

Table S1. Distribution of arrhenotoky, thelytoky and host-plant specialization among the seed-feeding wasps of the *Megastigmus* genus. Data were compiled from reviews of Grissell (1999), Roques & Skrzypczynska (2003), Roques *et al.* (2003) and Auger-Rozenberg *et al.* (2006). Species in bold were specifically studied in this paper. A: arrhenotokous parthenogenesis. T: thelytokous parthenogenesis. * Ratio of males to females; balanced: consideration of similar frequencies of females and males by authors; NA: no specific mention on sex ratio by authors but numbers of males mentioned in samples (thelytoky being associated with “males unknown” or “males are scarce” specific mentions). † These species are currently under description by MAAR and AR.

Host-plant taxon	Species	Reproduction	Sex ratio*	Main host-plant genus	Native area
Gymnosperms					
<u>Cupressaceae</u>	<i>M. chamaecyparidis</i>	A	balanced	<i>Chamaecyparis</i>	Palaearctic
	<i>M. thyoides</i>	A	balanced	<i>Chamaecyparis</i>	Nearctic
	<i>M. atlanticus</i>	A	balanced	<i>Cupressus</i>	Palaearctic
	<i>M. carinus</i>	A	NA	<i>Cupressus</i>	Palaearctic
	<i>M. cupressi</i>	A	NA	<i>Cupressus</i>	Oriental
	<i>M. duclouxiana</i>	A	NA	<i>Cupressus</i>	Palaearctic
	<i>M. watchli</i>	A	0.5-1.7	<i>Cupressus</i>	Palaearctic
	<i>M. amicum</i>	A	balanced	<i>Juniperus</i>	Palaearctic
	<i>M. bipunctatus</i>	A	balanced	<i>Juniperus</i>	Palaearctic
	<i>M. certus</i>	A	NA	<i>Juniperus</i>	Palaearctic
	<i>M. formosana</i>	A	balanced	<i>Juniperus</i>	East-Asia
	<i>M. fidus</i>	A	balanced	<i>Juniperus</i>	Palaearctic
	<i>M. gravis</i>	A	balanced	<i>Juniperus</i>	Palaearctic
	<i>M. juniperi</i>	A	NA	<i>Juniperus</i>	Palaearctic
	<i>M. pingii</i>	A	NA	<i>Juniperus</i>	East-Asia
	<i>M. procerae</i>	A	NA	<i>Juniperus</i>	Palaearctic
	<i>M. rigidae</i>	A	NA	<i>Juniperus</i>	Palaearctic
	<i>M. sabinae</i>	A	balanced	<i>Juniperus</i>	Palaearctic
	<i>M. somaliensis</i>	A	NA	<i>Juniperus</i>	Afrotropical
	<i>M. thuriferana</i>	A	balanced	<i>Juniperus</i>	Palaearctic
	<i>M. validus</i>	A	balanced	<i>Juniperus</i>	Palaearctic
	<i>M. thuyopsis</i>	A	NA	<i>Thuyopsis</i>	Palaearctic
<u>Pinaceae</u>	<i>M. firmae</i>	A	NA	<i>Abies</i>	Palaearctic
	<i>M. lasiocarpae</i>	A	balanced	<i>Abies</i>	Nearctic
	<i>M. milleri</i>	A	balanced	<i>Abies</i>	Nearctic
	<i>M. pinus</i>	A	0.4-0.5	<i>Abies</i>	Nearctic
	<i>M. rafni</i>	A	0.4	<i>Abies</i>	Nearctic
	<i>M. specularis</i>	A	balanced	<i>Abies</i>	Nearctic
	<i>M. schimitscheki</i>	A	0.4-0.5	<i>Cedrus</i>	Palaearctic
	<i>M. laricis</i>	A	balanced	<i>Larix</i>	Nearctic
	<i>M. atedius</i>	A	0.19-0.66	<i>Picea</i>	Palaearctic
	<i>M. ezomatsuanus</i>	A	0.5	<i>Picea</i>	Palaearctic
	<i>M. likiangensis</i>	A	NA	<i>Picea</i>	East-Asia
	<i>M. strobilobius</i>	A	0.4-0.6	<i>Picea</i>	Palaearctic

