**Additional File 5.** Comparison of *Wolbachia* infected individuals sampled from equatorial and tropical regions for (A) *B. bryoniae* (B), *B. frauenfeldi* (C) *B. neohumeralis*, (D) *B. strigifinis* and (E) *B. tryoni* over five collection years. Fisher's exact test shows overall no significant (ns) temporal effect at  $\alpha = 0.05$ , except for *B. tryoni*, but this did not represent an overall increase or decrease in prevalence.

Collection year	<b>A</b> B. bryoniae	<b>B</b> B. frauenfeldi	<b>C</b> B. neohumeralis	<b>D</b> B. strigifinis	<b>E</b> B. tryoni
1998	1/8	0/3	2/14	3/12	0/23
2001	1/2	2/6	1/4	0/3	2/5
2012		0/5	10/64	0/4	
2013	2/28	3/20		2/18	2/65
p value	p = 0.222	p=0.612	p = 0.848	p = 0.610	p = 0.019*