

Supplementary materials

Table S1. The best model for each partition of the five datasets.

Datasets	Partition names	Best model
PCG123	CO3_pos1, CO1_pos1, ATP6_pos1, CytB_pos1, CO2_pos1	GTR+I+G
	CO3_pos2, CO2_pos2, CO1_pos2, ATP6_pos2	F81+I
	ATP6_pos3, ND3_pos3, CytB_pos3, CO3_pos3, ATP8_pos3, CO2_pos3	HKY+I+G
	ATP8_pos2, ATP8_pos1, ND6_pos1	HKY+I+G
	CO1_pos3, ND2_pos3, ND6_pos3	HKY+G
	ND4L_pos2, ND4L_pos1, CytB_pos2, ND1_pos2, ND3_pos2, ND4_pos2, ND6_pos2, ND5_pos2, ND2_pos2	GTR+I+G
	ND3_pos1, ND2_pos1, ND1_pos1, ND5_pos1, ND4_pos1	GTR+I+G
	ND4L_pos3, ND4_pos3, ND1_pos3, ND5_pos3	GTR+G
PCG12R	16S, 12S	HKY+G
	ATP6, CO1, CO2, CytB, CO3	GTR+I+G
	ATP8, ND2, ND6	HKY+I+G
	ND4L, ND1, ND3, ND5, ND4	GTR+I+G
PCG123R	16S, 12S	HKY+G
	CO3_pos1, CO1_pos1, ATP6_pos1, CO2_pos1, CytB_pos1	GTR+I+G
	CO2_pos2, CO3_pos2, ATP6_pos2, CO1_pos2	F81+I
	CO3_pos3, CytB_pos3, CO2_pos3, ATP8_pos3, ATP6_pos3, ND2_pos3, ND6_pos3	HKY+I+G
	ATP8_pos2, ATP8_pos1, ND6_pos1	HKY+I+G
	CO1_pos3, ND3_pos3	HKY+G
	ND4L_pos1, ND4L_pos2, CytB_pos2, ND3_pos2, ND1_pos2, ND4_pos2, ND6_pos2, ND2_pos2, ND5_pos2	GTR+I+G
	ND1_pos1, ND5_pos1, ND4_pos1, ND2_pos1, ND3_pos1	GTR+I+G
	ND4L_pos3, ND1_pos3, ND4_pos3, ND5_pos3	GTR+G
AA	CO1, CO2, ND4L, ATP6	MTMAM+I+G
	ATP8, ND6	MTREV+I+G
	CO3, ND2, ND3, ND4, CytB, ND1	MTMAM+I+G
	ND5	MTMAM+I+G

Table S2. Nucleotide composition of mitochondrial genomes of 15 *Chironomus* species.