	<i>M. albifrons</i>	A	balanced	<i>Pinus</i>	Nearctic
	<i>M. strobiformis</i>	A	NA	<i>Pinus</i>	Nearctic
	<i>M. pseudotsugae</i>	A	NA	<i>Pseudotsuga</i>	East-Asia
	<i>M. spermotrophus</i>	A	balanced	<i>Pseudotsuga</i>	Nearctic
	<i>M. hoffmeyerii</i>	A	balanced	<i>Tsuga</i>	Nearctic
	<i>M. tsugae</i>	A	balanced	<i>Tsuga</i>	Nearctic
	<i>M. tsugaphilus</i>	A	NA	<i>Tsuga</i>	Palaeartic
	<i>M. borriesi</i>	T	♂<0.1%	<i>Abies</i>	East-Asia
	<i>M. suspectus</i>	T	♂<0.2%	<i>Abies</i>	Palaeartic
	<i>M. pinsapinis</i>	T	♂<0.1%	<i>Cedrus</i>	Palaeartic
	<i>M. pictus</i>	T	♂<1%	<i>Larix</i>	Palaeartic
<u>Taxodiaceae</u>	<i>M. cryptomeriae</i>	A	balanced	<i>Cryptomeria</i>	East-Asia
Angiosperms					
<u>Anacardiaceae</u>	<i>M. thomseni</i>	A	NA	<i>Heeria</i>	Afrotropical
	<i>M. kenyensis</i> [†]	A	NA	<i>Ozoroa</i>	Afrotropical
	<i>M. rhusi</i>	A	NA	<i>Rhus</i>	Afrotropical
	<i>M. transvaalensis</i>	A	0.5	<i>Schinus</i>	Afrotropical
	<i>M. pistaciae</i>	T	♂<0.1%	<i>Pistacia</i>	Palaeartic
<u>Fabaceae</u>	<i>M. albizziae</i>	A	NA	<i>Albizia</i>	Oriental
<u>Hamameliaceae</u>	<i>M. distylii</i>	A	NA	<i>Distylium</i>	Palaeartic
<u>Myrtaceae</u>	<i>M. ophelinii</i>	A	NA	<i>Eucalyptus</i>	Australasia
<u>Proteaceae</u>	<i>M. hakeae</i> [†]	A	NA	<i>Hakea</i>	Australasia
<u>Rhamnaceae</u>	<i>M. helinae</i> [†]	A	NA	<i>Helinus</i>	Afrotropical
<u>Rosaceae</u>	<i>M. amelanchieris</i>	A	NA	<i>Amelanchier</i>	Nearctic
	<i>M. cotoneastri</i>	A	NA	<i>Cotoneaster</i>	Palaeartic
	<i>M. mali</i>	A	NA	<i>Malus</i>	Palaeartic
	<i>M. pourthiaceae</i>	A	0.6	<i>Photinia</i>	Palaeartic
	<i>M. physocarpus</i>	A	NA	<i>Physocarpus</i>	Nearctic
	<i>M. fangii</i>	A	NA	<i>Rosa</i>	East-Asia
	<i>M. nigrovariegatus</i>	A	balanced	<i>Rosa</i>	Nearctic
	<i>M. yunnanensis</i>	A	NA	<i>Rosa</i>	East-Asia
	<i>M. aculeatus</i>	T	♂<4%	<i>Rosa</i>	Palaeartic
	<i>M. aculeatus nigroflavus</i>	T	♂<1%	<i>Rosa</i>	Palaeartic
	<i>M. rosae</i>	T	♂<1%	<i>Rosa</i>	Palaeartic
	<i>M. rosae var. alba</i>	T	♂<1%	<i>Rosa</i>	Palaeartic
	<i>M. brevicaudis</i>	T	♂<1%	<i>Sorbus</i>	Palaeartic

Table S2. Primer pairs used for endosymbiont PCR screening in *Megastigmus* spp.

