Ecological Dynamics of *Aedes aegypti-Wolbachia pipientis* *w*AlbB Strain Interactions

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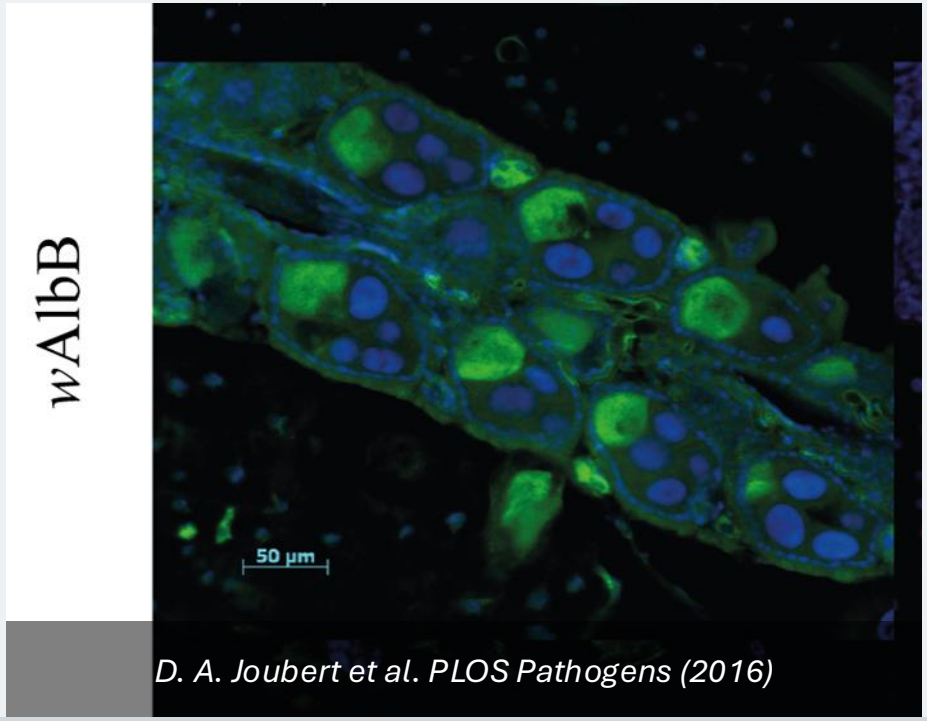
EMORY, ENVS DEPARTMENT

PROKOPEC LABORATORY

WRBU



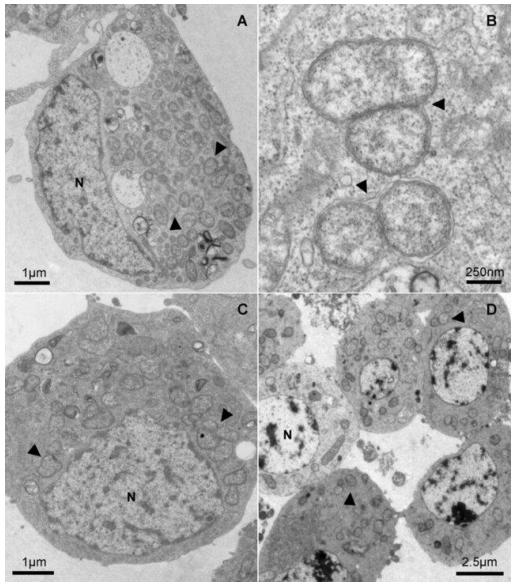
D. A. Joubert et al. PLOS Pathogens (2016)

Wolbachia pipientis

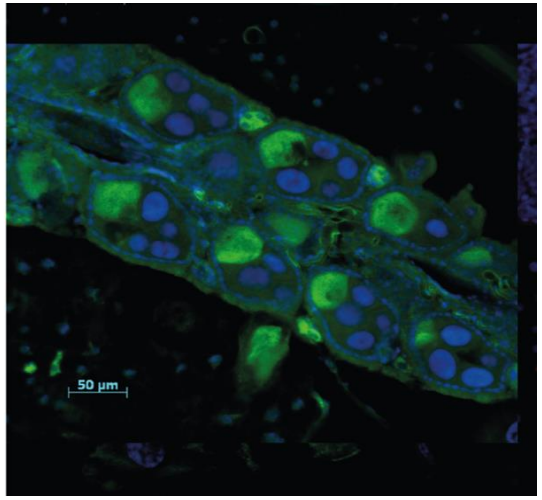
- *Cx. pipiens* – incompatibility
  - 1924, Hertig and Wolbach

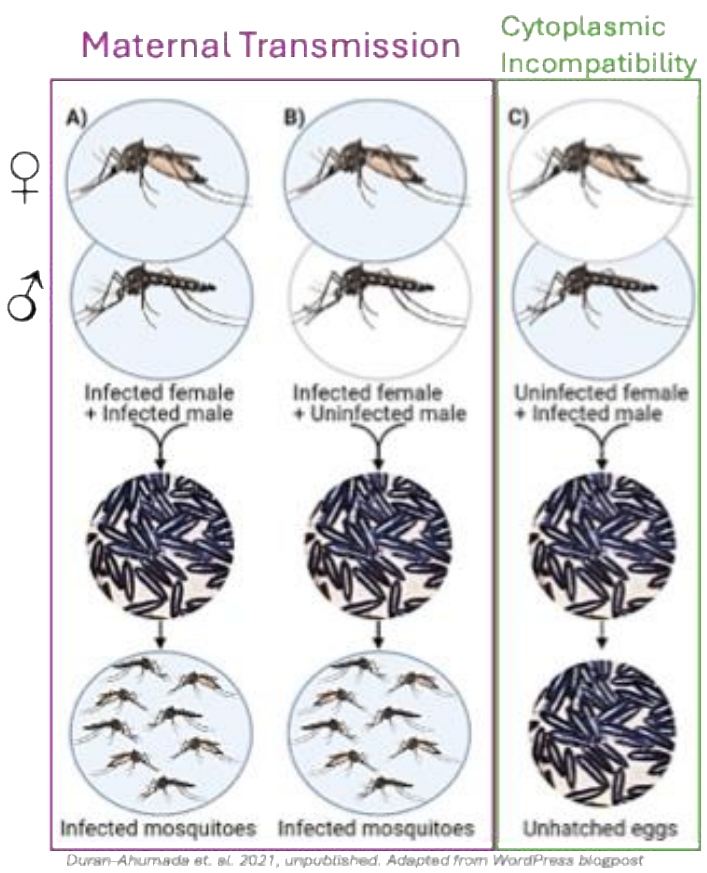
- Insects
  - Est. 60-70% of insect species

- Strains of interest interest for control of *Aedes*
  - *w*Mel
  - *w*MelPop
  - *w*AlbB
  - *w*AlbA

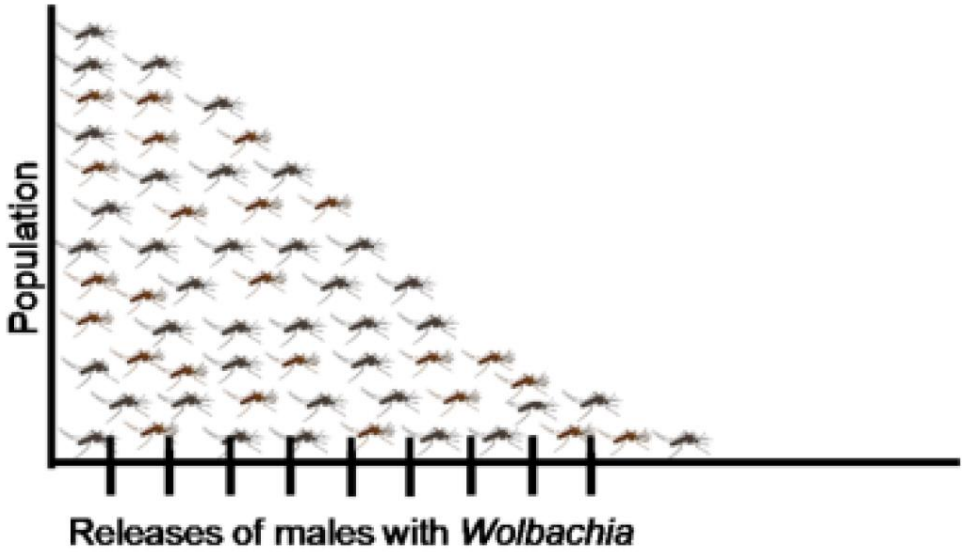


*w*AlbB





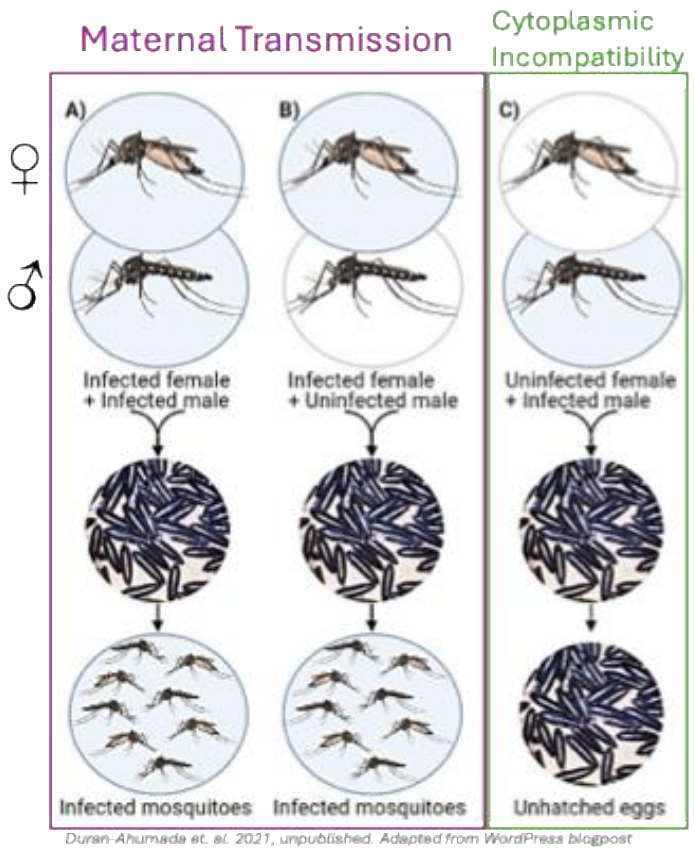
Duran-Ahumada et. al. 2021, unpublished. Adapted from WordPress blogpost



