

# Effects of temperature and diet on development and interspecies competition in *Aedes aegypti* and *Aedes albopictus*

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1 **Title:**

2 **Effect of temperature and diet on the development and interspecific competition of *Aedes***  
3 ***aegypti* and *Aedes albopictus* (Diptera: Culicidae)**

4 **Short title: Population growth of *Ae. aegypti* and *Ae. albopictus***

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11  
12 **Abstract**

13 We asked whether climate change might affect the geographic distributions of *Aedes aegypti* (L.)  
14 and *Aedes albopictus* (Skuse) (Diptera: Culicidae). We tested the effects of temperature, diet, and  
15 the presence of congeneric species on the immature stage performance of these two aedine  
16 species in the laboratory. Mosquitoes in three different species-density combinations were reared  
17 at four constant temperatures (20, 25, 30, and 35°C) with low or high diets. Among the four  
18 temperatures tested, mortality increased only at 35°C in both species. Mortality was higher on  
19 the high diet than on the low one at 35°C, but not at other temperatures. Presence of the  
20 congeneric species had a significant positive effect on the mortality of *Ae. albopictus*, but not in  
21 case of *Ae. aegypti*. Both species developed more quickly at higher temperatures within the range  
22 20–30°C; development was not enhanced at 35°C. Population growth of *Ae. albopictus* was more  
23 stable, regardless of diet and temperature; that of *Ae. aegypti* varied more with these two factors.

24 These species-specific attributes may help explain the latitudinal distribution of the mosquitoes  
25 and degree of species dominance where they are sympatric.

26

27 **Key words:** *Aedes aegypti*, *Aedes albopictus*, interspecific competition, temperature, diet

28

## 29 **Introduction**

30 The effect of global climate change on the geographic distributions of vectors and vector-borne  
31 infectious diseases is an important issue. Dengue fever and dengue hemorrhagic fever are  
32 expanding problems in tropical and subtropical regions, and are now the most frequent arboviral  
33 diseases worldwide (Gubler, 2002). Two common mosquito species, *Aedes aegypti* (L.) and  
34 *Aedes albopictus* (Skuse), are competent vectors in Asian countries. Southeast Asia is the native  
35 habitat of *Ae. albopictus*, but *Ae. aegypti* has spread throughout the region and is increasing in  
36 abundance (Rudnick & Hammon, 1960; Giloira *et al.*, 1967; Jueco & Cabrera, 1969; Russell *et*  
37 *al.*, 1969; Ho *et al.*, 1973; Hawley, 1988). *Aedes albopictus* is a known vector of the dengue  
38 virus in parts of Asia (Smith, 1956; Russell *et al.*, 1969; Chan *et al.*, 1971a) where *Ae. aegypti* is  
39 rare or absent (Sunarto *et al.*, 1979; Metselaar *et al.*, 1980). In Asia, *Ae. aegypti* is more closely  
40 associated with human environments in which indoor and outdoor artificial containers, such as  
41 drums, tires, buckets, flowerpots, and vases, retain water and provide habitats for aquatic larval  
42 development (Focks *et al.*, 1981; Service, 1992; Focks & Chadee, 1997; Gubler, 1998). In  
43 contrast, the immature stages of *Ae. albopictus* inhabit natural water containers such as  
44 bromeliads, bamboo stumps, and tree holes in addition to artificial containers (Hawley, 1988).  
45 *Aedes aegypti* is most prevalent in urbanized areas, whereas *Ae. albopictus* occurs in rural,  
46 suburban, and vegetated urban areas in Calcutta, India (Giloira *et al.*, 1967). In both urban and

47 rural areas in Singapore, it is uncommon for the two species to share breeding habitats (Chan *et*  
48 *al.*, 1971b). It has been hypothesized that the displacement of *Ae. albopictus* by *Ae. aegypti* in  
49 certain Asian cities is the result of urbanization (Chan *et al.*, 1971; Hawley, 1988). In contrast to  
50 Asia, the abundances of the two species are similar in most suburban areas in southeastern Brazil  
51 and in Florida, United States (Braks *et al.*, 2003).

52 *Aedes albopictus* was introduced into Texas in the 1980s (Hawley, 1988) and has since  
53 spread across southern North America (Hobbs *et al.*, 1991; Mekuria & Hyatt, 1995), replacing  
54 *Ae. aegypti*, which was present in the southeastern United States for more than 100 years before  
55 the arrival of *Ae. albopictus* (Christophers, 1960; Lounibos, 2002). Regional photoperiod and  
56 temperature regimes may determine the distributions of the two mosquitoes (Hawley, 1988), and  
57 the degree of urbanization (Chan *et al.*, 1971a) or vegetation/detritus type (Murrell & Juliano,  
58 2008) may affect the outcome of interspecific competition where they are sympatric.

59 *Aedes albopictus* is one of the most common mosquito species on the main (Honshu) and  
60 southern (Kyushu) islands of Japan. *Aedes aegypti* is absent on these islands; although one report  
61 stated that it was temporarily present in a small southern Japanese town during the period 1944–  
62 1947 (Hotta, 1998). Unfortunately, we are unaware of the conditions that allowed the invasion of  
63 *Ae. aegypti* to occur or the reason why the species did not become successfully established,  
64 although interactions with *Ae. albopictus* may have played a crucial role.