Primer pair (5'-3')	Target group	Target gene (fragment size)	Annealing temperature (°C)
wsp81F TGGTCCAATAAGTGATGAAGAAAC ^a wsp691R AAAAATTAAACGCTACTCCA ^a	<i>Wolbachia pipientis</i>	<i>wsp</i> (610 bp)	59
Ch-F TACTGTAAGAATAAGCACCGGC ^b Ch-R GTGGATCACTTAACGCTTTTCG ^b	<i>Cardinium</i> sp.	16S rRNA (~900 bp)	57
Ars23S-1 CGTTTGATGAATTCATAG TCAAA ^c Ars23S-2 GGTCCTCCAGTTAGTGTTACCCAAC ^c	<i>Arsenophonus nasoniae</i>	23S rDNA (650 bp)	60
27f GAGAGTTTGATCCTGGCTCAG ^d 1495r CTACGGCTACCTTGTTACGA ^d	<i>Rickettsia</i> sp.	16S rRNA (1500 bp)	50
LCO1490 GGTCAACAAATCATAAAGATATTGG ^e HCO2198 TAAACTTCAGGGTGACCAAAAAATCA ^e	Arthropods	COI (658 bp)	48

^aBraig *et al.* (1998)

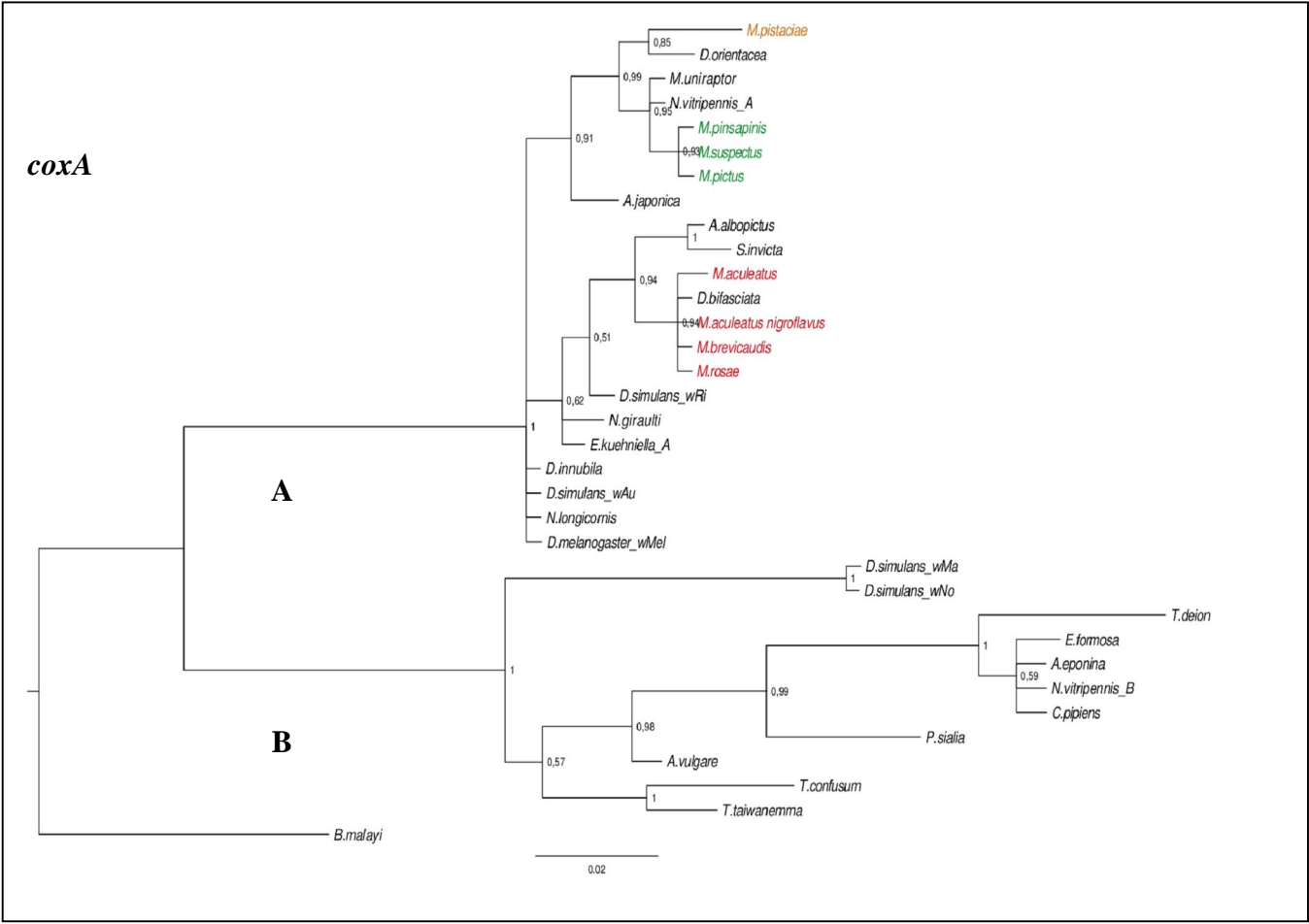
^bZchori-Fein and Perlman (2004)