	Species	Whole Genome	Protein Coding Genes	1 st Codon Position	2 nd Codon Position	3 rd Codon Position	tRNA Genes	12S rRNA	16S rRNA	Control Region
A+T %	<i>Chironomus anthracinus</i>	75.31	71.66	66.81	67.69	80.46	78.85	82.89	84.35	91.29
	<i>Chironomus nipponensis</i>	76.13	72.68	67.93	67.80	82.31	79.13	82.95	84.79	92.11
	<i>Chironomus flaviplumus</i>	78.47	76.00	69.13	67.72	91.15	79.21	83.25	84.88	95.38
	<i>Chironomus plumosus</i>	76.20	72.93	67.74	67.66	83.38	79.43	83.99	84.89	92.81
	<i>Chironomus tentans</i>	76.94	74.06	68.17	67.85	86.16	79.44	82.82	85.24	93.57
	<i>Chironomus novosibiricus</i>	76.19	72.72	67.58	67.56	83.03	79.13	82.80	85.18	93.36
	<i>Chironomus annularius</i>	75.56	72.33	67.91	67.75	81.33	79.10	83.07	84.70	91.90
	<i>Chironomus agilis</i>	75.64	72.34	67.44	67.79	81.80	79.17	82.97	85.01	92.40
	<i>Chironomus nippodorsalis</i>	76.40	73.40	68.12	67.85	84.23	79.16	83.54	84.73	92.32
	<i>Chironomus tepperi</i>	76.92	74.33	67.75	67.67	87.58	79.02	82.60	84.30	93.00
	<i>Chironomus claggi</i>	77.83	75.26	68.76	67.88	89.15	79.08	82.74	84.52	94.29
	<i>Chironomus kiiensis</i>	76.94	74.21	71.88	67.99	82.75	79.23	82.72	84.66	90.87
	<i>Chironomus transvaalensis</i>	78.54	75.92	69.24	67.82	90.70	79.88	83.42	85.57	95.96
	<i>Chironomus circumdatus</i>	78.44	75.87	69.27	67.72	90.62	79.71	83.15	85.15	94.75
	<i>Chironomus javanus</i>	77.37	74.57	68.04	67.93	87.73	79.59	82.97	85.03	95.15
AT-skew	<i>Chironomus anthracinus</i>	0.03	-0.20	-0.08	-0.40	-0.14	0.03	-0.01	0.00	-0.09
	<i>Chironomus nipponensis</i>	0.03	-0.20	-0.09	-0.40	-0.13	0.04	0.01	0.01	-0.10
	<i>Chironomus flaviplumus</i>	0.01	-0.20	-0.10	-0.40	-0.11	0.02	0.00	0.03	-0.03
	<i>Chironomus plumosus</i>	0.03	-0.20	-0.08	-0.40	-0.13	0.03	0.02	0.00	-0.06

	<i>Chironomus tentans</i>	0.02	-0.19	-0.09	-0.40	-0.10	0.04	0.01	0.02	-0.06
	<i>Chironomus novosibiricus</i>	0.03	-0.20	-0.09	-0.40	-0.13	0.05	0.01	0.01	-0.07
	<i>Chironomus annularius</i>	0.03	-0.20	-0.09	-0.40	-0.12	0.03	0.02	0.01	-0.09
	<i>Chironomus agilis</i>	0.03	-0.20	-0.08	-0.40	-0.12	0.04	0.01	0.01	-0.10
	<i>Chironomus nippodorsalis</i>	0.02	-0.19	-0.09	-0.41	-0.10	0.04	0.02	0.00	-0.06
	<i>Chironomus tepperi</i>	0.02	-0.19	-0.09	-0.40	-0.12	0.04	0.01	0.00	-0.10
	<i>Chironomus claggi</i>	0.01	-0.19	-0.10	-0.40	-0.11	0.03	-0.01	-0.01	-0.10
	<i>Chironomus kiiensis</i>	0.02	-0.18	-0.08	-0.36	-0.13	0.04	0.01	-0.02	-0.10
	<i>Chironomus transvaalensis</i>	0.01	-0.19	-0.10	-0.40	-0.11	0.04	0.00	0.01	-0.12
	<i>Chironomus circumdatus</i>	0.01	-0.20	-0.10	-0.40	-0.13	0.04	0.00	0.01	-0.07
	<i>Chironomus javanus</i>	0.01	-0.20	-0.08	-0.40	-0.14	0.03	0.01	0.00	-0.11
GC-skew	<i>Chironomus anthracinus</i>	-0.20	-0.04	0.15	-0.17	-0.18	0.13	0.36	0.39	-0.23
	<i>Chironomus nipponensis</i>	-0.21	-0.02	0.19	-0.18	-0.13	0.12	0.37	0.37	-0.35
	<i>Chironomus flaviplumus</i>	-0.17	0.01	0.23	-0.18	-0.03	0.15	0.36	0.36	-0.25
	<i>Chironomus plumosus</i>	-0.23	-0.04	0.17	-0.18	-0.18	0.14	0.33	0.39	-0.55
	<i>Chironomus tentans</i>	-0.21	-0.04	0.18	-0.18	-0.22	0.12	0.36	0.38	-0.50
	<i>Chironomus novosibiricus</i>	-0.19	-0.03	0.18	-0.17	-0.15	0.11	0.35	0.36	-0.29
	<i>Chironomus annularius</i>	-0.22	-0.02	0.19	-0.18	-0.10	0.13	0.35	0.39	-0.32
	<i>Chironomus agilis</i>	-0.22	-0.05	0.15	-0.18	-0.18	0.12	0.37	0.39	-0.33
	<i>Chironomus nippodorsalis</i>	-0.20	-0.03	0.18	-0.18	-0.13	0.13	0.34	0.37	-0.15
	<i>Chironomus tepperi</i>	-0.18	-0.01	0.19	-0.18	-0.12	0.14	0.37	0.36	-0.43
	<i>Chironomus claggi</i>	-0.18	-0.01	0.22	-0.18	-0.13	0.06	-0.36	-0.35	-0.31

