regulation of intracellular transport            | 0.012            | 0.26         | 2 / 4 | 2 / 2799 | 309 / 14776|

Table S10. (continued on next page)
| GO Term | Definition | P-value | Adj. P | Study | Family | Population |
|---------|------------|---------|--------|-------|--------|------------|
| **Manacus vitellinus** | | | | | | |
| GO:0030239 | myofibril assembly | 7.5 × 10⁻⁷ | 5.5 × 10⁻⁴ | 6 / 66 | 10 / 1340 | 56 / 14776 |
| GO:0030048 | actin filament-based movement | 8.8 × 10⁻⁷ | 5.5 × 10⁻⁴ | 8 / 66 | 15 / 2182 | 137 / 14776 |
| GO:0010927 | cellular component assembly involved in morphogenesis | 8.9 × 10⁻⁵ | 5.5 × 10⁻⁴ | 6 / 66 | 15 / 3419 | 86 / 14776 |
| GO:0055001 | muscle cell development | 2.5 × 10⁻⁶ | 0.0011 | 7 / 66 | 10 / 1778 | 149 / 14776 |
| GO:0003012 | muscle system process | 1.0 × 10⁻⁵ | 0.0038 | 14 / 66 | 18 / 1407 | 384 / 14776 |
| GO:0003004 | muscle structure development | 2.8 × 10⁻⁵ | 0.0085 | 11 / 66 | 24 / 4364 | 506 / 14776 |
| GO:0003004 | muscle filament sliding | 1.9 × 10⁻⁴ | 0.0461 | 7 / 66 | 14 / 304 | 33 / 14776 |
| GO:0003002 | actin filament-based process | 2.0 × 10⁻⁴ | 0.0461 | 11 / 66 | 51 / 11575 | 705 / 14776 |
| GO:0003002 | muscle cell differentiation | 3.4 × 10⁻⁴ | 0.0698 | 8 / 66 | 21 / 3154 | 292 / 14776 |
| GO:1903115 | system process | 5.5 × 10⁻⁴ | 0.092 | 18 / 66 | 33 / 5318 | 1407 / 14776 |
| GO:0030048 | regulation of membrane depolarization | 5.7 × 10⁻⁴ | 0.092 | 3 / 66 | 32 / 7750 | 40 / 14776 |
| GO:0090131 | mesenchyme migration | 6.0 × 10⁻⁴ | 0.092 | 2 / 66 | 3 / 316 | 5 / 14776 |
| **Pseudopipra pipra** | | | | | | |
| GO:0030048 | actin filament-based movement | 4.3 × 10⁻⁷ | 6.8 × 10⁻⁴ | 8 / 43 | 14 / 2182 | 137 / 14777 |
| GO:0032501 | multicellular organismal process | 1.5 × 10⁻⁶ | 0.0012 | 32 / 43 | 39 / 11976 | 5318 / 14777 |
| GO:0003008 | system process | 8.4 × 10⁻⁵ | 0.044 | 19 / 43 | 32 / 5318 | 1407 / 14776 |
| GO:0030029 | actin filament-based process | 1.8 × 10⁻⁴ | 0.062 | 9 / 43 | 35 / 11575 | 705 / 14776 |
| GO:0003012 | muscle system process | 2.0 × 10⁻⁴ | 0.063 | 13 / 43 | 19 / 1407 | 384 / 14776 |
| GO:1903115 | regulation of actin filament-based movement | 4.0 × 10⁻⁴ | 0.103 | 4 / 43 | 12 / 1100 | 36 / 14777 |
| GO:0044057 | regulation of system process | 4.6 × 10⁻⁴ | 0.103 | 11 / 43 | 22 / 2764 | 483 / 14777 |
| GO:0006928 | movement of cell or subcellular component | 7.3 × 10⁻⁴ | 0.143 | 14 / 43 | 35 / 11575 | 1897 / 14777 |
| GO:0030049 | muscle filament sliding | 0.001 | 0.17 | 6 / 43 | 13 / 304 | 33 / 14777 |
| GO:0032971 | regulation of muscle filament sliding | 0.0013 | 0.17 | 2 / 43 | 10 / 451 | 3 / 14777 |
| GO:0032879 | regulation of localization | 0.0013 | 0.17 | 15 / 43 | 30 / 9414 | 2201 / 14777 |
| GO:0035637 | multicellular organismal signalling | 0.0013 | 0.17 | 5 / 43 | 35 / 6943 | 166 / 14777 |

Table S10. (continued on next page)
| GO Term                                                                 | Definition                                                                 | P-value       | Adj. P      | Study       | Family     | Population   |
|------------------------------------------------------------------------|-----------------------------------------------------------------------------|---------------|-------------|-------------|------------|--------------|
| **Ceratopipra cornuta**                                                 |                                                                             |               |             |             |            |              |
| GO:0030048 actin filament-based movement                                |                                                                             | $4.3 \times 10^{-7}$ | $6.8 \times 10^{-4}$ | 8 / 43     | 14 / 2182  | 137 / 14777  |
| GO:0032501 multicellular organismal process                            |                                                                             | $1.5 \times 10^{-6}$ | 0.0012      | 32 / 43     | 39 / 11976 | 5318 / 14777 |
| GO:0003008 system process                                              |                                                                             | $8.4 \times 10^{-5}$ | 0.044       | 19 / 43     | 32 / 5318  | 1407 / 14777 |
| GO:0030029 actin filament-based process                                |                                                                             | $1.8 \times 10^{-4}$ | 0.062       | 9 / 43      | 35 / 11575 | 705 / 14777  |
| GO:0003012 muscle system process                                       |                                                                             | $2.0 \times 10^{-4}$ | 0.063       | 13 / 43     | 19 / 1407  | 384 / 14777  |
| GO:1903115 regulation of actin filament-based movement                 |                                                                             | $4.0 \times 10^{-4}$ | 0.103       | 4 / 43      | 12 / 1100  | 36 / 14777   |
| GO:0044057 regulation of system process                                |                                                                             | $4.6 \times 10^{-4}$ | 0.103       | 11 / 43     | 22 / 2764  | 483 / 14777  |
| GO:0006928 movement of cell or subcellular component                   |                                                                             | $7.3 \times 10^{-4}$ | 0.143       | 14 / 43     | 35 / 11575 | 1897 / 14777 |
| GO:0030049 muscle filament sliding                                     |                                                                             | 0.001         | 0.17        | 6 / 43      | 13 / 304   | 33 / 14777   |
| GO:0032971 regulation of muscle filament sliding                        |                                                                             | 0.0013        | 0.17        | 2 / 43      | 10 / 451   | 3 / 14777    |
| GO:0032879 regulation of localization                                  |                                                                             | 0.0013        | 0.17        | 15 / 43     | 30 / 9414  | 2201 / 14777 |
| GO:0035637 multicellular organismal signaling                           |                                                                             | 0.0013        | 0.17        | 5 / 43      | 35 / 6943  | 166 / 14777  |
| **Ceratopipra mentalis**                                               |                                                                             |               |             |             |            |              |
| GO:0006928 movement of cell or subcellular component                   |                                                                             | $4.5 \times 10^{-5}$ | 0.052       | 16 / 40     | 35 / 11575 | 1897 / 14776 |
| GO:0030049 muscle filament sliding                                     |                                                                             | $6.3 \times 10^{-5}$ | 0.052       | 5 / 40      | 6 / 304    | 33 / 14776   |
| GO:0030048 actin filament-based movement                                |                                                                             | $3.8 \times 10^{-4}$ | 0.213       | 6 / 40      | 17 / 2182  | 137 / 14776  |
| GO:0030029 actin filament-based process                                |                                                                             | $9.8 \times 10^{-4}$ | 0.305       | 8 / 40      | 35 / 11575 | 705 / 14776  |
| GO:0052363 catabolism by organism of protein in other organism involved in symbiotic interaction |                                                                             | 0.0013        | 0.305       | 1 / 40      | 1 / 797    | 1 / 14776    |
| GO:0052361 catabolism by organism of macromolecule in other organism involved in symbiotic interaction |                                                                             | 0.0016        | 0.305       | 1 / 40      | 2 / 1219   | 1 / 14776    |
| GO:0009611 response to wounding                                        |                                                                             | 0.0017        | 0.305       | 6 / 40      | 9 / 2907   | 523 / 14776  |
| GO:0033275 actin-myosin filament sliding                                |                                                                             | 0.002         | 0.305       | 5 / 40      | 5 / 109    | 33 / 14776   |
| GO:0052214 metabolism of substance in other organism involved in symbiotic interaction |                                                                             | 0.0021        | 0.305       | 1 / 40      | 3 / 1441   | 1 / 14776    |
| GO:0044035 multi-organism catabolic process                             |                                                                             | 0.0022        | 0.305       | 1 / 40      | 5 / 2242   | 1 / 14776    |
| GO:0052418 metabolism by organism of protein in other organism involved in symbiotic interaction |                                                                             | 0.0022        | 0.305       | 1 / 40      | 10 / 4484  | 1 / 14776    |
| GO:0003012 muscle system process                                       |                                                                             | 0.0023        | 0.305       | 7 / 40      | 9 / 1407   | 384 / 14776  |

Table S10. SH-PEC Pairwise DGE Gene Ontology. Top Gene Ontology terms from the differential expressed genes between SH and PEC for each species, showing the $P$-value of overrepresentation, the BH-adjusted $P$-value, the proportion of genes represented by the term in the target study set, the number of terms in the total GO term family, and the total proportion of the GO term in the total population.
| Gene                | Max cpm | Best Model | M6 model BF (SH) | Best Model | M6 model BF (PEC) | Annotation                                                                 |
|---------------------|---------|------------|------------------|------------|------------------|---------------------------------------------------------------------------|
| **M6 Ancestor Branch Model – SH and PEC**                  |         |            |                  |            |                  |                                                                           |
| LRSAM1              | 109.3   | M6         | 1.79 (+)         | M6         | 1.92 (+)         | leucine rich repeat and sterile alpha motif containing 1                 |
| LOC114000536        | 183.2   | M6         | 1.72 (+)         | M6         | 2.13 (+)         | uncharacterized LOC114000536                                             |
| PKP                 | 249.1   | M6         | 1.6 (+)          | M6         | 1.63 (+)         | phosphofructokinase, platelet                                            |
| **M6 Ancestor Branch Model – SH**                          |         |            |                  |            |                  |                                                                           |
| LOC113995244        | 17.2    | M6         | 1.82 (+)         | M6         | 1.48 (+)         | uncharacterized LOC113995244                                            |
| CIAPIN1             | 1255    | M6         | 1.71 (+)         | M6         | 0.91 (+)         | cytokine induced apoptosis inhibitor 1                                    |
| HARBII              | 174.8   | M6         | 1.70 (+)         | M6         | 1.22 (+)         | harbinger transposase derived 1                                           |
| YARS2               | 108.1   | M6         | 1.67 (+)         | M6         | 0.59 (+)         | tyrosyl-tRNA synthetase 2                                                |
| PDK2                | 1865    | M6         | 1.63 (+)         | M6         | 1.11 (+)         | pyruvate dehydrogenase kinase 2                                           |
| TMED5               | 241.7   | M6         | 1.63 (+)         | M6         | 0.92 (+)         | transmembrane p24 trafficking protein 5                                   |
| **M6 Ancestor Branch Model – PEC**                         |         |            |                  |            |                  |                                                                           |
| LOC113986517        | 116.3   | M6         | 1.53 (+)         | M6         | 0.37 (+)         | alpha-soluble NSF attachment protein-like                                 |
| LOC113988691        | 6.8     | M6         | 1.6 (+)          | M6         | 1.38 (+)         | transmem. 9 superfamily member 2-like                                     |
| SLC25A19            | 172.5   | M6         | 1.59 (+)         | M6         | 0.96 (+)         | solute carrier family 25 member 19                                       |
| LOC113993369        | 1387    | M6         | 1.57 (+)         | M6         | 1.41 (+)         | lysine-specific demethylase 5C-like                                       |
| IGBP2               | 115.6   | M6         | 1.54 (+)         | M6         | 1.19 (+)         | insulin like growth factor binding prtn. 2                               |
| CIB2                | 326.7   | M6         | 1.53 (+)         | M6         | 1.19 (+)         | calcium and integrin binding family member 2                              |
| LOC113991411        | 430.6   | M6         | 1.46 (+)         | M6         | 2.06 (+)         | malignant fibrous histiocytoma-amplified sequence 1 homolog              |
| LOC113990764        | 131.6   | M6         | 1.01 (+)         | M6         | 1.93 (+)         | Schwann cell myelin protein-like                                          |
| LOC113988445        | 21.2    | M6         | 0.72 (+)         | M6         | 1.83 (+)         | C-type lectin dmn. fam. 2 member B-like                                   |
| LOC113989785        | 5.5     | LC         | −0.15 (–)        | M6         | 1.80 (–)         | von Willebrand factor A domain-containing protein 5A-like                |
| LOC113990255        | 620.8   | M6         | 0.58 (+)         | M6         | 1.76 (+)         | 6-phosphofructo-2-kinase/fructose-2,6-bisphosphatase                      |
| ZNF532              | 70.2    | M6         | 1.42 (+)         | M6         | 1.72 (+)         | zinc finger protein 532                                                  |
| LOC113992038        | 439.2   | M6         | 0.91 (+)         | M6         | 1.71 (–)         | uncharacterized LOC113992038                                             |
| MRPL34              | 77      | null       | −0.05 (–)        | M6         | 1.7 (+)          | mitochondrial ribosomal protein L34                                      |
| LOC113994638        | 13.4    | M6         | 1.02 (+)         | M6         | 1.67 (+)         | uncharacterized LOC113994638                                             |
| LOC113989700        | 278     | M6         | 0.05 (+)         | M6         | 1.67 (+)         | excitatory amino acid transporter 4-like                                 |
| NEFL                | 815.9   | LC         | −0.29 (–)        | M6         | 1.66 (+)         | neurofilament light                                                     |
| TRMT10B             | 24      | M6         | 0.79 (+)         | M6         | 1.66 (+)         | tRNA methyltransferase 10B                                                |
| LOC113994118        | 347.6   | M6         | 1.32 (+)         | M6         | 1.65 (+)         | laminin subunit beta-2-like                                              |
| SLC27A3             | 16.4    | M6         | 1.13 (+)         | M6         | 1.64 (+)         | solute carrier family 27 member 3                                       |
| LOC113995895        | 51      | M6         | 0.9 (+)          | M6         | 1.63 (+)         | F-box only protein 6-like                                                |
| PPIC                | 95.1    | M6         | 1.46 (+)         | M6         | 1.61 (+)         | peptidylprolyl isomerase C                                               |
| MBLAC2              | 127.9   | M6         | 0.71 (–)         | M6         | 1.60 (+)         | metallo-beta-lactamase dmn. containing 2                                 |
| CLPX                | 534.2   | M6         | 0.94 (+)         | M6         | 1.54 (+)         | caseinolytic mitochondrial matrix peptidase chaperone subunit            |
| OPRL1               | 86.6    | M6         | 0.61 (–)         | M6         | 1.54 (–)         | opioid related nociceptin receptor 1                                     |
| PLA2G12A            | 369.7   | M6         | 1.17 (–)         | M6         | 1.54 (–)         | phospholipase A2 group XIIA                                              |
| LOC113999006        | 18.3    | M6         | 0.52 (+)         | M6         | 1.54 (+)         | uncharacterized LOC113999006                                            |
| AMBRA1              | 140.9   | M6         | 0.84 (–)         | M6         | 1.53 (–)         | autophagy and beclin 1 regulator 1                                       |

Table S11. (continued on next page)
| Gene          | Max cpm | Best Model SH | M5 model BF (SH) | Best Model PEC | M5 model BF (PEC) | Annotation                                      |
|--------------|---------|---------------|-----------------|---------------|-----------------|------------------------------------------------|
| LOC113986884 | 5       | M5            | 2.17 (+)        | M5            | 1.62 (+)        | uncharacterized LOC113986884                   |
| SLC44A3      | 694.7   | M5            | 1.87 (+)        | M5            | 1.74 (+)        | solute carrier family 44 member 3             |
| LOC113991394 | 10.2    | M5            | 1.84 (+)        | M5            | 1.64 (+)        | multidrug resistance protein 1-like           |

**M5 Ancestor Branch Model – SH**

| Gene       | Max cpm | Best Model SH | M5 model BF (SH) | Best Model PEC | M5 model BF (PEC) | Annotation                                      |
|------------|---------|---------------|-----------------|---------------|-----------------|------------------------------------------------|
| CCDC15     | 32.3    | M5            | 2.02 (+)        | M4            | 0.37 (+)        | coiled-coil domain containing 15              |
| LOC113993297 | 16.3   | M5            | 1.98 (+)        | M5            | 1.03 (+)        | syncytin-2-like                                |
| CACFD1     | 29      | M5            | 1.91 (+)        | M4            | 0.74 (+)        | calcium channel flower domain containing 1    |
| LAPTMB4    | 100.1   | M5            | 1.79 (+)        | M5            | 1.16 (+)        | lysosomal protein transmembrane 4 beta        |
| EAF2       | 16.2    | M5            | 1.77 (+)        | M5            | 1.09 (+)        | ELL associated factor 2                       |
| LOC113990152 | 49.1   | M5            | 1.73 (+)        | M5            | 1.11 (+)        | uncharacterized LOC113990152                 |
| LOC113983157 | 5.2    | M5            | 1.71 (+)        | M5            | 0.86 (+)        | collagen alpha-1(VII) chain-like             |
| RETREG1    | 1638.6  | M5            | 1.65 (+)        | M6            | 0.14 (+)        | reticulophagy regulator 1                    |