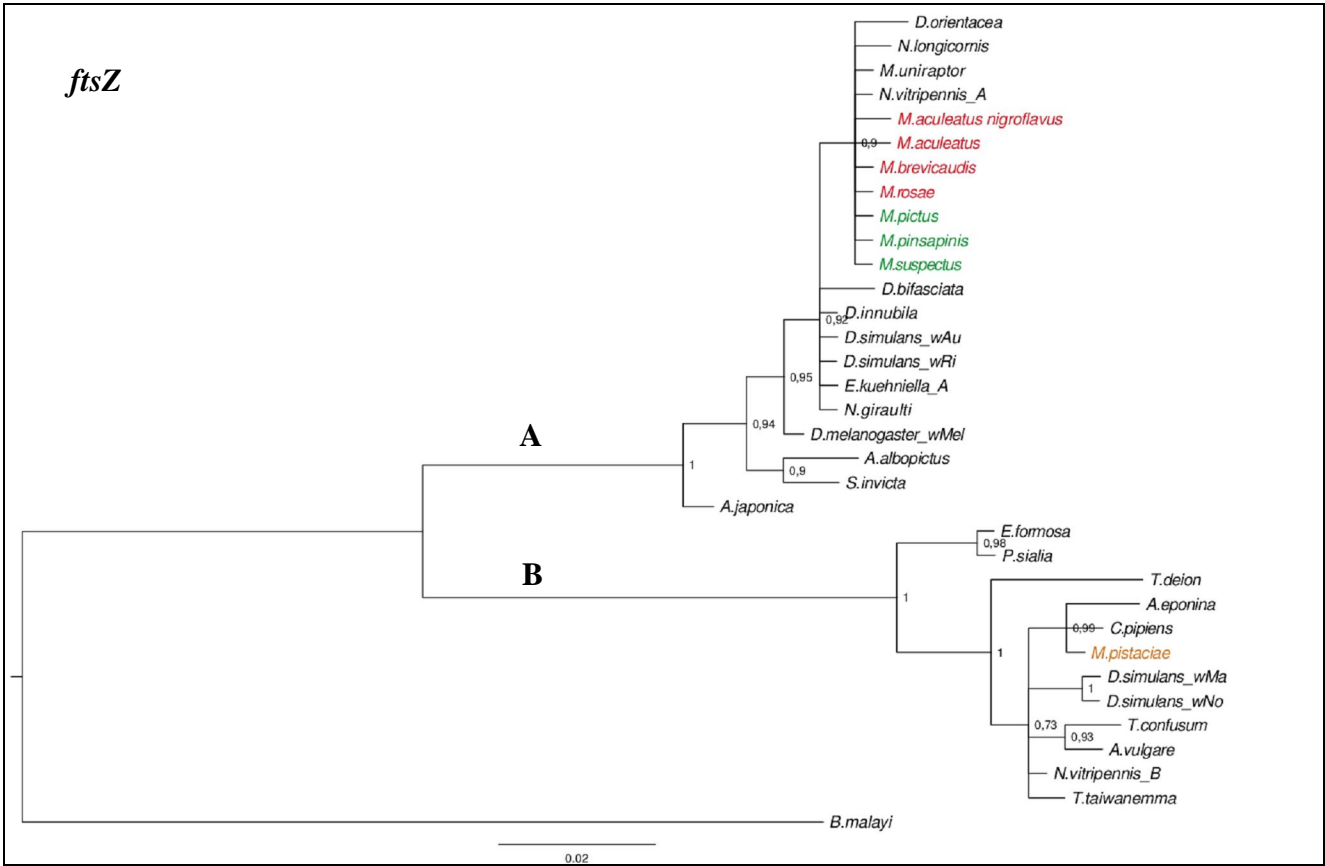