	<i>Chironomus kiiensis</i>	-0.17	-0.02	0.15	-0.10	-0.13	0.13	0.35	0.35	-0.25
	<i>Chironomus transvaalensis</i>	-0.17	0.01	0.22	-0.18	-0.03	0.15	0.35	0.36	-0.14
	<i>Chironomus circumdatus</i>	-0.18	0.01	0.22	-0.18	-0.03	0.14	0.36	0.38	-0.41
	<i>Chironomus javanus</i>	-0.19	-0.01	0.18	-0.17	-0.08	0.15	0.37	0.34	-0.36

Table S3 Start and stop codons of 13 PCGs in the mitogenomes of 15 *Chironomus* species.

Feature	Start/Stop codon												
	<i>ATP8</i>	<i>ATP6</i>	<i>COI</i>	<i>CO2</i>	<i>CO3</i>	<i>CytB</i>	<i>ND1</i>	<i>ND2</i>	<i>ND3</i>	<i>ND4</i>	<i>ND4</i> <i>L</i>	<i>ND5</i>	<i>ND6</i>
<i>Chironomus tepperi</i>	ATC/ TAA	ATG/ TAA	TTG/ TAG	ATG/ TAA	ATG/ TAA	ATG/ TAA	ATT/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus flaviplumus</i>	ATT/ TAA	ATG/ TAA	ATA/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAG	ATT/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus kiiensis</i>	ATC/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	ATT/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAG	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus transvaalensis</i>	ATT/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus circumdatus</i>	ATC/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus javanus</i>	ATT/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAG	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus anthracinus</i>	ATC/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAG	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus nipponensis</i>	ATT/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TA-	ATT/ TAA	ATG/ TAG	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus claggi</i>	ATT/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus plumosus</i>	ATT/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus tentans</i>	ATT/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus novosibiricus</i>	ATC/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAG	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus annularius</i>	ATC/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA

<i>Chironomus agilis</i>	ATT/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA
<i>Chironomus nipodorsalis</i>	ATT/ TAA	ATG/ TAA	TTG/ TAA	ATG/ TAA	ATG/ TAA	ATG/ TAA	TTG/ TAA	ATT/ TAA	ATT/ TAA	ATG/ TAA	ATG/ TAA	GTG/ TAA	ATT/ TAA