**M5 Ancestor Branch Model – PEC**

| Gene      | Max cpm | Best Model SH | M5 model BF (SH) | Best Model PEC | M5 model BF (PEC) | Annotation                                      |
|-----------|---------|---------------|-----------------|---------------|-----------------|------------------------------------------------|
| FOXO3     | 1324.3  | M5            | 1.59 (-)        | M5            | 0.89 (-)        | forkhead box O3                                |
| LOC113991453 | 13.2   | M5            | 1.56 (+)        | M5            | 0.26 (+)        | semaphorin-3D-like                             |
| DHPS      | 13.5    | M5            | 1.55 (-)        | M5            | 1.37 (-)        | deoxyhypusine synthase                        |
| PTPRCAP   | 371.2   | M5            | 1.54 (-)        | M6            | 0.35 (-)        | protein tyrosine phosphatase, receptor type C associated protein |
| FOXF1     | 26      | M5            | 1.53 (+)        | M4            | 0.45 (+)        | forkhead box F1                               |
| ADAMTS9   | 78.2    | M5            | 1.53 (+)        | M5            | 1.21 (+)        | ADAM metallopeptidase with thrombospondin type 1 motif 9 |
| UBE3B     | 121.9   | M5            | 1.46 (-)        | M5            | 1.44 (-)        | ubiquitin protein ligase E3B                   |
| LOC113994576 | 183.4  | M5            | 1.22 (-)        | M5            | 1.93 (-)        | ATP-dependent RNA helicase DDX19B             |
| LOC113984085 | 94.5   | M4            | 0.56 (+)        | M5            | 1.79 (+)        | uncharacterized LOC113984085                 |
| COMMD2    | 130.8   | M5            | 1.39 (+)        | M5            | 1.63 (+)        | COMM domain containing 2                      |
| TOMM5     | 45.8    | M5            | 0.76 (+)        | M5            | 1.59 (+)        | translocase of outer mitochondrial membrane 5 |

Table S11. (continued on next page)
| Gene             | Max cpm | Best Model SH | M4 model BF (SH) | Best Model PEC | M4 model BF (PEC) | Annotation                                           |
|------------------|---------|---------------|------------------|----------------|------------------|-----------------------------------------------------|
| LOC113998187     | 449.7   | M4            | 3.57 (+)         | M4             | 4.61 (+)         | uncharacterized LOC113998187                        |
| LOC113989298     | 20.8    | M4            | 2.65 (+)         | M4             | 2.97 (+)         | uncharacterized LOC113989298                        |
| DPYSL5           | 426.5   | M4            | 2.18 (+)         | M4             | 3.44 (+)         | dihydropyrimidinase like 5                          |
| LOC113983437     | 5       | M4            | 1.83 (+)         | M4             | 1.42 (+)         | uncharacterized LOC113983437                        |
| PSMA7            | 186.6   | M4            | 1.79 (+)         | M4             | 1.34 (+)         | proteasome subunit alpha 7                          |
| ZNF395           | 20.6    | M4            | 1.78 (–)         | M4             | 0.85 (–)         | zinc finger protein 395                             |
| STUM             | 131.4   | M4            | 1.78 (+)         | M5             | 0.17 (+)         | stom, mechanosensory transduction mediator homolog   |
| FBXO4            | 20      | M4            | 1.71 (–)         | M4             | 0.52 (–)         | F-box protein 4                                     |
| FAM169B          | 34.9    | M4            | 1.71 (+)         | M4             | 0.5 (+)          | family with sequence similarity 169 member B        |
| LOC114003417     | 12409   | M4            | 1.66 (+)         | M4             | 0.3 (+)          | sarcoplasmic/endoplasmic reticulum calcium ATPase 1 |
| GRM5             | 60.5    | M4            | 1.58 (–)         | M5             | 0.06 (–)         | glutamate metabotropic receptor 5                   |
| KDM4B            | 38.2    | M4            | 1.57 (+)         | M5             | 0.11 (+)         | lysine demethylase 4B                               |
| LOC113996660     | 20.7    | M4            | 1.57 (–)         | M4             | 0.34 (–)         | mothers against decapentaplegic homolog 4-like       |
| CPS1             | 41.9    | M4            | 1.55 (+)         | M4             | 0.04 (+)         | carbamoyl-phosphate synthase 1                      |
| LOC113998417     | 19.1    | M4            | 1.55 (–)         | M5             | 0.11 (–)         | biogenesis of lysosome-related organelles complex 1 subunit 1-like |
| ATP2A3           | 10344   | M4            | 1.5 (+)          | M4             | 0.93 (+)         | ATPase sarcoplasmic/endoplasmic reticulum Ca2+ transporting 3 |
| LOC113992280     | 12.2    | M4            | 1.5 (–)          | PP             | –0.3 (+)         | alpha-1-antiproteinase 2-like                       |
| PSMA2            | 158.6   | M4            | 1.43 (+)         | MV             | 0.39 (+)         | proteasome subunit alpha 2                          |
| DDRGK1           | 218.2   | M4            | 1.42 (+)         | M6             | 0.03 (+)         | DDRGK domain containing 1                           |
| RPAP2            | 52.3    | M4            | 1.42 (–)         | M4             | 0.88 (–)         | RNA polymerase II associated protein 2               |
| LOC113986682     | 30.5    | LC            | –0.42 (–)        | M4             | 2 (–)            | double-headed protease inhibitor, submandibular gland-like |
| SGCE             | 105.3   | null          | –0.49 (–)        | M4             | 1.9 (–)          | sarcoglycan epsilon                                 |
| PIGR             | 40.9    | LC            | 0 (–)            | M4             | 1.8 (–)          | polymeric immunoglobulin receptor                   |
| ZCCHC9           | 23.6    | M4            | 0.28 (–)         | M4             | 1.73 (–)         | zinc finger CCHC-type containing 9                   |
| CIAO2B           | 133.8   | LC            | 0.07 (+)         | M4             | 1.69 (+)         | cytosolic iron-sulfur assembly component 2B          |
| LOC113982339     | 7       | LC            | 0.34 (–)         | M4             | 1.66 (–)         | 1,25-dihydroxyvitamin D(3) 24-hydroxylase, mitochondrial |
| SNX4             | 71.3    | M4            | 0.77 (–)         | M4             | 1.66 (–)         | sorting nexin 4                                    |
| HPSE2            | 35.7    | M4            | 0.97 (+)         | M4             | 1.64 (+)         | heparanase 2 (inactive)                             |
| KCNH7            | 22.5    | PP            | –0.26 (–)        | M4             | 1.62 (–)         | potassium voltage-gated channel subfamily H member 7 |
| LOC113989406     | 7.9     | null          | –0.1 (–)         | M4             | 1.54 (–)         | alpha-2-macroglobulin-like protein 1                |

Table S11. (continued on next page)
| Gene          | Max cpm | Best Model SH | R41 model BF (SH) | Best Model PEC | R41 model BF (PEC) | Annotation                                                                 |
|--------------|---------|---------------|-------------------|----------------|-------------------|-----------------------------------------------------------------------------|
| LOC113998963 | 38.8    | R41           | 1.62 (+/−)        | PP             | 0.71 (+/−)        | uncharacterized LOC113998963                                             |
| LOC113989513 | 15.5    | R41           | 1.6 (+/−)         | null           | −0.89 (+/+        | mucin-5B                                                                   |
| UBA2         | 77      | R41           | 1.59 (+/−)        | PP             | 0.89 (+/−)        | UBA domain containing 2                                                    |
| LOC113999394 | 31.2    | R41           | 1.58 (+/−)        | M4             | 0.53 (+/−)        | alpha-globin transcription factor CP2-like                                  |
| LOC113994824 | 564.7   | R41           | 1.4 (+/−)         | null           | −0.29 (+/−)       | ryanodine receptor 1-like                                                  |
| SPPL2A       | 174.3   | R41           | 1.23 (+/−)        | null           | −0.98 (+/−)       | signal peptide peptidase like 2A                                           |
| CA2          | 367     | R41           | 1.15 (+/−)        | PP             | 0.56 (+/+)        | carbonic anhydrase 2                                                       |
| CUL1         | 110.6   | R41           | 1.15 (+/−)        | MV             | −0.56 (+/−)       | cullin 1                                                                   |
| LOC114001935 | 794.4   | R41           | 1.12 (+/−)        | null           | −0.62 (+/−)       | ryanodine receptor 1-like                                                  |
| MPHOSPH8     | 92.9    | R41           | 1.08 (+/−)        | M4             | −0.3 (+/−)        | M-phase phosphoprotein 8                                                  |
| SNAPIN       | 42.1    | R41           | 1 (+/−)           | R41            | 0.59 (+/−)        | SNAP associated protein                                                   |
| NRDE2        | 54.2    | R41           | 0.89 (+/−)        | CE             | −0.01 (+/−)       | NRDE-2, necessary for RNA interference, domain containing                  |
| PSMC3        | 336.3   | R41           | 0.87 (+/−)        | R41            | 0.05 (+/−)        | proteasome 26S subunit, ATPase 3                                           |
| LOC113983589 | 397.3   | MC1           | 0.86 (+/−)        | R41            | 0.8 (+/−)         | eukaryotic translation initiation factor 4 gamma 1-like                    |
| LOC114003174 | 74.5    | R41           | 0.84 (+/−)        | null           | −0.5 (+/−)        | sodium/calcium exchanger 3                                                |
| TRDN         | 896.8   | R41           | 0.84 (+/−)        | null           | −0.73 (+/−)       | triadin                                                                    |
| LOC113997739 | 8.1     | R41           | 0.77 (+/−)        | PP             | 0.4 (+/−)         | zinc finger CCCH domain-containing protein 3-like                          |
| EIF4EBP2     | 140.9   | R41           | 0.76 (+/−)        | MV             | −0.23 (+/−)       | eukaryotic translation initiation factor 4E binding protein 2              |
| TTN          | 19375   | R41           | 0.76 (+/−)        | null           | −0.98 (+/−)       | titin                                                                       |
| SLC49A4      | 215.4   | R41           | 0.73 (+/−)        | A6             | −0.51 (+/−)       | solute carrier family 49 member 4                                          |
| LOC113996621 | 129.5   | R41           | 0.72 (+/−)        | null           | −0.57 (+/−)       | zinc finger protein 883-like                                               |
| LOC113985889 | 775.5   | R41           | 0.61 (+/−)        | null           | −0.82 (+/−)       | ryanodine receptor 1-like                                                  |
| FOXRED1      | 47.8    | R41           | 0.55 (+/−)        | null           | −0.29 (+/−)       | FAD dependent oxidoreductase domain containing 1                           |
| PEX3         | 72.9    | R41           | 0.54 (+/−)        | null           | −0.95 (+/−)       | peroxisomal biogenesis factor 3                                             |
| NEK9         | 149.6   | R41           | 0.86 (+/−)        | PP             | 2.17 (+/−)        | NIMA related kinase 9                                                      |
| PDGFC        | 19.6    | LC            | −1.38 (+/−)       | R41            | 1.66 (+/−)        | platelet derived growth factor C                                           |
| TARS2L       | 118.7   | CE            | 0.19 (+/−)        | R41            | 1.65 (+/−)        | threonyl-tRNA synthetase like 2                                             |
| GDF1         | 95      | R51           | 0.17 (+/−)        | R41            | 1.65 (+/−)        | growth differentiation factor 1                                           |
| GHITM        | 2413.6  | LC            | −0.75 (+/−)       | R41            | 1.59 (+/−)        | growth hormone inducible transmembrane protein                            |
| PREPL        | 93.2    | R41           | 0.44 (+/−)        | R41            | 1.41 (+/−)        | prolyl endopeptidase like                                                 |
| ZDHHC14      | 75.1    | MV            | −0.11 (+/−)       | R41            | 1.38 (+/−)        | zinc finger DHHC-type containing 14                                         |
| FXR1         | 641.7   | R41           | 0.97 (+/−)        | R41            | 1.28 (+/−)        | FMR1 autosomal homolog                                                     |
| CARMIL1      | 101.9   | M4            | −0.2 (+/−)        | R41            | 1.22 (+/−)        | capping protein regulator and myosin 1 linker 1                             |

Table S11. (continued on next page)
| Gene                  | M1 Max cpm | Best Model | M1 model BF (SH) | Best Model | M1 model BF (PEC) | Annotation                                         |
|-----------------------|------------|------------|-----------------|------------|-----------------|---------------------------------------------------|
| **MC1 Parallel Branches Model — PEC and SH** |            |            |                 |            |                 |                                                   |
| BCO2                  | 64.5       | MV         | 1.56 (-/-)      | MV         | 1.79 (-/+       | beta-carotene oxygenase 2                         |
| **MC1 Parallel Branches Model — SH** |            |            |                 |            |                 |                                                   |
| ZDHHC12               | 35.8       | MV         | 2.03 (+/-)      | MV         | 0.57 (+/-       | zinc finger DHHC-type containing 12               |
| CCDC47                | 156.3      | MC1        | 1.84 (+/+       | MV         | -0.15 (+/+      | coiled-coil domain containing 47                 |
| LOC113983902          | 24.8       | MV         | 1.75 (-/-)      | MV         | 1.34 (-/-       | uncharacterized LOC113983902                     |
| LOC113994595          | 7.6        | MV         | 1.73 (-/-       | MV         | 1.48 (-/-       | uncharacterized LOC113994595                     |
| PSMC6                 | 284.4      | MC1        | 1.64 (+/+       | MV         | 0.35 (+/+       | proteasome 26S subunit, ATPase 6                  |
| RAC3                  | 38.9       | MV         | 1.54 (+/-)      | PP         | -0.77 (+/+      | Rac family small GTPase 3                        |
| ASZ1                  | 6.2        | MV         | 1.45 (+/-       | MV         | 0.79 (+/+       | ankyrin repeat, SAM and basic leucine zipper domain containing 1 |
| LOC114003700          | 76.2       | MV         | 1.42 (-/+       | null       | -0.43 (-/-      | helicase SRCAP-like                               |
| LOC113983589          | 397.3      | MC1        | 1.36 (+/+       | R41        | 0.12 (+/+       | eukaryotic translation initiation factor 4 gamma 1-like |
| UFL1                  | 76.2       | MC1        | 1.3 (+/+        | M6         | -0.51 (+/-      | UFM1 specific ligase 1                            |
| LOC113999226          | 35.8       | CE         | 1.23 (-/-       | PP         | -0.12 (-/-      | ATP-dependent DNA helicase Q4-like                |
| LOC113994824          | 564.7      | R41        | 1.23 (+/+       | null       | -0.32 (+/+      | ryanodine receptor 1-like                         |
| MTDH                  | 334.3      | MV         | 1.22 (+/+       | M5         | -0.44 (+/+      | metadherin                                       |
| LOC113989613          | 342.2      | MC1        | 0.98 (+/+       | PP         | -0.28 (+/+      | uncharacterized LOC113989613                     |
| SRL                   | 1171.8     | MC1        | 0.92 (+/+       | null       | -0.4 (+/+       | socalumenin                                       |
| LOC114002148          | 60.6       | MVP        | 0.72 (+/+       | M5         | -1.16 (+/+      | inactive tyrosine-protein kinase transmembrane receptor ROR1 |
| AR                    | 83.9       | MC1        | 0.34 (+/+       | M5         | -0.36 (+/+      | androgen receptor                                 |
| **MC1 Parallel Branches Model — PEC** |            |            |                 |            |                 |                                                   |
| VRK1                  | 16.8       | MV         | -0.37 (+/+      | MV         | 2.14 (+/-      | VRK serine/threonine kinase 1                     |
| LOC113987315          | 5.2        | MV         | 0.48 (+/+       | MV         | 2.05 (+/-      | uncharacterized LOC113987315                     |
| LOC113987316          | 8.7        | MV         | 1.3 (+/+        | MC1        | 1.7 (+/+       | uncharacterized LOC113987316                     |
| LOC113994137          | 11.2       | null       | -1.03 (+/+      | M5         | 1.69 (+/+      | borealin-2-like                                   |
| LOC113990618          | 215.7      | MV         | 1.53 (+/-       | MV         | 1.63 (+/+      | lysosomal alpha-glucosidase-like                  |
| PIWIL1                | 16.4       | LC         | -1.31 (-/-      | MV         | 1.51 (+/-      | piwi like RNA-mediated gene silencing 1           |
| LPAR3                 | 83         | MV         | -0.42 (+/-      | MV         | 1.45 (+/+      | lysophosphatidic acid receptor 3                  |
| SEL1L                 | 327.7      | MC1        | 0.71 (+/+       | MV         | 1.33 (+/-      | SEL1L, ERAD E3 ligase adaptor subunit             |
| EIF1B                 | 113        | M4         | -0.23 (-/-      | CE         | 1.32 (-/-      | eukaryotic translation initiation factor 1B       |
| HDLBP                 | 2028.2     | R41        | 0.08 (+/-       | MC1        | 1.32 (+/-      | high density lipoprotein binding protein          |
| GHITM                 | 2413.6     | LC         | -0.69 (+/+      | R41        | 1.28 (+/+      | growth hormone inducible transmembrane protein   |
| SRRM4                 | 28         | LC         | -1.17 (-/-      | MV         | 1.25 (-/-      | serine/arginine repetitive matrix 4              |
| EPPRD1                | 285.1      | MC1        | 0.11 (+/+       | MC1        | 1.25 (+/+      | ependym related 1                                |
| DCXR                  | 36         | MV         | 0.53 (+/-       | MV         | 1.24 (-/-      | dicarbonyl and L-xylulose reductase              |
| CCDC13                | 13         | M5         | -1.34 (+/+      | MV         | 1.22 (+/-      | coiled-coil domain containing 13                 |

