

**Table S1.** The primers and reaction program for PCR detection of endosymbionts in Asia II1 *Bemisia tabaci*.

Symbiont	Primer name	Primer sequence (5'-3')	amplicon size (bp)	Reaction program
<i>Portiera</i> <sup>a</sup>	28 F	TGCAAGTCGAGCGGCATCAT	1000	95°C 1min; 60°C 1min, 72°C 1min (5 cycles); 95°C 1min, 58°C 1min, 72°C 1min (25 cycles); 72°C 20min
	1098 R	AAAGTTCCCGCCTTATGCGT		
<i>Hamiltonella</i> <sup>b</sup>	Ham F	TGAGTAAAGTCTGGAATCTGG	700	95°C 1min; 60°C 1min, 72°C 1min (5 cycles); 95°C 1min, 58°C 1min, 72°C 1min (30 cycles); 72°C 20min
	Ham R	AGTTCAAGACCGCAACCTC		
<i>Cardinium</i> <sup>c</sup>	CFB F	GCGGTGTAAAATGAGCGTG	400	94°C 4min; 94°C 40s, 57°C 40s, 72°C 45s (35 cycles); 72°C 5min
	CFB R	ACCTMTTCTTAACTCAAGCCT		
<i>Rickettsia</i> <sup>d</sup>	Rb F	GCTCAGAACGAACGCTATC	900	95°C 2min; 92°C 30s, 58°C 30s, 72°C 30s (30 cycles); 72°C 5min
	Rb R	GAAGGAAAGCATCTCTGC		
<i>Arsenophonus</i> <sup>e</sup>	Ars23S-1	CGTTTGATGAATTCATAGTCAAA	600	94°C 5min; 94°C 30s, 52°C 30s, 72°C 1min (35 cycles); 72°C 5min
	Ars23S-2	GGTCCTCCAGTTAGTGTACCCAA C		
<i>Fritschea</i> <sup>f</sup>	U23F	GATGCCTTGGCATTGATAGGCGAT	600	95°C 5min; 94°C 1min, 55°C 1min, 72°C 1.5min (35 cycles); 72°C 5min
	23SIGR	GAAGGA TGGCTCATCATGCAAAAGGCA		
<i>Hemipteriphilus</i> <sup>g</sup>	OLO F	GCTCAGAACGAACGCTRKC	700	94°C 4min; 94°C 30s, 60°C 1min, 72°C 2min (35 cycles); 72°C 5min
	OLO R	TTCGCCACTGGTGTTCCTC		
<i>Wolbachia</i> <sup>h</sup>	wsp-81F	TGGTCCAATAAGTGATGAAGAAAC	600	95°C 3min; 94°C 35s, 55°C 30s, 72°C 30s (40 cycles); 72°C 10min
	wsp-691 R	AAAAATTAAACGCTACTCCA		

<sup>a</sup> Sequences obtained from Zchori-Fein and Brown. (2002) [41].

<sup>b</sup> Sequences obtained from Chiel et al. (2007) [42].

<sup>c</sup> Sequences obtained from Weeks et al. (2003) [43].

<sup>d</sup> Sequences obtained from Gottlieb et al. (2006) [26].

<sup>e</sup> Sequences obtained from Thao and Baumann (2004) [44].

<sup>f</sup> Sequences obtained from Everett et al. (2005) [45].

<sup>g</sup> Sequences obtained from Bing et al. (2013) [46].

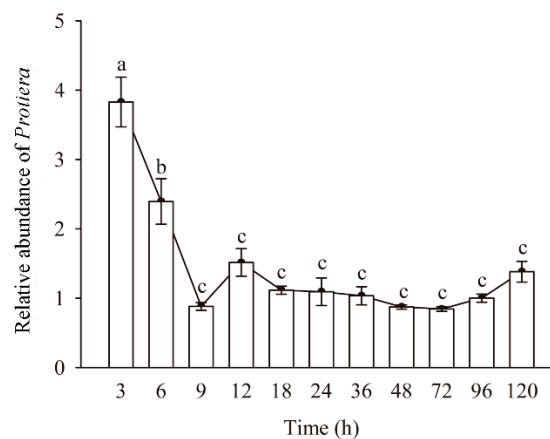
<sup>h</sup> Sequences obtained from Zhou et al. (1998) [27].

