Effects of larval rearing temperature on immature development and West Nile virus vector competence of *Culex tarsalis*

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**Abstract**

**Background:** Temperature is known to induce changes in mosquito physiology, development, ecology, and in some species, vector competence for arboviruses. Since colonized mosquitoes are reared under laboratory conditions that can be significantly different from their field counterparts, laboratory vector competence experiments may not accurately reflect natural vector-virus interactions.

**Methods:** We evaluated the effects of larval rearing temperature on immature development parameters and vector competence of two *Culex tarsalis* strains for West Nile virus (WNV).

**Results:** Rearing temperature had a significant effect on mosquito developmental parameters, including shorter time to pupation and emergence and smaller female body size as temperature increased. However, infection, dissemination, and transmission rates for WNV at 5, 7, and 14 days post infectious feeding were not consistently affected.

**Conclusions:** These results suggest that varying constant larval rearing temperature does not significantly affect laboratory estimates of vector competence for WNV in *Culex tarsalis* mosquitoes.

**Keywords:** Mosquito, West Nile virus, Global climate change, Transmission, Development

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**Background**

Temperature is a key factor in vector-borne pathogen transmission. It can significantly affect insect behavior and physiology [1,2] including the extrinsic incubation period of vector-borne pathogens [3]. Studies have measured the effects of temperature on the aquatic immature stages of mosquitoes and other insects [4-6]. Fluctuating temperatures can be experienced by immature mosquitoes residing in containers, irrigation pools and similar ephemeral habitats [7]. In many cases, extremely high temperatures can cause rapid mortality, but moderately high temperatures can also cause thermal wounding at immature stages that affect future adult stages [1]. Low temperatures pose a different set of challenges than high temperatures, invoking complex changes to cell integrity, tissue morphogenesis, reproduction and sex ratio in a variety of insects [8].

*Culex tarsalis* is a primary vector of West Nile virus (WNV), St Louis encephalitis virus (SLEV), and Western equine encephalitis virus (WEEV) in the western United States [9-12]. California field populations have been found persisting in water temperatures as low as 5°C in winter duck ponds or mid-day upper thermal limits of >40°C [13,14]. These conditions are markedly different from those utilized in laboratory settings for colony rearing and vector competence experiments. Adults reared in the laboratory are significantly larger than those collected in the field [15] and lab mosquitoes maintained on sucrose produce and store significantly more glycogen and lipids than their counterparts in the field [16]. Colonization can lead to reduced genotype and phenotype variation [17]. Together, these studies suggest that vector competence experiments with laboratory mosquitoes may not accurately reflect natural vector-virus interactions.

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Several studies have shown that the influence of extrinsic factors on immature stages extends to the adult stage, possibly affecting susceptibility to pathogens in a species-specific manner. Changes in larval water quality affected *Cx. tarsalis* development parameters including body size, but did not affect vector competence for WEEV or SLEV [18]. Similarly, changes in larval rearing temperature or diet did not alter susceptibility of *Culex spp.* to Rift Valley fever virus (RVFV) or Murray Valley encephalitis virus (MVEV) [19,20]. By contrast, *Aedes* adults resulting from low rearing temperatures were found to be more susceptible to infection with RVFV, Venezuelan equine encephalitis virus (VEEV) and Chikungunya virus (CHIKV) [5,21]. Others have singly investigated various environmental factors in *Aedes* mosquitoes such as nutrition deprivation, intra- and interspecific competition, or malathion exposure and found increased susceptibility to La Crosse (LACV), dengue (DENV) and Sindbis (SINV) viruses [4,22,23]. Correlations between small adult *Aedes* females and increased vector competence for various arboviruses have also been examined [24,25].

Our previous work with *Cx. tarsalis* demonstrated that decreased larval nutrition impacted several development variables including body size, but did not result in consistent changes in adult vector competence for WNV [26]. Immature development, adult size and survivorship have been shown to vary seasonally in field populations of *Cx. tarsalis* [14]. In addition, incidence of WNV, as well as SLEV and WEEV is highly variable seasonally [27-29]. In this study, we investigated the relationship between larval rearing temperatures and vector competence of adult *Cx. tarsalis* for WNV. We also studied impacts of temperature on various development parameters and body size for *Cx. tarsalis*.