**Table S11.** (continued on next page)
| Gene                | Max cpm | Best Model | BF for SH | Best Model | BF for PEC | Annotation                                                                 |
|---------------------|---------|------------|-----------|------------|------------|-----------------------------------------------------------------------------|
| MV Manacus vitellinus Branch Model — SH and PEC                   |         |            |           |            |            |                                                                             |
| UBXN1               | 51.1    | MV         | 5.78 (-)  | MV         | 5.6 (-)    | UBX domain protein 1                                                        |
| LOC113987757        | 6.8     | MV         | 3.94 (+)  | MV         | 6.45 (+)   | uncharacterized LOC113987757                                              |
| LOC113994630        | 18      | MV         | 3.64 (+)  | MV         | 5.91 (+)   | uncharacterized LOC113994630                                              |
| LOC113983963        | 98      | MV         | 3.23 (-)  | MV         | 3.53 (-)   | polypeptide N-acetylgalactosaminyltransferase-like 6                        |
| LOC113989357        | 5.2     | MV         | 3.15 (+)  | MV         | 6.02 (+)   | uncharacterized LOC113989357                                              |
| LOC113997237        | 428.9   | MV         | 3.08 (+)  | MV         | 2.23 (+)   | cytochrome P450 11B, mitochondrial-like                                      |
| LOC113994595        | 7.6     | MV         | 2.82 (-)  | MV         | 2.49 (-)   | uncharacterized LOC113994595                                              |
| GRM8                | 19.5    | MV         | 2.78 (-)  | MV         | 3.65 (-)   | glutamate metabotropic receptor 8                                           |
| LOC114001824        | 173.6   | MV         | 2.77 (+)  | MV         | 3.57 (+)   | transcription factor ReIB homolog                                           |
| ALKBH1              | 60      | MV         | 2.69 (-)  | MV         | 3.09 (-)   | alkB homolog 1, histone H2A dioxygenase                                    |
| HEMK1               | 30.6    | MV         | 2.67 (-)  | MV         | 2.14 (-)   | HemK methyltransferase family member 1                                     |
| OPN3                | 153     | MC1        | 2.57 (+)  | MV         | 5.7 (+)    | opsin 3                                                                     |
| LOC113992750        | 16.1    | MC1        | 2.48 (+)  | MV         | 4.59 (+)   | uncharacterized LOC113992750                                              |
| STAU2               | 332.5   | MV         | 2.38 (+)  | MV         | 2.26 (+)   | staufen double-stranded RNA binding protein 2                               |
| LOC113983902        | 24.8    | MV         | 2.27 (-)  | MV         | 1.62 (-)   | uncharacterized LOC113983902                                              |
| LOC114003641        | 17      | MV         | 2.14 (+)  | MV         | 3.24 (+)   | uncharacterized LOC114003641                                              |
| LOC113985529        | 50.8    | MV         | 2.08 (-)  | MV         | 3.48 (-)   | chondroitin sulfate proteoglycan 4-like                                     |
| LOC113990474        | 171.6   | MV         | 2.02 (+)  | MV         | 2.37 (+)   | C-C motif chemokine 3-like                                                 |
| LOC114003428        | 88.8    | MV         | 1.96 (+)  | MV         | 2.16 (+)   | actin cytoskeleton-regulatory complex protein PAN1-like                     |
| ANK2                | 764     | MV         | 1.95 (-)  | MV         | 1.63 (-)   | ankyrin 2                                                                   |
| LOC113987527        | 28      | MV         | 1.94 (+)  | MV         | 1.86 (+)   | uncharacterized LOC113987527                                              |
| DBI                 | 133.8   | MV         | 1.93 (-)  | MV         | 1.72 (-)   | diazepam binding inhibitor, acyl-CoA binding protein                       |
| LOC113994629        | 11.5    | MV         | 1.83 (+)  | MV         | 2.01 (+)   | uncharacterized LOC113994629                                              |
| LOC113987316        | 8.7     | MV         | 1.82 (+)  | MC1        | 1.6 (+)    | uncharacterized LOC113987316                                              |
| TOP1MT              | 5.7     | MV         | 1.81 (-)  | MV         | 2.01 (-)   | DNA topoisomerase I mitochondrial                                           |
| LOC113994659        | 46.2    | MV         | 1.73 (-)  | MV         | 1.54 (-)   | uncharacterized LOC113994659                                              |
| BCO2                | 64.5    | MV         | 1.72 (-)  | MV         | 3 (-)      | beta-carotene oxygenase 2                                                  |
| POU4F2              | 18.7    | MC1        | 1.57 (+)  | MV         | 2.7 (+)    | POU class 4 homeobox 2                                                      |
| LOC113984041        | 46.2    | MV         | 1.51 (+)  | MV         | 1.73 (+)   | staphylococcal nuclease domain-containing protein 1                         |
| THAP1               | 67.6    | MV         | 1.5 (-)   | MV         | 1.69 (-)   | THAP domain containing 1                                                   |

*Table S11.* (continued on next page)
| Gene                      | Max cpm | Best Model | MV model SH | Best Model PEC | MV model PEC | Annotation                                                                 |
|---------------------------|---------|------------|-------------|----------------|--------------|-----------------------------------------------------------------------------|
| **MV Manacus vitellinus**  |         |            |             |                |              | Branch Model — SH                                                          |
| TPRKB                     | 105     | MV         | 3.71 (+)    | MV             | 0.82 (+)     | TP53RK binding protein                                                     |
| ZDHHC12                   | 35.8    | MV         | 3.13 (+)    | MV             | 1.31 (+)     | zinc finger DHHC-type containing 12                                         |
| HACD1                     | 279.2   | MV         | 2.9 (+)     | MV             | 0.07 (+)     | 3-hydroxyacyl-CoA dehydratase 1                                            |
| TTL4                      | 195.5   | MV         | 2.7 (+)     | MV             | 1.38 (+)     | tubulin tyrosine ligase like 4                                             |
| LOC113996114              | 6.9     | MV         | 2.65 (-)    | MV             | 0.06 (-)     | uncharacterized LOC113996114                                              |
| CLRN1                     | 44.4    | MV         | 2.49 (+)    | MV             | 1.17 (+)     | clarin 1                                                                   |
| LOC113994076              | 20.9    | MV         | 2.42 (+)    | null           | -0.09 (+)    | uncharacterized LOC113994076                                              |
| LTA4H                     | 52.2    | MV         | 2.4 (+)     | MV             | 0.21 (+)     | leukotriene A4 hydrolase                                                   |
| AGFG1                     | 195.7   | MV         | 2.29 (-)    | MV             | 0.93 (-)     | ArfGAP with FG repeats 1                                                   |
| UTP4                      | 17.6    | MV         | 2.28 (-)    | MV             | 1.38 (-)     | UTP4, small subunit processome component                                   |
| ARFGAP3                   | 185.3   | MV         | 2.25 (+)    | MC1            | 1.5 (+)      | ADP ribosylation factor GTPase activating protein 3                         |
| LOC11402120               | 7.1     | MV         | 2.12 (-)    | M5             | -0.18 (-)    | uncharacterized LOC11402120                                               |
| KIF17                     | 13.1    | MV         | 2.06 (-)    | null           | -0.42 (-)    | kinesin family member 12                                                   |
| ASZ1                      | 6.2     | MV         | 2.03 (+)    | MV             | 1.46 (+)     | ankyrin repeat, SAM and basic leucine zipper domain containing 1           |
| ORC4                      | 31.3    | MV         | 2.02 (-)    | MV             | 0.03 (-)     | origin recognition complex subunit 4                                      |
| RAC3                      | 38.9    | MV         | 1.97 (+)    | PP             | -0.9 (+)     | Rac family small GTPase 3                                                  |
| POLC2                     | 49      | MV         | 1.97 (-)    | MV             | 0.8 (-)      | PQ loop repeat containing 2                                                |
| RARS2                     | 36.6    | MV         | 1.96 (+)    | null           | -0.22 (+)    | arginyl-tRNA synthetase 2, mitochondrial                                    |
| ZNF346                    | 16.4    | MV         | 1.95 (-)    | MV             | 0.26 (-)     | zinc finger protein 346                                                    |
| ZBTB40                    | 95.3    | MV         | 1.9 (+)     | MV             | 1.32 (+)     | zinc finger and BTB domain cntng. 40                                       |
| LOC114003700              | 76.2    | MV         | 1.86 (-)    | null           | -0.3 (-)     | helicase SRCAI-like                                                        |
| PHF5A                     | 123.4   | MV         | 1.86 (-)    | MV             | 0.85 (-)     | PHD finger protein 5A                                                       |
| AMACR                     | 28.8    | MV         | 1.85 (+)    | MV             | 0.88 (+)     | alpha-methylacyl-CoA racemase                                               |
| LOC11399215               | 5       | MV         | 1.82 (+)    | MV             | 1.23 (+)     | uncharacterized LOC11399215                                               |
| TMEM240                   | 27.1    | MV         | 1.78 (-)    | MV             | 0.85 (-)     | transmembrane protein 240                                                  |
| LOC114000575              | 19      | MV         | 1.68 (+)    | M6             | -0.46 (+)    | uncharacterized LOC114000575                                              |
| PON2                      | 83.8    | MV         | 1.66 (+)    | PP             | -0.88 (+)    | paraoxonase 2                                                              |
| LOC11399489               | 6.4     | MV         | 1.66 (+)    | null           | -0.07 (-)    | apoptosis-inducing factor 3-like                                           |
| DENND1B                   | 96.7    | MV         | 1.65 (+)    | MV             | 1.44 (+)     | DENN domain containing 1B                                                 |
| CCDC47                    | 156.3   | MC1        | 1.64 (+)    | MV             | 0.5 (+)      | coiled-coil domain containing 47                                           |
| RAB4A                     | 98.8    | MV         | 1.62 (-)    | MV             | 1.33 (-)     | RAB4A, member RAS oncogene family                                          |
| IQC                       | 39.7    | MV         | 1.61 (+)    | MV             | 0.77 (+)     | IQ motif containing C                                                       |
| PSMC8                     | 284.4   | MC1        | 1.61 (+)    | MV             | 0.81 (+)     | proteasome 26S subunit, ATPase 6                                           |
| TSR1                      | 58.1    | MV         | 1.6 (+)     | MV             | 0.23 (+)     | TSR1, ribosome maturation factor                                           |
| FRMD7                     | 5.6     | MV         | 1.59 (-)    | MC2            | 1.16 (-)     | FERM domain containing 7                                                   |
| CUNH15orf48               | 37.9    | MV         | 1.56 (-)    | MV             | 0.75 (-)     | chromosome unknown C15orf48 homolog                                        |
| DUSP28                    | 276.8   | MC1        | 1.55 (+)    | MV             | 0.88 (+)     | dual specificity phosphatase 28                                             |
| LOC113996059              | 11.7    | MV         | 1.55 (-)    | MV             | 0.22 (-)     | GDNF family receptor alpha-4-like                                          |
| LONRF1                    | 112.6   | MV         | 1.52 (-)    | null           | -0.3 (-)     | LON peptidase N-terminal domain and ring finger 1                          |
| UTP20                     | 43.2    | MV         | 1.52 (-)    | M5             | 0.2 (-)      | UTP20, small subunit processome component                                  |
| LOC113987868              | 26.3    | MV         | 1.5 (+)     | MC1            | -0.15 (+)    | uncharacterized LOC113987868                                              |

Table S11. (continued on next page)
| Gene | Max cpm | Best Model SH | MV model BF (SH) | Best Model PEC | MV model BF (PEC) | Annotation |
|------|---------|---------------|------------------|----------------|------------------|------------|
| LOC113987146 | 35.6 | CE | –0.24 (+) | MV | 5.62 (+) | uncharacterized LOC113987146 |
| LOC114003534 | 36.3 | MV | 0.23 (+) | MV | 3.94 (+) | uncharacterized LOC114003534 |
| LOC113987756 | 158.9 | MC1 | 1.49 (+) | MV | 3.53 (+) | uncharacterized LOC113987756 |
| VRK1 | 16.8 | MV | 0.46 (+) | MV | 3.35 (+) | VRK serine/threonine kinase 1 |
| LOC113987315 | 5.2 | MV | 1.28 (+) | MV | 3.25 (+) | uncharacterized LOC113987315 |
| RUBCNL | 179.6 | MV | 0.53 (+) | MV | 3.19 (+) | rubicon like autophagy enhancer |
| PPP1R11 | 31.6 | MV | 0.24 (–) | MV | 3.15 (–) | protein phosphatase 1 regulatory inhibitor subunit 11 |
| LOC113995174 | 5.7 | LC | –0.44 (+) | MV | 3.13 (+) | uncharacterized LOC113995174 |
| PRLR | 369.3 | MV | 0.87 (+) | MV | 3.12 (+) | prolactin receptor |
| EXO1 | 62.9 | null | –0.28 (+) | MV | 2.97 (+) | exonuclease 1 |
| SGTB | 141.5 | MV | 1.39 (–) | MV | 2.95 (–) | small glutamine rich tetra-tricopeptide repeat containing beta |
| UROC1 | 18.8 | LC | –0.71 (+) | MV | 2.92 (+) | urocanate hydratase 1 |
| LOC113994137 | 11.2 | null | –0.77 (+) | MV | 2.71 (+) | borealin-2-like |
| UCP3 | 1232.9 | MV | 0.55 (–) | MV | 2.69 (–) | uncoupling protein 3 |
| LOC113989356 | 8.5 | MV | 0.98 (+) | MV | 2.67 (+) | ran-binding protein 17-like |
| LOC113997196 | 8.5 | MV | 0.32 (–) | MV | 2.65 (–) | butyrophilin subfamily 1 member A1-like |
| AKNA | 29.8 | MV | 0.43 (+) | MV | 2.58 (+) | AT-hook transcription factor |
| WDR64 | 76 | MV | 0.16 (+) | MV | 2.56 (+) | WD repeat domain 64 |
| SNRPB | 38.1 | MV | 0.77 (–) | MV | 2.53 (–) | small nuclear ribonucleoprotein polypeptides B and B1 |
| RTKN2 | 31 | MV | 0.66 (+) | MV | 2.46 (+) | rhotekin 2 |
| LOC113996018 | 215.7 | MV | 0.35 (+) | MV | 2.39 (+) | lysosomal alpha-glucosidase-like |
| KIF20B | 17.3 | null | –0.84 (–) | MV | 2.38 (–) | kinesin family member 20B |
| GATB | 266.6 | M5 | –0.28 (–) | MV | 2.38 (–) | glutamyl-tRNA amidotransferase subunit B |
| FAM118A | 62.3 | MV | 0.8 (+) | MV | 2.34 (+) | family with sequence similarity 118 member A |
| SLC20A1 | 196 | LC | –0.86 (+) | MV | 2.31 (+) | solute carrier family 20 member 1 |
| LOC114002601 | 17.5 | MV | 0.6 (+) | MV | 2.28 (+) | cytochrome P450 2J2-like |
| LPAR3 | 83 | MV | 0.48 (+) | MV | 2.21 (+) | lysophosphatidic acid receptor 3 |
| MICU3 | 396.4 | MV | 1.04 (+) | MV | 2.2 (–) | mitochondrial calcium uptake family member 3 |
| RSL1D1 | 63.6 | MC2 | 0.79 (–) | MV | 2.19 (–) | ribosomal L1 domain containing 1 |
| COL5A1 | 82.9 | MV | 0.19 (–) | MV | 2.18 (–) | collagen type V alpha 1 chain |
| FARS2 | 147.4 | LC | –0.19 (–) | MV | 2.17 (–) | phenylalanyl-tRNA synthetase subunit beta |
| TMEM154 | 9.1 | null | –0.52 (–) | MV | 2.13 (–) | transmembrane protein 154 |
| HDAC2 | 133.7 | MV | 0.25 (+) | MV | 2.1 (–) | histone deacetylase 2 |
| MPP7 | 61.4 | MV | 0.18 (–) | MV | 2.1 (–) | membrane palmitoylated protein 7 |
| ACTA1 | 33933 | PP | –0.94 (–) | MV | 2.01 (–) | actin, alpha 1, skeletal muscle |

Table S11. (continued on next page)
### CE Ceratopipra Ancestor Branch Model — SH and PEC

| Gene          | Max  | Best Model | CE model BF (SH) | Best Model | CE model BF (PEC) | Annotation                                      |
|---------------|------|------------|------------------|------------|-------------------|------------------------------------------------|
| LOC113990672 | 5315.2 | CE         | 7.69 (+)         | CE         | 2.67 (+)          | parvalbumin, thymic                             |
| LOC113990673 | 64.2  | CE         | 5.09 (+)         | CE         | 1.82 (+)          | parvalbumin beta-like                           |
| LOC113989134 | 5.3   | CE         | 2.6 (+)          | CE         | 3.2 (+)           | uncharacterized LOC113989134                   |
| IL18          | 55.4  | CE         | 2.2 (+)          | CE         | 3.12 (+)          | interleukin 18                                  |
| NAALADL2     | 50.1  | CE         | 2.01 (+)         | CE         | 2.18 (+)          | N-acetylated alpha-linked acidic dipeptidase like 2 |
| LOC113983196 | 22.8  | MC1        | 1.97 (+)         | CE         | 2.56 (+)          | uncharacterized LOC113983196                   |
| MAD1L1       | 53.4  | CE         | 1.74 (+)         | CE         | 2.41 (+)          | mitotic arrest deficient 1 like 1             |
| LOC113998906 | 41.9  | CE         | 1.64 (-)         | CE         | 3.28 (-)          | interferon-induced very large GTPase 1-like   |
| LOC113990771 | 6.8   | CE         | 1.63 (-)         | CE         | 1.6 (-)           | WD repeat and SOCS box-containing protein 2-like |
| BRINP2       | 42.3  | PP         | 1.59 (+)         | CE         | 2.09 (+)          | BMP/retinoic acid inducible neural specific 2   |

### CE Ceratopipra Ancestor Branch Model — SH

| Gene          | Max  | Best Model | CE model BF (SH) | Best Model | CE model BF (PEC) | Annotation                                      |
|---------------|------|------------|------------------|------------|-------------------|------------------------------------------------|
| LOC113983894 | 5.2   | CE         | 3.62 (+)         | CE         | 0.31 (+)          | uncharacterized LOC113983894                   |
| LOC113991466 | 23.4  | CE         | 2.66 (-)         | CE         | 1.08 (-)          | sodium channel protein type 5 subunit alpha-like |
| PSKH1        | 78.3  | CE         | 2.46 (+)         | CE         | 1.18 (+)          | protein serine kinase H1                       |
| STYK1        | 34.6  | CE         | 2.32 (-)         | CE         | 1.06 (-)          | serine/threonine/tyrosine kinase 1             |
| TSEN15       | 51.5  | CE         | 2.27 (+)         | CE         | 0.03 (+)          | tRNA splicing endonuclease subunit 15          |
| LOC113999226 | 35.8  | CE         | 2.08 (-)         | PP         | –0.42 (-)         | ATP-dependent DNA helicase Q4-like             |
| RNF141       | 108.5 | CE         | 1.95 (+)         | CE         | 0.34 (+)          | ring finger protein 141                        |
| GINS1        | 7.1   | CE         | 1.93 (+)         | LC         | 0.13 (+)          | GINS complex subunit 1                         |
| LOC113995569 | 11.7  | CE         | 1.93 (+)         | CE         | 0.96 (+)          | scavenger receptor cysteine-rich type 1 protein M130-like |
| Ninjurin      | 30.5  | CE         | 1.75 (+)         | CE         | 0.67 (+)          | ninjurin 2                                     |
| VPS18        | 60.7  | CE         | 1.7 (+)          | CE         | 0.74 (+)          | VPS18, CORVET/HOPS core subunit                |
| UPF1         | 327.7 | PP         | 1.64 (+)         | CE         | 1.05 (+)          | UPF1, RNA helicase and ATPase                  |
| LARP7        | 71.7  | CE         | 1.64 (+)         | CE         | 1.04 (+)          | La ribonucleoprotein domain family member 7    |
| LOC113997729 | 10.7  | CE         | 1.63 (-)         | null       | –0.41 (-)         | uncharacterized LOC113997729                   |
| CRIP1        | 42.7  | CE         | 1.6 (+)          | CE         | 0.78 (+)          | CXXC repeat containing interactor of PDZ3 domain |
| MAPKAP1      | 75.4  | CE         | 1.6 (+)          | CE         | 1.4 (+)           | mitogen-activated protein kinase associated protein 1 |
| LOC114002587 | 13.5  | CE         | 1.55 (+)         | null       | –0.45 (+)         | uncharacterized LOC114002587                   |
| PAX7         | 30.6  | CE         | 1.54 (+)         | CE         | 1.36 (+)          | paired box 7                                   |
| HIPK3        | 309.1 | CE         | 1.53 (+)         | CE         | 0.51 (+)          | homeodomain interacting protein kinase 3       |
| LOC113996835 | 18.9  | CE         | 1.52 (+)         | CE         | 1.47 (+)          | uncharacterized LOC113996835                   |
| STX2         | 122.4 | CE         | 1.51 (+)         | M6         | –0.26 (+)         | syntaxin 2                                     |