65 The main environmental factors affecting mosquito population growth are temperature  
66 (Clements, 1992; Atkinson, 1994) and nutrition (Merritt *et al.*, 1992). Higher temperatures  
67 shorten the developmental duration of *Aedes* species (Tun-lin *et al.*, 2000; Alto & Juliano, 2001a;  
68 2001b; Delatte *et al.*, 2009), leading to the production of smaller adults (Reuda *et al.*, 1990; Rae,  
69 1990; Tun-lin *et al.*, 2000). Nutrition and density dependent factors also influence developmental

70 time and survivorship in *Ae. aegypti* (Christophers, 1960) and *Ae. albopictus* (Teng & Apperson,  
71 2000). Immature development in *Ae. aegypti* is governed by a combination of temperature and  
72 nutrition (Tun-lin *et al.*, 2000).

73 Considerable literature has accumulated regarding competition between *Ae. aegypti* and *Ae.*  
74 *albopictus*. Several studies have suggested that *Ae. albopictus* has a competitive advantage over  
75 *Ae. aegypti* (Barrera *et al.*, 1996; O'Meara *et al.*, 1995; Juliano, 1998; Daugherty *et al.*, 2000;  
76 Lounibos, 2002; Braks *et al.*, 2004; Juliano *et al.*, 2004; Murrell & Juliano, 2008). Other studies  
77 report the competitive superiority of *Ae. aegypti* (Moore & Fisher, 1969; Sucharit *et al.*, 1978;  
78 Serpa *et al.*, 2008). Although these studies support the hypothesis that negative interactions exist  
79 between the two species, their relative superiority is variable depending on nutrition conditioning  
80 and the stability of breeding sites. We need to monitor invasions of alien species and their  
81 establishment under on-going global warming. However, the distributions of the two species are  
82 apparently not governed by a single factor, such as temperature. In this study, we manipulated  
83 nutrition and temperature to represent the geographic and local variation found among aedine  
84 breeding sites. This experiment allowed us to evaluate the outcome of competition between the  
85 two species by studying population growth rates of immature stages in the laboratory.

86

## 87 **Materials and Methods**

### 88 *Mosquitoes*

89 Laboratory stocks of *Ae. aegypti* (collected in Moshi, Tanzania, 3°20'38"S and 37°20'76"E) and  
90 *Ae. albopictus* (collected in Nagasaki, Japan, 32°46'20.35"N and 129°52'9.86"E) were used. Both  
91 colonies had been maintained at the Institute of Tropical Medicine, Nagasaki University, for over  
92 3 years before they were transferred to our laboratory at Kanazawa University. Adults were

93 maintained at  $25\pm 1^{\circ}\text{C}$  and 70–90% relative humidity under 14L/10D photoperiod conditions.  
94 They were fed with a 3% sucrose solution daily and blood fed on rats once weekly.

95

#### 96 *Experimental Design*

97 Twenty-four (4 temperatures  $\times$  2 diets  $\times$  3 larval densities) treatment combinations were  
98 established. One experimental unit included 20 first instars (*Ae. aegypti* and *Ae. albopictus* in the  
99 following proportions: 0:20, 10:10, 20:0). Twenty larvae were transferred to a 500-ml vessel  
100 containing 200 ml dechlorinated tap water. Five replicates of single-species treatments and 10  
101 replicates of mixed-species treatments made up 100 individual per species per treatment.  
102 Experimental vessels were kept at constant temperatures of 20, 25, 30, or  $35^{\circ}\text{C}$  under a 14L/10D  
103 cycle photoperiod. Larvae were fed a mixture of rat food (CE-2, CLEA Japan, Inc. Tokyo) and  
104 yeast extract powder (Ebios, Mitsubishi Tanabe Pharma Corporation, Osaka ) (1:1 in weight).  
105 Two levels of the diet factor were tested: high and low. First and second instars were fed 0.2 mg  
106 and 0.05 mg/larva/day of larval food as the high and low diets, respectively; the respective  
107 feeding rates for third and fourth instars were 0.5 mg and 0.1 mg/larva/day as the high and low  
108 diets. Larval development, survival, molting, and pupation, were checked daily. Dead larvae  
109 were removed and rearing water was changed every second day to prevent scum formation and  
110 accumulation of metabolites. Pupae were isolated in vials until they emerged. Emerged adults  
111 were killed by freezing to allow measurement of one wing per insect using a micrometer under a  
112 stereomicroscope. The wing was measured from the distal end of the axial inclusion to the apical  
113 margin, not including the fringe (Van Den Heuvel, 1963).