Wolbachia pipientis

Incompatible Insect Technique

- Male releases ≈ SIT
  - IIT-SIT
- Local suppression/elimination
- Ideally n.s. fitness/performance effects on released males



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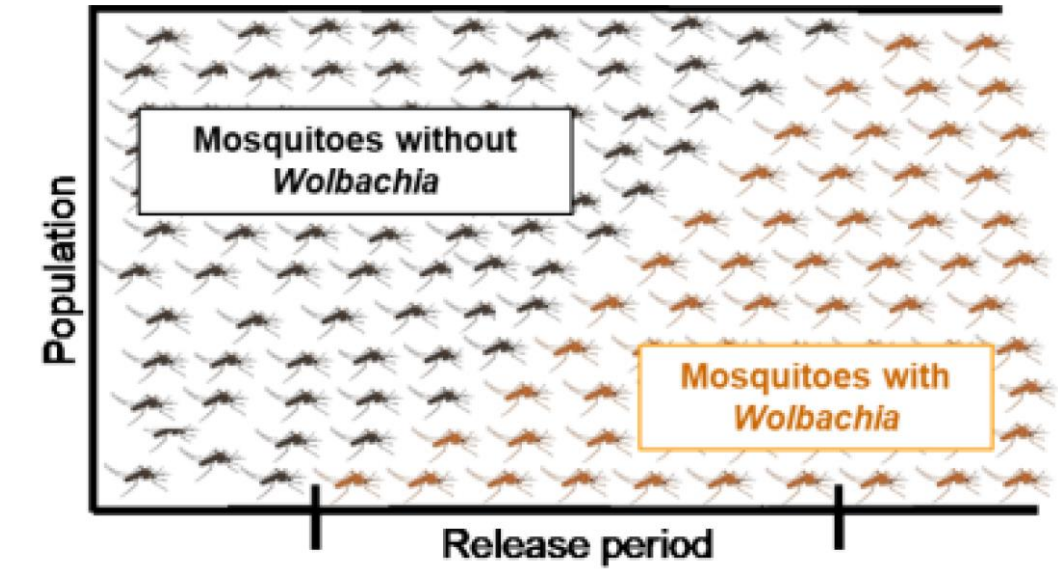


Wolbachia pipientis

Pop. Replacement

- Increased and self-sustained *Wolbachia* frequencies
- Ideally n.s. fitness/performance effects
- WMPB*-based decreased transmission risk
  - **Wolbachia* mediated pathogen blocking
    - DENV
    - ZKV, CHKV

*w*AlbB based replacement to tilt the scale?!

Trials - Malaysia

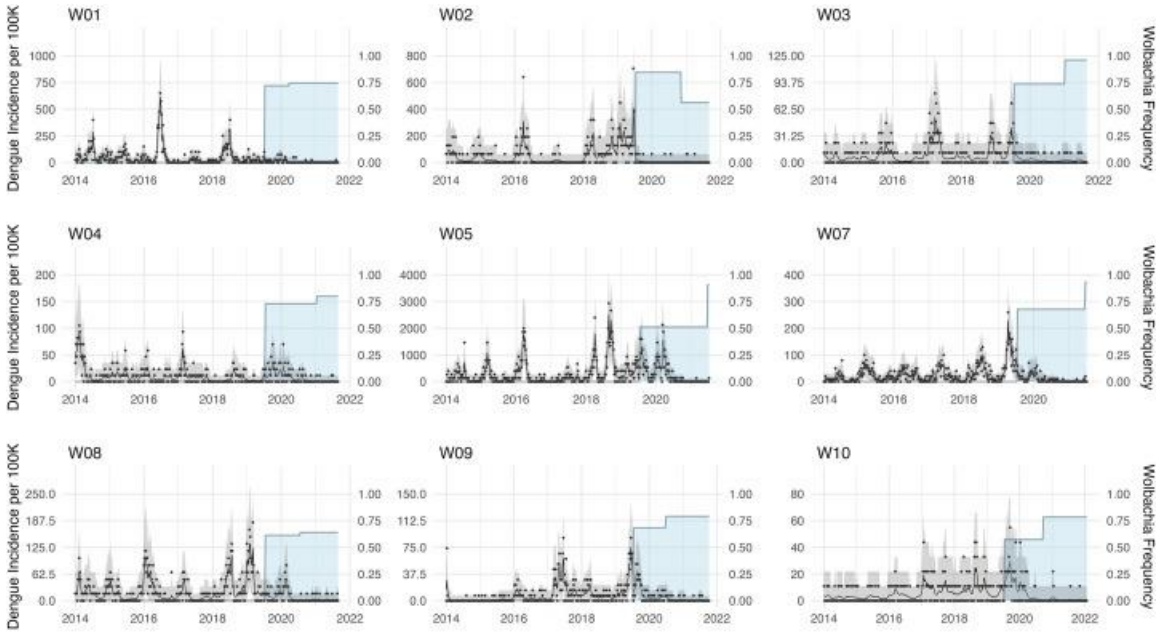


Figure 3. Incidence of dengue in release sites, 9/20
Data are black dots, the posterior mean predicted incidence is represented by the black lines, with the 95% predictive interval in gray. The period of *Wolbachia* release and mean frequency of *Wolbachia* are also provided (blue region; first level is the release period, second is after releases finished).

Hoffmann AA, Ahmad NW, Keong WM, Ling CY, Ahmad NA, Golding N, Tierney N, Jelip J, Putit PW, Mokhtar N, et al. 2024. Introduction of Aedes aegypti mosquitoes carrying wAlbB Wolbachia sharply decreases dengue incidence in disease hotspots. iScience. 27(2):108942.

Vs. other strains (fertility)

Donor *sp.* ->

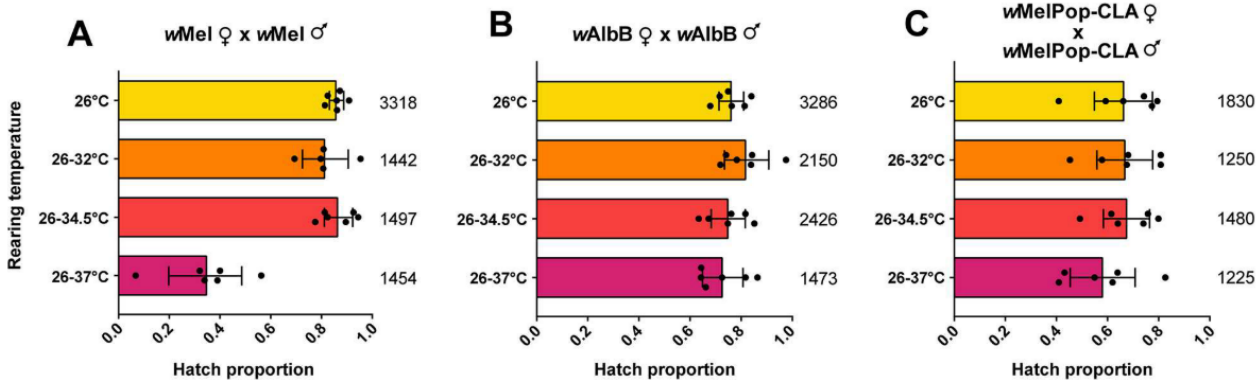
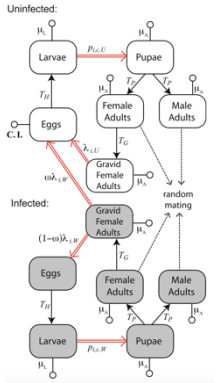


Fig 1. Proportion of eggs hatched from *Wolbachia*-infected *Ae. aegypti* reared at cyclical temperatures. *Wolbachia*-infected females were crossed to *Wolbachia*-infected males reared at cyclical temperatures for the (A) *w*Mel, (B) *w*AlbB and (C) *w*MelPop-CLA infections. Both sexes were reared under the same temperature regime and then crossed together at 26˚C. Each data point shows the proportion of eggs hatched from a cage of 7 females and 7 males (n = 6 replicates per cross). Numbers for each bar denote the total number of eggs scored per cross. Error bars show 95% confidence intervals.