Table S11. (continued on next page)
| Gene            | Max cpm | Best Model SH | Best Model BF (SH) | Best Model PEC | CE model BF (PEC) | Annotation                                                                 |
|-----------------|---------|---------------|-------------------|----------------|------------------|-----------------------------------------------------------------------------|
| HFM1            | 21.2    | PP            | 0.72 (+)          | CE             | 3.62 (+)         | HFM1, ATP dependent DNA helicase homolog                                  |
| EIF2D           | 209.4   | CE            | 0.63 (−)          | CE             | 2.32 (−)         | eukaryotic translation initiation factor 2D                                |
| MORC2           | 76.5    | CE            | 1.25 (+)          | CE             | 2.26 (+)         | MORC family CW-type zinc finger 2                                          |
| WDR11           | 66.1    | PP            | 0.34 (+)          | CE             | 2.23 (+)         | WD repeat domain 11                                                        |
| LOC113998234    | 75.1    | LC            | −0.52 (+)         | CE             | 2.16 (+)         | uncharacterized LOC113998234                                              |
| KLHL15          | 63.4    | null          | −0.36 (−)         | CE             | 2.11 (−)         | kelch like family member 15                                                |
| TNNT1           | 289.9   | R51           | 0.44 (+)          | CE             | 2.06 (+)         | troponin T1, slow skeletal type                                            |
| STAR10          | 43.4    | CE            | 0.5 (−)           | CE             | 2.04 (−)         | STAR related lipid transfer domain containing 10                           |
| SYTL1           | 11.9    | null          | −0.3 (−)          | CE             | 2.03 (−)         | synaptotagmin like 1                                                       |
| PIEZO1          | 137.1   | CE            | 1.03 (−)          | CE             | 2.01 (−)         | piezo type mechanosensitive ion channel component 1                        |
| TPD52L1         | 43      | CE            | 0.6 (+)           | CE             | 1.91 (+)         | TPD52 like 1                                                              |
| UFSP2           | 110.6   | CE            | 1.36 (+)          | CE             | 1.88 (+)         | UFM1 specific peptidase 2                                                  |
| LOC113998901    | 20.8    | CE            | 0.5 (−)           | CE             | 1.86 (−)         | interferon-induced very large GTPase 1-like                                |
| LOC11399965     | 19.7    | null          | −0.33 (−)         | CE             | 1.85 (−)         | poly [ADP-ribose] polymerase 12-like                                       |
| HGD             | 29      | CE            | 0.7 (+)           | CE             | 1.81 (+)         | homogentisate 1,2-dioxygenase                                              |
| TNFRSF11A       | 25      | R51           | 0 (+)             | CE             | 1.81 (+)         | TNF receptor superfamily member 11a                                       |
| TPD52L2         | 117.3   | LC            | −0.71 (−)         | CE             | 1.73 (−)         | TPD52 like 2                                                              |
| SEMA6A          | 73.4    | null          | −0.59 (−)         | CE             | 1.7 (−)          | semaphorin 6A                                                             |
| LOC113998631    | 75      | LC            | −0.73 (−)         | CE             | 1.68 (−)         | dnaJ homolog subfamily B member 1-like                                     |
| SYBU            | 56.9    | PP            | 0.43 (+)          | CE             | 1.64 (+)         | syntabulin                                                                |
| ZNF687          | 12      | LC            | −0.66 (−)         | CE             | 1.62 (−)         | zinc finger protein 687                                                    |
| LOC114001289    | 11.3    | CE            | 1.14 (+)          | CE             | 1.6 (+)          | uncharacterized LOC114001289                                              |
| MMP16           | 22.9    | PP            | −0.01 (−)         | CE             | 1.58 (+)         | matrix metallopeptidase 16                                                 |
| LOC113984982    | 6.6     | M4            | 0.24 (+)          | CE             | 1.58 (+)         | uncharacterized LOC113984982                                              |
| LOC113984741    | 36.6    | PP            | −0.52 (+)         | CE             | 1.57 (+)         | retinol dehydrogenase 8-like                                               |
| CP              | 22.1    | CE            | 0.68 (+)          | CE             | 1.55 (+)         | ceruloplasmin                                                              |
| TRIM9           | 66.7    | CE            | 0.37 (+)          | CE             | 1.55 (+)         | tripartite motif containing 9                                             |
| TEX12           | 7.4     | M4            | −0.1 (−)          | CE             | 1.54 (−)         | testis expressed 12                                                        |
| EIF1B           | 113     | M4            | −0.41 (−)         | CE             | 1.53 (−)         | eukaryotic translation initiation factor 1B                                |
| MMEL1           | 93.8    | M5            | −0.2 (−)          | CE             | 1.52 (−)         | membrane metalloendopeptidase 1                                            |

Table S11. Top PhyDGET genes for each model for SH and PEC. Each gene indicated as BF $\geq 1.5$ by PhyDGET for each model, gene maximum counts-per-million (cpm), the best likelihood model for SH and PEC, Bayes Factor for SH and PEC, and the gene reference annotation from the *P. filicauda* reference genome annotation.
| GO Term                                   | Definition                                                                 | P-value | Adj. P-value | Study | Family | Population |
|-------------------------------------------|---------------------------------------------------------------------------|---------|--------------|-------|--------|------------|
| **M6 Ancestor Branch Model — SH**        |                                                                           |         |              |       |        |            |
| GO:0065002                                | intracellular protein transmembrane transport                            | 0.0016  | 0.88         | 4 / 160 | 15 / 1285 | 47 / 14795 |
| GO:006457                                 | protein folding                                                          | 0.0017  | 0.88         | 7 / 160 | 109 / 11581 | 183 / 14795 |
| GO:0045494                                | photoreceptor cell maintenance                                           | 0.0022  | 0.88         | 3 / 160 | 38 / 5318  | 37 / 14795  |
| GO:0071806                                | protein transmembrane transport                                          | 0.003   | 0.88         | 4 / 160 | 25 / 2198  | 55 / 14795  |
| GO:0051131                                | chaperone-mediated protein complex assembly                              | 0.0043  | 0.88         | 2 / 160 | 6 / 900   | 16 / 14795  |
| GO:0044090                                | positive regulation of vacuole organization                               | 0.0044  | 0.88         | 2 / 160 | 6 / 658   | 12 / 14795  |
| GO:0006626                                | protein targeting to mitochondrion                                        | 0.0045  | 0.88         | 5 / 160 | 11 / 791   | 90 / 14795  |
| GO:1901223                                | negative regulation of NIK/NF-kappaB signaling                            | 0.0046  | 0.88         | 2 / 160 | 3 / 565   | 23 / 14795  |
| GO:0006002                                | fructose 6-phosphate metabolic process                                   | 0.0049  | 0.88         | 2 / 160 | 26 / 2889  | 12 / 14795  |
| GO:0007529                                | establishment of synaptic specificity at neuromuscular junction           | 0.0054  | 0.88         | 1 / 160 | 1 / 369   | 2 / 14795   |
| GO:0033210                                | leptin-mediated signaling pathway                                         | 0.0055  | 0.88         | 2 / 160 | 6 / 484   | 10 / 14795  |
| GO:0046890                                | regulation of lipid biosynthetic process                                 | 0.0058  | 0.88         | 5 / 160 | 26 / 3433  | 158 / 14795 |
| **M6 Ancestor Branch Model — PEC**        |                                                                           |         |              |       |        |            |
| GO:0031099                                | regeneration                                                             | 2.8 × 10⁻⁵  | 0.03      | 4 / 56 | 6 / 4366  | 165 / 14806 |
| GO:0031102                                | neuron projection regeneration                                           | 6.8 × 10⁻⁴  | 0.39      | 3 / 56 | 8 / 2499  | 60 / 14806  |
| GO:0031103                                | axon regeneration                                                        | 0.0013  | 0.39         | 3 / 56 | 3 / 499   | 55 / 14806  |
| GO:0051402                                | neuron apoptotic process                                                 | 0.0019  | 0.39         | 3 / 56 | 3 / 1534  | 190 / 14806 |
| GO:1903937                                | response to acrylamide                                                   | 0.0021  | 0.39         | 1 / 56 | 3 / 1409  | 1 / 14806   |
| GO:0046835                                | carbohydrate phosphorylation                                             | 0.0022  | 0.39         | 2 / 56 | 3 / 1820  | 50 / 14806  |
| GO:0043523                                | regulation of neuron apoptotic process                                   | 0.0024  | 0.39         | 3 / 56 | 3 / 1185  | 160 / 14806 |
| GO:0043524                                | negative regulation of neuron apoptotic process                          | 0.0028  | 0.4          | 3 / 56 | 3 / 758   | 108 / 14806 |
| GO:0045105                                | intermediate filament polymerization or depolymerization                 | 0.0036  | 0.43         | 1 / 56 | 3 / 1686  | 2 / 14806   |
| GO:0016829                                | lyase activity                                                           | 0.0039  | 0.43         | 4 / 56 | 7 / 2574  | 285 / 14806 |
| GO:0042699                                | follicle-stimulating hormone signaling pathway                           | 0.005   | 0.43         | 1 / 56 | 2 / 800   | 2 / 14806   |
| GO:0070997                                | neuron death                                                             | 0.005   | 0.43         | 3 / 56 | 3 / 1662  | 285 / 14806 |

Table S12. (continued on next page)
| GO Term | Definition |   |   |   |   |
|---------|------------|---|---|---|---|
| GO:0060749 | mammary gland alveolus development | $1.5 \times 10^{-4}$ | 0.2 | 3 / 100 | 32 / 4367 | 15 / 14783 |
| GO:0061377 | mammary gland lobe development | $1.5 \times 10^{-4}$ | 0.2 | 3 / 100 | 32 / 4367 | 15 / 14783 |
| GO:2000026 | regulation of multicellular organismal development | $4.2 \times 10^{-4}$ | 0.37 | 15 / 100 | 29 / 4736 | 1044 / 14783 |
| GO:0002541 | activation of plasma proteins involved in acute inflammatory response | $7.2 \times 10^{-4}$ | 0.41 | 2 / 100 | 3 / 223 | 4 / 14783 |
| GO:0010641 | positive regulation of platelet-derived growth factor receptor signaling pathway | 0.0016 | 0.41 | 2 / 100 | 10 / 1224 | 8 / 14783 |
| GO:0050921 | positive regulation of chemotaxis | 0.0018 | 0.41 | 4 / 100 | 6 / 943 | 104 / 14783 |
| GO:2000232 | regulation of rRNA processing | 0.0019 | 0.41 | 2 / 100 | 20 / 3547 | 12 / 14783 |
| GO:0035306 | positive regulation of dephosphorylation | 0.002 | 0.41 | 3 / 100 | 7 / 1142 | 47 / 14783 |
| GO:2000145 | regulation of cell motility | 0.0021 | 0.41 | 11 / 100 | 11 / 1351 | 772 / 14783 |
| GO:0198738 | cell-cell signaling by wnt | 0.0024 | 0.41 | 7 / 100 | 8 / 1379 | 455 / 14783 |
| GO:0046777 | protein autophosphorylation | 0.0025 | 0.41 | 7 / 100 | 14 / 1331 | 206 / 14783 |
| GO:0046467 | membrane lipid biosynthetic process | 0.0026 | 0.41 | 5 / 100 | 35 / 4586 | 128 / 14783 |
| GO:1903184 | L-dopa metabolic process | 0.0036 | 0.5 | 1 / 88 | 1 / 278 | 1 / 14784 |
| GO:1902883 | negative regulation of response to oxidative stress | 0.0041 | 0.5 | 2 / 88 | 8 / 1567 | 20 / 14784 |
| GO:1900453 | negative regulation of long-term synaptic depression | 0.0042 | 0.5 | 1 / 88 | 18 / 4244 | 1 / 14784 |
| GO:1904708 | MAP kinase kinase activity | 0.0044 | 0.5 | 3 / 88 | 5 / 724 | 58 / 14784 |
| GO:2000284 | positive regulation of cellular amino acid biosynthetic process | 0.0045 | 0.5 | 1 / 88 | 7 / 1552 | 1 / 14784 |
| GO:1903181 | positive regulation of dopamine biosynthetic process | 0.0047 | 0.5 | 1 / 88 | 7 / 1503 | 1 / 14784 |
| GO:2000282 | regulation of cellular amino acid biosynthetic process | 0.0048 | 0.5 | 1 / 88 | 14 / 2942 | 1 / 14784 |
| GO:1903179 | regulation of dopamine biosynthetic process | 0.0048 | 0.5 | 1 / 88 | 14 / 2896 | 1 / 14784 |
| GO:0035330 | regulation of hippo signaling | 0.0048 | 0.5 | 2 / 88 | 9 / 1555 | 19 / 14784 |
| GO:0035814 | negative regulation of renal sodium excretion | 0.0049 | 0.5 | 1 / 88 | 4 / 818 | 1 / 14784 |
| GO:1903363 | negative regulation of cellular protein catabolic process | 0.0049 | 0.5 | 3 / 88 | 8 / 1421 | 68 / 14784 |
| GO:1903204 | negative regulation of oxidative stress-induced neuron death | 0.005 | 0.5 | 2 / 88 | 2 / 191 | 14 / 14784 |

Table S12. (continued on next page)
| GO Term                                | Definition                                                                 | \( P \)-value | Adj. \( P \)-value | Study | Family | Population |
|----------------------------------------|---------------------------------------------------------------------------|----------------|---------------------|-------|--------|------------|
| GO:0007060                            | male meiosis chromosome segregation                                       | \( 1.8 \times 10^{-4} \) | 0.32                | 2 / 102 | 2 / 259 | 4 / 14803  |
| GO:0046967                            | cytosol to endoplasmic reticulum transport                                 | \( 2.5 \times 10^{-4} \) | 0.32                | 2 / 102 | 15 / 2859 | 5 / 14803   |
| GO:1903515                            | calcium ion transport from cytosol to endoplasmic reticulum                | \( 5.3 \times 10^{-4} \) | 0.42                | 2 / 102 | 4 / 260  | 3 / 14803   |
| GO:0032278                            | positive regulation of gonadotropin secretion                              | \( 6.7 \times 10^{-4} \) | 0.42                | 2 / 102 | 8 / 1108 | 6 / 14803   |
| GO:0014075                            | response to amine                                                          | 0.0026         | 0.65                | 3 / 102 | 7 / 848  | 38 / 14803  |
| GO:0003360                            | brainstem development                                                      | 0.0034         | 0.65                | 2 / 102 | 30 / 4368 | 13 / 14803  |
| GO:0032276                            | regulation of gonadotropin secretion                                       | 0.0039         | 0.65                | 2 / 102 | 3 / 206  | 8 / 14803   |
| GO:0048731                            | system development                                                         | 0.0041         | 0.65                | 30 / 102 | 30 / 4368 | 3639 / 14803 |
| GO:0000378                            | RNA exon ligation                                                          | 0.0042         | 0.65                | 1 / 102 | 14 / 3306 | 1 / 14803   |
| GO:1905304                            | regulation of cardiac myofibril assembly                                    | 0.0047         | 0.65                | 1 / 102 | 2 / 854  | 2 / 14803   |
| GO:0099170                            | postsynaptic modulation of chemical synaptic transmission                    | 0.0048         | 0.65                | 2 / 102 | 3 / 410  | 17 / 14803  |
| GO:0070858                            | negative regulation of bile acid biosynthetic process                       | 0.005          | 0.65                | 1 / 102 | 3 / 1184 | 2 / 14803   |