114

#### 115 *Per Capita Performance*

116 We determined the condition-specific population performance parameter “*I*” of Livdahl and  
117 Sugihara (1984), with some modifications:

$$118 \quad \text{Ln}(1/N_0)(\sum w_x^3)$$
$$119 \quad I = \frac{\quad}{\quad}$$
$$120 \quad \frac{\sum x w_x^3}{\sum w_x^3}$$

121

122 where  $N_0$  is the initial number of larvae in an experimental treatment and  $w_x$  is the wing length of  
123 females that emerged on day  $x$ . The cubic value of the female wing length,  $w_x^3$ , a dimensionless  
124 expression of body volume, represents fecundity; this is a meaningful value for scaling metabolic  
125 parameters (Schmidt-Nielsen, 1984). A well documented positive correlation exists between  
126 wing length and fecundity, but the formula varies across the data range and by the method of  
127 data collection, possibly due to the trade-off between egg size and egg number (Berrigan, 1991;  
128 Hard & Bradshaw, 1993). Cubic values of wing length represent potential blood meal size and  
129 may be considered appropriate proxies of fecundity without considering the trade-offs between  
130 egg number and size. The definition of  $N_0$  in the original paper (Livdahl and Sugihara 1984) is  
131 the initial number of females; however, it is impossible to know the initial number of female  
132 unless all of eggs emerged into adult stages. To deal with the problem some studies define  $N_0$  as  
133 a half of eggs/larvae applied assuming even sex ratio in mosquitoes to determine “*I*” or similar  
134 population growth index (Livdahl & Sugihara, 1984; Lounibos *et al.*, 2002; Alto *et al.*, 2005). To  
135 our knowledge there was no data to judge if the assumption is rational, therefore, we determined  
136 “*I*” in two ways, one is to apply absolute data and the other is to calculate using average of wing  
137 length of females emerged day  $x$ , under the assumption of even sex ratio.

138

139 | *Statistical analysis:*

140 | Effects of temperature, diet and species size (single or mixed) on the mortality rate of *Ae. aegypti*  
141 | and *Ae. albopictus* were analyzed with ANOVA. Mortality ratios were arcsin square-root  
142 | transformed to meet the assumptions of normality and homogeneity of variances. We applied  
143 | MANOVA to analyze the effect of temperature and diet on the development time and wing size  
144 | of the aedine species respectively in mixed species treatment. Since we did not record the  
145 | development time by sex in single species treatment, we did not analyze the data further except  
146 | for mortality. By the same reason we calculate index “*T*” in mixed species but not in single  
147 | species treatment.

148 | To compare the mortality, development time and wing size among four temperature groups, we  
149 | used Tukey’s HSD test. We compared these parameters between high-low diet conditions by *t*-  
150 | test adjusted by Bonferroni correction. We compared sex-specific wing size between single  
151 | species and mixed species treatments using *t*-test adjusted by Bonferroni correction again.  
152 | Statistical analyses were performed using JMP version 5.0.1.2 (SAS Institute, Cary, NC, USA).

153 |

## 154 | **Results**

155 | Most mortality occurred in the late larval stages: 44.0% of the total mortality at the fourth instars  
156 | and 40.7% at the pupal stages in *Ae. aegypti*, and 31.0% at the fourth instars and 52.5% at the  
157 | pupal stages in *Ae. albopictus*. The ANOVA results showed that species interactions had a  
158 | significant negative effect of *Ae. aegypti* on *Ae. albopictus*, but not of *Ae. albopictus* on *Ae.*  
159 | *aegypti* (Table 1). The interaction of temperature × diet was significant in both species, but that  
160 | of temperature × species was only significant in *Ae. albopictus* (Table 1). Mortality rates were  
161 | significantly higher at 35°C than at lower temperatures for both species ( $P < 0.05$ ; Fig. 1). The



162 effect of diet level was significant only at 35°C for both species, where more mortality occur  
163 under high diet conditions (Fig. 1A, C). *Aedes albopictus* showed significantly higher mortality  
164 under mixed species treatment at 35°C (Fig. 1D). Comparisons of the effects of single/mixed-  
165 species treatment on mortality revealed complex effects. No effect of *Ae. albopictus* on *Ae.*  
166 *aegypti* was observed except under the treatment combination of high diet × 35°C ( $P < 0.01$ ).  
167 The mortality of *Ae. albopictus* was higher in the presence of *Ae. aegypti*, but it was condition  
168 specific, i.e., mortality was reduced under the mixed species × low diet × 25°C ( $P < 0.01$ )  
169 treatment combination, and mortality was elevated under the mixed species × low diet × 30°C  
170 and the mixed species × high diet × 30°C treatments ( $P < 0.01$ ).

171 We recorded developmental duration by sex in the mixed-species treatment only (Fig. 2A,  
172 B). Males generally had shorter development times than females. Increasing temperature reduced  
173 developmental duration in the temperature range of 20–30°C, but an increase in developmental  
174 duration was observed at 35°C in *Ae. albopictus* males, which took longer to develop at 35°C  
175 than at 30°C (Fig. 2B). Development times were reduced in both sexes of both species with the  
176 high diet at all temperatures (Fig. 2A, B).