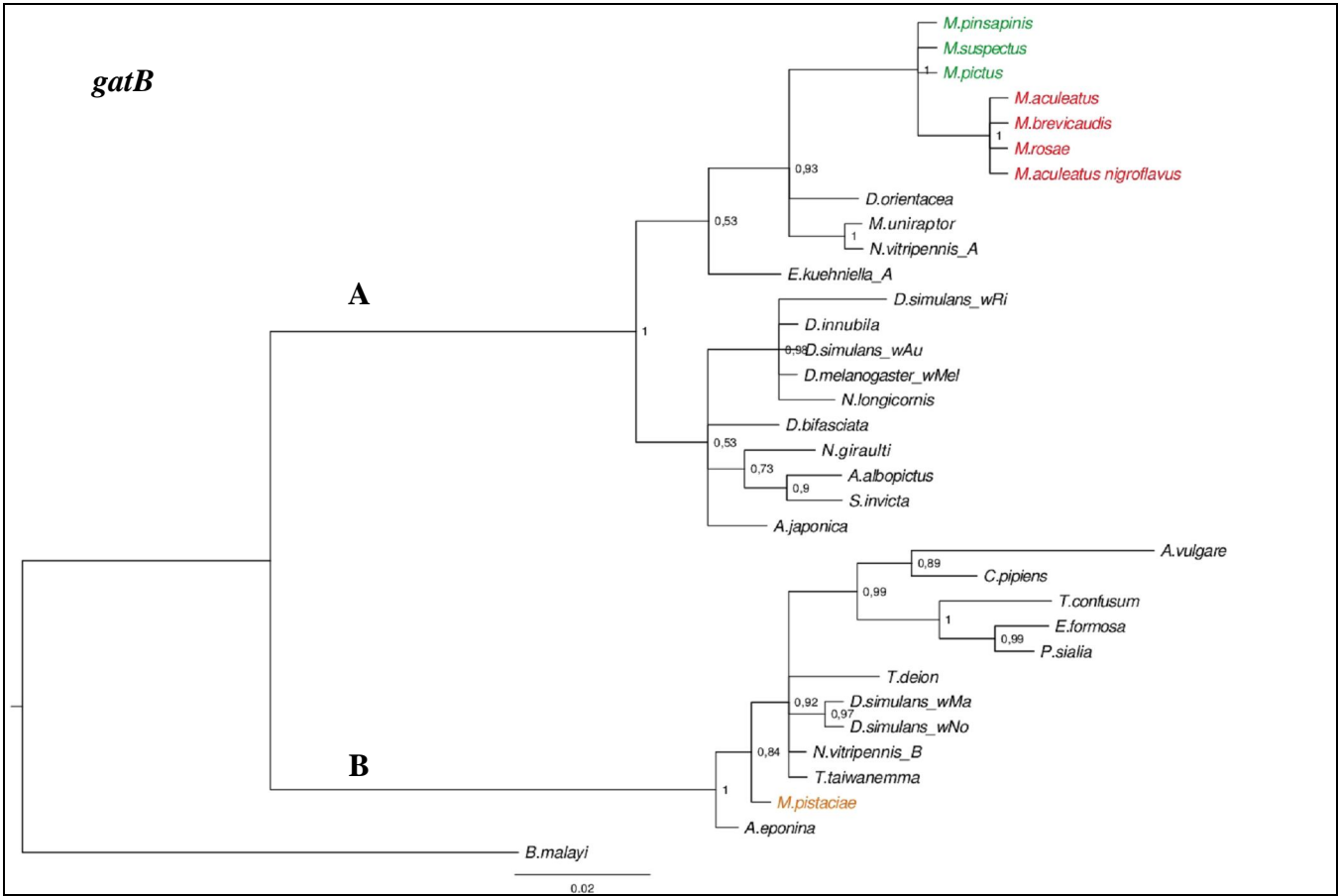
^cThao and Baumann (2004)

