## Supplementary Materials

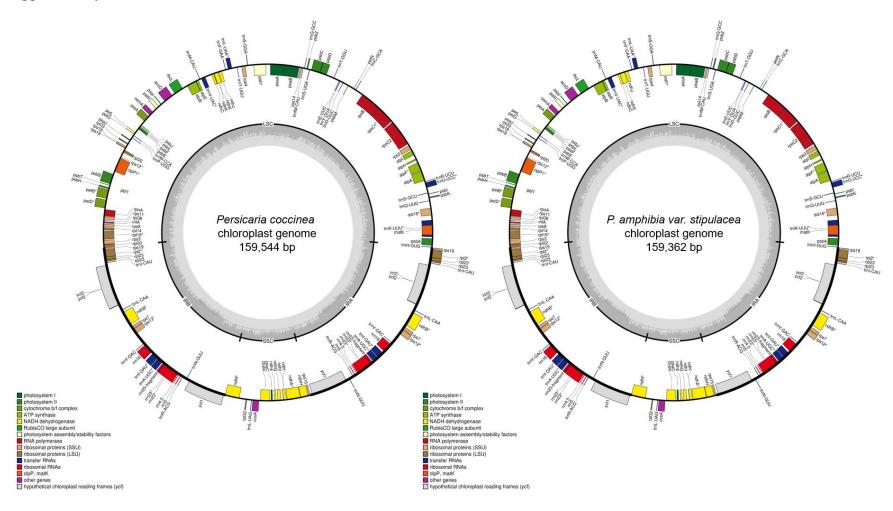


Figure S1. The structure of plastid genome of *P. coccinea* (= *P. amphibia* var. *emersa*) from Potsdam population (left) and *P. amphibia* var. *stipulacea* from Watertown (right). Two representative plastome structures were shown, as no structural variations were observed among the individuals investigated here.

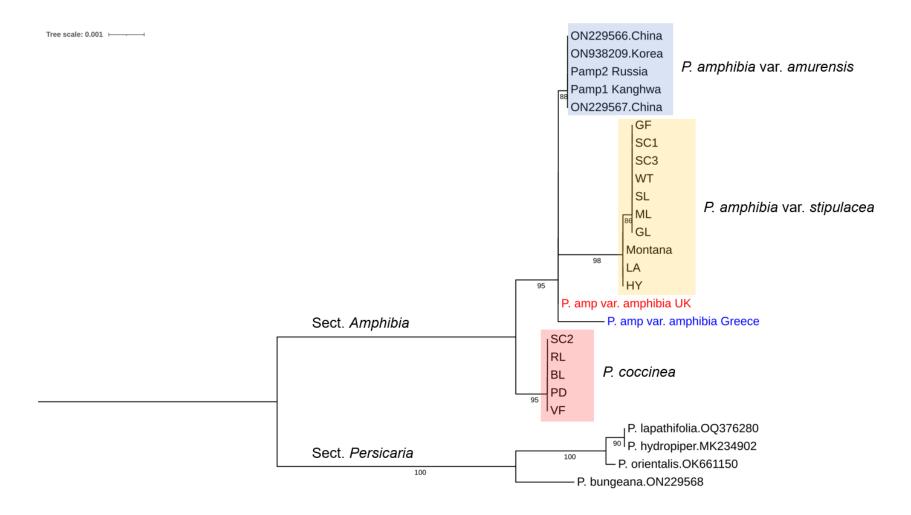


Figure S2. A maximum-likelihood tree inferred from the partial chloroplast sequences with the best fit model of K3Pu+F. The sequences of Greece individual were obtained from Gitsopoulos *et al.* (2013)'s study. The aligned length was 3,897 bp, of which 97.7% (3,808 bp) were constant across taxa and 79 sites were parsimony informative. The number below the branch shows the bootstrap support value. Two European elements (blue and red text) failed to form one clade.

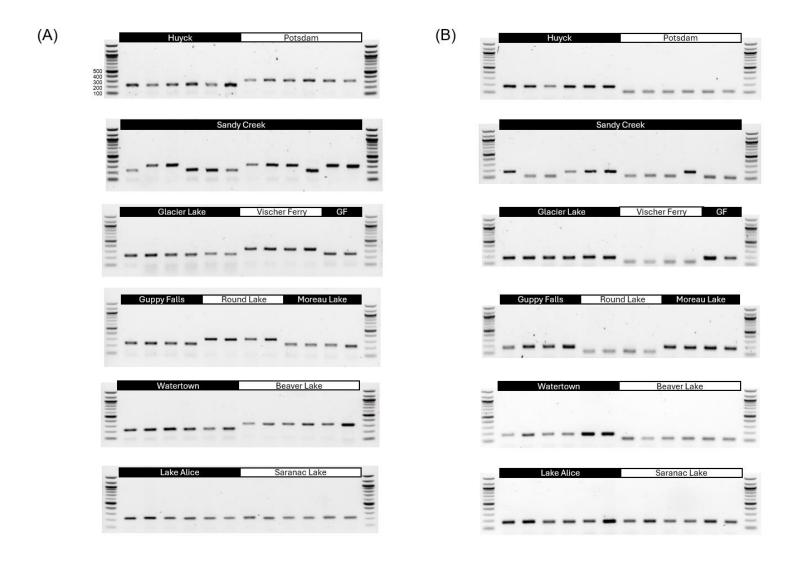


Figure S3. Genotyping results using two chloroplast regions. (A) *rpl20-rpl12* intergenic spacer (240 in *P. amphibia* var. *stipulacea* vs. 311 bp in *P. coccinea*), (B) *rpl33-rps18* intergenic spacer (171 in *P. amphibia* var. *stipulacea* vs. 108 bp in *P. coccinea*). Note that Sandy Creek population shows co-occurrence of two chloroplast genomes.

Table S1. Seven chloroplast regions which show differences between two American elements. Asterisk (\*) indicates the chloroplast regions used in genotyping each population.

Chloroplast	Primer sequences (5'->3')	Amplicon size (bp) in	Amplicon size (bp) in <i>P</i> .	Difference in
region		P. coccinea	amphibia var. stipulacea	amplicon sizes
psbA-trnK IGS	F: CAGTATAACATGACTTATATGC	130	124	6 bp
	R: TCATATCGAAGTCATATGTG			
atpF intron	F: TGCTATGGTTCTTCCATATG	127	120	7 bp
	R: CAAGAATAGACTGGATTCAAC			
rpoC1 intron	F: ACTACTCCTTACTCAAGTTC	131	123	8 bp
	R: GGAAGTAGACTACTCAAG			
trnE-trnA IGS	F: CTTGGTTCATTGAAGTGATAG	159	144	15 bp
	R: CTATGCGGATAAGCTATCTC			
trnT-trnL IGS	F: CATTCCAATCCTAACAATTAG	172	148	24 bp
	R: GATAGAACTCATAATTGAATTCA			
$rpl20$ - $rpl12~\mathrm{IGS}^*$	F: TCGAACGTCTGTATATCAC	311	240	71 bp
	R: AGATCCGACCGAAATAGG			
<i>rpl33-rps18</i> IGS*	F: TCATACGCAATAGATGAATATG	108	171	63 bp
	R: GCCGAGGAACATGTACTAG			