177 Wing size was significantly reduced by increasing temperature (Fig. 3). Larger *Ae. aegypti*  
178 emerged under the high diet regardless of temperature or single/mixed species condition ( Fig.  
179 3A, B). However, the high diet did not contribute to larger *Ae. albopictus* at 35°C, except for  
180 males in the single-species treatment (Fig. 3C, D). Wing size was greatest under the low  
181 temperature × high diet combination, and it was smallest under the high temperature × low diet  
182 combination (Fig. 3). Each species was influenced by the presence of the other at 25°C and 30°C  
183 under the high diet; the effect was stronger under the low diet for *Ae. aegypti* than for *Ae.*  
184 *albopictus*. Both male and female *Ae. aegypti* were larger under mixed-species combinations

185 than under single species treatments at 20, 25, and 30°C (Fig. 3A, B). *Aedes albopictus* females  
186 were larger under mixed-species conditions at 20°C, whereas males were larger at 20°C and  
187 35°C in mixed populations (Fig. 3C, D).

188 MANOVA indicated that the interaction of temperature × diet had a significant effect on  
189 the development time and wing size of females of both species and on males of *Ae. albopictus*  
190 (Table 2).

191 The overall female ratio of *Ae. aegypti* was 0.502, with no significant departure from 0.5 in  
192 any treatment ( $\chi^2$  test,  $P > 0.1$ ), whereas in *Ae. albopictus*, the ratio was 0.469, which was  
193 significantly different from 0.5 ( $\chi^2$  test,  $P = 0.03$ ). Furthermore, the sex ratio differed  
194 considerably among treatments. More females of *Ae. albopictus* emerged with low diets than  
195 with high diets (female ratio: 0.502 vs. 0.435,  $\chi^2$  test,  $P < 0.01$ ) and under the mixed-species  
196 treatment than under the single-species treatment (female ratio: 0.525 vs. 0.419,  $\chi^2$  test,  $P <$   
197 0.001).

198 We calculated the per capita performance index,  $I$ , to integrate the effects of  
199 temperature and diet on population growth under a mixed-species treatment. This would further  
200 allow us to determine which species would be favored by the presence of the other species  
201 (Figure 4). The  $I$  values of the two species increased with temperature up to 30 °C; however, the  
202  $I$  values declined at higher temperatures and were the lowest at 35 °C (Fig. 4). Diet also affected  
203 the  $I$  values of the two species. In *Ae. aegypti*, the  $I$  values were 61.2–93.4% higher with a high  
204 diet than with a low diet at the same temperature, while they were 48.3–56.7% higher with a high  
205 diet in *Ae. albopictus*. The  $I$  values of the two species were similar on a low diet (Fig. 4A). For  
206 *Ae. aegypti*, the  $I$  values were 35.1% higher at 30 °C (absolute sex ratio) than at 25 °C, whereas  
207 for *Ae. albopictus* the  $I$  values were only 4.4% higher (Fig. 4B). If we assume asymmetric diet

208 conditions, the species on a high diet would overwhelm that on a low diet at temperatures  
209 between 20 and 30 °C (Fig. 4C, D).

210

## 211 **Discussion**

212 We showed that temperature and diet combinations had variable effects on the population  
213 growth of the two species, and that interspecific competitive superiority can be condition  
214 dependent. Although *Ae. aegypti* performed better at temperatures higher than 25 °C on a high  
215 diet, the population growth in both species was quite similar. Therefore, there should be no  
216 difference in their population growth under the same breeding conditions. Nevertheless, there is  
217 striking sympatric variation in water temperatures in the field, ranging from 10 to 30 °C (Tun-  
218 Lin *et al.*, 2000; Tuno *et al.*, 2005). There is also drastic variation in the larval survival rate at  
219 similar water temperatures, indicating large variation in terms of larval diet (Tun-Lin *et al.*,  
220 2000; Tuno *et al.*, 2005). The variable temperatures and diet observed in the field may enhance  
221 species coexistence, since both factors markedly affect the population growth of the two species.

222 Brakes *et al.* (2003) reported that *Ae. aegypti* was most prevalent in highly urbanised  
223 areas, although the habitats used by the two species are remarkably similar in most suburban  
224 areas in southeastern Brazil and Florida, despite their hypothesis of habitat segregation. The  
225 paradoxical distribution of the two species cannot be explained by the three factors examined in  
226 our study: temperature, nutrition, and the presence of other species. The stability of breeding  
227 sites may be an important factor to consider (Alto & Juliano, 2001b). *Aedes aegypti* has been  
228 reported to be prevalent in highly urbanised areas. Highly urbanised areas may be interpreted as  
229 a highly disturbed unpredictable environment. *Aedes aegypti* has higher population growth than  
230 *Ae. albopictus* if conditions allow. Based on our observations, the former species always hatches

231 together, while the latter species hatches irregularly when the eggs were soaked in water. These  
232 features, *i.e.*, immediate hatching and rapid population growth, may enable *Ae. aegypti* to  
233 establish colonies in highly disturbed environments in urban areas, in addition to their closer  
234 association to humans (Hawley, 1988). By the end of the 21st century, world temperature will  
235 have increased by 1.1 to 6.4 °C (IPCC 2007). Based on our data we predict that, with an elevated  
236 world temperature, the population growth of *Ae. aegypti* will increase in regions where the  
237 average temperature is in the range of 20 to 30 °C. With global climate change, the distribution  
238 of both species will likely expand when temperature is considered alone. However, climate  
239 change also affects precipitation patterns (IPCC 2007) and interspecies competition (Ives and  
240 Gilchrist, 1993), which may affect the distributions of both species. In addition, the combined  
241 effect of temperature and diet makes it difficult to predict the expansion of these two species.