**Methods**

**Mosquitoes, cells and virus**

To control for variation among mosquito genotypes and to account for vector competence changes that possibly occur during the colonization process, two independent *Culex tarsalis* strains were used for experiments. All mosquitoes were reared at the Wadsworth Center Arbovirus Laboratory. Colonies were originally established from field mosquitoes collected in Coachella Valley, CA (“CV”) and Yolo County, CA (“YC”) in 2003 and 2009, respectively, by WK Reisen. Colony larvae, pupae and adult mosquitoes were reared and maintained under the following controlled conditions: **27°C ± 1°C**, 16:8 h light:dark diurnal cycle with approximately 45% relative humidity. Newly emerged adults were removed to **4.3 L cartons** and held at **27°C** until blood feeding. The number of pupae, males, and females were recorded six days per week. Adult mosquito wings were measured from the alular notch to the distal margin, excluding the fringe, using Axiovision software and a Zeiss microscope. Time to pupation, time to adulthood, percent pupation, percent emergence and wing length were compared using chi-squared and t-tests.

**Vector competence**

Five to ten-day old female mosquitoes were starved of sugar for four hours prior to blood feeding. Stock WN02-1956 was added to 5 mL defibrinated bovine blood (Hema-Resource & Supply, Aurora, OR) with 2.5% sucrose solution to average titers of 3.4E+08 PFU/ml (Table 1). Mosquitoes were fed an infectious blood meal via a Hemotek membrane feeding system (Discovery Workshops, Accrington, UK) for approximately one hour according to the manufacturer’s specifications. After feeding, fully engorged females were separated from the unfed under CO₂ anesthesia on a FlyPad (Flystuff, San Diego, CA) and placed in 4.3 L cardboard.
containers. Blood fed adults were provided with 10% sucrose ad lib and held for up to 14 days after feeding at 27°C, 16:8 light:dark photoperiod in a growth chamber. *In vitro* transmission assays were performed as previously described [26] with modifications at 5, 7 and 14 days post feeding. Female mosquitoes were anesthetized with triethylamine (Sigma, St. Louis, MO), legs were removed and placed in 1 mL mosquito diluent (MD: 20% heat-inactivated fetal bovine serum (FBS) in Dulbecco’s phosphate-buffered saline, 50 ug/mL penicillin/streptomycin, 50 ug/mL gentamicin and 2.5 ug/mL fungizone) per mosquito. For each mosquito, the proboscis was placed in a capillary tube containing 10 uL of a 1:1 solution of 50% sucrose and FBS. After 30 min, the contents were expelled into 0.3 mL MD. One wing per mosquito was removed and retained for measurement for use as an indication of body size. Bodies were placed individually into 1 mL MD. Mosquito body, legs and salivary secretion samples were stored at -70°C until tested for virus. Mosquito bodies and legs were homogenized utilizing a mixer mill at 24 cycles/second and clarified by centrifugation for one minute. Mosquito bodies, legs and salivary secretions were tested for WNV infectious particles via plaque assay on Vero cell culture [30]. Infection was defined as the proportion of mosquitoes with WNV positive bodies. Dissemination and transmission were defined as the proportion of infected mosquitoes with positive legs and salivary secretions, respectively. Proportions were compared using Fisher’s exact test.

**Results**

**Mosquito development**

To examine the consequences of various larval rearing temperatures on *Cx. tarsalis*, development time, numbers of immature and adult mosquitoes, and adult wing length were evaluated (Table 2). Generally, mosquitoes reared at 19°C took the longest time to pupate and emerge, followed by 25°C and 31°C, (Table 2). Unsurprisingly, temperature affected pupation and emergence rates. In general, larvae reared at 19°C and 25°C were more likely to develop into adults than larvae reared at 31°C. YC mosquitoes used in experiments 3 and 4 were more susceptible to larval and pupal death than CV mosquitoes when reared at 31°C. For all experiments, there was a significant inverse relationship between temperature and wing length, with the largest adults produced at 19°C, and the smallest at 31°C.

