Supplementary Materials:

Detailed DNA Extraction Methods

Lice were collected in the field and transferred to ethanol in a 2-ml screw-cap tube for storage. Lice were then stored long-term at -80°C. Total genomic DNA was extracted from each louse species in Table S1. The extraction protocol was modified from the "Purification of total DNA from animal tissues" protocol found in the Qiagen DNeasy Blood and Tissues Handbook provided with the Qiagen DNA micro-extraction kit. First, louse specimens were allowed to dry at room temperature to prevent ethanol contamination of DNA extraction buffers. The lice were then incubated in 180 µl of ATL buffer for 5 minutes at room temperature in a sterile 1.5-ml snap cap tube. After incubation, the lice were crushed in the ATL buffer using a sterile pestle. After crushing, 25 µl of proteinase K was added to the solution. The solution was incubated for 48 hours at 55°C, with occasional pulse vortexing. Following the two-day incubation, 200µl of AL buffer was added and the solution was incubated for 10 minutes at 70°C. Then 200 µl of 95% ethanol was added to the solution and mixed by pulse vortexing. The solution was then transferred to a Qiagen spin column by pipet. The column was centrifuged at 8000 rpm for 1 minute using an Eppendorf 5414D centrifuge and column flow-through was discarded. 500 µl of AW1 buffer was pipetted onto the column. The column was centrifuged at 8000 rpm for 1 minute and flow-through was discarded. 500 µl of AW2 was pipetted on to the column. The column was centrifuged at 14000 rpm for 3 minutes using the Eppendorf 5415C centrifuge and the flow-through was discarded. Finally the DNA was eluted from the column by applying 40 µl of elution buffer (EB) to the column,

incubating the column for 5 minutes at 70°C, and then centrifuging the column at 8000 rpm for 5 minutes using the Eppendorf 5415D centrifuge. Three microliters of the resulting EB with gDNA was used to quantify the concentration of DNA in the EB using the Life Technology Qubit 3.0 fluorometer, using the High Sensitivity kit.

Commands

Estimating UPP alignment:

python run_upp.py -A 10 -B 1000 -M -1 -m amino -s <input_gene_sequences> -x 4 -o <output_prefix>

Estimating the RAxML ML gene trees under GTR+gamma, with 100 bootstrap replicates:

raxmlHPC-PTHREADS-SSE3 -N 100 -f a -T 8 --no-bfgs -s <input_alignment> -n <output_prefix> -m GTRGAMMA -p 1111 -x 2222

Computing bootstrap replicate concatenated alignments using RAxML:

raxmlHPC-PTHREADS-SSE3 -N 100 -f j -m GTRGAMMA -s <input_alignment> -n <output_prefix> -T 8 -b 1111

Estimating the starting concatenated FastTree ML tree under the GTR+CAT model:

fasttreeMP –nt –gtr <output_tree> <input_alignment>

Estimating concatenated RAxML ML tree under GTR+gamma:

raxmlHPC-PTHREADS-SSE3 -T 8 -q <partition_file> -s <input_alignment> -p 1111 -m GTRGAMMA -n <output_prefix> -t <fasttree_ml_tree>

Estimating coalescence ASTRAL tree:

java -jar astral.4.9.7.jar -i <input_gene_trees> -o <output_tree>

Estimating coalescence ASTRID tree:

ASTRID -i <input_gene_trees> -o <output_tree>

Estimating local posterior probabilities for coalescence trees:

java -jar astral.4.9.9.jar -i <input_gene_trees> -q <input_tree> >

<input_tree_with_support>

Figure S1. Percent of GC bases by taxon and position in codon.