242 Counter to assumptions in previous studies, the sex ratio of the *Ae. albopictus* population was  
243 not even. This result emphasises the need to evaluate the absolute sex ratio and to explore  
244 whether environmental conditions affect aedine sex ratios for precise estimation of population  
245 growth rates.

246 High nutrition reduced fitness at 35 °C, despite the fact that a high diet contributed to greater  
247 fitness when the temperature was 20–30 °C. Across the four temperatures (20, 25, 30, and 35 °C),  
248 mortality differed (was higher) only in the 35 °C treatment group in both species. Most deaths in  
249 this study occurred at the fourth instar and pupal stages at 35 °C. Holometabolous insects, such  
250 as mosquitoes, must attain a critical mass during larval development before pupation (Clements,  
251 1992). They also require sufficient mass for emergence, and the potential attainable mass  
252 decreases with increasing temperature (Chambers & Klowden, 1990). The effect of the high diet  
253 on mortality was reversed at 35 °C in both species, and the developmental duration in *Ae.*

254 *albopictus* males was also reversed at this high temperature. Larval feeding duration affects  
255 ecdysteroid levels and nutritional reserves, which are crucial factors in the initiation of  
256 metamorphic moult in mosquito larvae (Telang *et al.*, 2007). The high mortality rates among  
257 fourth instars and pupae on high diets at 35 °C may be explained by the early triggering of  
258 metamorphosis by elevated nutritional reserves. This early onset would not allow sufficient time  
259 for ecdysteroid accumulation to reach a level that would permit the completion of  
260 metamorphosis. Nevertheless, it is difficult to explain why the developmental duration was  
261 prolonged at 35 °C in *Ae. albopictus* males, although the cues triggering metamorphosis may be  
262 species specific (e.g., specific nutritional reserve or ecdysteroid levels).

263 We demonstrated that the competitive status of the two mosquito species is affected by  
264 temperature and nutrition. The performances of the two species are quite similar under the same  
265 conditions. We predict that considerable variation in their breeding sites enables coexistence of  
266 the two species. We also speculate that temperature and nutrition are not sufficient to explain the  
267 puzzling distribution of the two species. We need to consider other parameters, such as  
268 environmental stability, to better understand the location-specific dominance outcomes in the  
269 two species.

270

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275

276

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430 **Table 1. Summary of ANOVA on the mortality rate from hatching to emergence in single**  
 431 **and mixed species treatment.**

Source	<i>Ae. aegypti</i>			<i>Ae. albopictus</i>		
	df	F Ratio	P	df	F Ratio	P
Temperature	3	84.6	<0.0001	3	115.2	<0.0001
Diet	1	3.4	0.068	1	2.9	0.090
Mixed species	1	3.3	0.070	1	4.1	0.045
Temperature*Diet	3	12.1	0.001	3	8.3	0.005
Temperature*Mixed species	3	1.6	0.214	3	8.0	0.006
Diet*Mixed species	1	0.6	0.453	1	0.1	0.801
Temperature*Diet*Mixed species	3	0.9	0.345	3	0.2	0.676
C. total	119			119		

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441 **Table 2: Summary of MANOVA on the sex wise developing time and wing size in mixed species treatment.**

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		<i>Ae. aegypti</i>				<i>Ae. albopictus</i>			
Analysis	Source	F value	NumDF	DenDF	P	F value	NumDF	DenDF	P
Female									
	Temperature	540.9	2	71	<0.0001	353.9	2	67	<0.0001
	Diet	199.8	2	71	<0.0001	162.2	2	67	<0.0001
	Temperature*Diet	5.1	2	71	0.0087	9.0	2	67	0.0003
Male									
	Temperature	455.2	2	68	<0.0001	164.0	2	69	<0.0001
	Diet	131.2	2	68	<0.0001	242.5	2	69	<0.0001
	Temperature*Diet	0.1	2	68	0.8831	9.0	2	69	0.0003

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444

445 Figure legends.

446 Fig. 1. Mortality rate (%) from second instars to emergence maintained at four temperatures: 20,  
447 25, 30, and 35. (A) *Aedes aegypti* – Low vs high diet (B) *Ae. aegypti*-Single vs 2 species (C) *Ae.*  
448 *albopictus*- Low vs high diet (D) *Ae. albopictus* - Single vs 2 species. Different letters indicates  
449 significant differences between temperature treatments ( $P < 0.05$ ; Tukey's HSD test). Significant  
450 differences between poor and rich nutritious conditions are indicated by asterisk ( $P < 0.05$ , Paired  
451 *t*-test, Bonferroni corrected).

452 Fig. 2. Developing time from hatching to emergence (days) of male and female (A) *Ae. aegypti*  
453 and (B) *Ae. albopictus* in mixed species treatment.

454 Fig. 3. Wing length (mm) of *Ae. aegypti*, (A) female, (B) male, and *Ae. albopictus*, (C) female,  
455 (D) male at 20, 25, 30 and 35 °C.