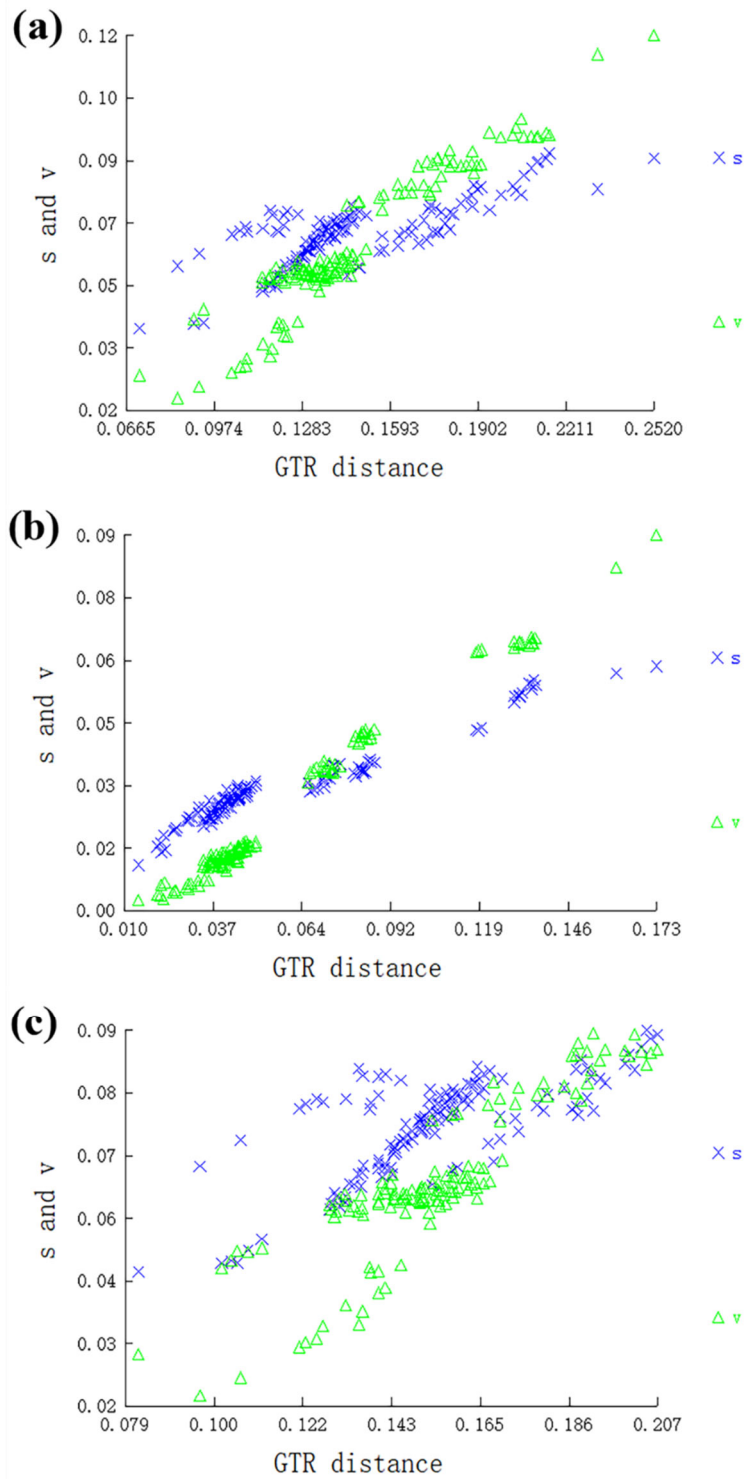


Figure S1. Substitution patterns of the PCG123R (a), PCG12R (b) and PCG123 (c) datasets. The graphs represent the increase in GTR distance.