**Table S2** The information of oligonucleotide primers used in quantitative PCR in AsiaII1 *Bemisia tabaci*

Gene	Primer name	Primer sequence	Reference
<i>Wolbachia</i>	Cox14F	GCCAGTATTTGGTTATATGGGAATG	[28]
<i>coxA</i>	Cox14R	CTCGCTAAGCCCAACAGTA	
<i>Rickettsia</i>	gltA-F	CGGATTGCTTTACTTAC	[30]
<i>gltA</i>	gltA-R	AAATACGCCACCTCTA	
$\beta$ -Actin	Actin-QF	TCTTCCAGCCATCC TTCTTG	[29]
	Actin-QR	CGGTGATTTCCTTCTGCA TT	

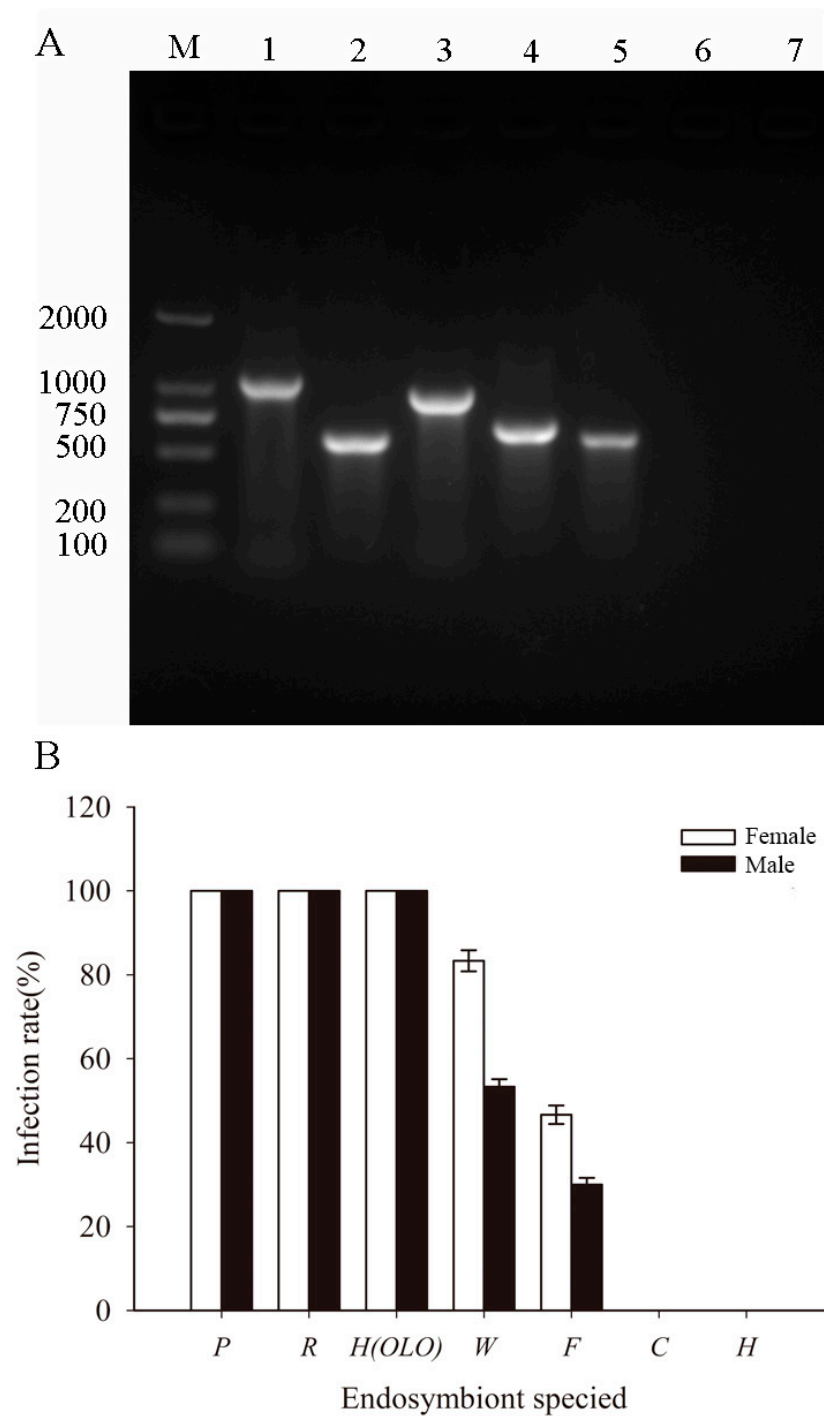
### PCR detection of secondary endosymbionts in Asia II1 whitefly