456 Fig. 4. Estimated per capita performance index *I* of *Ae. albopictus* and *Ae. aegypti* with observed  
457 sex ratio and with assumed even sex ratio under combinations diet (low or high) and rearing  
458 temperatures(20, 25, 30 and 35 °C) in mixed species treatment. (A) *Ae. aegypti*-Low diet & *Ae.*  
459 *albopictus*-Low diet (B) *Ae. aegypti* -High diet & *Ae. albopictus* –High diet (C) *Ae. aegypti*-High  
460 diet & *Ae. albopictus* –Low diet (D) *Ae. aegypti* -Low diet & *Ae. albopictus* -High diet.  
461 Abbreviations in the figure: *Ae. aegypti* (aeg), *Ae. albopictus* (albo), low diet (L), High diet (H),  
462 1:1 sex ratio assumed (1:1).

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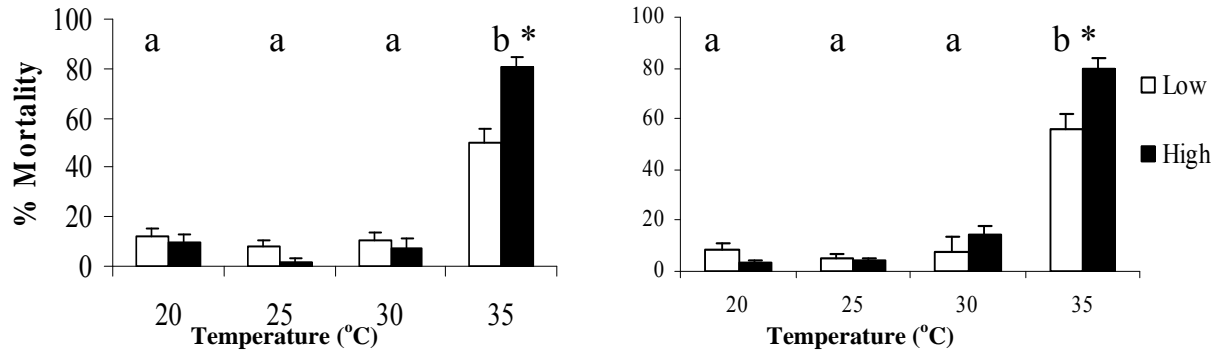
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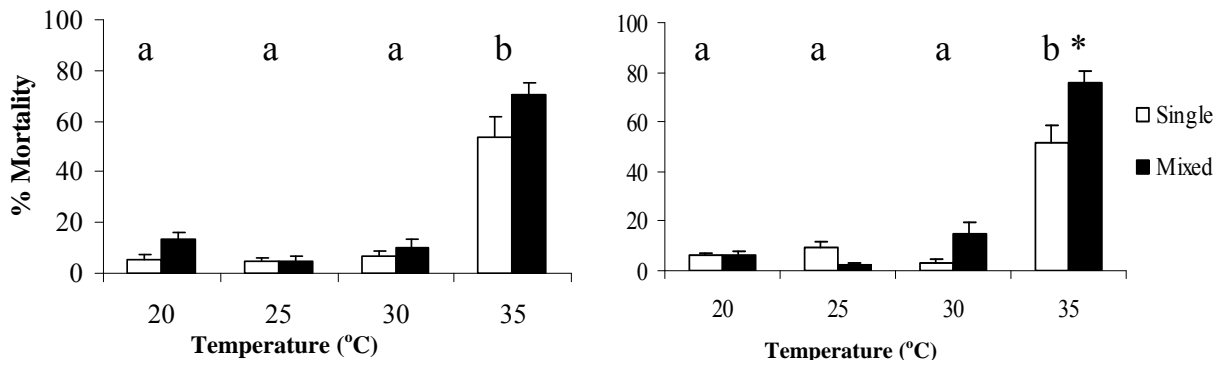
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468 **Fig. 1:**



(A)

(C)



(B)

(D)