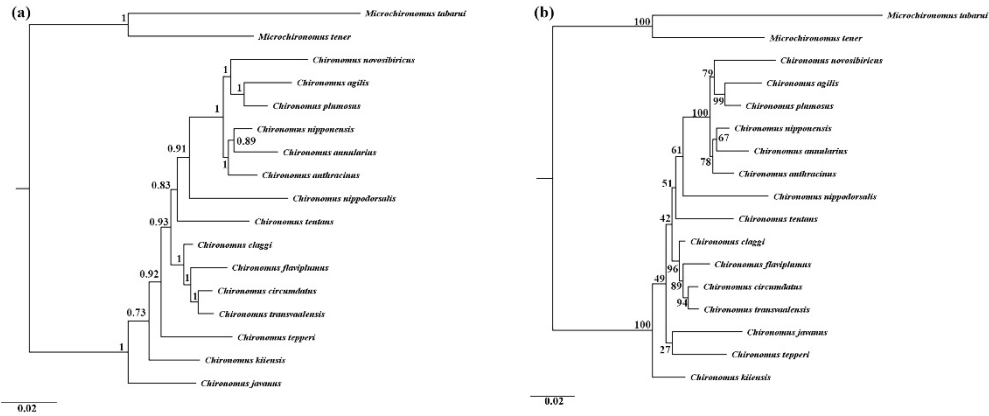


Figure S2. Phylogenetic trees of *Chironomus* inferred from the AA dataset. (a) BI tree. Numbers at the nodes are BI posterior probabilities. (b) ML tree. Numbers at the nodes are ML bootstrap values.

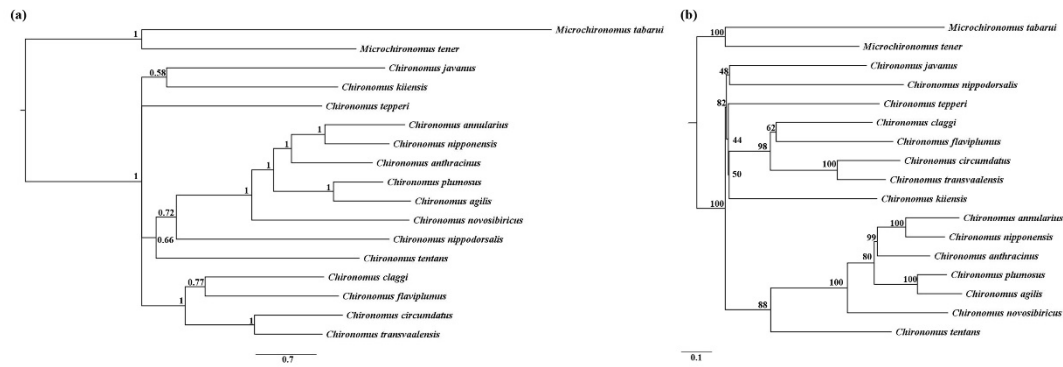


Figure S3. Phylogenetic trees of *Chironomus* inferred from the PCG123 dataset. (a) BI tree. Numbers at the nodes are BI posterior probabilities. (b) ML tree. Numbers at the nodes are ML bootstrap values.

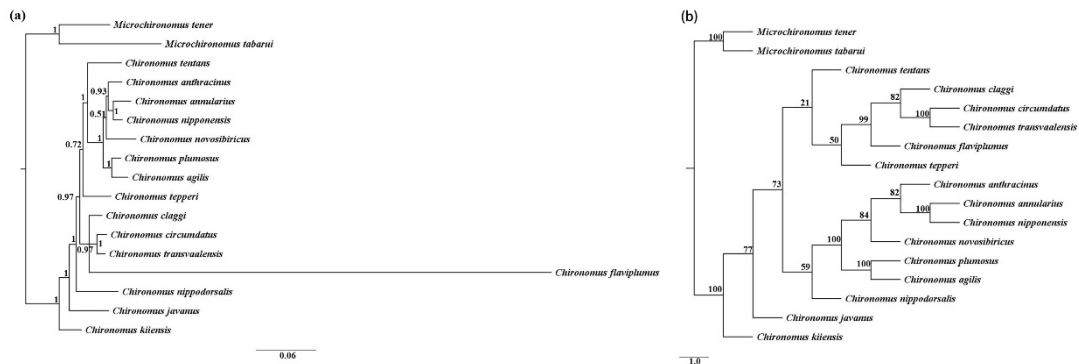


Figure S4. Phylogenetic trees of *Chironomus* inferred from the PCG12R dataset. (a) BI tree. Numbers at the nodes are BI posterior probabilities. (b) ML tree. Numbers at the nodes are ML bootstrap values.