**Vector competence**

Effects of rearing temperature on *Cx. tarsalis* vector competence was evaluated by infection, dissemination, and transmission of WNV at 5, 7 and 14 days post feeding (dpf) (Table 3). In experiment 1 (CV mosquitoes), infection and dissemination rates among 19°C treatment group were significantly higher at 7 dpf than the mosquitoes reared at 25°C. In experiment 2 (CV mosquitoes), infection rate of 19°C treatment at 5 dpf was significantly higher than 31°C treatment, and the infection rates at 7 dpf of 19°C and 25°C treatments were both significantly higher than those reared at 31°C. Additionally in experiment 2, 14 dpf transmission rate was significantly lower in the 19°C treatment than those reared at either 25°C or 31°C. In experiment 4 (YC mosquitoes), mosquitoes reared at 25°C had a significantly

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**Table 1 Final WNV (WN02-1956) blood meal titers in experiments (PFU/mL)**

| Temperature | Exp 1 (CV) | Exp 2 (CV) | Exp 3 (YC) | Exp 4 (YC) |
|-------------|------------|------------|------------|------------|
| 19°C        | 5.0E + 08  | 4.7E + 08  | 2.4E + 08  | 1.4E + 08  |
| 25°C        | 5.8E + 08  | 4.1E + 08  | 1.8E + 08  | 1.3E + 08  |
| 31°C        | 3.8E + 08  | 5.1E + 08  | 2.0E + 08  | 1.4E + 08  |

YC Yolo County, CV Coachella Valley.

**Table 2 Effect of rearing temperature on *Cx. tarsalis* development parameters**

| Treatment Temperature | Days to first pupation^1 | Days to first emergence^1 | % pupation^2 | % pupal emergence^2 | % emergence larvae to adult^2 | Adult winglength (mm)^1 |
|-----------------------|--------------------------|---------------------------|--------------|--------------------|------------------------------|-------------------------|
| 19°C                  | 11.6^a                   | 12.5^a                    | 11^a         | 13.6^a             | 13.6^a                      | 2.8^a                   |
| 25°C                  | 7.6^b                   | 8.8^b                    | 7.6^b         | 7^b                | 7^b                          | 3.0^b                   |
| 31°C                  | 7.2^c                   | 7.8^c                    | 6.6^c         | 6.6^c              | 6.6^c                       | 2.8^c                   |

^1 t-test. ^2 χ² - test. Data with different superscripted letters in each development parameter are significantly different within each experiment. YC Yolo County, CV Coachella Valley.
higher infection rate at 14 dpf than at 19°C. However, there were no consistent statistically significant differences in WNV infection, dissemination or transmission rates at any temperature for either mosquito strain (Table 3).

**Discussion**

Investigating effects of environmental conditions on mosquito life cycle parameters is important considering that laboratory rearing protocols are usually not representative of the temperature, nutrition, and density fluctuations immature larval stages experience in the field. Generally, mosquitoes collected from the field are significantly smaller than those utilized in laboratory experiments, and studies have shown various other differences, including adult nutritional reserves [15,16]. Several studies postulate that climate change may alter infectious disease dynamics, which is influenced by numerous vector and host factors [31,32]. The abundance, distribution, and competence of disease vectors are of particular concern because they are easily influenced by climate and have the potential to impact human health through altered pathogen distribution [31]. Most recently, outbreaks of arboviruses including CHIKV, RVFV, dengue and WNV have resurfaced or emerged in areas where they were previously undetected, suggesting a further increase in vector-borne disease [32-35].