**Table S12.** (continued on next page)
| GO Term | Definition | P-value | Adj. P-value | Study | Family | Population |
|---------|------------|---------|--------------|-------|--------|------------|
| GO:0032849 | positive regulation of cellular pH reduction | 0.0014 | 0.35 | 1 / 24 | 2 / 4330 | 3 / 14781 |
| GO:0035037 | sperm entry | 0.0021 | 0.35 | 1 / 24 | 3 / 1412 | 1 / 14781 |
| GO:1903801 | L-leucine import across plasma membrane | 0.0025 | 0.35 | 1 / 24 | 1 / 788 | 2 / 14781 |
| GO:0032847 | regulation of cellular pH reduction | 0.0033 | 0.35 | 1 / 24 | 5 / 7639 | 5 / 14781 |
| GO:0060357 | regulation of leucine import | 0.0035 | 0.35 | 1 / 24 | 1 / 283 | 1 / 14781 |
| GO:0044265 | cellular macromolecule catabolic process | 0.0036 | 0.35 | 3 / 24 | 3 / 6690 | 1027 / 14781 |
| GO:0044257 | cellular protein catabolic process | 0.0038 | 0.35 | 3 / 24 | 3 / 4281 | 668 / 14781 |
| GO:0016192 | vesicle-mediated transport | 0.0048 | 0.4 | 5 / 24 | 8 / 11621 | 1960 / 14781 |
| GO:0006508 | proteolysis | 0.0093 | 0.62 | 4 / 24 | 4 / 4484 | 1393 / 14781 |
| GO:0060736 | prostate gland growth | 0.014 | 0.62 | 1 / 24 | 1 / 715 | 10 / 14781 |
| GO:0060358 | negative regulation of leucine import | 0.0147 | 0.62 | 1 / 24 | 1 / 68 | 1 / 14781 |

| GO Term | Definition | P-value | Adj. P-value | Study | Family | Population |
|---------|------------|---------|--------------|-------|--------|------------|
| GO:0055017 | cardiac muscle tissue growth | 0.0016 | 0.71 | 3 / 53 | 3 / 611 | 72 / 14787 |
| GO:0040020 | regulation of meiotic nuclear division | 0.0017 | 0.71 | 2 / 53 | 3 / 766 | 19 / 14787 |
| GO:0060419 | heart growth | 0.0029 | 0.71 | 3 / 53 | 3 / 526 | 76 / 14787 |
| GO:0008202 | steroid metabolic process | 0.004 | 0.71 | 4 / 53 | 15 / 5228 | 241 / 14787 |
| GO:0060025 | regulation of synaptic activity | 0.004 | 0.71 | 1 / 53 | 2 / 497 | 1 / 14787 |
| GO:0051445 | regulation of meiotic cell cycle | 0.0044 | 0.71 | 2 / 53 | 4 / 1148 | 32 / 14787 |
| GO:2000813 | negative regulation of barbed-end actin filament capping | 0.0045 | 0.71 | 1 / 53 | 3 / 674 | 1 / 14787 |
| GO:0045321 | leukocyte activation | 0.0055 | 0.71 | 8 / 53 | 9 / 2038 | 859 / 14787 |
| GO:1905643 | positive regulation of DNA methylation | 0.0085 | 0.71 | 1 / 53 | 1 / 234 | 2 / 14787 |
| GO:0090367 | negative regulation of mRNA modification | 0.0097 | 0.71 | 1 / 53 | 1 / 103 | 1 / 14787 |
| GO:0035281 | pre-miRNA export from nucleus | 0.0099 | 0.71 | 1 / 53 | 1 / 202 | 2 / 14787 |
| GO:1905962 | glutamatergic neuron differentiation | 0.01 | 0.71 | 1 / 53 | 4 / 1200 | 3 / 14787 |

Table S12. (continued on next page)
| GO Term | Definition | P-value | Adj. P-value | Study | Family | Population |
|---------|------------|---------|--------------|-------|--------|------------|
| GO:0036309 | protein localization to M-band | 0.0033 | 0.75 | 1 / 28 | 2 / 1200 | 2 / 14779 |
| GO:0099172 | presynapse organization | 0.0039 | 0.75 | 2 / 28 | 12 / 5917 | 47 / 14779 |
| GO:0034976 | response to endoplasmic reticulum stress | 0.0043 | 0.75 | 4 / 28 | 6 / 1741 | 241 / 14779 |
| GO:1903337 | positive regulation of vacuolar transport | 0.0045 | 0.75 | 1 / 28 | 10 / 4413 | 2 / 14779 |
| GO:1903335 | regulation of vacuolar transport | 0.0083 | 0.75 | 1 / 28 | 16 / 7690 | 4 / 14779 |
| GO:0061919 | process utilizing autophagic mechanism | 0.0101 | 0.75 | 4 / 28 | 21 / 11576 | 482 / 14779 |
| GO:0120253 | hydrocarbon catabolic process | 0.0106 | 0.75 | 1 / 28 | 5 / 1865 | 4 / 14779 |
| GO:0007029 | endoplasmic reticulum organization | 0.0107 | 0.75 | 2 / 28 | 9 / 4427 | 80 / 14779 |
| GO:1905535 | regulation of eukaryotic translation initiation factor 4F complex assembly | 0.0111 | 0.75 | 1 / 28 | 2 / 360 | 2 / 14779 |
| GO:0097010 | eukaryotic translation initiation factor 4F complex assembly | 0.0133 | 0.75 | 1 / 28 | 3 / 900 | 4 / 14779 |
| GO:0043435 | response to corticotropin-releasing hormone | 0.0139 | 0.75 | 1 / 28 | 1 / 359 | 5 / 14779 |
| GO:0051098 | regulation of binding | 0.014 | 0.75 | 3 / 28 | 21 / 11977 | 294 / 14779 |
| GO:1904158 | axonemal central apparatus assembly | 0.0116 | 0.73 | 1 / 20 | 5 / 4703 | 11 / 14778 |
| GO:1902659 | regulation of glucose mediated signaling pathway | 0.012 | 0.73 | 1 / 20 | 5 / 2482 | 6 / 14778 |
| GO:1902661 | positive regulation of glucose mediated signaling pathway | 0.0124 | 0.73 | 1 / 20 | 3 / 1202 | 5 / 14778 |
| GO:009756 | carbohydrate mediated signaling | 0.0127 | 0.73 | 1 / 20 | 6 / 4224 | 9 / 14778 |
| GO:1901159 | xylulose 5-phosphate biosynthetic process | 0.0129 | 0.73 | 1 / 20 | 2 / 926 | 6 / 14778 |

Table S12. (continued on next page)
| GO Term | Definition | P-value | Adj. P-value | Study | Family | Population |
|---------|------------|---------|--------------|-------|--------|------------|
| GO:0030054 | cell junction | $1.1 \times 10^{-12}$ | $3.3 \times 10^{-9}$ | 36 / 223 | 69 / 4515 | 701 / 14884 |
| GO:0098793 | presynapse | $7.6 \times 10^{-11}$ | $1.2 \times 10^{-7}$ | 20 / 223 | 69 / 4515 | 230 / 14884 |
| GO:0001505 | regulation of neurotransmitter levels | $1.6 \times 10^{-9}$ | $1.7 \times 10^{-6}$ | 18 / 223 | 57 / 3209 | 191 / 14884 |
| GO:0006836 | neurotransmitter transport | $1.3 \times 10^{-8}$ | $9.9 \times 10^{-6}$ | 17 / 223 | 73 / 4256 | 192 / 14884 |
| GO:0007610 | presynapse | $1.6 \times 10^{-7}$ | $7.4 \times 10^{-5}$ | 21 / 223 | 102 / 7876 | 448 / 14884 |
| GO:0001505 | regulation of neurotransmitter levels | $1.6 \times 10^{-7}$ | $7.4 \times 10^{-5}$ | 21 / 223 | 102 / 7876 | 448 / 14884 |
| GO:0030182 | neuron differentiation | $6.1 \times 10^{-7}$ | $2.4 \times 10^{-4}$ | 39 / 223 | 54 / 3108 | 1212 / 14884 |
| GO:0045103 | intermediate filament-based process | $8.4 \times 10^{-7}$ | $2.9 \times 10^{-4}$ | 7 / 223 | 140 / 11622 | 44 / 14884 |
| GO:0032989 | membrane assembly | $1.6 \times 10^{-6}$ | $5.0 \times 10^{-4}$ | 25 / 223 | 91 / 7082 | 705 / 14884 |
| GO:0045104 | intermediate filament cytoskeleton organization | $1.9 \times 10^{-6}$ | $5.3 \times 10^{-4}$ | 7 / 223 | 22 / 1403 | 43 / 14884 |
| GO:0140352 | export from cell | $2.0 \times 10^{-6}$ | $5.3 \times 10^{-4}$ | 32 / 223 | 140 / 11668 | 1110 / 14884 |
| GO:0031424 | keratinization | $2.7 \times 10^{-19}$ | $8.4 \times 10^{-16}$ | 16 / 201 | 80 / 5326 | 43 / 14825 |
| GO:0016755 | transferase activity, transferring amino-acyl groups | $5.3 \times 10^{-17}$ | $7.0 \times 10^{-14}$ | 15 / 201 | 28 / 1374 | 43 / 14825 |
| GO:0070268 | cornification | $6.7 \times 10^{-17}$ | $7.02 \times 10^{-14}$ | 15 / 201 | 34 / 1553 | 39 / 14825 |
| GO:0043588 | skin development | $6.9 \times 10^{-16}$ | $5.46 \times 10^{-13}$ | 27 / 201 | 57 / 2602 | 207 / 14825 |
| GO:0016746 | transferase activity, transferring acyl groups | $2.6 \times 10^{-15}$ | $1.7 \times 10^{-12}$ | 15 / 201 | 26 / 1163 | 51 / 14825 |
| GO:0071709 | membrane assembly | $9.8 \times 10^{-15}$ | $5.1 \times 10^{-12}$ | 15 / 201 | 52 / 3698 | 71 / 14825 |
| GO:0044091 | membrane biogenesis | $6.2 \times 10^{-14}$ | $2.8 \times 10^{-12}$ | 15 / 201 | 43 / 2806 | 76 / 14825 |
| GO:0008544 | epidermis development | $1.8 \times 10^{-12}$ | $7.1 \times 10^{-10}$ | 29 / 201 | 49 / 1509 | 238 / 14825 |
| GO:0007009 | plasma membrane organization | $2.1 \times 10^{-10}$ | $7.3 \times 10^{-8}$ | 16 / 201 | 33 / 1881 | 143 / 14825 |
| GO:0098888 | tissue development | $1.2 \times 10^{-19}$ | $3.6 \times 10^{-7}$ | 49 / 201 | 70 / 4371 | 1509 / 14825 |
| GO:1903575 | cornified envelope assembly | $1.4 \times 10^{-9}$ | $4.1 \times 10^{-7}$ | 15 / 201 | 16 / 163 | 41 / 14825 |
| GO:0030855 | epithelial cell differentiation | $4.1 \times 10^{-9}$ | $1.1 \times 10^{-6}$ | 29 / 201 | 64 / 3219 | 480 / 14825 |

Table S12. (continued on next page)
| GO Term | Definition | P-value | Adj. P-value | Study | Family | Population |
|---------|------------|---------|--------------|-------|--------|------------|
| GO:0034660 | ncRNA metabolic process | $8.1 \times 10^{-5}$ | 0.12 | 8 / 90 | 14 / 3305 | 411 / 14801 |
| GO:1902743 | regulation of lamellipodium organization | $3.7 \times 10^{-4}$ | 0.27 | 3 / 90 | 3 / 614 | 45 / 14801 |
| GO:1902745 | positive regulation of lamellipodium organization | $9.1 \times 10^{-4}$ | 0.32 | 3 / 90 | 3 / 362 | 411 / 14801 |
| GO:0021576 | hindbrain formation | 0.0011 | 0.32 | 1 / 90 | 1 / 908 | 1 / 14801 |
| GO:0021588 | cerebellum formation | 0.0011 | 0.32 | 1 / 90 | 1 / 908 | 1 / 14801 |
| GO:0060061 | Spemann organizer formation | 0.0022 | 0.38 | 1 / 90 | 1 / 901 | 2 / 14801 |
| GO:0030031 | cell projection assembly | 0.0026 | 0.38 | 6 / 90 | 9 / 3147 | 611 / 14801 |
| GO:1905920 | positive regulation of CoA transferase activity | 0.0033 | 0.38 | 1 / 90 | 1 / 615 | 2 / 14801 |
| GO:0022004 | midbrain-hindbrain boundary maturation during brain development | 0.0034 | 0.38 | 1 / 90 | 18 / 5320 | 1 / 14801 |
| GO:0010591 | regulation of lamellipodium assembly | 0.0036 | 0.38 | 3 / 90 | 3 / 216 | 34 / 14801 |
| GO:0045184 | establishment of protein localization | 0.0038 | 0.38 | 10 / 90 | 12 / 4727 | 1961 / 14801 |
| GO:0021551 | central nervous system morphogenesis | 0.0039 | 0.38 | 1 / 90 | 5 / 2530 | 2 / 14801 |
| GO:0016782 | transferase activity, transferring sulfur-containing groups | $6.1 \times 10^{-4}$ | 0.79 | 2 / 178 | 6 / 1162 | 8 / 14815 |
| GO:1904158 | axonemal central apparatus assembly | 0.0007 | 0.79 | 2 / 178 | 19 / 2595 | 6 / 14815 |
| GO:1903363 | negative regulation of cellular protein catabolic process | 0.0019 | 0.79 | 4 / 178 | 12 / 1422 | 69 / 14815 |
| GO:0045599 | negative regulation of fat cell differentiation | 0.0022 | 0.79 | 3 / 178 | 5 / 608 | 39 / 14815 |
| GO:0038161 | prolactin signaling pathway | 0.0033 | 0.79 | 1 / 178 | 2 / 604 | 1 / 14815 |
| GO:1990542 | mitochondrial transmembrane transport | 0.0049 | 0.79 | 4 / 178 | 12 / 1435 | 90 / 14815 |
| GO:0060998 | regulation of dendritic spine development | 0.0053 | 0.79 | 3 / 178 | 12 / 1947 | 61 / 14815 |
| GO:0011979 | response to organonitrogen compound | 0.0054 | 0.79 | 11 / 178 | 16 / 2452 | 848 / 14815 |
| GO:0060897 | negative regulation of ubiquitin-specific protease activity | 0.0055 | 0.79 | 1 / 178 | 1 / 364 | 2 / 14815 |
| GO:0021576 | hindbrain formation | 0.0066 | 0.79 | 1 / 178 | 6 / 908 | 1 / 14815 |
| GO:0021588 | cerebellum formation | 0.0066 | 0.79 | 1 / 178 | 6 / 908 | 1 / 14815 |
| GO:0042177 | negative regulation of protein catabolic process | 0.0069 | 0.79 | 4 / 178 | 12 / 1549 | 107 / 14815 |

**Table S12.** (continued on next page)
| GO Term | Definition | P-value | Adj. P-value | Study | Family | Population |
|---------|------------|---------|--------------|-------|--------|------------|
| SH — CE Ceratopipra Ancestor Branch Model |
| GO:0048278 | vesicle docking | 0.0011 | 0.66 | 2 / 45 | 2 / 1966 | 67 / 14793 |
| GO:1902296 | DNA strand elongation involved in cell cycle DNA replication | 0.0016 | 0.66 | 1 / 45 | 2 / 1285 | 1 / 14793 |
| GO:0034103 | regulation of tissue remodeling | 0.0019 | 0.66 | 2 / 45 | 3 / 2096 | 53 / 14793 |
| GO:1902983 | DNA strand elongation involved in mitotic DNA replication | 0.0022 | 0.66 | 1 / 45 | 2 / 895 | 1 / 14793 |
| GO:0010157 | response to chlorate | 0.0041 | 0.66 | 1 / 45 | 3 / 1461 | 2 / 14793 |
| GO:0034287 | detection of monosaccharide stimulus | 0.0062 | 0.66 | 1 / 45 | 1 / 162 | 1 / 14793 |
| GO:0009732 | detection of hexose stimulus | 0.0064 | 0.66 | 1 / 45 | 1 / 156 | 1 / 14793 |
| GO:0051594 | detection of glucose | 0.0066 | 0.66 | 1 / 45 | 1 / 152 | 1 / 14793 |
| GO:0071351 | cellular response to interleukin-18 | 0.0069 | 0.66 | 1 / 45 | 1 / 724 | 5 / 14793 |
| GO:1905380 | regulation of snRNA transcription by RNA polymerase II | 0.0072 | 0.66 | 1 / 45 | 4 / 1655 | 3 / 14793 |
| GO:0032148 | activation of protein kinase B activity | 0.0082 | 0.66 | 2 / 45 | 2 / 305 | 28 / 14793 |
| GO:0009730 | detection of carbohydrate stimulus | 0.0093 | 0.66 | 1 / 45 | 1 / 214 | 2 / 14793 |
| PEC — CE Ceratopipra Ancestor Branch Model |
| GO:0044848 | biological phase | 0.0023 | 0.73 | 3 / 53 | 35 / 11980 | 91 / 14785 |
| GO:0046449 | creatinine metabolic process | 0.0028 | 0.73 | 1 / 53 | 21 / 7554 | 1 / 14785 |
| GO:0120077 | angiogenic sprout fusion | 0.0033 | 0.73 | 1 / 53 | 3 / 900 | 1 / 14785 |
| GO:0120078 | cell adhesion involved in sprouting angiogenesis | 0.0036 | 0.73 | 1 / 53 | 4 / 1111 | 1 / 14785 |
| GO:0043254 | regulation of protein-containing complex assembly | 0.0047 | 0.73 | 5 / 53 | 10 / 2831 | 360 / 14785 |
| GO:0010157 | response to chlorate | 0.0055 | 0.73 | 1 / 53 | 4 / 1461 | 2 / 14785 |
| GO:0034103 | regulation of tissue remodeling | 0.0058 | 0.73 | 2 / 53 | 5 / 2095 | 52 / 14785 |
| GO:0044087 | regulation of cellular component biogenesis | 0.0061 | 0.73 | 7 / 53 | 23 / 8378 | 847 / 14785 |
| GO:0034287 | detection of monosaccharide stimulus | 0.0062 | 0.73 | 1 / 53 | 1 / 162 | 1 / 14785 |
| GO:0009732 | detection of hexose stimulus | 0.0064 | 0.73 | 1 / 53 | 1 / 156 | 1 / 14785 |
| GO:0051594 | detection of glucose | 0.0066 | 0.73 | 1 / 53 | 1 / 152 | 1 / 14785 |
| GO:0106088 | regulation of cell adhesion involved in sprouting angiogenesis | 0.007 | 0.73 | 1 / 53 | 4 / 573 | 1 / 14785 |