Ross PA, Wiwatanaratanabutr I, Axford JK, White VL, Endersby-Harshman NM, Hoffmann AA. 2017. Wolbachia Infections in Aedes aegypti Differ Markedly in Their Response to Cyclical Heat Stress. PLoS Pathog. 13(1):1–17.

Ecological Dynamics of *Aedes aegypti-Wolbachia pipientis wAlbB* Strain Interactions



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Aim 2	To assess the population level effects of *w*AlbB and the stability of *w*AlbB in semi-field conditions – multiple generation approach
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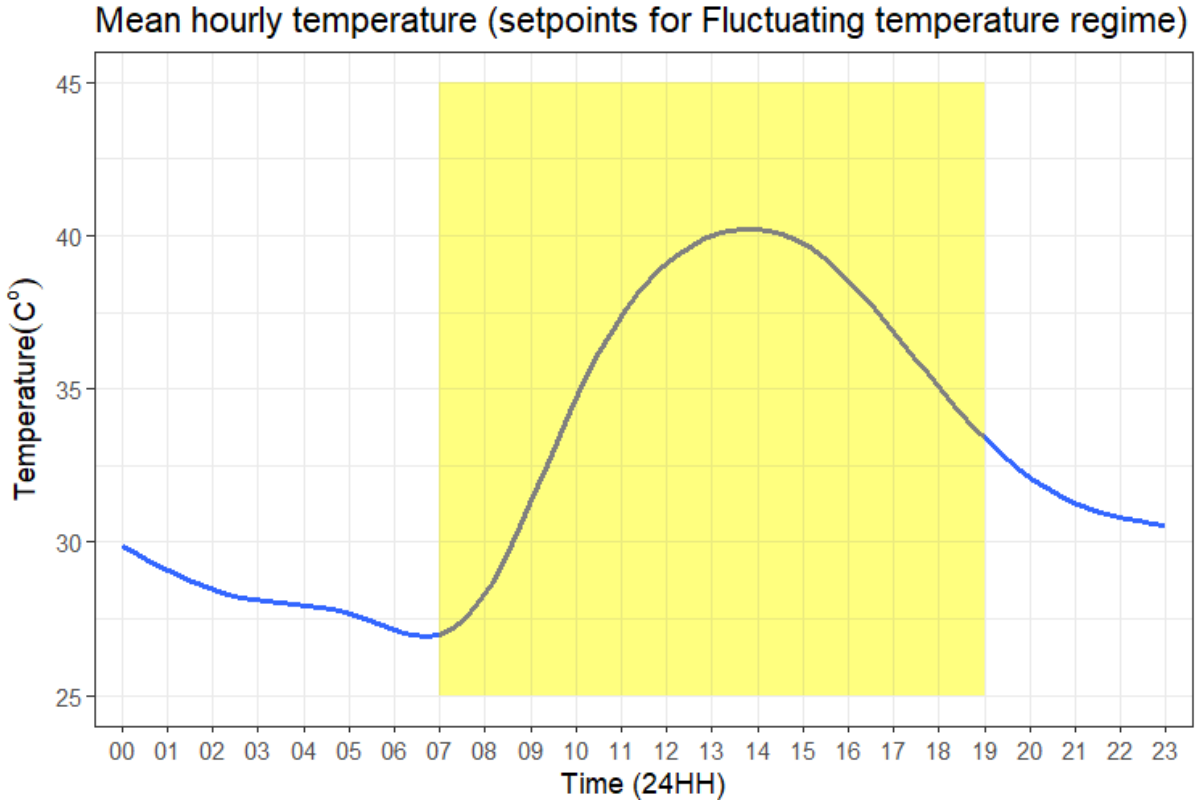
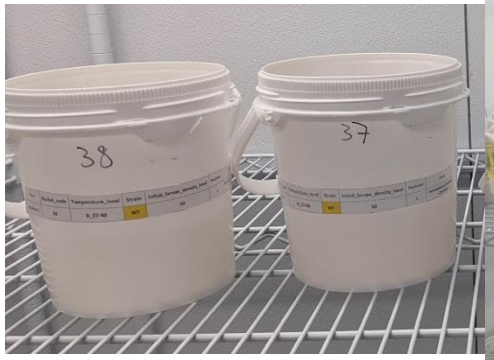
Experimental Design

Temperature regime	*Aedes aegypti* line	Initial larvae density
Constant temperature (27° C)	wMID	Low - 50 larvae High - 500 larvae
	MID	Low High
Fluctuating temperature (Daily fluctuation, 27 - 40° C)	wMID	Low High
	MID	Low High

4 buckets constituted a complete treatment, N=32 buckets



- 2-liter plastic buckets, 1 liter of RO water
- 0.1 gr every 2 days, liquid diet
  - Sifted beef liver powder (ϕ<120 micron)
- Relative Humidity 60%
- 12:12 light dark cycle

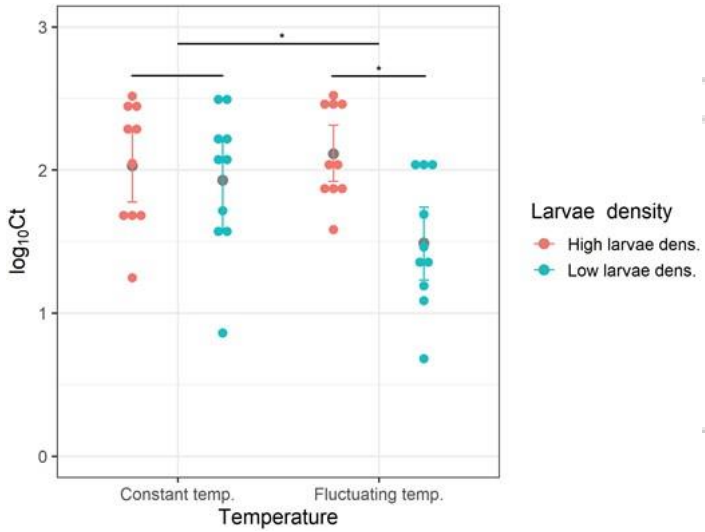


Stability of *w*AlbB and CI offset

Rel. [*w*AlbB] in females

Post-emergence *w*AlbB relative density.

n=60.

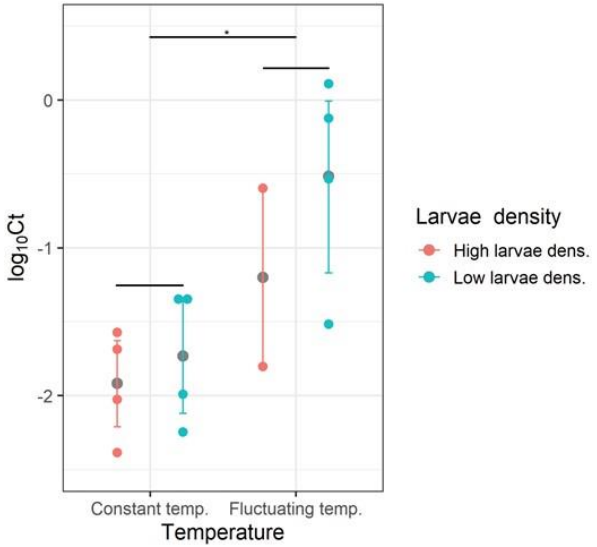


♀ Rel [wAlbB]	A) Complete Set			B) Constant temperature			C) Fluctuating temperature		
Predictors	Estimate	$CI_{95\%}$	p value	Estimate	$CI_{95\%}$	p value	Estimate	$CI_{95\%}$	p value
(Intercept)	1.93	1.66 - 2.20	<0.001*	1.93	1.64 - 2.22	<0.001*	1.49	1.24 - 1.73	<0.001*
Temperature	-0.44	-0.82 - -0.06	0.024*	-	-	-	-	-	-
Density	0.10	-0.28 - 0.48	0.60	0.10	0.31 - 0.52	0.628	0.62	0.28 - 0.97	<0.001*
Temp. * Dens.	0.52	-0.02 - 1.06	0.058	-	-	-	-	-	-
Observations		40			20			20	
AIC		52.80			30.57			23.64	
R^2		0.253			0.013			0.408	

Rel. [*w*AlbB] in progeny

Samples from egg batches laid by experimental females.

10 eggs/pool, n=14 pools.



