

Supplementary Material

MK188349 <i>Trissolcus japonicus</i>	TAATTGGATCAGCAATAAGAATATTAAATCCGAATAGAACTAAGAGTTCCAGGTATATTAATTGGTAATGA 70
MK188350 <i>Trissolcus japonicus</i>	TAATTGGATCAGCAATAAGAATATTAAATCCGAATAGAACTAAGAGTTCCAGGTATATTAATTGGTAATGA 70
MK188360 <i>Trissolcus japonicus</i>	TAATTGGATCAGCAATAAGAATATTAAATCCGAATAGAACTAAGAGTTCCAGGTATATTAATTGGTAATGA 70

MK188349 <i>Trissolcus japonicus</i>	CCAAATTTATAATTCTATTGTAACTTCACATGCATTATTATAATCTTTTTATAGTTATACCAATTATA 140
MK188350 <i>Trissolcus japonicus</i>	CCAAATTTATAATTCTATTGTAACTTCACATGCATTATTATAATCTTTTTATAGTTATACCAATTATA 140
MK188360 <i>Trissolcus japonicus</i>	CCAAATTTATAATTCTATTGTAACTTCACATGCATTATTATAATCTTTTTATAGTTATACCAATTATA 140

MK188349 <i>Trissolcus japonicus</i>	CTTGGAGGATTTGGAAATTTGATTAATTCCTTTAATAATTAATGCCCCAGATATAGCATTTCACGATTAA 210
MK188350 <i>Trissolcus japonicus</i>	CTTGGAGGATTTGGAAATTTGATTAATTCCTTTAATAATTAATGCCCCAGATATAGCATTTCACGATTAA 210
MK188360 <i>Trissolcus japonicus</i>	CTTGGAGGATTTGGAAATTTGATTAATTCCTTTAATAATTAATGCCCCAGATATAGCATTTCACGATTAA 210

TJ234F	
MK188349 <i>Trissolcus japonicus</i>	ATAATATAAGATTCTGATTACTAATCCCATCATTAAATTTATTAATCTATAGGAATATCTTTGGATCAGG 280
MK188350 <i>Trissolcus japonicus</i>	ATAATATAAGATTCTGATTACTAATCCCATCATTAAATTTATTAATCTATAGGAATATCTTTGGATCAGG 280
MK188360 <i>Trissolcus japonicus</i>	ATAATATAAGATTCTGATTACTAATCCCATCATTAAATTTATTAATCTATAGGAATATCTTTGGATCAGG 280

MK188349 <i>Trissolcus japonicus</i>	AAC TGGAACTGGATGAACAGTATACCCCTCTTATCTACTCAATTAATCCCTCTATTGATTTAACTATT 350
MK188350 <i>Trissolcus japonicus</i>	AAC TGGAACTGGATGAACAGTATACCCCTCTTATCTACTCAATTAATCCCTCTATTGATTTAACTATT 350
MK188360 <i>Trissolcus japonicus</i>	AAC TGGAACTGGATGAACAGTATACCCCTCTTATCTACTCAATTAATCCCTCTATTGATTTAACTATT 350

MK188349 <i>Trissolcus japonicus</i>	TTTTCCTACATATAGCAGGAATTTTCATCTATTCTTAGATCAATCAATTTCTTATGTACAATTATTAATA 420
MK188350 <i>Trissolcus japonicus</i>	TTTTCCTACATATAGCAGGAATTTTCATCTATTCTTAGATCAATCAATTTCTTATGTACAATTATTAATA 420
MK188360 <i>Trissolcus japonicus</i>	TTTTCCTACATATAGCAGGAATTTTCATCTATTCTTAGATCAATCAATTTCTTATGTACAATTATTAATA 420