Table S12. GO Terms overrepresented in top gene sets from each PhyDGET models Gene Ontology terms using showing the P-value of overrepresentation, the BH-adjusted P-value, the proportion of genes represented by the term in the target study set, the number of terms in the total GO term family, and the total proportion of the GO term in the total population.
| Gene                 | log$_2$ FC | log$_2$ avg. exp. | P-value       | Adjusted P-value | Mod. | BF | Annotation                                |
|---------------------|-----------|-------------------|---------------|------------------|------|----|-------------------------------------------|
| LOC113999394        | 3.67      | 3.11              | 5.1 x 10$^{-11}$ | 6.1 x 10$^{-7}$ | R41  | 1.48 | alpha-globin transcription factor         |
| LOC113989613        | 5.06      | 5.73              | 7.8 x 10$^{-11}$ | 6.1 x 10$^{-7}$ | MC1  | 0.98 | uncharacterized LOC113989613             |
| LOC113990940        | 3.44      | 3.62              | 3.2 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | M4   | 0.88 | uncharacterized LOC113990940             |
| SEC14L5             | 4.64      | 6.37              | 4.0 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | null | N/A | SEC14 like lipid binding 5               |
| PREPL               | 4.62      | 4.81              | 4.1 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | R41  | 0.44 | prolyl endopeptidase like                |
| LOC113998190        | 3.56      | 4.72              | 4.3 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | null | N/A | dynein heavy chain 5, axonemal-like      |
| MAP3K15             | 4.26      | 5.64              | 6.7 x 10$^{-10}$ | 1.2 x 10$^{-6}$ | null | N/A | mitogen-activated protein kinase kinase 15 |
| STRIP2              | 3.24      | 4.12              | 6.9 x 10$^{-10}$ | 1.2 x 10$^{-6}$ | null | N/A | striatin interacting protein 1           |
| PLGRKT              | 3.87      | 5.59              | 7.1 x 10$^{-10}$ | 1.2 x 10$^{-6}$ | null | N/A | plasminogen receptor with a C-terminal lysine |
| CEP131              | 3.12      | 3.7               | 1.1 x 10$^{-9}$ | 1.2 x 10$^{-6}$ | MC1  | 0.37 | centrosomal protein 131                  |
| LOC113998077        | 3.23      | 2.83              | 1.2 x 10$^{-9}$ | 1.2 x 10$^{-6}$ | M5   | 0.15 | fer-1-like protein 4                     |
| PHH1D1              | 3.64      | 4.36              | 1.2 x 10$^{-9}$ | 1.2 x 10$^{-6}$ | MV   | 0.81 | PIH1 domain containing 1                 |
| GDF1                | 4.32      | 1.69              | 2.3 x 10$^{-12}$ | 3.5 x 10$^{-6}$ | R41  | 1.64 | growth differentiation factor 10         |
| PUDP                | 3.33      | 4.11              | 1.4 x 10$^{-10}$ | 1.0 x 10$^{-6}$ | M4   | 0.03 | pseudouridine 5'-phosphatase             |
| IL18                | 4.25      | 2.22              | 2.2 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | CE   | 3.2  | interleukin 18 binding protein           |
| PIH1D1              | 3.89      | 4.32              | 3.0 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | null | N/A | PIH1 domain containing 1                 |
| ACOT7               | 3.26      | 4.26              | 5.3 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | null | N/A | acyl-CoA thioesterase 7                  |
| GAA                 | 3.85      | 3.9               | 5.6 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | CE   | 0.96 | glucosidase alpha, acid                  |
| SFRP4               | 4.1       | 3.87              | 6.1 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | null | N/A | secreted frizzled related protein 4       |
| PREPL               | 4.07      | 4.32              | 6.2 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | R41  | 1.41 | prolyl endopeptidase like                |
| TTC31               | 3.38      | 4.33              | 8.0 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | LC   | 1.08 | tetratricopeptide repeat domain 31       |
| LOC113998860        | 2.9       | 3.27              | 8.2 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | M4   | 0.25 | uncharacterized LOC113998860             |
| MSANTD4             | 3.09      | 4.04              | 8.3 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | M4   | 0.63 | Myb/SANT DNA binding domain containing 4 with coiled-coils |
| IQCB1               | 3.25      | 3.9               | 9.1 x 10$^{-10}$ | 1.1 x 10$^{-6}$ | M4   | 0.23 | IQ motif containing B1                   |

Table S13. Top 12 differentially expressed genes in contrasts for each tissue for RWM species versus non-RWM species. List of top genes (by P-value) identified by limma as differentially expressed using a flat comparison of all RWM versus all non-RWM individuals. The log2(fold change), log2 average expression, P-value, Benajmini-Hochberg adjusted P-value, best fitting PhyDGET model and Bayes Factor, and reference annotation are shown.
### Calcium Transport and Signaling

| Gene                  | Model Name | Notes on Function                                                                                                                                                                                                                                                                                                                                 |
|-----------------------|------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| **CIB2** (−)          | M6         | **Calcium And Integrin Binding Family Member 2**: “responsible for maintaining Ca^{2+} homeostasis in cells and interacting with integrins-transmembrane receptors essential for cell adhesion, migration, and activation of signaling pathways. . . Within the muscle, Cib2 is expressed in sarcolemma, enriched in the myotendinous junctions and neuromuscular junctions” (40). |
| **CALM2** [+ ]        | M5         | **Calmodulin 2**: “mediates the control of a large number of enzymes, ion channels, aquaporins and other proteins through calcium-binding” (UniProtKB). Calmodulin activates ryanodine receptor (41).                                                                                              |
| **ATP2A3** (+) and "ATP2A1-like" LOC114003417 (+) | M4         | **ATPase Sarcoplasmic/Endoplasmic Reticulum Ca^{2+} Transporting 3 and ATPase Sarcoplasmic/Endoplasmic Reticulum Ca^{2+} Transporting 1** (a.k.a. SERCA): “encodes one of the SERCA Ca(2+)-ATPases, which are intracellular pumps located in the sarcoplasmic or endoplasmic reticula of muscle cells. This enzyme catalyzes the hydrolysis of ATP coupled with the translocation of calcium from the cytosol to the sarcoplasmic reticulum lumen, and is involved in muscular excitation and contraction. Mutations in this gene cause some autosomal recessive forms of Brody disease, characterized by increasing impairment of muscular relaxation during exercise” (NCBI RefSeq). Domes-ticated chicken and turkeys studies indicate association with RYR and strong impact on muscle properties (42, 43). |
| **RYR1-like-1** [+/−] LOC113994824 and **RYR1-like-2** [+/−] LOC114001935 | R41       | **Ryanodine Receptor 1**: Calcium channel that mediates the release of Ca(2+) from the sarcoplasmic reticulum into the cytoplasm and thereby plays a key role in triggering muscle contraction following depolarization of T-tubules. Repeated very high-level exercise increases the open probability of the channel and leads to Ca(2+) leaking into the cytoplasm” (UniProtKB). Studies in domesticated chicken have shown this gene is associated with ATP2A1 and the expression has strong impact on muscle properties (44, 43). |
| **TRDN** [+/−]        | R41       | **Triadin**: “Contributes to the regulation of lumenal Ca^{2+} release via the sarcoplasmic reticulum calcium release channels RYR1 and RYR2, a key step in triggering skeletal and heart muscle contraction. Required for normal organization of the triad junction, where T-tubules and the sarcoplasmic reticulum terminal cisternae are in close contact. Required for normal skeletal muscle strength” (UniProtKB). Known to interact with RYR1 and Parvalbumin as part of the muscle contractile system. |
| **CCDC47** (+)        | MC1        | **Coiled-Coil Domain Containing 47**: Involved in calcium ion maintenance in the endoplasmic reticulum (45).                                                                                                                                                                                                                                           |
| **SRL** [+ ]          | MC1        | **Sarcalumenin**: “SAR significantly contributes to Ca^{2+} buffering and the maintenance of the SERCA protein in the skeletal muscle SR” (46). Also interacts with RYR1 and ATP2A1.                                                                                                                                                                   |
| **OBSCN** [+ ]        | MC1        | **Obscurin**: “Structural component of striated muscles which plays a role in myofibrillogenesis. Probably involved in the assembly of myosin into sarcomeric A bands in striated muscle.” (UniProtKB)                                                                                                                                                                   |
| **CLRN1** (+)         | MV         | **Clarin 1**: “encodes a protein that contains a cytosolic N-terminus, multiple helical transmembrane domains, and an endoplasmic reticulum membrane retention signal, TKGH, in the C-terminus” (NCBI RefSeq). Reorganized actin filaments and created extracellular lamellipodia (47). |
| **PVALB-like-1** (+) LOC113990673 | CE         | **Parvalbumin, Avian Thymic Hormone**: An parvalbumin first identified in chicken (48). Parvalbumins in manakins and zebrafinch “enhance cellular Ca(2+) cycling and hypertrophy of skeletal muscle fibers” (49).                                                                                                                                 |
| **PVALB-like-2** (+) LOC113990672 | CE         | **Parvalbumin**: “In muscle, parvalbumin is thought to be involved in relaxation after contraction. It binds two calcium ions.” (UniProtKB). Parvalbumins in manakins and zebrafinch “enhance cellular Ca(2+) cycling and hypertrophy of skeletal muscle fibers” (49). |

Table S14. (continued on next page)
| Gene         | Model Name | Notes on Function                                                                                                                                                                                                 |
|--------------|------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| **Metabolism and Oxidative Stress**                                                                                                                                  |
| **PDK2 (+)** | M6         | **Pyruvate Dehydrogenase Kinase 2:** "Key role in the regulation of glucose and fatty acid metabolism and homeostasis via phosphorylation of the pyruvate dehydrogenase subunits PDHA1 and PDHA2. Mediates cellular responses to insulin. Plays an important role in maintaining normal blood glucose levels and in metabolic adaptation to nutrient availability" (UniProtKB). |
| **PFKP (+)** | M6         | **Phosphofructokinase, Platelet:** "Catalyzes the phosphorylation of D-fructose 6-phosphate to fructose 1,6-bisphosphate by ATP, the first committing step of glycolysis" (UniProtKB; 50). |
| **YARS2 (–)**| M6         | **Tyrosyl-TRNA Synthetase 2:** "... a mitochondrial protein that catalyzes the attachment of tyrosine to tRNA(Tyr). Mutations in this gene are associated with myopathy with lactic acidosis and sideroblastic anemia type 2." (NCBI RefSeq) |
| **PP1R3C [+]**| M5         | **Protein Phosphatase 1 Regulatory Subunit 3C:** "... a carbohydrate binding protein that is a subunit of the protein phosphatase 1 (PP1) complex. PP1 catalyzes reversible protein phosphorylation, which is important in a wide range of cellular activities. The encoded protein affects glycogen biosynthesis by activating glycogen synthase and limiting glycogen breakdown by reducing glycogen phosphorylase activity" (NCBI RefSeq; 51). |
| **PPARGC1B [+]**| M5         | **Peroxisome Proliferator-Activated Receptor Gamma, Coactivator 1 Beta:** "... fatty acids differentially regulated expression of the genes encoding the PGC-1 isoforms. ... accompanied by significant changes in mitochondrial activity, ... fatty acid-induced regulation of expression of these genes plays an important role in muscle oxidative metabolism" (52). |
| **MLH1 (+)** | M4         | **MutL Homolog 1:** "Heterodimerizes with PMS2 to form MutL alpha, a component of the post-replicative DNA mismatch repair system (MMR). ... Also implicated in DNA damage signaling, a process which induces cell cycle arrest and can lead to apoptosis in case of major DNA damages" (UniProtKB). |
| **BCO2 (–)** | MC1        | **Beta-Carotene Oxygenase 2:** "BCO2 is a carotene-cleaving enzyme that localizes to mitochondria and catalyzes the 9priime,109priimeoxidative cleavage of carotenoids, an es-sential step in carotenoid degradation" (quote from Gazda 2020; Amengual 2011, dela Seña 2016). Also responsible for plumage coloration in some Passerine birds (53). |
| **ARFGAP3 (+)** | MV         | **ADP Ribosylation Factor GTPase Activating Protein 3:** "GTPase-activating protein (GAP) for ADP ribosylation factor 1 (ARF1). Hydrolysis of ARF1-bound GTP may lead to dissociation of coatamer from Golgi-derived membranes to allow fusion with target membranes" (UniProtKB). Found to be induced by oxidative stress response in mouse smooth muscles (54). |
| **HEMK1 (–)** | MV         | **HemK Methyltransferase Family Member 1:** "N5-glutamine methyltransferase responsible for the methylation of the glutamine residue in the universally conserved GGQ motif of the mitochondrial translation release factor MTRF1L" (UniProtKB). |
| **LTA4H (–)** | MV         | **Leukotriene A4 Hydrolase:** "The protein encoded by this gene is an enzyme that contains both hydrolase and aminopeptidase activities. The hydrolase activity is used in the final step of the biosynthesis of leukotriene B4, a proinflammatory mediator." (NCBI RefSeq). |
| **PON2 (+)** | MV         | **Paraoxonase 2:** "This gene encodes a member of the paraoxonase gene family, which includes three known members located adjacent to each other on the long arm of chromosome 7. The encoded protein is ubiquitously expressed in human tissues, membrane-bound, and may act as a cellular antioxidant, protecting cells from oxidative stress." (NCBI RefSeq) Highly concentrated in the endoplasmic reticulum and can have an interplay with calcium levels (55) |

Table S14. (continued on next page)
| Gene            | Model Name | Notes on Function                                                                                                                                                                                                                                                                                                                                 |
|-----------------|------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| **Muscle Tone and Repair**                                                                                                                                                                                                                                                                                                                                 |
| *CIAPIN1* (+)   | M6         | **Cytokine Induced Apoptosis Inhibitor 1**: "Component of the cytosolic iron-sulfur (Fe-S) protein assembly machinery required for the maturation of extramitochondrial Fe-S proteins. Part of an electron transfer chain functioning in an early step of cytosolic Fe-S biogenesis" (UniProtKB). |
| *FOXF1* (+)     | M5         | **Forkhead Box F1**: Transcription factor active in the lungs and blood vessels. Regulates contractile proteins in smooth muscle (56).                                                                                                                                                                                                            |
| *NAA35* (–)     | M5         | **N-Alpha-Acetyltransferase 35, NatC Auxiliary Subunit**: "Auxiliary component of the N-terminal acetyltransferase C (NatC) complex which catalyzes acetylation of N-terminal methionine residues. Involved in regulation of apoptosis and proliferation of smooth muscle cells" (UniProtKB). |
| *SLC44A3* (+)   | M5         | **Solute Carrier Family 44 Member 3**: Choline-like transporter family protein of uncertain function. Identified as a gene associated with high muscle stress response in rats (57) and humans (58).                                                                                     |
| *RPAP2* [–]     | M4         | **RNA Polymerase-associated Protein 2**: "Protein phosphatase that displays CTD phosphatase activity and regulates transcription of snRNA genes" (UniProtKB). Abnormal expression has been associated with myopathy (59).                                                      |
| *TFCP2* (+/–)   | R41        | **Alpha-Globin Transcription Factor CP2**: "transcription factor that binds the alpha-globin promoter and activates transcription of the alpha-globin gene. The encoded protein regulates erythroid gene expression, plays a role in the transcriptional switch of globin gene promoters." (NCBI RefSeq). |
| *CUL1* [+/-]    | R41        | **Cullin 1**: "Core component of multiple cullin-RING-based SCF (SKP1-CUL1-F-box protein) E3 ubiquitin-protein ligase complexes, which mediate the ubiquitination of proteins involved in cell cycle progression, signal transduction and transcription" (UniProtKB). Related to ubiquitination of SMAD4, Fbox proteins. |
| *TTN* [+/-]     | R41        | **Titin**: "Key component in assembly and functioning of vertebrate striated muscles. By providing connections at the level of individual microfilaments, it contributes to the fine balance of forces between the two halves of the sarcomere" (UniProtKB).                                                                 |
| "*PREPL* [+/-]  | R41        | **Prolyl Endopeptidase Like**: "Serine peptidase whose precise substrate specificity remains unclear. Does not cleave peptides after a arginine or lysine residue. Regulates trans-Golgi network morphology and sorting by regulating the membrane binding of the AP-1 complex. May play a role in the regulation of synaptic vesicle exocytosis" (UniProtKB; see also 60, 61) |
| "MYH3-like" LOC113989613 [+]| MC1       | **Myosin Heavy Chain 3**: "Major contractile protein which converts chemical energy into mechanical energy through the hydrolysis of ATP." (NCBI RefSeq)                                                                                                                           |
| "ROR1-like" LOC114002148 [+]| MC1       | **Receptor Tyrosine Kinase Like Orphan Receptor 1**: "receptor tyrosine kinase-like orphan receptor that modulates neurite growth in the central nervous system." Shown to play a key role muscle regeneration after injury (62).                                                                 |
| Gene            | Model Name | Notes on Function                                                                                                                                 |
|-----------------|------------|--------------------------------------------------------------------------------------------------------------------------------------------------|
| **Muscle Tone and Repair (continued)**                                                                                                                         |
| ANK2 (-)        | MV         | Ankyrin Repeat 2: “In skeletal muscle, required for localization of DMD and DCTN4 and for the formation and/or stability of a special subset of microtubules associated with costameres and neuromuscular junctions” (UniProtKB). |
| STAU2 (+)       | MV         | Staufen Double-Stranded RNA Binding Protein 2: “Stau1 and Stau2 are key components of the postsynaptic apparatus in muscle, and that they contribute to the maturation and plasticity of the neuromuscular junction” (63). |
| MYBPH (+)       | MV         | Myosin Binding Protein H: “Binds to myosin; probably involved in interaction with thick myofilaments in the A-band” (UniProtKB).                     |
| NINJ2 (+)       | CE         | Ninjurin 2: “Ninjurin (for nerve injury induced) family. Cell surface adhesion protein upregulated in Schwann cells surrounding the distal segment of injured nerve, and promotes neurite outgrowth, thus may have a role in nerve regeneration after nerve injury.” (NCBI RefSeq) Found to be a regulator of cells after endothelial inflammation and blood vessel after injuries (64) |
| STX2 (+)        | CE         | Syntaxin 2: “The syntaxins are a large protein family implicated in the targeting and fusion of intracellular transport vesicles. The product of this gene regulates epithelial-mesenchymal interactions and epithelial cell morphogenesis and activation” (NCBI RefSeq) |
| **Endoplasmic Reticulum Stress**                                                                                                                              |
| FOXO3 (-)       | M5         | Forkhead Box O3: “Acts as a positive regulator of autophagy in skeletal muscle: in starved cells, enters the nucleus following dephosphorylation and binds the promoters of autophagy genes, such as GABARAP1L, MAP1LC3B and ATG12, thereby activating their expression, resulting in proteolysis of skeletal muscle proteins” (UniProtKB). |
| RETREG1 (+)     | M5         | Reticulophagy Regulator 1 (a.k.a. FAM134B): “Endoplasmic reticulum-anchored autophagy receptor that mediates ER delivery into lysosomes through sequestration into autophagosomes” (UniProtKB). Involved also in ER turnover during stress (65). |
| DDRGK1 (+)      | M4         | DDRGK Domain Containing 1: “UFMylation, a ubiquitin-like post-translational modification, is required for ER-phagy. The protein DDRGK1 recruits UFMylation machinery to the ER surface in a striking parallel to the mitophagic recruitment of Parkin by PINK1. DDRGK1 is specifically required for the ER-phagy of ER sheets, including ER-phagy mediated by LIR/GIM receptors located on these subdomains. Unbiased proteomics identified Ribophorin 1 (RPN1), an ER-localized quality control factor, as an ER sheet-localized target of DDRGK1-dependent UFMylation” (66). |
| SPPL2A (+)      | R41        | Signal Peptide Peptidase Like 2A: “Intramembrane-cleaving aspartic protease (L-CLIP) that cleaves type II membrane signal peptides in the hydrophobic plane of the membrane. Functions in FASLG, ITM2B and TNF processing” (UniProtKB). The SPP family alters ER shape by degradation of morphogenic proteins (67). |
| TPRKB (+)       | MV         | TP53 Regulating Kinase Binding Protein: “Component of the EKC/KEOPS complex that is required for the formation of a threonylcarbamoyl group on adenosine at position 37 (t(6)A37) in tRNAs that read codons beginning with adenine” (UniProtKB). Associated with chronic ER stress and defects in actin (68). |
| UFL1 (+)        | MC1        | Ubiquitin fold modifier 1 Specific Ligase: “E3 protein ligase that mediates ufmylation, the covalent attachment of the ubiquitin-like modifier UFM1 to lysine residues on target proteins, and which plays a key role in reticulophagy (also called ER-phagy) induced in response to endoplasmic reticulum stress” (UniProtKB). |