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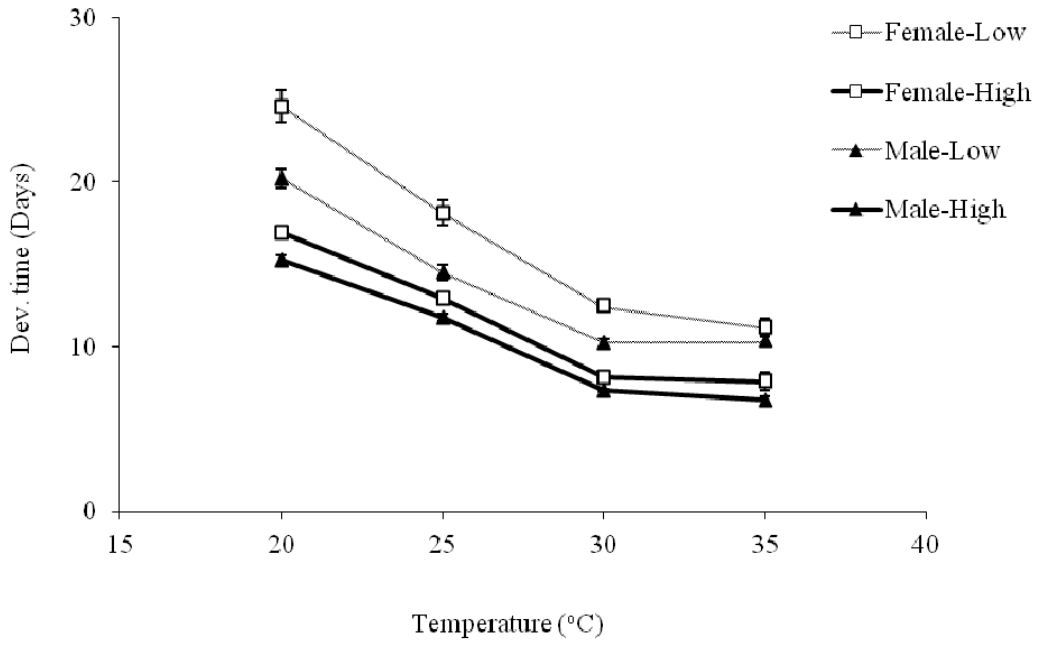
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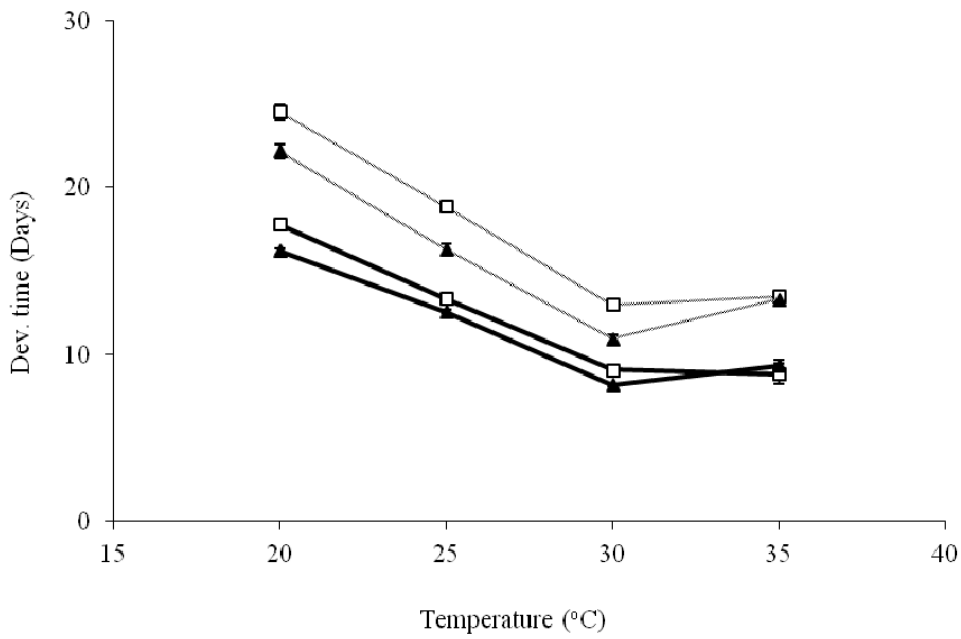
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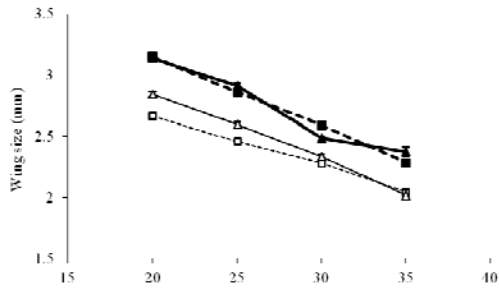




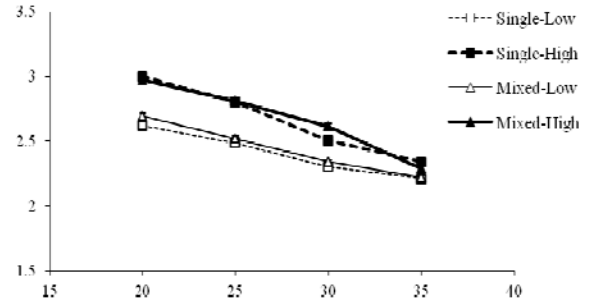
(A)



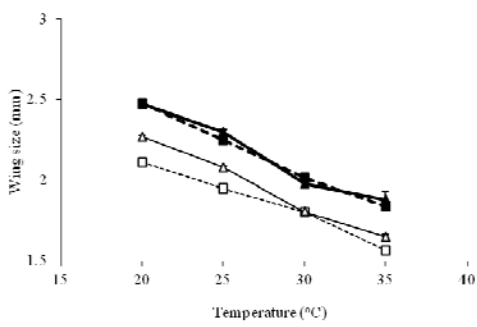
(B)