MK188349 <i>Trissolcus japonicus</i>	TAAGAAATATATCAATTAATAATTGAACGTTATTACATGATCAATTTAATTACAACAATTTTATTACT 490
MK188350 <i>Trissolcus japonicus</i>	TAAGAAATATATCAATTAATAATTGAACGTTATTACATGATCAATTTAATTACAACAATTTTATTACT 490
MK188360 <i>Trissolcus japonicus</i>	TAAGAAATATATCAATTAATAATTGAACGTTATTACATGATCAATTTAATTACAACAATTTTATTACT 490

TJ460R	
MK188349 <i>Trissolcus japonicus</i>	TCTATCTTTACCAGTTTTCAGGGGCAATCACTATAATTTTATCAGATCGAAATCTTAATACATCCTTT 560
MK188350 <i>Trissolcus japonicus</i>	TCTATCTTTACCAGTTTTCAGGGGCAATCACTATAATTTTATCAGATCGAAATCTTAATACATCCTTT 560
MK188360 <i>Trissolcus japonicus</i>	TCTATCTTTACCAGTTTTCAGGGGCAATCACTATAATTTTATCAGATCGAAATCTTAATACATCCTTT 560

MK188349 <i>Trissolcus japonicus</i>	TTTAATCCAGCAGGA 575
MK188350 <i>Trissolcus japonicus</i>	TTTAATCCAGCAGGA 575
MK188360 <i>Trissolcus japonicus</i>	TTTAATCCAGCAGGA 575

Fig. S1. Alignment of TJ234F/TJ460R primers against three *T. japonicus* sequences. CO1 sequence alignment of three North American *T. japonicus* specimens used for primer design and location of primers TJ234F and TJ460R within the sequence.

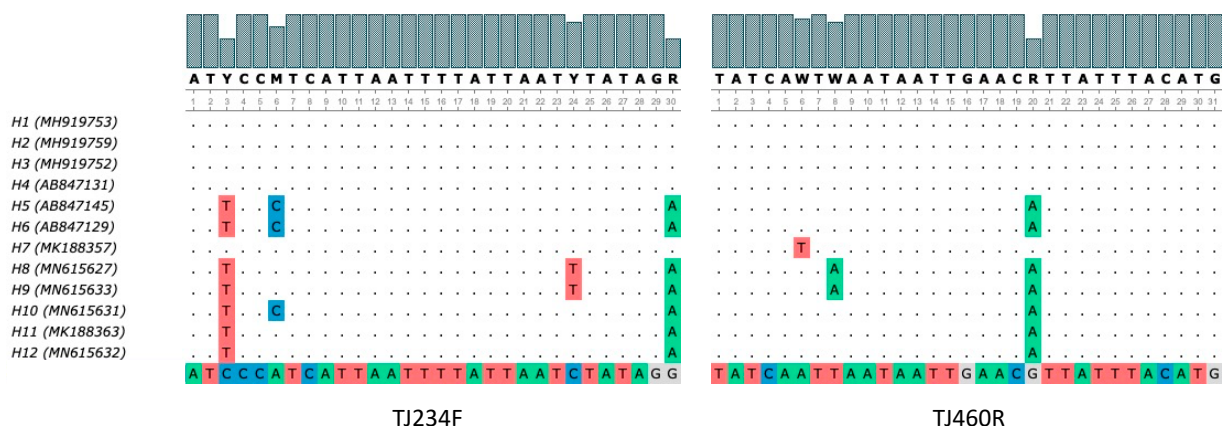


Fig. S2. Alignment between *T. japonicus* CO1 haplotypes and TJ234F/TJ460R primer mismatches. Primers show total homology with ca. 71% of the sequences in the NCBI database, the ones belonging to

haplotypes H1-H4. These haplotypes include all the identified specimens from the US, Canada, Switzerland and Italy, plus variable proportions of Asian *T. japonicus* populations. Following the name of the haplotype, a representative NCBI accession number is specified. The haplotype names are based on Stahl et al. [55] and analysis in this study.