Table S14. (continued on next page)
| Gene                  | Model Name | Notes on Function                                                                                                                                 |
|----------------------|------------|--------------------------------------------------------------------------------------------------------------------------------------------------|
| 26S Proteasome Complex |            |                                                                                                                                                   |
| UBE3B [-]            | M5         | Ubiquitin Conjugating Enzyme E2 B: “Accepts ubiquitin from the E1 complex and catalyzes its covalent attachment to other proteins. In association with the E3 enzyme BRE1 (RNF20 and/or RNF40), it plays a role in transcription regulation by catalyzing the monoubiquitination of histone H2B.” (Uniprot) Loss of this protein is associated with myofibrillar protein loss (69). |
| FBXO4 (-)            | M4         | F-Box Protein 4: “Encodes a member of the F-box protein family which is characterized by an approximately 40 amino acid motif, the F-box. The F-box proteins constitute one of the four subunits of the ubiquitin protein ligase complex called SCFs.” (NCBI RefSeq). SCF complex catalyzes ubiquitination for the 26S Proteasome complex. |
| PSMA7 (+)            | M4         | Proteasome 20S Subunit Alpha 7: 20S subunit of the 26S Proteasome.                                                                                   |
| PSMA2 (+)            | M4         | Proteasome 20S Subunit Alpha 2: 20S subunit of the 26S Proteasome.                                                                                   |
| PSMC3 (-/+           | R41        | Proteasome 26S Subunit, ATPase 3: ATPase subunit of the 26S Proteasome.                                                                            |
| PSMC6 (+)            | MC1        | Proteasome 20S Subunit Alpha 6: 20S subunit of the 26S Proteasome.                                                                                   |
| UBXN1 (-)            | MV         | UBX Domain Protein 1: “Ubiquitin-binding protein that plays a role in the modulation of innate immune response. Blocks both the RIG-I-like receptors (RLR) and NF-kappa-B pathways. ... Component of a complex required to couple deglycosylation and proteasome-mediated degradation of misfolded proteins in the endoplasmic reticulum that are retrotranslocated in the cytosol” (UniProtKB). Reported in association follistatin induced muscle hypertrophy (70). |
| Cell-cell Junctions and Extracellular Matrix |            |                                                                                                                                                   |
| "ERVFRD-1-like" (+)  | LOC113993297 | Endogenous Retrovirus Group FRD Member 1, Envelope (a.k.a. syncitin 2): “This endogenous retroviral envelope protein has retained its original fusogenic properties and participates in trophoblast fusion and the formation of a syncytium during placenta morphogenesis” (UniProtKB). Associated with the placenta usually, but has also been observed forming syncitia in muscle cells, and with high levels causing unusual muscle phenotypes (71, 72). |
| ADAMTS9 (+)          | M5         | ADAM Metallopeptidase With Thrombospondin Type 1 Motif 9: This gene has been shown to alter the sensitivity of skeletal muscles to insulin signaling by changes to the extracellular matrix surrounding myocytes (73). |
| DPYSL5 (+)           | M4         | Dihydropyrimidinase Like 5 (a.k.a. ULIP6): glycine transporter interacting protein that is implicated as involved in neurological response in ‘startle’ syndromes (74). |
| STUM (+)             | M4         | Stum, Mechanosensory Transduction Mediator Homolog (a.k.a. stumble): “required for transduction of mechanical stimuli in a specific subpopulation of Drosophila proprioceptive neurons that sense joint angles.” and “dendritic stretching also elicited elevation of cellular Caty2+ levels — not seen in stum mutants” (75). The same study also noted that mouse homologs rescued deletion, indicating conservation of sequence. |
| UBAC2 (+/-)          | R41        | UBA Domain Containing 2 (a.k.a. TM4): “ Restricts trafficking of FAF2 from the endoplasmic reticulum to lipid droplets” (UniProtKB). Insulin-sensitive and associated with inflammatory disease Behçet’s syndrome. |
| HACD1 (+)            | MV         | 3-Hydroxyacyl-CoA Dehydratase 1: “involved in the elongation of the very long chain fatty acids, in muscle fibre formation. In humans and dogs, HACD1 deficiency leads to a congenital myopathy with fibre size disproportion associated with a generalized muscle weakness.” “promotes myoblast fusion during muscle development and regeneration” “catalyzes lipid modifications correlate with a reduction in plasma membrane rigidity” (76, 77) |

Table S14. (continued on next page)
| Gene                | Model Name | Notes on Function                                                                                                                                                                                                                                                                                                                                                                                                                                                                                           |
|---------------------|------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| **Androgen Signaling** |            |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                |
| "KDM5C-like" (–)    | M6         | **Lysine Demethylase 5C**: "Histone demethylase that specifically demethylates 'Lys-4' of histone H3, thereby playing a central role in histone code. Among its related pathways are Chromatin organization and Activated PKN1 stimulates transcription of AR (androgen receptor) regulated genes KLK2 and KLK3" (UniProtKB).                                                                 |
| LOC113993369        |            |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                |
| **EAF2** (+)         | M5         | **ELL Associated Factor 2**: “EAF2-deficiency on either background was associated with an increase in epithelial cell proliferation, the development of mPIN lesions as well as an associated increased incidence in reactive stroma and increased vascularity” (78). Gene expression stimulated by androgen (79).                                                                                                       |
| **KDM4B** (+)        | M4         | **Lysine Demethylase 4B**: "Histone demethylase that specifically demethylates 'Lys-9' of histone H3" (UniProtKB). Known to be a key molecule in androgen receptor signaling and promotes turnover of AR (80).                                                                                                                                         |
| **AR** [+]           | MC1        | **Androgen Receptor**: Direct evidence in manakins associates androgen receptor signaling with rapid wing movement both in neural cells and muscular fibers. (81, 82, 83).                                                                                                                                                                                                                                                                             |
| **HIPK3** (+)        | CE         | **Homeodomain Interacting Protein Kinase 3**: “Serine/threonine-protein kinase involved in transcription regulation, apoptosis and steroidogenic gene expression. Phosphorylates JUN and RUNX2. Seems to negatively regulate apoptosis by promoting FADD phosphorylation. Enhances androgen receptor-mediated transcription. May act as a transcriptional corepressor for NK homeodomain transcription factors. The phosphorylation of NR5A1 activates SF1 leading to increased steroidogenic gene expression upon cAMP signaling pathway stimulation” (UniProtKB). |

Table S14. Functional information for the genes highlighted in Figure 3. Basic database descriptions from UniProtKB and NCBI human RefSeq are included for most genes. Additional references are cited specific to birds or muscles, where available and appropriate. Model codes are shown in Fig. S4.
| Gene         | Len | $d_N$ | $d_S$ | $d_N/d_S$ | $P$ | BF SH | BF PEC | Annotation                                      |
|--------------|-----|-------|-------|-----------|-----|-------|--------|------------------------------------------------|
| THAP9        | 111 | 0.06  | 0.117 | 0.51      | 1   | 1.31  | 0.48   | THAP domain containing 9                       |
| RESF1        | 1723| 0.056 | 0.094 | 0.6        | 0.89| -0.39 | -0.2   | retroelement silencing factor 1                |
| LOC11399513  | 386 | 0.053 | 0.078 | 0.68      | 0.98| -0.49 | -0.5   | uncharacterized LOC113995132                  |
| NDUFC1       | 143 | 0.053 | 0.1    | 0.53      | 1   | -0.57 | -0.39  | NADH:ubiquinone oxidoreductase subunit C1      |
| LOC113998632 | 138 | 0.051 | 0.464 | 0.11      | 0.25| -0.47 | -0.03  | very-long-chain enoyl-CoA reductase-like       |
| SERPINF2     | 207 | 0.037 | 0.036 | 1.05      | 0.79| -0.43 | -0.38  | serpin family F member 2                      |
| CUNH19orf44  | 151 | 0.037 | 0.025 | 1.52      | 0.02| -0.5  | -0.39  | chromosome unknown C19orf44 homolog            |
| LOC11398634  | 110 | 0.035 | 0.033 | 1.04      | 1   | -0.35 | -0.28  | ras-related and estrogen-regulated growth inhibitor-like |
| CD164        | 165 | 0.034 | 0.018 | 1.87      | 0.06| -0.38 | 0.6    | CD164 molecule                                 |
| LOC113983299 | 120 | 0.033 | 0.029 | 1.13      | 0.17| -0.15 | -0.49  | skin secretory protein xP2-like                |
| CCDC15       | 270 | 0.033 | 0.098 | 0.33      | 0.39| 2.02  | 0.37   | coiled-coil domain containing 15               |
| KNOB1        | 610 | 0.03  | 0.026 | 1.19      | 0.24| 0.59  | 1.44   | lysine rich nucleolar protein 1                |
| SLC16A4      | 159 | 0.03  | 0.029 | 1.05      | 0.56| -0.26 | -0.14  | solute carrier family 16 member 4             |
| ETAA1        | 121 | 0.024 | 0.018 | 1.29      | 0.24| 0.05  | -0.03  | ETAA1, ATR kinase activator                   |
| LOC114000836 | 150 | 0.023 | 0.021 | 1.08      | 0.17| -0.47 | -0.55  | uncharacterized LOC114000836                  |
| LOC114004335 | 193 | 0.022 | 0.012 | 1.79      | 0.38| -0.21 | -0.36  | zinc finger protein RFP-like                  |
| FABP7        | 103 | 0.021 | 0.017 | 1.23      | 1   | -0.57 | -0.51  | fatty acid binding protein 7                  |
| SYNJ2BP      | 146 | 0.016 | 0.016 | 1.02      | 0.12| -0.51 | -0.11  | synaptojanin 2 binding protein                |
| CCDC173      | 109 | 0.014 | 0.014 | 1.03      | 1   | -0.4  | -0.58  | coiled-coil domain containing 173             |
| LOC113991506 | 137 | 0.013 | 0.011 | 1.16      | 0.42| -0.42 | -0.52  | uncharacterized LOC113991506                  |
| EFCA1        | 111 | 0.011 | 0.006 | 2.01      | 0.33| -0.5  | -0.49  | EF-hand calcium binding domain 1              |
| FAM174A      | 117 | 0.01  | 0.01  | 1.05      | 0.21| -0.41 | -0.6   | family with sequence similarity 174 member A  |
| LOC113983346 | 111 | 0.01  | 0.009 | 1.15      | 0.12| 0.37  | 0.4    | sodium channel modifier 1-like               |
| LOC114000280 | 207 | 0.007 | 0.005 | 1.3       | 1   | 0.15  | 0.56   | UPF0488 protein C8orf33 homolog               |
| EAF2         | 207 | 0    | 0.01  | 0.03     | 0.6 | 1.77  | 1.09   | ELL associated factor 2                      |
| RETREG1      | 372 | 0.004 | 0.021 | 0.2       | 0.03| 1.65  | 0.14   | reticulophagy regulator 1                    |
| FOXO3        | 601 | 0    | 0.015 | 0.02     | 0.67| 1.59  | 0.89   | forkhead box O3                              |
| LOC113991453 | 151 | 0.001 | 0.054 | 0.02      | 0.36| 1.56  | 0.26   | semaphorin-3D-like                           |
| DHPS         | 379 | 0.001 | 0.04  | 0.02      | 0.99| 1.55  | 1.37   | deoxyhypusine synthase                       |
| ADAMTS9      | 923 | 0    | 0.001 | 0.05     | 0.29| 1.53  | 1.21   | ADAM metalloproteinase with thrombospondin type 1 motif 9 |
| COMM2D       | 199 | 0.001 | 0.004 | 0.19      | 0.99| 1.39  | 1.63   | COMM domain containing 2                     |
| LOC113994576 | 479 | 0.001 | 0.04  | 0.01      | 0.82| 1.22  | 1.93   | ATP-dependent RNA helicase DDX19B             |

Table S15. (continued on next page)
| Gene                     | Len | $d_N$ | $d_S$ | $d_N/d_S$ | $P$ | BF SH | BF PEC | Annotation                                                                 |
|-------------------------|-----|-------|-------|-----------|-----|-------|--------|---------------------------------------------------------------------------|
| LOC114001961            | 102 | 0.022 | 0.046 | 0.48      | 0.59| −0.71 | −0.7   | aldo-keto reductase family 1 member B10-like                               |
| MRPL4                   | 225 | 0.016 | 0.103 | 0.15      | 0.89| −0.46 | −0.61  | mitochondrial ribosomal protein L4                                        |
| IRAK1BP1                | 125 | 0.014 | 0      | ∞         | 0.07| −0.15 | −0.24  | interleukin 1 receptor associated kinase 1 binding protein 1              |
| ALDH16A1                | 311 | 0.013 | 0.028 | 0.47      | 0.19| −0.42 | −0.57  | aldehyde dehydrogenase 16 family member A1                                |
| LOC113998672            | 110 | 0.012 | 0      | ∞         | 0.06| 0.38  | 0.04   | NADH dehydrogenase 1 subunit C2-like                                      |
| CISD3                   | 106 | 0.011 | 0      | ∞         | 0.18| −0.71 | −0.71  | CDGSH iron sulfur domain 3                                               |
| NDUFS5                  | 106 | 0.01  | 0      | ∞         | 0.1 | −0.49 | −0.26  | NADH:ubiquinone oxidoreductase subunit S5                                 |
| TIGAR                   | 276 | 0.01  | 0      | ∞         | 0   | −0.69 | −0.55  | TP53 induced glycolysis regulatory phosphatase                            |
| FBXL15                  | 198 | 0.01  | 0.006 | 1.66      | 0.25| −0.62 | −0.02  | F-box and leucine rich repeat protein 15                                   |
| LOC114003559            | 132 | 0.01  | 0      | ∞         | 0.04| −0.63 | −0.27  | fatty acid-binding protein, adipocyte                                     |
| FAM204A                 | 242 | 0.01  | 0      | ∞         | 0.02| −0.67 | −0.6   | family with sequence similarity 204 member A                              |
| DNAJC9                  | 258 | 0.01  | 0      | ∞         | 0   | 0.16  | 0.39   | DnaJ heat shock protein family (Hsp40) member C9                           |
| CD164                   | 165 | 0.01  | 0      | ∞         | 0.18| 0.62  | −0.15  | CD164 molecule                                                            |
| PPCS                    | 173 | 0.009 | 0      | ∞         | 0   | −0.58 | −0.51  | phosphopantetheinecysteine synthetase                                     |
| SLC35G1                 | 308 | 0.009 | 0.004 | 2.24      | 0.03| −0.64 | −0.32  | solute carrier family 35 member G1                                       |
| DGCR2                   | 519 | 0.009 | 0.003 | 3.42      | 0.01| −0.47 | −0.63  | DiGeorge syndrome critical region gene 2                                  |
| LOC113984987            | 218 | 0.009 | 0.007 | 1.35      | 0.15| −0.25 | −0.35  | glutathione S-transferase-like                                            |
| ELP6                    | 256 | 0.008 | 0.007 | 1.19      | 0.05| −0.08 | −0.24  | elongator acetyltransferase complex subunit 6                             |
| NAF1                    | 328 | 0.008 | 0      | ∞         | 0.03| −0.64 | −0.27  | nuclear assembly factor 1 ribonucleoprotein                              |
| MRPS6                   | 121 | 0.008 | 0      | ∞         | 0.07| −0.22 | −0.34  | mitochondrial ribosomal protein S6                                        |
| VGILL3                  | 139 | 0.008 | 0      | ∞         | 0.02| −0.66 | −0.6   | vestigial like family member 3                                            |
| MPC2                    | 129 | 0.008 | 0      | ∞         | 0.01| −0.18 | −0.22  | mitochondrial pyruvate carrier 2                                          |
| CMBL                    | 245 | 0.007 | 0.008 | 0.96      | 0.21| −0.52 | −0.33  | carboxymethylenebutenolidase homolog                                     |
| CTSO                    | 395 | 0.007 | 0.003 | 2.37      | 0.05| 0.09  | −0.02  | cathepsin D                                                               |
| FUNDC2                  | 152 | 0.006 | 0      | ∞         | 0.02| 0.57  | 0.81   | FUN14 domain containing 2                                                |
| NIPSAP3A                | 250 | 0.006 | 0      | ∞         | 0.05| −0.34 | −0.68  | nipsnap homolog 3A                                                        |
| GNL3                    | 580 | 0.006 | 0.005 | 1.29      | 0.19| −0.16 | −0.51  | G protein nucleolar 3                                                     |
| LOC113988940            | 140 | 0.006 | 0      | ∞         | 0.42| −0.63 | −0.71  | class I histocompatibility antigen, F10 alpha chain-like                   |
| LOC113990807            | 221 | 0.006 | 0      | ∞         | 0.36| −0.03 | −0.38  | glutathione S-transferase-like                                            |