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**Table 3 Effect of rearing temperature on adult Cx. tarsalis vector competence for WNV (WN02-1956)**

| Treatment Temperature | Day post feeding | Exp 1 (CV) | Exp 2 (CV) | Exp 3 (YC) | Exp 4 (YC) |
|-----------------------|-----------------|-----------|-----------|-----------|-----------|
|                       |                 | % Infected |           |           |           |
| 19°C                  | 5               | 92(50)    | 83(41)a   | 100(30)   | 100(30)   |
| 25°C                  | 8               | 86(50)    | 80(50)    | 97(30)    | 98(50)    |
| 31°C                  | 7               | 86(50)    | 60(40)b   | -         | 95(20)    |
| 19°C                  | 7               | 94(50)    | 81(36)b   | 87(30)    | 100(35)   |
| 25°C                  | 7               | 76(50)c   | 82(50)c   | 83(30)    | 100(38)   |
| 31°C                  | 8               | 88(50)    | 44(50)d   | 95(20)    | 92(24)    |
| 19°C                  | 14              | 92(50)    | 76(34)    | 89(38)    | 88(33)    |
| 25°C                  | 14              | 86(50)    | 76(50)    | 100(13)   | 100(40)c  |
| 31°C                  | 14              | 82(50)    | 64(50)    | -         | -         |

|               | % Disseminated1 |           |           |           |
|---------------|-----------------|-----------|-----------|-----------|
| 19°C          | 5               | 35        | 59        | 63        | 50        |
| 25°C          | 5               | 37        | 40        | 76        | 63        |
| 31°C          | 5               | 30        | 46        | -         | 47        |
| 19°C          | 7               | 74b       | 90        | 96        | 86        |
| 25°C          | 7               | 53c       | 80        | 80        | 84        |
| 31°C          | 7               | 73        | 73        | 84        | 91        |
| 19°C          | 14              | 98        | 100       | 100       | 100       |
| 25°C          | 14              | 88        | 95        | 92        | 98        |
| 31°C          | 14              | 95        | 97        | -         | -         |

|               | % Transmitted1 |           |           |           |
|---------------|----------------|-----------|-----------|-----------|
| 19°C          | 5               | 4         | 3         | 10        | 7         |
| 25°C          | 5               | 0         | 3         | 10        | 2         |
| 31°C          | 5               | 2         | 4         | -         | 0         |
| 19°C          | 7               | 15        | 7         | 15        | 9         |
| 25°C          | 7               | 8         | 22        | 12        | 16        |
| 31°C          | 7               | 9         | 27        | 21        | 14        |
| 19°C          | 14              | 61        | 27        | 65        | 72        |
| 25°C          | 14              | 51        | 63d       | 54        | 70        |
| 31°C          | 14              | 56        | 78        | -         | -         |

1 Calculated based on number of infected mosquitoes following incubation at 27°C.
Values in parentheses indicate number of mosquitoes tested.
Percentages with different superscripted letters in each infection parameter are significantly different within each experiment.
YC Yolo County, CV Coachella Valley.
Significance tested by Fisher’s exact test.
Seasonal and yearly fluctuations of Cx. tarsalis vector competence for WEEV and SLEV have been correlated with changing temperature in Kern County, California, indicating that climate change could facilitate the northward expansion of these viruses [28,29,31]. Our results indicate that larval rearing temperature had significant consequences on development, including shorter development time and smaller female body size as temperature increased. Similarly, other larval environment studies have revealed that temperature influences many Cx. tarsalis life history parameters, including decreased adult survivorship, adult body size, reproductive effort and immature development time, which was correlated with increasing temperature [14,37]. Additional larval environment conditions have been examined with a variety of other mosquito species and revealed comparable trends [4,23,38,39].

Many studies in Aedes mosquitoes have found that altering environmental parameters not only influences immature development, but also vector competence for arboviruses [5,22,24,39]. We observed no consistent consequences of rearing temperature on the ability of Cx. tarsalis to become infected, disseminate or transmit WNV. Previously, it has been shown that changes in rearing conditions had no effect on the vector competence of Cx. annulirostris for MVEV, Cx. pipiens for RVFV or Cx. tarsalis for either SLEV or WEEV [18-20]. However, it is important to note that in our study, the abundance of adult mosquitoes was significantly affected by rearing temperature. There were significantly fewer adults emerging from the larval group reared at 31°C than at 19°C or 25°C in the majority of replicates, providing fewer mosquitoes that are able to be infected and transmit virus. Though rearing temperature has no direct effect on vector competence, it has the potential to affect adult mosquito abundance and thus vectorial capacity, although this phenomenon will be influenced by density dependent regulation.