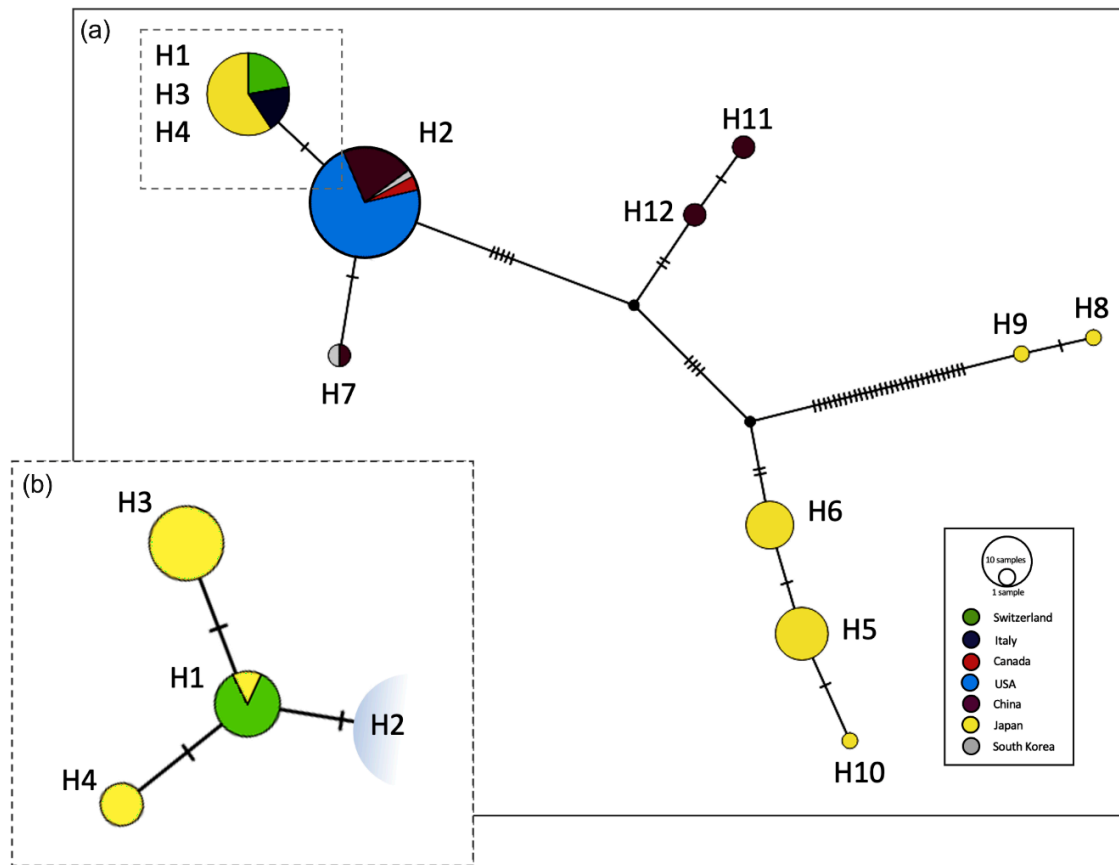


Fig. S3. Barcode haplotype network of the *T. japonicus* CO1 sequences analyzed in this study. Each circle corresponds to one haplotype. Circle sizes are proportional to the number of sequences belonging to the haplotype, and each color distinguishes a different geographical origin. Black dots are nodes and hatch marks show the number of mutations between haplotypes. The phylogenetic relationships among haplotypes were established using statistical parsimony in TCS as implemented in PopART software [57]. The analyzed sequences had different lengths and were trimmed before the analysis. In (a), the haplotype network was constructed with 103 *T. japonicus* CO1 sequences of 373 bp. The region of the analyzed CO1 sequences did not allow distinguishing between the haplotypes H1, H3 and H4 described by Stahl et al. [55], who used longer CO1 sequences for the analysis. In (b) the relationship between haplotypes H1, H3, and H4 described by Stahl et al. [55] is represented. In this case, the compared sequences are 423 bp long, and exclude the Italian and Japanese sequences whose short length did not allow their haplotype (H1, H3 or H4) to be elucidated.