In order to evaluation the infection species and status of endosymbionts in the Asia II1 whitefly , newly emerged adults were randomly selected in the experiment population were measured by PCR with special primers of 16S *rRNA*, 23S *rRNA* and *wsp* genes of endosymbionts. Results revealed that the experiment population of Asia II1 whitefly was infected with one primary endosymbiont (*Portiera*) and four secondary endosymbionts (*Rickettsia*, *Wolbachia*, *Hemipteriphilus*, *Fritschea*), but not infected with *Cardinium* and *Hamiltonella*. The infection percentages of *Rickettsia* and *Hemipteriphilus* in whitefly adults were 100%, while The infection percentages of *Wolbachia* and *Fritschea* were respectively 83.3% and 46.67%, and the infection rate of males was higher than that of females.



**Figure S1** The relative abundance of *Portiera* in different times eggs of *B. tabaci*.

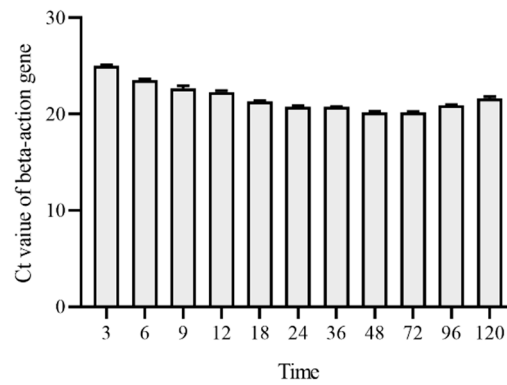
Error bars represent the standard error of the mean; Means marked with different letters are significantly different from each other ( $P \leq 0.05$ ).  $n= 3$  biological replicates. A  $\beta$  - actin whitefly gene was selected as an internal control for data standardization and quantification



**Figure S2** The endosymbionts information of Asia II 1 whitefly *B. tabaci*

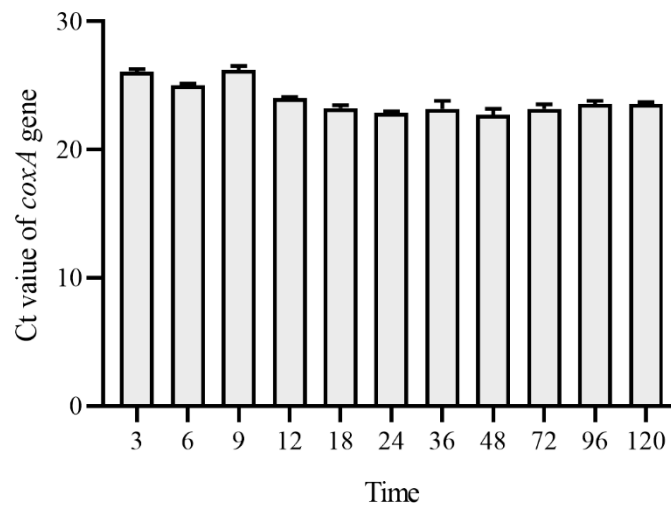
Note: A: The endosymbiont species infected in whitefly, M: DNA marker; 1: *Portiera*; 2: *Wolbachia*; 3: *Rickettsia*; 4: *Hemipteriphilus*; 5: *Fritschea*; 6: *Cardinium*; 7: *Hamiltonella*. B: The endosymbionts infection rates of whitefly. P: *Portiera*; R: *Rickettsia*; H(OLO): *Hemipteriphilus*; W: *Wolbachia*; F: *Fritschea*; C: *Cardinium*; H: *Hamiltonella*. all error bars