Egg batches Rel [wAlbB]	A) Complete Set		
Predictors	Estimate	$CI_{95\%}$	p value
(Intercept)	-1.73	-2.3 - -1.17	<0.001*
Temperature	1.22	0.42 - 2.01	0.003*
Density	-0.18	-0.98 - 0.61	0.65
Temp.* Dens.	-0.5	-1.76 - 0.76	0.438
Observations		14	
R^2		0.587	
AIC		29.554	

Cytoplasmic incompatibility

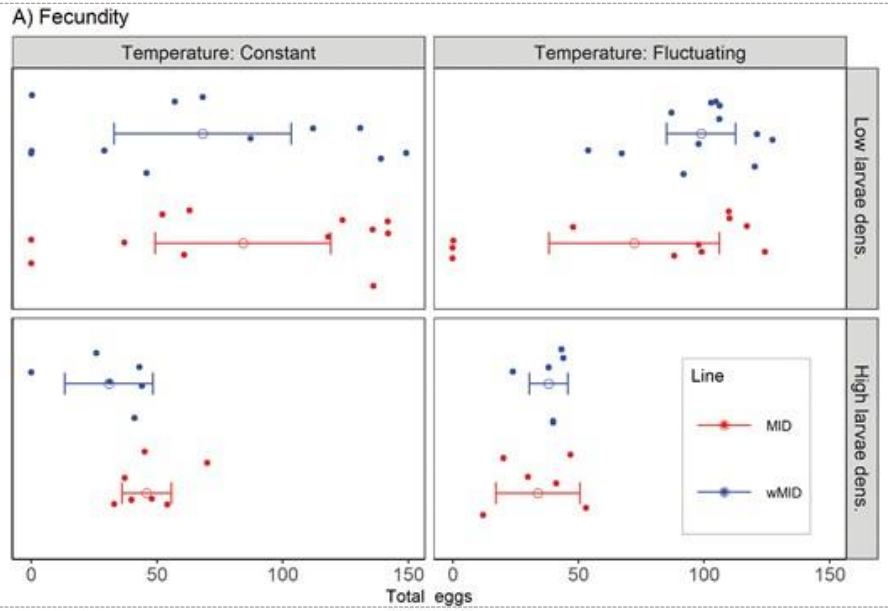
All hatch rates were equal to 0

Mosquito fitness I

Fecundity

Eggs/female

n=73



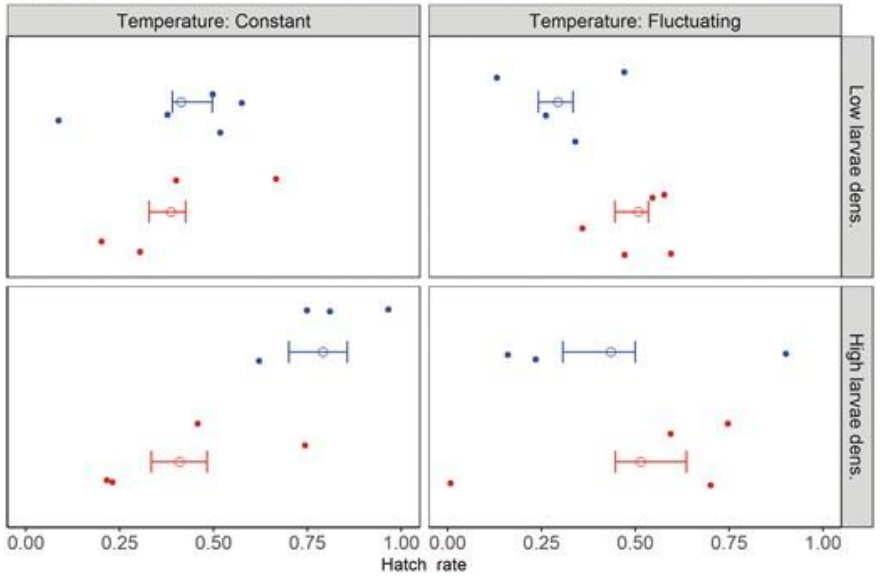
Fecundity	Aj Complete set			Bj Constant temperature			Cj Fluctuating temperature		
Predictors	IRR	CI95%	p value	IRR	CI95%	p value	IRR	CI95%	p value
(Intercept)	84.25	79.16 - 89.55	<0.001*	84.25	79.16 - 89.55	<0.001*	72.18	67.39 - 77.32	<0.001*
Temperature	0.99	0.78 - 0.94	0.001*	-	-	-	-	-	-
Density	0.54	0.48 - 0.61	<0.001*	0.54	0.48 - 0.61	<0.001*	0.47	0.40 - 0.55	<0.001*
Line	0.81	0.74 - 0.89	<0.001*	0.81	0.74 - 0.89	<0.001*	1.37	1.25 - 1.50	<0.001*
Temp. * Dens.	0.99	0.71 - 1.05	0.132	-	-	-	-	-	-
Temp. * Line	1.69	1.49 - 1.93	<0.001*	-	-	-	-	-	-
Dens. * Line	0.83	0.69 - 1.01	0.060	0.83	0.69 - 1.01	0.060	0.82	0.67 - 1.02	0.070
Temp. * Dens. * Line	0.99	0.74 - 1.33	0.955	-	-	-	-	-	-
Observations		73			36			35	
R² Nagelkerke		1.000			0.999			1.000	
AIC		2344.815			1468.137			853.678	