		Position of Haplotype Markers																									
Haplotype	Representative sequence	5	16	19	29	34	37	55	61	67	73	74	76	91	94	100	103	106	109	115	154	155	160	161	175		
H1	MH919753	T	A	A	C	C	A	C	G	C	A	T	A	A	A	A	C	T	T	T	C	C	T	A	A		
H2	MH919759		
H3	MH919744		
H4	AB847131		
H5	AB847144	T	C	.	A	G	.	T	T	.	G	.		
H6	AB847129	T	C	.	A	T	T	.	G	.		
H7	MK188357		
H8	MN615627	C	G	G	T	T	.	T	A	T	G	.	C	.	.	T	.	A	A	A	A	T	C	G	T		
H9	MN615633	C	G	G	T	T	.	T	A	T	G	.	C	.	.	T	T	A	A	A	A	T	C	G	T		
H10	MN615631	T	C	.	A	G	.	T	T	.	G	.		
H11	MK188363	T	.	.	A	.	.	G	.	G	G	.		
H12	MN615632	T	.	.	A	G	G	.		
Haplotype	Representative sequence	178	187	196	217	233	235	247	278	281	284	286	289	290	310	313	319	329	343	349	355	358	364	367	370		
H1	MH919753	T	A	T	T	A	T	G	A	T	T	A	T	C	C	G	C	T	T	T	C	T	T	A	A		
H2	MH919759	A		
H3	MH919744		
H4	AB847131		
H5	AB847144	A	.	.	.	C	.	A	A	T	.	C	.	.	.		
H6	AB847129	A	.	.	.	C	.	A	A	T	.	C	.	.	.		
H7	MK188357	T	A	T	.	C	.	.	.		
H8	MN615627	A	G	C	C	.	A	A	.	C	C	T	C	T	A	A	T	C	C	C	.	.	C	G	T		
H9	MN615633	A	G	C	C	.	A	A	.	C	C	T	C	T	A	A	T	C	C	C	.	.	C	G	T		
H10	MN615631	A	.	.	.	C	.	.	A	A	T	C	C	.	.		
H11	MK188363	A	G	A	A		
H12	MN615632	A	G	A	A		
Haplotype	Representative sequence	374	375	376	377	378	379	380	381	382	383	384	385	386	387	388	389	390	391	392	393	394	395	396	397		
H1	MH919753	G	G	A	G	G	A	G	A	T	C	C	A	G	T	A	C	T	A	T	A	C	C	A	A		
H2	MH919759		
H3	MH919744		
H4	AB847131		
H5	AB847144	T		
H6	AB847129	T		
H7	MK188357		
H8	MN615627		
H9	MN615633		
H10	MN615631		
H11	MK188363		
H12	MN615632		
Haplotype	Representative sequence	398	399	400	401	402	403	404	405	406	407	408	409	410	411	412	413	414	415	416	417	418	419	420	421		
H1	MH919753	C	A	T	T	T	A	T	T	T	T	G	A	T	T	T	T	T	T	G	G	T	C	A	C		
H2	MH919759		
H3	MH919744	A	.	.	.		
H4	AB847131	T		
H5	AB847144		
H6	AB847129		
H7	MK188357		
H8	MN615627		
H9	MN615633		
H10	MN615631		
H11	MK188363		
H12	MN615632		

Fig. S4. Position of haplotype markers in *T. japonicus* CO1 sequence. For each haplotype, the NCBI accession number of a representative sequence is provided. Haplotype H1 was arbitrary selected as reference. Identical nucleotides are represented with dots. Differences between haplotypes H1-H6 were described by Stahl et al. [55] using CO1 aligned sequences of 423 nucleotides. Sequences representing haplotypes H7-H12 are shorter (dashes in a grey background denote non-available CO1 sequence regions) and can be differentiated from each other and from haplotypes H1-H6 by comparing a shorter region (373 bp).