represent standard error of mean.



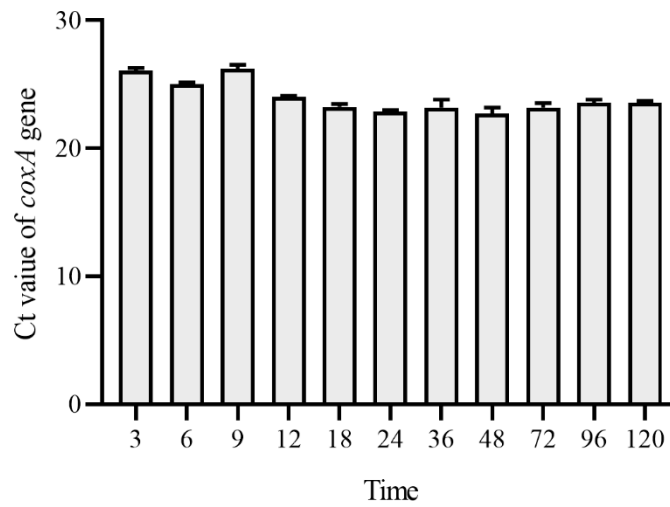
**Figure S3** Ct value of beta-actin gene in whitefly eggs at different time

Error bars represent the standard error of the mean;  $n=3$  biological replicates.



**Figure S4** Ct value of *coxA* gene in whitefly eggs at different time

Error bars represent the standard error of the mean;  $n=3$  biological replicates.



**Figure S5** Ct value of *glbA* gene in whitefly eggs at different time

Error bars represent the standard error of the mean;  $n=3$  biological replicates.

## Supplementary file 2

### Biological explanation of dynamic change of symbiotic bacteria titer

According to the biological characteristics of symbiotic bacteria, the reasons for the dynamic change of *Rickettsia* and *Wolbachia* titers were as follows: The titers of *Rickettsia* and *Wolbachia* decreased from 3 h to 9 h, and the fluorescence intensity of *Rickettsia* and *Wolbachia* also decreased from 3 h to 9 h by FISH detection. The main reason why the titer of *Rickettsia* and *Wolbachia* decreased was that the egg stalk could not provide a better cell environment, which *Rickettsia* and *Wolbachia* were intracellular symbiotic bacteria. When *Rickettsia* and *Wolbachia* enter the egg from the egg stalk, the titer and distribution range of *Rickettsia* and *Wolbachia* increase, which leads to

the decrease of fluorescence intensity of *Rickettsia* and *Wolbachia* per unit area, at the same time, the detection sensitivity of FISH was lower than that of qPCR, therefore, FISH analysis can not support the increase of the titers of *Rickettsia* and *Wolbachia* within 12-18 h. The reason why the titer of *Rickettsia* and *Wolbachia* decreased in 12-72 h was that it was lost with the transfer of *Rickettsia* and *Wolbachia* in eggs. Symbiotic bacteria may infect other tissues, such as Salivary gland, fat extraction and so on, in the process of transferring in egg, *Rickettsia* and *Wolbachia* was eventually fixed in the middle of the egg, and the titers of *Rickettsia* and *Wolbachia* increased with time.