IRR = Incidence rate ratio

Fertility

Hatch rate/batch

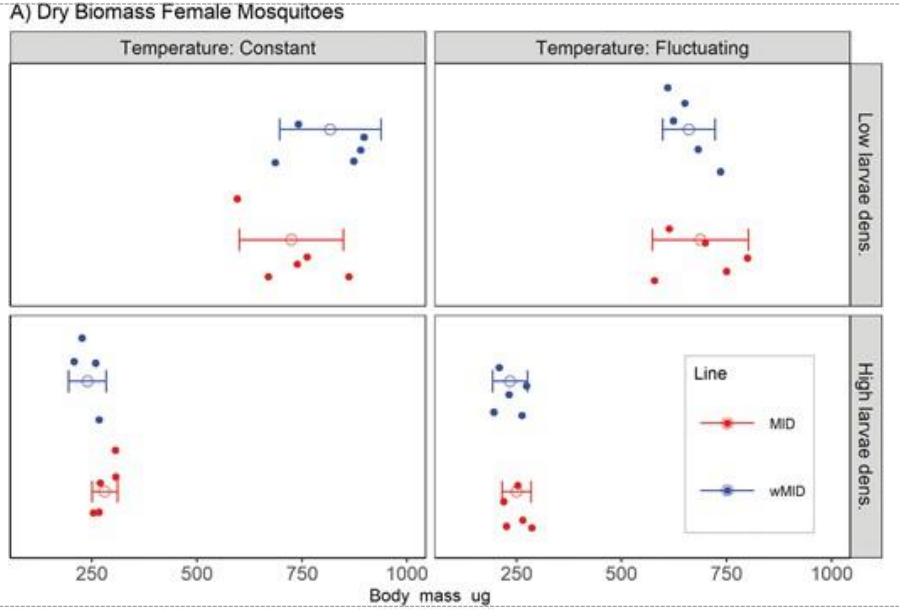
n=33

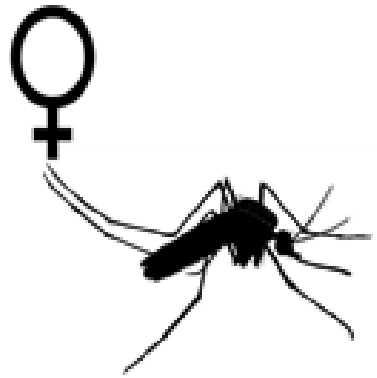


Mosquito fitness II

Female biomass

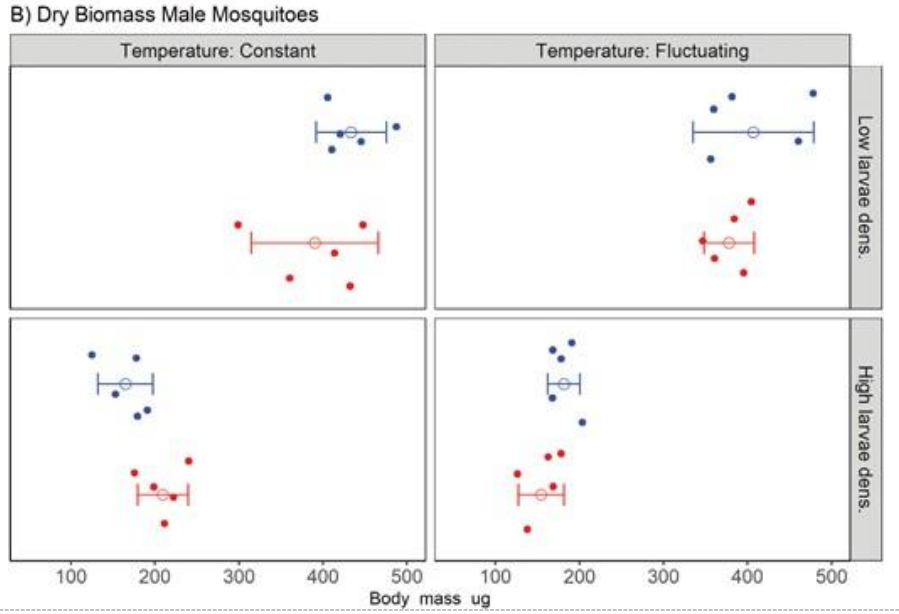
n=40

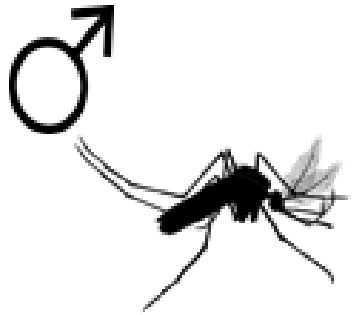




Male biomass

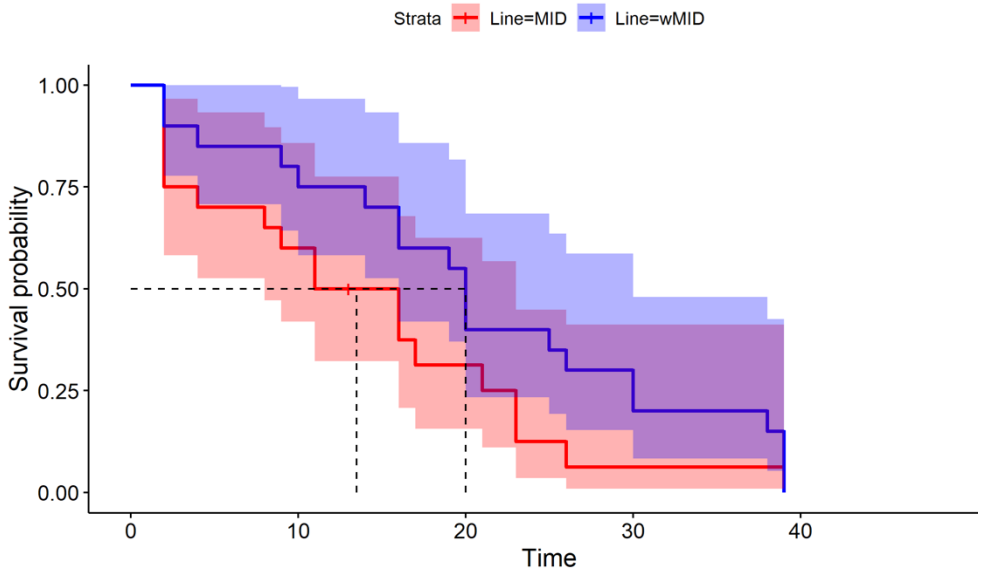
n=40





Mosquito performance I

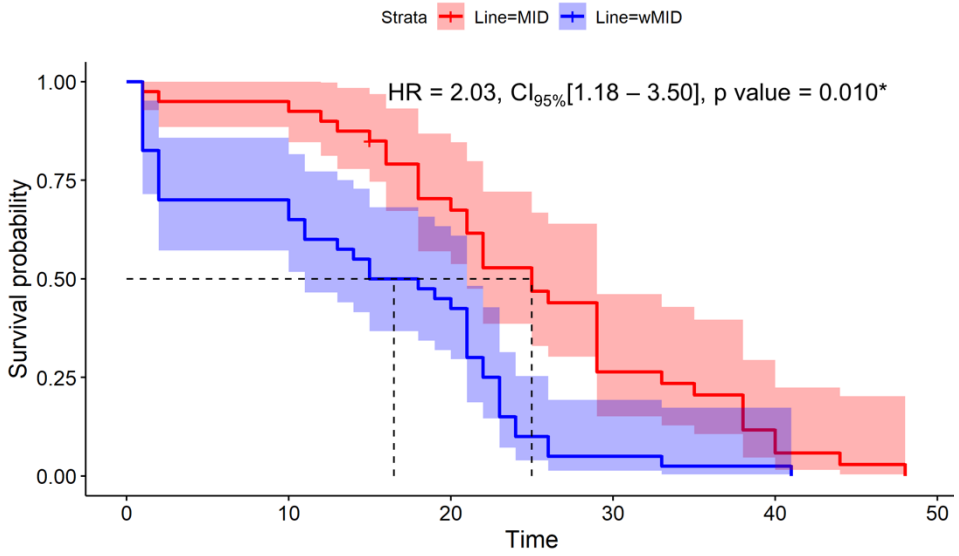
Female adult survival



♀ Survival	A) Constant temperature			B) Fluctuating temperature		
Predictors	HR	$CI_{95\%}$	p value	HR	$CI_{95\%}$	p value
Density	7.24	2.24 - 23.41	<0.001*	0.68	0.24 - 1.89	0.459
Line	0.66	0.26 - 1.68	0.381	0.29	0.10 - 0.80	0.017*
Dens. * Line	0.71	0.18 - 2.74	0.616	4.90	1.01 - 23.68	0.048*

HR=Hazard ratio

Male adult survival



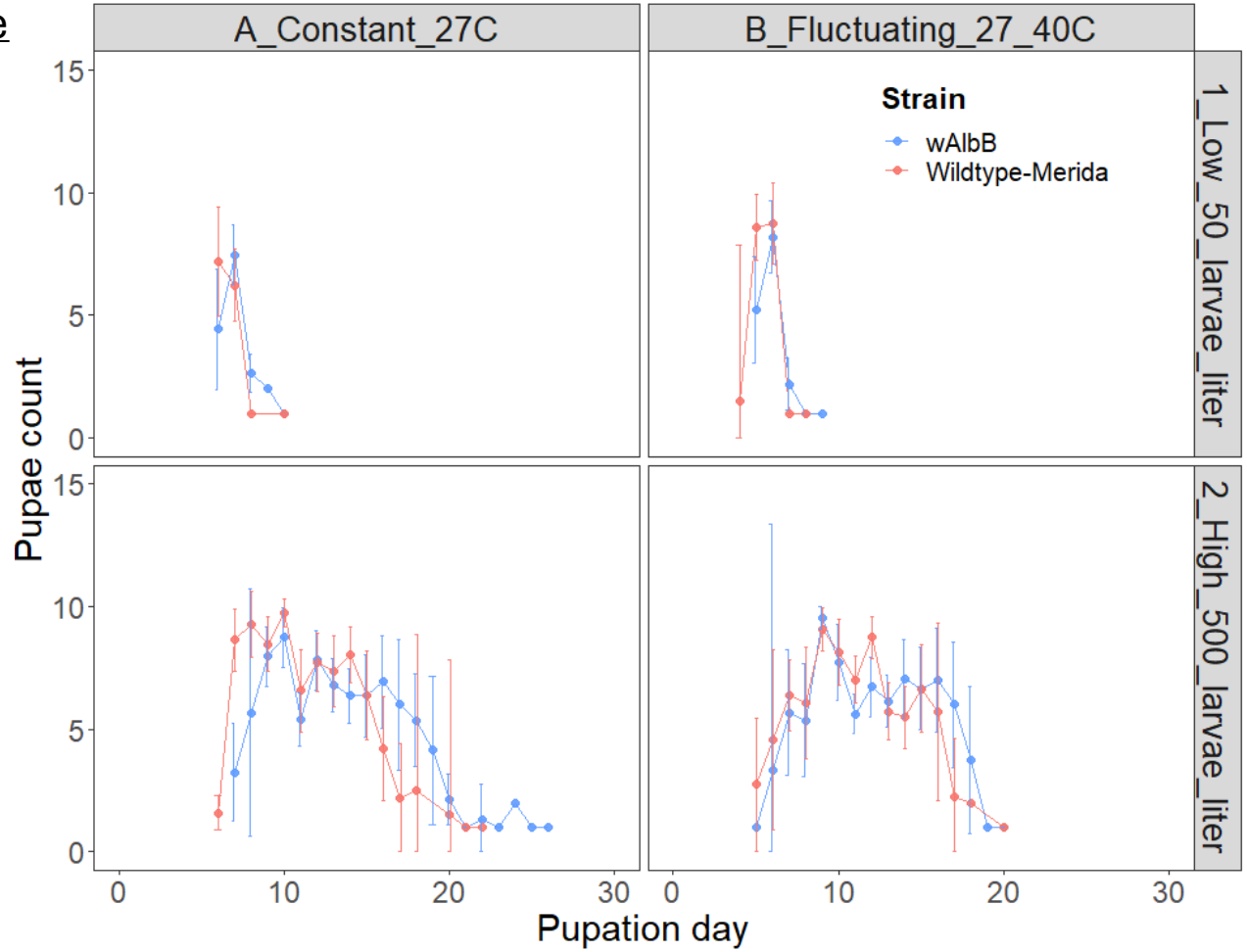
♂ Survival	A) Constant temperature			B) Fluctuating temperature		
Predictors	HR	$CI_{95\%}$	p value	HR	$CI_{95\%}$	p value
Density	2.24	0.84 - 6.00	0.108	0.73	0.32 - 1.68	0.461
Line	1.57	0.63 - 3.90	0.328	2.03	1.18 - 3.50	0.010*
Dens. * Line	2.48	0.65 - 9.42	0.182	2.83	0.93 - 8.60	0.068

HR=Hazard ratio

Mosquito performance II

Development time
(time to pupae)

mean pupae counts/treatment/line/day



Pupation time	A) Complete Set			B) Constant Temperature			B) Fluctuating Temperature		
Predictors	HR	$CI_{95\%}$	p value	HR	$CI_{95\%}$	p value	HR	$CI_{95\%}$	p value
Temperature	5.92	4.86 – 7.21	<0.001*	-	-	-	-	-	-
Density	0.03	0.03 – 0.04	<0.001*	0.03	0.02 – 0.03	<0.001*	0.01	0.01 – 0.01	<0.001*
Line	0.68	0.54 – 0.81	<0.001*	0.61	0.49 – 0.74	<0.001*	0.50	0.42 – 0.61	<0.001*
Temp. * Dens.	0.18	0.15 – 0.22	<0.001*	-	-	-	-	-	-
Temp. * Line	0.69	0.52 – 0.91	0.006*	-	-	-	-	-	-
Dens. * Line	0.92	0.74 – 1.14	0.431	1.01	0.82 – 1.26	0.899	1.61	1.32 – 1.97	<0.001*
Temp. * Dens. * Line	1.94	1.45 – 2.59	<0.001*	-	-	-	-	-	-

HR =Hazard ratios

Other metrics

- Proportion of pupae to emerge as adults
- Adult sex ratios

Conclusions

- Differential effects of assessed treatments (*Wolbachia* infection status, temperature, and larval density) across mosquito sexes and life stages.