Table S15. (continued on next page)
| Gene          | Len | $d_N$ | $d_S$ | $d_N/d_S$ | $P$   | BF SH | BF PEC | Annotation                                           |
|---------------|-----|-------|-------|-----------|-------|-------|---------|------------------------------------------------------|
| MV *Manacus vitellinus* Branch Model                        |     |       |       |           |       |       |         |                                                      |
| LOC11398940   | 140 | 0.054 | 0.057 | 0.95     | 0.77  | −1.04 | −1.06  | class I histocompatibility antigen, F10 alpha chain-like |
| LOC114003393 | 208 | 0.043 | 0.068 | 0.63     | 0.17  | −1.1  | −1.04  | class I histocompatibility antigen, F10 alpha chain-like |
| LOC113988548  | 184 | 0.043 | 0.129 | 0.33     | 0.9   | −0.43 | −0.18  | uncharacterized LOC113988548                         |
| LOC113991726  | 122 | 0.039 | 0.034 | 1.15     | 0.04  | −1    | −0.74  | major centromere autoantigen B-like                   |
| LOC114003600  | 206 | 0.038 | 0.076 | 0.5      | 0.1   | −1    | −1.02  | collagen alpha-1(I) chain-like                       |
| LOC113992639  | 161 | 0.037 | 0.02  | 1.82     | 0.26  | −0.9  | −0.76  | zinc finger protein 777-like                         |
| HAUST         | 280 | 0.026 | 0.026 | 1.03     | 0.82  | −0.98 | −1.04  | HAUS augmin like complex subunit 1                    |
| LRRC30        | 133 | 0.022 | 0.009 | 2.42     | 0.02  | −0.29 | −0.85  | leucine rich repeat containing 30                     |
| TGOLN2        | 427 | 0.021 | 0.018 | 1.15     | 0.38  | −0.98 | −1.09  | trans-golgi network protein 2                         |
| LOC113983346  | 111 | 0.021 | 0.008 | 2.67     | 0.51  | −0.98 | −0.94  | sodium channel modifier 1-like                       |
| ALKBH2        | 244 | 0.016 | 0.007 | 2.12     | 0.01  | −0.37 | 1.27   | alkB homolog 2, alpha-ketoglutarate dependent dioxygenase |
| HEMK1         | 314 | 0.011 | 0.01  | 1.06     | 0.29  | 2.67  | 2.14   | HemK methyltransferase family member 1               |
| GHR           | 610 | 0.008 | 0.008 | 0.99     | 0.02  | −0.26 | 0.57   | growth hormone receptor                               |
| LOC113984727  | 245 | 0.005 | 0.∞   | 0.15     | 0.65  | 1.19  |        | serum amyloid P-component-like                        |
| TPRKB         | 174 | 0.005 | 0.∞   | 0.07     | 3.71  | 0.82  |        | TP53RK binding protein                               |
| RPA2          | 266 | 0.005 | 0.∞   | 0.05     | 0.33  | 1.24  |        | replication protein A2                               |
| LARP4B        | 1316| 0.004 | 0.004 | 1.02     | 0.32  | 0.84  | 0.44   | La ribonucleoprotein domain family member 4B          |
| SLC25A26      | 267 | 0.004 | 0.004 | 0.96     | 0.21  | −0.7  | 0.78   | solute carrier family 25 member 26                    |
| PON2          | 354 | 0.003 | 0.∞   | 0.07     | 1.66  | −0.88 |        | paraoxonase 2                                        |

*Table S15.* (continued on next page)
| Gene       | Len | $d_N$ | $d_S$ | $d_N/d_S$ | $P$ | BF SH | BF PEC | Annotation                                      |
|------------|-----|-------|-------|-----------|-----|-------|--------|------------------------------------------------|
| **CE Ceratopipra Ancestral Branch Model** |     |       |       |           |     |       |        |                                                 |
| FABP7      | 103 | 0.014 | 0     | $\infty$ | 0.17| -0.8  | -0.67  | fatty acid binding protein 7                    |
| LOC114001961 | 102 | 0.012 | 0.025 | 0.49      | 0.72| 0.32  | 0.36   | aldo-keto reductase family 1 member B10-like   |
| ATP12A     | 140 | 0.011 | 0.093 | 0.12      | 0.04| 0.16  | -0.11  | ATPase H+K+ transporting non-gastric alpha2 subunit |
| LOC114001388 | 100 | 0.01  | 0.031 | 0.31      | 0.98| -0.85 | -0.58  | class I histocompatibility antigen, F10 alpha chain-like |
| CD164      | 165 | 0.009 | 0.006 | 1.5       | 0.94| -0.79 | -0.82  | CD164 molecule                                  |
| CUNH20orf85 | 110 | 0.008 | 0     | $\infty$ | 0.01| -0.68 | -0.78  | chromosome unknown C20orf85 homolog             |
| TMEM233    | 111 | 0.008 | 0     | $\infty$ | 0.12| -0.22 | 0.48   | transmembrane protein 233                      |
| MRPL51     | 156 | 0.008 | 0     | $\infty$ | 0.07| -0.33 | -0.46  | mitochondrial ribosomal protein L51            |
| DSN1       | 344 | 0.008 | 0.006 | 1.26      | 0.12| -0.73 | -0.55  | DSN1 homolog, MIS12 kinetochore complex component |
| LOC114003744 | 219 | 0.007 | 0.004 | 1.68      | 0.22| -0.42 | -0.14  | mucin-5AC-like                                  |
| GGCT       | 182 | 0.007 | 0     | $\infty$ | 0.05| -0.52 | -0.69  | gamma-glutamylcyclotransferase                 |
| CUNH19orf12 | 141 | 0.007 | 0     | $\infty$ | 0   | -0.7  | -0.68  | chromosome unknown C19orf12 homolog            |
| CUNH12orf43 | 199 | 0.005 | 0     | $\infty$ | 0.23| 0.59  | -0.69  | chromosome unknown C12orf43 homolog            |
| LOC113990771 | 185 | 0.004 | 0     | $\infty$ | 0.06| 1.63  | 1.6    | WD repeat and SOCS box-containing protein 2-like |
| NUDT13     | 305 | 0.004 | 0     | $\infty$ | 0.04| 1.13  | -0.3   | nudix hydrolase 13                              |
| ACS53      | 257 | 0.004 | 0     | $\infty$ | 0.02| -0.15 | 0.62   | acyl-CoA synthetase short chain family member 3 |
| DCAF4      | 468 | 0.003 | 0     | $\infty$ | 0.03| -0.39 | 0.6    | DDB1 and CUL4 associated factor 4              |
| COQ9       | 314 | 0.003 | 0     | $\infty$ | 0.04| 0.45  | 1.13   | coenzyme Q9                                    |
| SECISBP2   | 833 | 0.003 | 0     | $\infty$ | 0.01| 0.82  | -0.21  | SECIS binding protein 2                        |
| WDHR60     | 346 | 0.003 | 0     | $\infty$ | 0.1 | -0.8  | 0.56   | WD repeat domain 60                            |
| SRBD1      | 962 | 0.002 | 0.001 | 1.86      | 0.02| 0.68  | -0.29  | S1 RNA binding domain 1                        |
| PRRC1      | 446 | 0.002 | 0     | $\infty$ | 0.11| 0.69  | 0.14   | proline rich coiled-coil 1                     |
| PREPL       | 737 | 0.001 | 0     | $\infty$ | 0.11| 0.19  | 0.52   | prolyl endopeptidase like                      |
| FAN1       | 1024 | 0.001 | 0     | 2.77      | 0.33| 1.16  | 0.56   | FANCD2 and FANCI associated nuclease 1          |

**Table S15. Results of Tests of Positive Selection on Gene Sequences.** Genes with highest $d_N$ values indicating possible positive selection. Results of PAML branch-test for M5, M4, MV, and CE models (see Fig. S4). PhyDGET scores for PEC and SH are shown for comparison.
| Gene          | Model Name | Notes on Function                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |
|---------------|------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| THAP9         | M5         | THAP Domain Containing 9: “The studies reported here indicate that the human THAP9 gene encodes an active DNA transposase that can mobilize Drosophila P-element transposons in Drosophila and human cells” (84).                                                                                                                                                                                                                                                                                                      |
| RESF1         | M5         | Retroelement Silencing Factor 1: “Plays a role in the regulation of imprinted gene expression, regulates repressive epigenetic modifications associated with SETDB1. Required for the recruitment or accumulation of SETDB1 to the endogenous retroviruses (ERVs) and maintenance of repressive chromatin configuration, contributing to a subset of the SETDB1-dependent ERV silencing in embryonic stem cells” (UniProtKB).                                                                                                                                                                                                                           |
| KNOPT1        | M5         | Lysine Rich Nucleolar Protein 1: “The protein encoded by this gene is a nucleolar protein that interacts with zinc finger 106 protein. The encoded protein has several of the same characteristics as nucleostemin and may be involved in testis development” (NCBI RefSeq).                                                                                                                                                                                                                                       |
| TRIM27-like   | M5         | Tripartite Motif Containing 27: “E3 ubiquitin-protein ligase that mediates ubiquitination of PIK3C2B and inhibits its activity; mediates the formation of ‘Lys-48’-linked polyubiquitin chains; the function inhibits CD4 T-cell activation. . . . May function in male germ cell development” (UniProtKB).                                                                                                                                                                                                    |
| CD164         | M5/M4      | CD164 Molecule: “This gene encodes a transmembrane sialomucin and cell adhesion molecule that regulates the proliferation, adhesion and migration of hematopoietic progenitor cells. The encoded protein also interacts with the C-X-C chemokine receptor type 4 and may regulate muscle development” (NCBI RefSeq).                                                                                                                                                                                                                         |
| CTTNBP2       | M4         | Cortactin Binding Protein 2: “This gene encodes a protein with six ankyrin repeats and several proline-rich regions. A similar gene in rat interacts with a central regulator of the actin cytoskeleton” (NCBI RefSeq).                                                                                                                                                                                                                                                                                        |
| FUNDC2        | M4         | FUN14 Domain Containing 2:                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |
| IRAK1BP1      | M4         | Interleukin 1 Receptor Associated Kinase 1 Binding Protein 1:                                                                                                                                                                                                                                                                                                                                                                                                                                              |
| AKR1B10       | M4/MV      | Aldo-Keto Reductase Family 1 Member B10: “Catalyzes the NADPH-dependent reduction of a wide variety of carbonyl-containing compounds to their corresponding alcohols” (UniProtKB).                                                                                                                                                                                                                                                                                                                      |

Table S16. (continued on next page)
| Gene                     | Model Name | Notes on Function                                                                                                                                                                                                 |
|--------------------------|------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| **"HA1F-like"** LOC113988940 and LOC113988940 | MV         | Class I Histocompatibility Antigen F10 alpha chain: Involved in the presentation of foreign antigens to the immune system.                                                                                           |
| **COL1A1-like**          | MV         | Collagen Type I Alpha 1 Chain: “This gene encodes the pro-alpha1 chains of type I collagen whose triple helix comprises two alpha1 chains and one alpha2 chain. Type I is a fibril-forming collagen found in most connective tissues and is abundant in bone, cornea, dermis and tendon” (NCBI RefSeq). |
| **"CENPB-like" LOC113988940** | MV         | Centromere Protein B: This gene product is a highly conserved protein that facilitates centromere formation. It is a DNA-binding protein that is derived from transposases of the pogo DNA transposon family” (NCBI RefSeq). |
| **"WSB2-like" LOC113990771** | CE         | WD Repeat And SOCS Box Containing 2: “The encoded protein contains five WD-repeats spanning most of the protein and an SOCS box in the C-terminus. The SOCS box may act as a bridge between specific substrate-binding domains and E3 ubiquitin protein ligases” (NCBI RefSeq). |
| **FAN1**                 | CE         | FANCD2 And FANCI Associated Nuclease 1: “Nuclease required for the repair of DNA interstrand cross-links (ICL) recruited at sites of DNA damage by monoubiquitinated FANCD2. Specifically involved in repair of ICL-induced DNA breaks by being required for efficient homologous recombination, probably in the resolution of homologous recombination intermediates” (UniProtKB). |
| **FABP7**                | CE         | Fatty Acid Binding Protein 7: “F-BABP could be involved in the transport of a so far unknown hydrophobic ligand with potential morphogenic activity during CNS development. It is required for the establishment of the radial glial fiber system in developing brain, a system that is necessary for the migration of immature neurons to establish cortical layers” (UniProtKB). |
| **ATP12A**               | CE         | ATPase H+/K+ Transporting Non-Gastric Alpha2 Subunit: “Catalyzes the hydrolysis of ATP coupled with the exchange of H(+) and K(+) ions across the plasma membrane. Responsible for potassium absorption in various tissues” (UniProtKB). |
| **SECISBP2**             | CE         | Selenocysteine Insertion Sequence-Binding Protein 2: “Binds to the SE CIS element in the 3’-UTR of some mRNAs encoding selenoproteins” (UniProtKB).                                                                         |

Table S16. Functional information for the genes highlighted in Figure 4. Basic database descriptions from UniProtKB and NCBI human RefSeq are included for most genes. Additionally, muscle, avian, and avian-muscle references are cited, where available and appropriate. Model codes are shown in Fig. S4. Genes are not repeated if they also appear in Table S14.
| Gene          | #NON | #SYN | Total codons | Gene          | #NON | #SYN | Total codons |
|--------------|------|------|--------------|--------------|------|------|--------------|
| F8           | 2    |      | 1246         | TFAM         | 1    |      | 207          |
| CUNH10orf71  | 2    |      | 1532         | ANGEL1       | 1    |      | 644          |
| CMYA5        | 2    |      | 3980         | NAE1         | 1    |      | 528          |
| LOC113983397 | 2    |      | 293          | LOC114002103 | 1    |      | 87           |
| RAD50        | 2    |      | 787          | NOP10        | 1    |      | 64           |
| GBE1         | 1    |      | 702          | LOC113986238 | 1    |      | 444          |
| CLIP1        | 1    |      | 2184         | LRP12        | 1    |      | 855          |
| ZC3H14       | 1    |      | 684          | JSRP1        | 1    |      | 281          |
| LOC113997062 | 2    |      | 335          | LOC113985373 | 1    |      | 374          |
| PYGM         | 1    |      | 834          | LARP1B       | 1    |      | 1009         |
| AB13         | 1    |      | 313          | SCAF4        | 1    |      | 1059         |
| CALCOCO2     | 1    |      | 436          | PRRC2C       | 1    |      | 1785         |
| MTCL1        | 1    |      | 1673         | TMEM184B     | 1    |      | 418          |
| SERINC2      | 1    |      | 437          | LMO7         | 1    |      | 1186         |
| QRER1        | 1    |      | 1429         | TRPT1        | 1    |      | 95           |
| ANKRD49      | 1    |      | 235          | DDX10        | 1    |      | 765          |
| RAD54L2      | 1    |      | 1319         | MEPCE        | 1    |      | 412          |
| LDB3         | 1    | 1    | 195          | REXO1        | 1    |      | 1182         |
| TANC1        | 1    |      | 1562         | SMCHD1       | 1    |      | 155          |
| EMC1         | 1    |      | 986          | LOC113982770 | 1    |      | 394          |
| SFSWAP       | 1    |      | 962          | ZNF518A      | 1    |      | 416          |
| TRMT2A       | 1    |      | 563          | LOC113982172 | 1    |      | 562          |
| N4BP1        | 1    |      | 830          | PNPLA8       | 1    |      | 794          |
| PEPD         | 1    | 1    | 494          | MFN1         | 1    |      | 740          |
| PRR5L        | 1    |      | 365          | ZNF639       | 1    |      | 473          |
| LOC113992976 | 1    | 1    | 571          | PCNT         | 1    |      | 1636         |
| LOC113992904 | 1    |      | 192          | RINT1        | 1    |      | 764          |
| CLGN         | 1    |      | 618          | EC1          | 1    |      | 301          |
| CD59         | 1    |      | 117          | TARP1        | 1    |      | 907          |
| PUSL1        | 1    |      | 281          | MYPN         | 1    |      | 1336         |
| UHRF1BP1     | 1    |      | 1198         | PEX16        | 1    |      | 283          |
| CHKA         | 1    |      | 301          | GPAM         | 1    |      | 828          |
| CPA          | 1    |      | 1224         | PDK4         | 1    |      | 417          |
| STAT6        | 1    |      | 672          | ATG4C        | 1    |      | 458          |
| EIF2AK1      | 1    |      | 594          | LOC114002020 | 1    |      | 426          |
| MPP1         | 1    |      | 464          | KIDINS220    | 1    |      | 1735         |
| DKC1         | 1    |      | 509          | NOSIP        | 1    |      | 110          |
| AOX1         | 1    |      | 1160         | ACLY         | 1    |      | 1091         |
| ATG14        | 1    |      | 358          | LRIG2        | 1    |      | 395          |
| LOC113989432 | 1    |      | 69           | NAF1         | 1    |      | 262          |
| WIPF3        | 1    |      | 303          | NUP153       | 1    |      | 1430         |
| LOC113989152 | 1    |      | 39           | L2HGHD       | 1    |      | 410          |
| LAMC1        | 1    |      | 1358         | RSL1D1       | 1    |      | 425          |
| MIS18BP1     | 1    |      | 442          | MRPS9        | 1    |      | 368          |
| ARM68        | 1    |      | 480          | HHATL        | 1    |      | 501          |
| CAST         | 1    |      | 548          | RSF1         | 1    |      | 898          |
| PTCD3        | 1    |      | 671          | FAN1         | 1    |      | 948          |
| ACSS2        | 1    |      | 695          | LOC113998311 | 1    |      | 437          |
| ABCB10       | 1    |      | 644          | SAMHD1       | 1    |      | 527          |
| HLX          | 1    |      | 193          | SEC31B       | 1    | 2    | 1213         |

**Table S17. Genes with snap species-specific amino acids.** Genes where *Manacus* and *Ceratopipra* (the snap species) have a separate amino acid from the other four species. The number of non-synonymous and synonymous substitutions are shown (","=zeros).
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