^dHagimori *et al.* (2006)

^eFolmer *et al.* (1994)

Figure S1. Phylogenetic placement of *Wolbachia* infecting the seed-specialized wasps *Megastigmus* spp., among other *Wolbachia* strains belonging to A and B supergroups, based on the sequences of each of the four MLST genes used in this study (*coxA*, *gatB*, *ftsZ* and *hcpA*). Bayesian likelihood inference phylogenies are shown, while maximum likelihood analyses gave substantially the same results. *Wolbachia* sequences are labelled with the name of their host. *Wolbachia* of *Brugia malayi* was used as an outgroup. Posterior probability values are indicated at each node. Each tree represents phylogenetic reconstruction based on 34 *Wolbachia* strains for *coxA* (402 bp), *gatB* (369 bp), *ftsZ* (435 bp) and *hcpA* (444 bp). Three lineages are revealed (green, red and light brown), two belonging to the A supergroup while the position of the *Wolbachia* infecting *M. pistaciae* suggests a recombination event between A and B *Wolbachia* supergroups. Indeed, this strain belongs to the B supergroup based on all sequences genes except for *coxA* for which it is placed in the B supergroup.

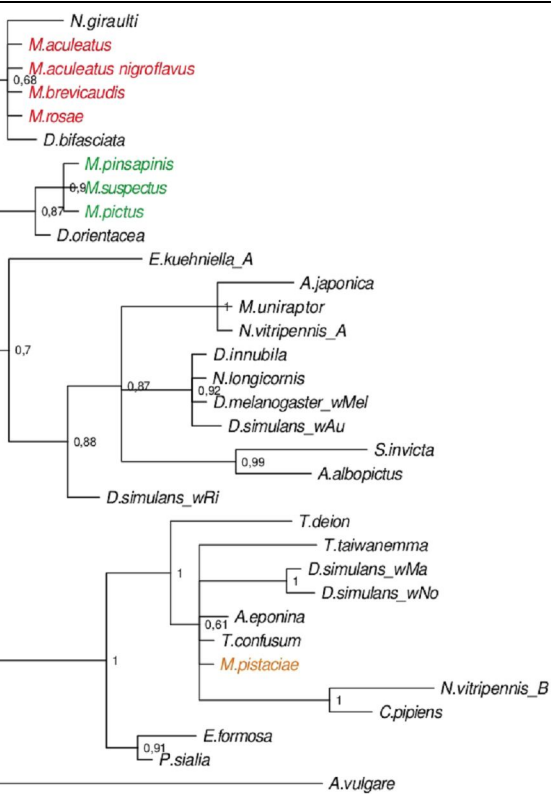




hcpA

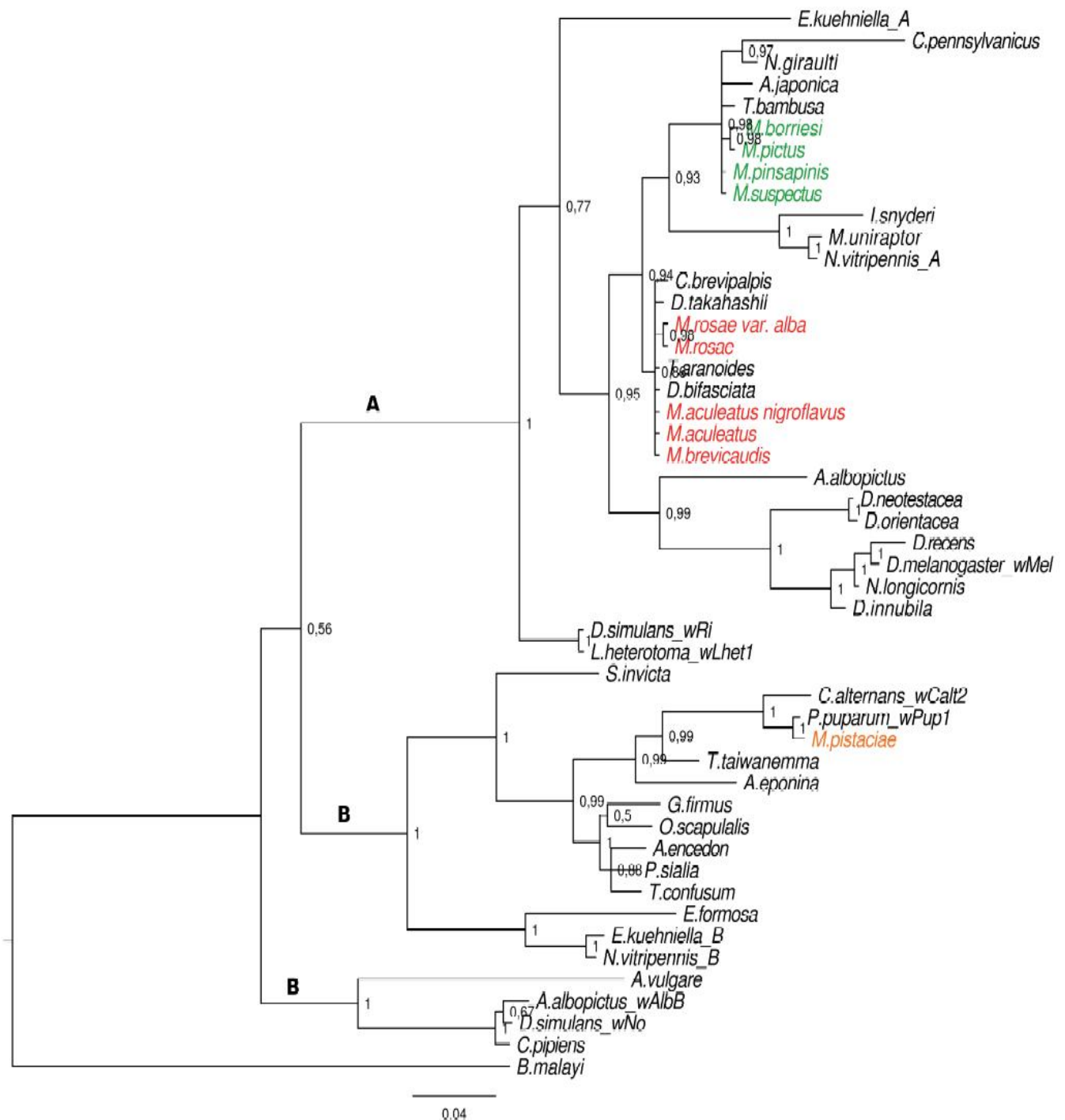
A

B



0.02

Figure S2. Phylogenetic placement of the *Wolbachia* strains infecting the seed-specialized wasps *Megastigmus* spp., among other *Wolbachia* strains belonging to A and B supergroups, based on wsp sequences of 49 strains (530 bp). Bayesian likelihood inference phylogenies are shown, while maximum likelihood analyses gave substantially the same results. *Wolbachia* sequences are labelled with the name of their host. *Wolbachia* of *Brugia malayi* was used as an outgroup. Posterior probability values are indicated at each node.



References

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