- Realistic conditions may not impact dramatically the stability of *w*AlbB infection in *A. aegypti*.

- Understanding the ecological consequence of *A. aegypti-wAlbB* interaction is complex
  - Life history tradeoffs under conditions faced by natural populations

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JOURNAL ARTICLE

Wolbachia pipientis (Rickettsiales: Rickettsiaceae) mediated effects on the fitness and performance of *Aedes aegypti* (Diptera: Culicidae) under variable temperatures and initial larval densities

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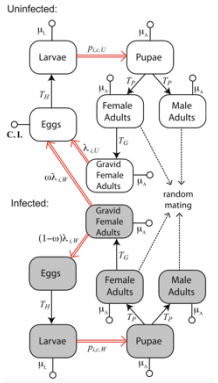

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Methods

Line / Treatment	Low dens. house	High dens. house
WMID	1 pop	1 pop
MID	1 pop	1 pop

Questions

- How do population **dynamics** compare between infected and uninfected lines?
- What happens to ***w*AlbB** at the end of the experiment?

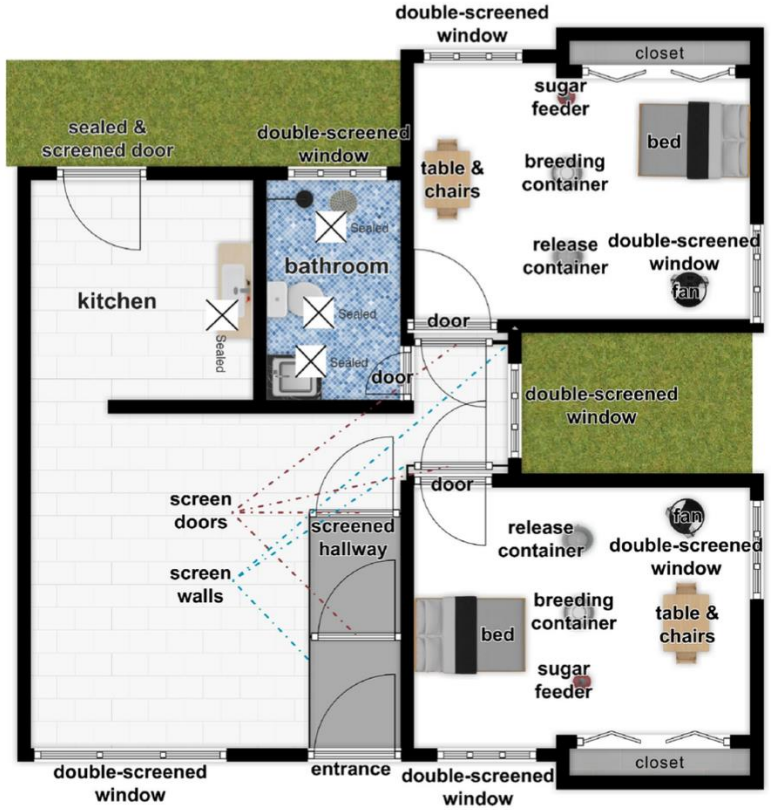
N_0:

- Low; 5♀, 5♂, 50 eggs
- High; 30♀, 30♂, 150eggs

Bloodfeeding:

- 3 days/week
- 30 minutes per day

A)



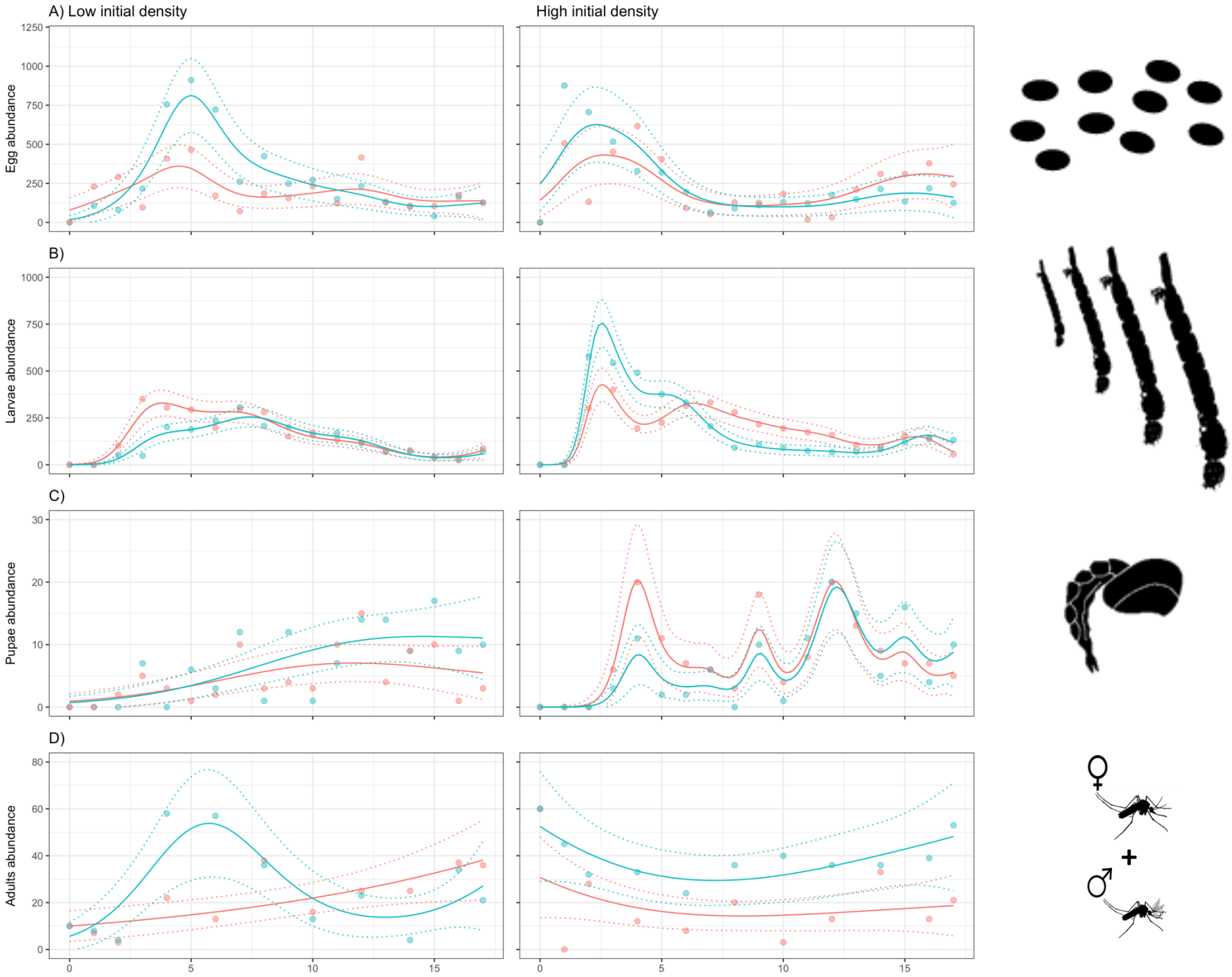
B)

C)

D)