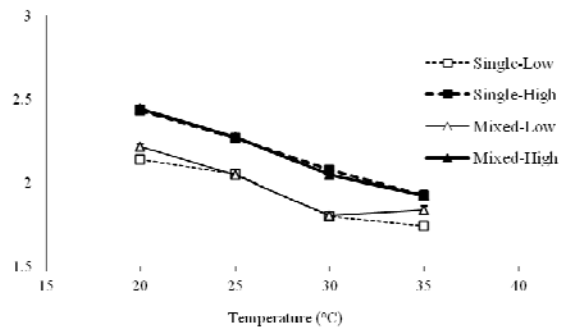
(A)



(C)



(B)



(D)

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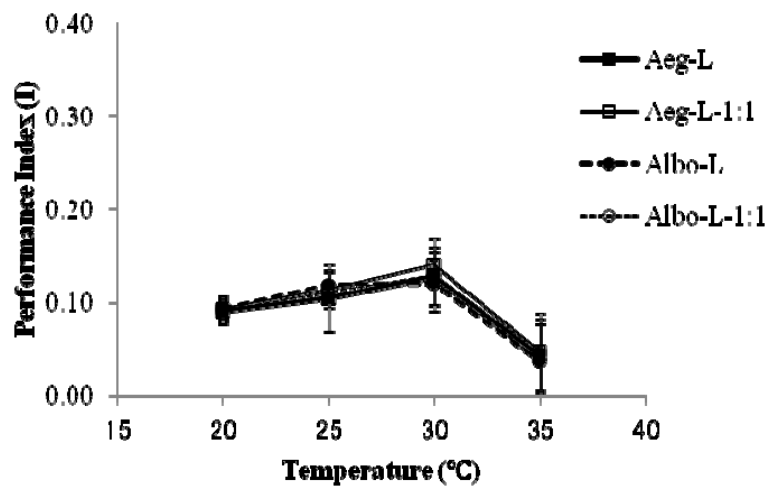
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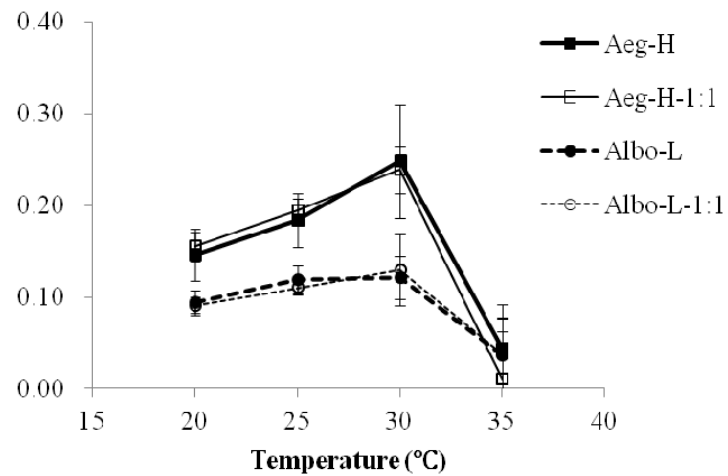
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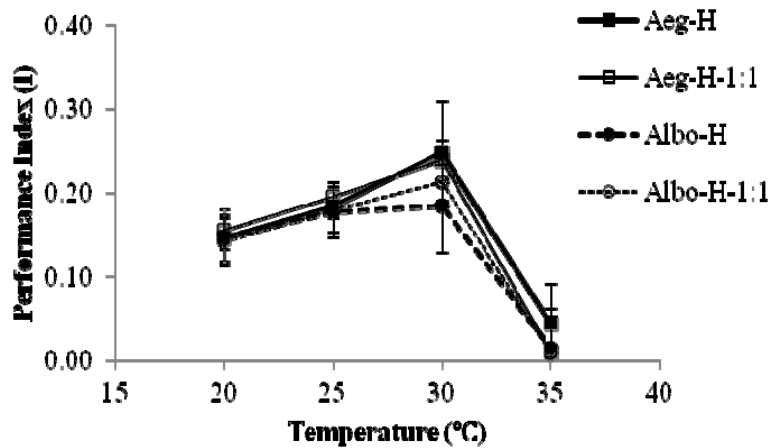
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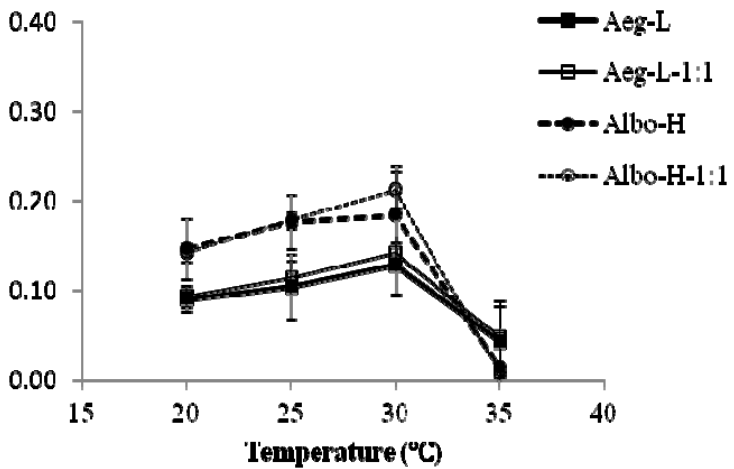
(A)



(C)



(B)



(D)