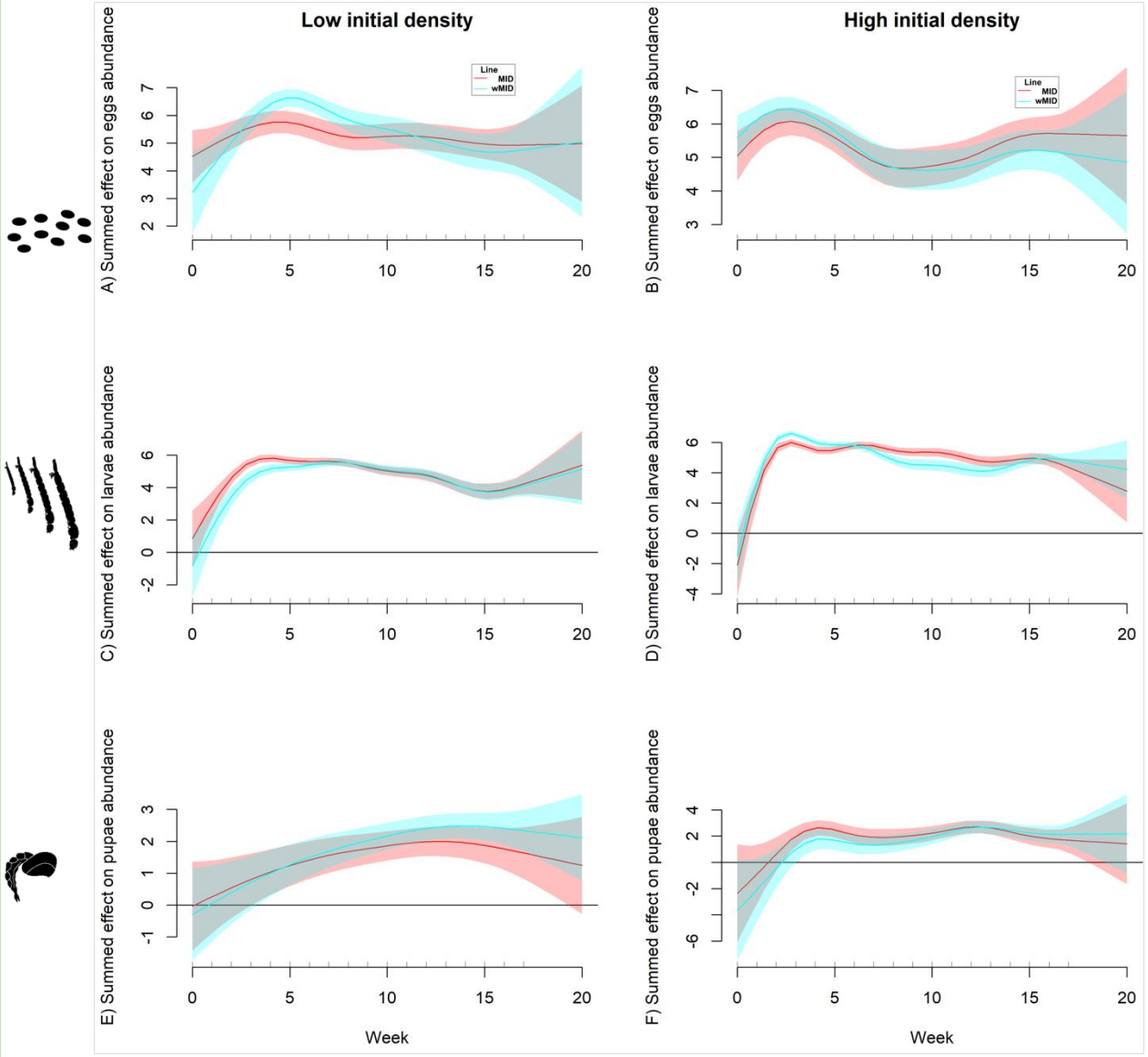
E)

Abundances

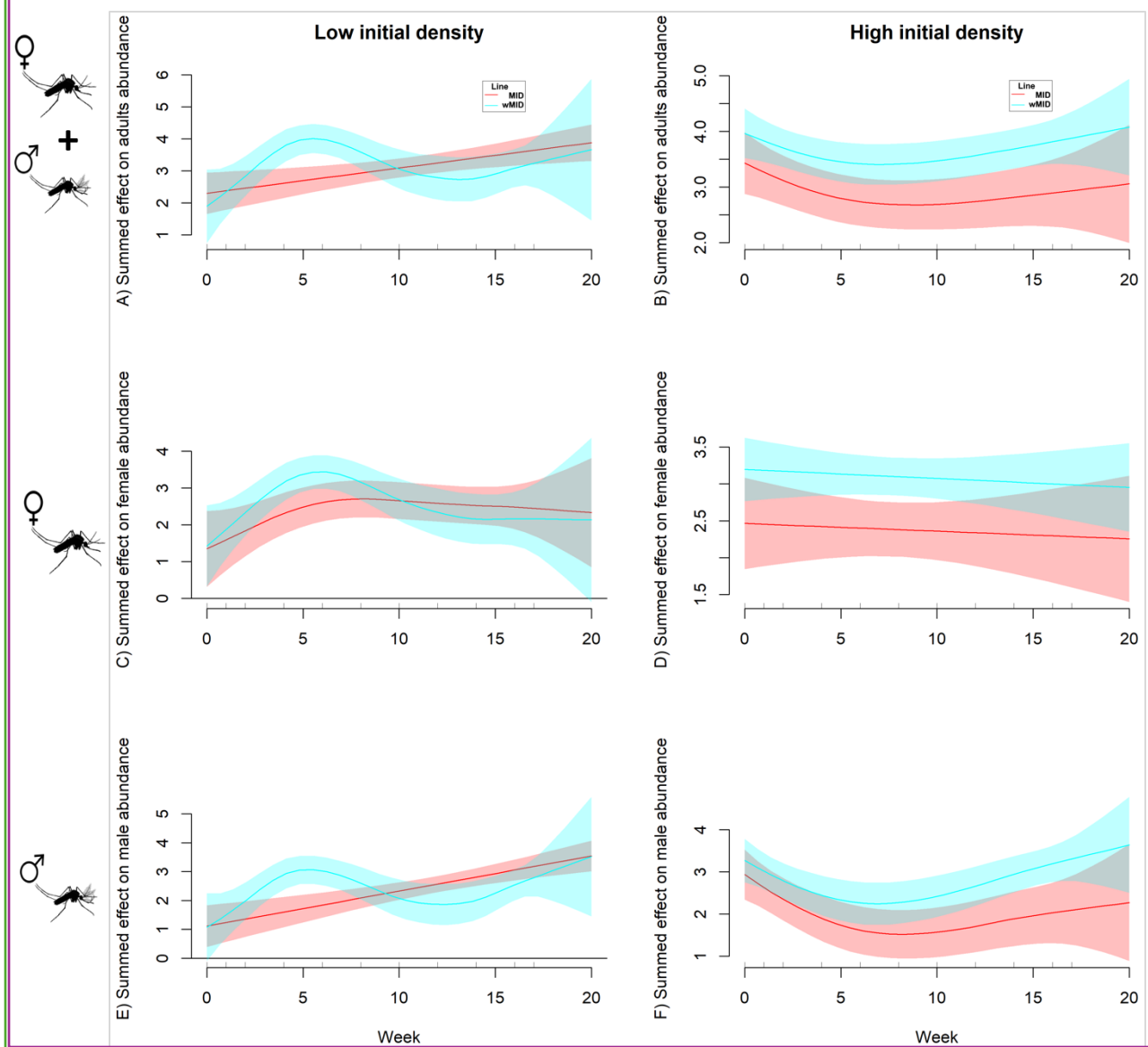

A) Low initial density
High initial density
Egg abundance
B)
Larvae abundance
C)
Pupae abundance
D)
Adults abundance
0
250
500
750
1000
1250
10
20
30
40
60
80
5
15

Abundances - Effect plots

Immature forms

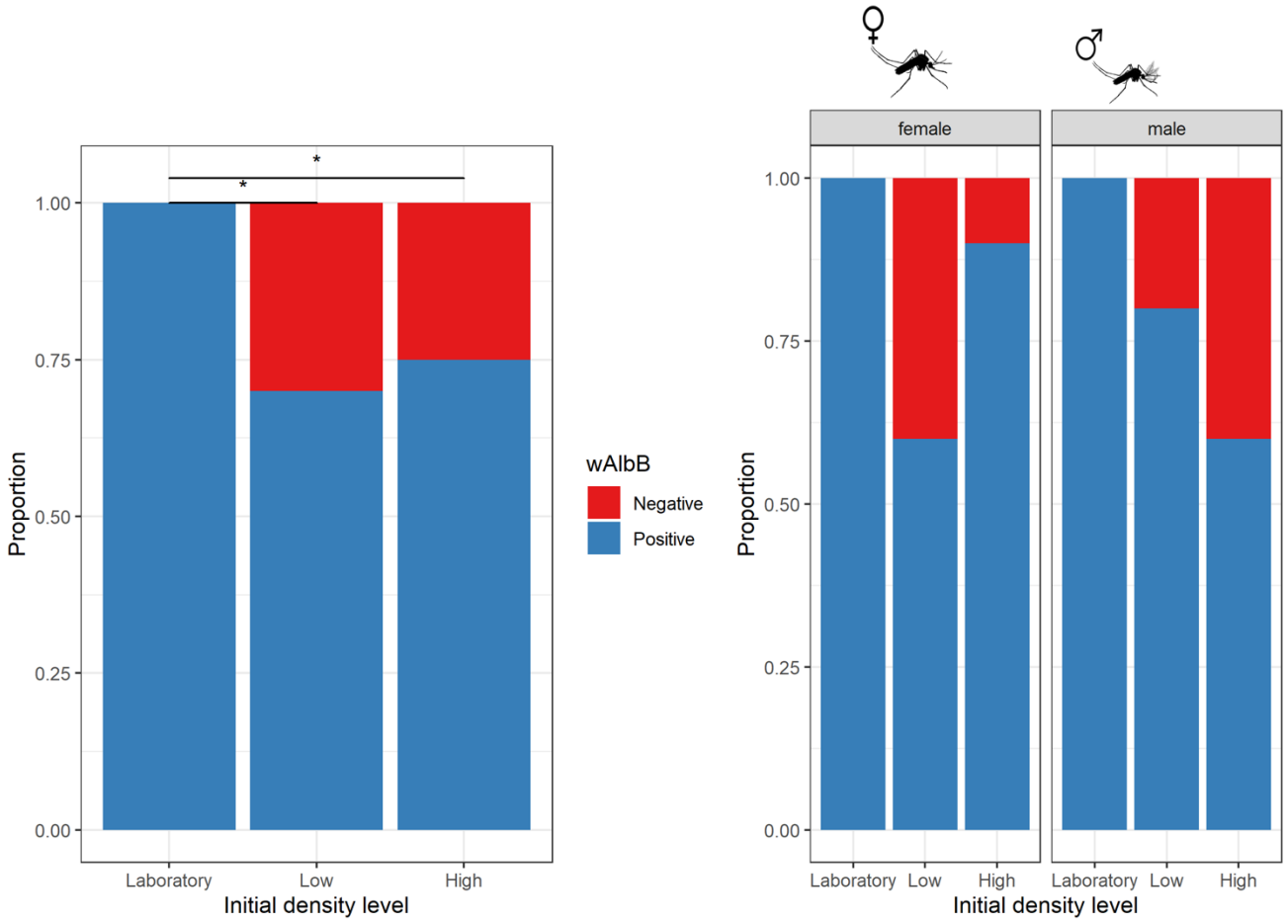


Adults

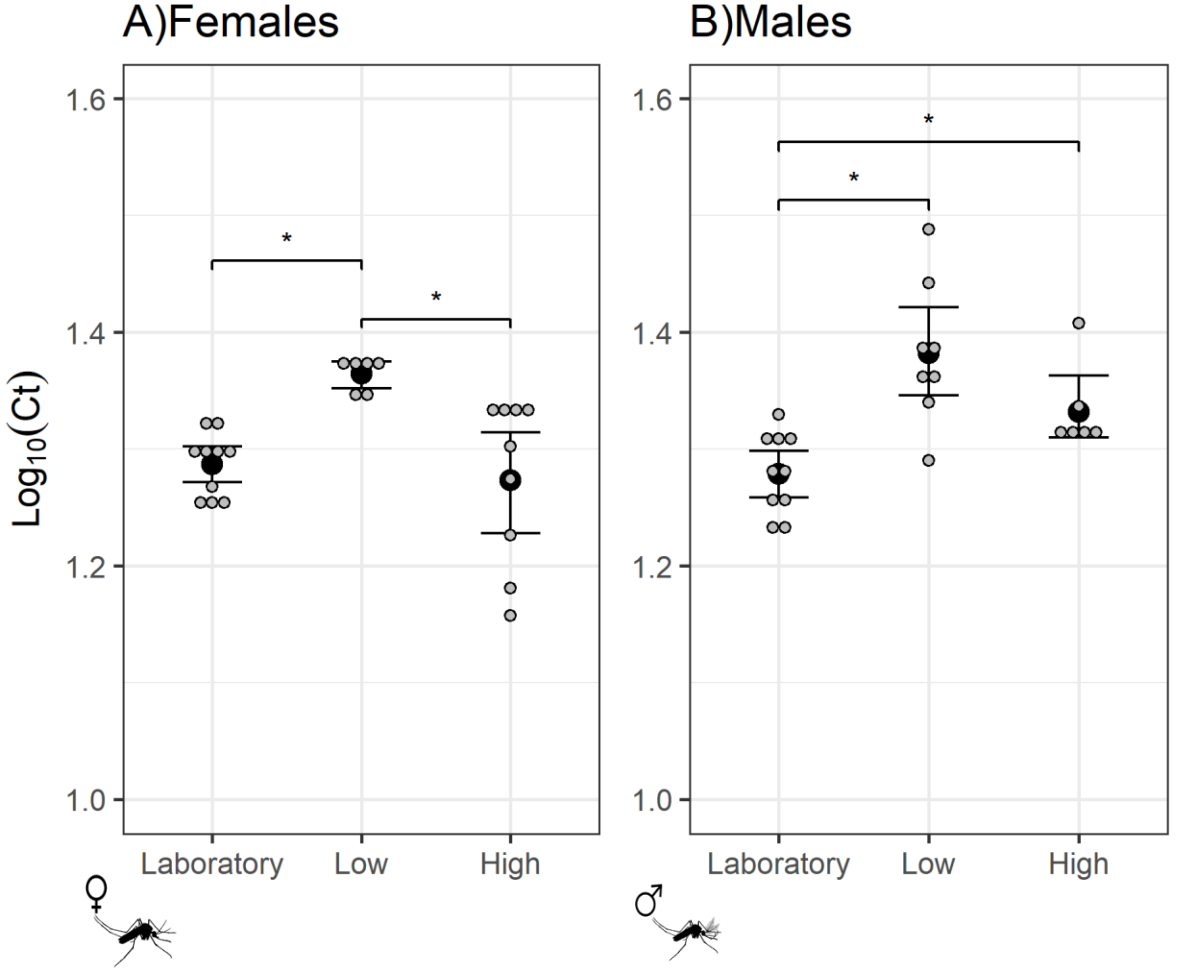


*w*AlbB stability in adults

*w*AlbB Final Frequency



*w*AlbB Relative density



Conclusions

Population dynamics

Transient effects on abundances

- Potential self compensatory effects leading to convergence in abundances and life-stage distributions

***w*AlbB persistence in adults**

Decreased frequencies

- Potential for intergenerational loss
- Monitoring WPR programs -> priority

Equal or increased relative densities

- Might aid in maintaining WMPB* and CI (high fidelity mech.)
  - Despite suspected decreased MT

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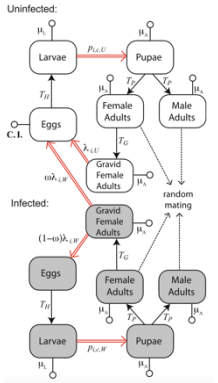

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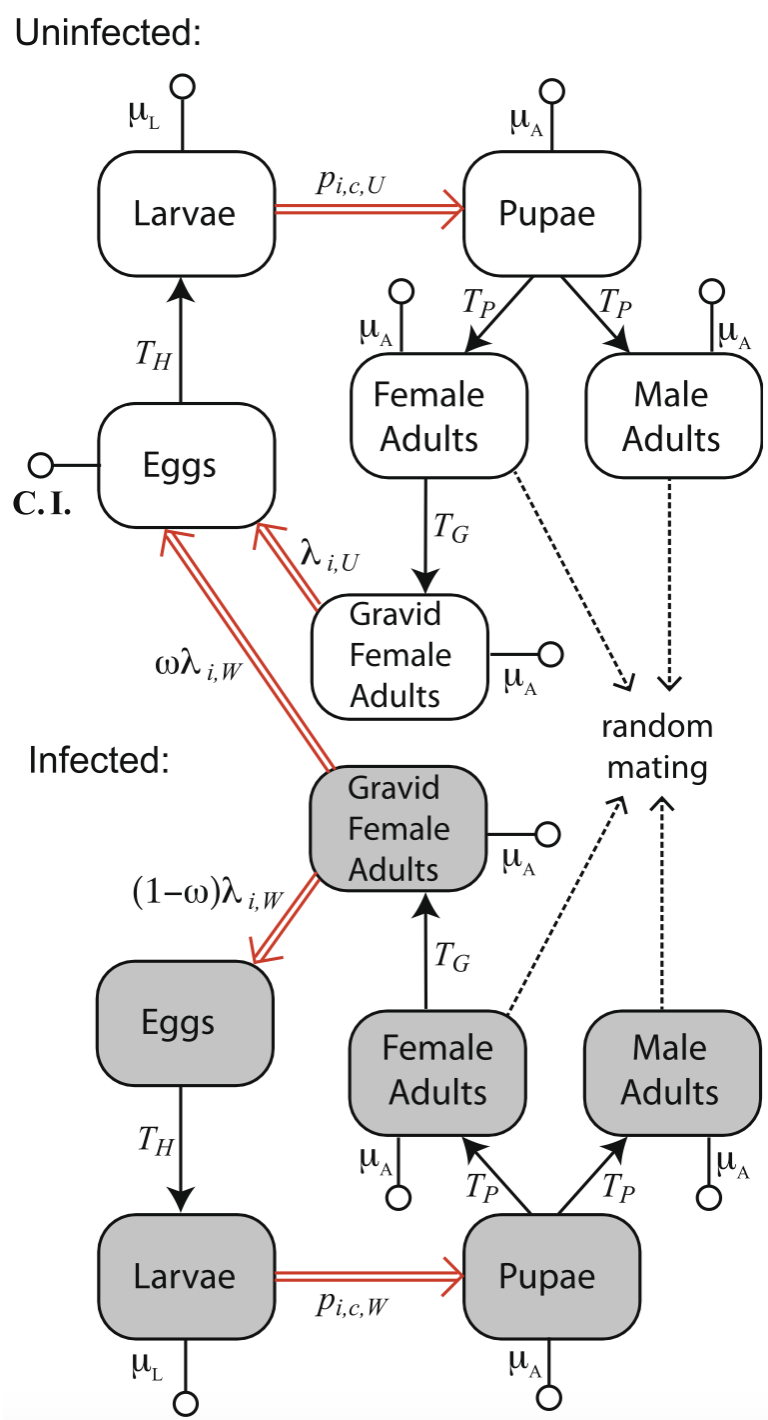
Aim 2	To assess the population level effects of *w*AlbB and the stability of *w*AlbB in semi-field conditions – multiple generation approach
Aim 3	To assess the long-term consequence of observed *w*AlbB-mediated effects on its invasion dynamics withing WPR through modelling - integration through established model

Methods

- Density dependent demographic traits
  - *w*Mel model -> Hancock et al 2016
- Adapting to *w*AlbB
  - CI – higher fidelity values
  - **Further delayed larvae development**
  - MT - increased loss
- Implications over
  - Release ratios and release schemes
- Optimization within WPR entomological goals -> *w*AlbB invasion



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