Conclusions
Our larval studies, in combination with results from other groups, suggest that varying constant larval rearing temperature has no effect on Culex species vector competence. It remains to be seen whether fluctuating rearing temperature may affect this phenotype [40]. In addition, temperature was altered while holding other parameters constant; further studies should be conducted to determine whether changing multiple factors simultaneously will affect WNV vector competence in Cx. tarsalis.

Abbreviations
WNV: West Nile virus; WEEV: Western Equine Encephalitis virus; SLEV: Saint Louis Encephalitis virus; RVFV: Rift Valley Fever virus; MVEV: Murray Valley Encephalitis virus; VEEV: Venezuelan Equine Encephalitis virus; CHIKV: Chikungunya virus; DENV: Dengue virus; SNV: Sindbis virus; LACV: LaCrosse Encephalitis virus; PFU: Plaque forming units; CV: Coachella Valley strain; YC: Yolo County strain; FBS: Fetal bovine serum; MD: Mosquito diluent; dpf: Days post feeding.

Competing interests
The authors declare that they have no competing interests.

Authors’ contributions
BLD participated in the study design, carried out laboratory experiments, participated in data analysis and participated in drafting the manuscript. LDK participated in the study design and participated in drafting the manuscript. JLR participated in the study design, participated in data analysis and participated in drafting the manuscript. All authors read and approved the final version of the manuscript.

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References
1. Denlinger DL, Yocum GD: Physiology of heat sensitivity. In Temperature sensitivity in insects in integrated pest management. Edited by Hallman GJ, Denlinger DL. Boulder: Westview Press; 1998:7–57.
2. Hardy JL, Houk EJ, Kramer LD, Reeves WC: Intrinsic factors affecting vector competence of mosquitoes for arboviruses. Annu Rev Entomol 1983, 28:229–262.
3. Chamberlain RW, Sudia WD: The effects of temperature on the extrinsic incubation of eastern equine encephalitis in mosquitoes. Am J Hyg 1955, 62:295–305.
4. Muturi EJ, Alto BW: Larval environmental temperature and insecticide exposure alter Aedes aegypti competence for arboviruses. Vector Borne Zoonotic Dis 2011, 11:1157–1163.
5. Westbrook CJ, Reskind MH, Pesko KN, Lounibos LE, Greene LP: Larval environmental temperature and the susceptibility of Aedes aegypti to Chikungunya virus. Vector Borne Zoonotic Dis 2010, 10:241–247.
6. Padmanabha H, Bokler B, Lord CC, Rubio C, Lounibos LP: Food availability alters the effects of larval temperature on Aedes aegypti growth. J Med Entomol 2011, 48:974–984.
7. Bailey SJ, Geake PA: A study of the effect of water temperatures on rice field mosquito development. Proc Papers Annu Conf Calif Mosq Control Assoc 1968, 26:53–61.
8. Denlinger DL, Lee RE: Physiology of cold sensitivity. In Temperature sensitivity in insects in integrated pest management. Edited by Hallman GJ, Denlinger DL. Boulder: Westview Press; 1998:855–95.
9. Venkatesan M, Ranson H: Population genetic data suggest a role for mosquito-mediated dispersal of West Nile virus across the western United States. Mol Ecol 2010, 19:1573–1584.
10. Hayes EB, Komar N, Nasci RS, Montgomery SP, O’Leary DR, Campbell GL: Epidemiology and transmission dynamics of West Nile virus disease. Emerg Infec Dis 2005, 11:1167–1173.
11. Meyer RP, Hardy JL, Presser SB: Comparative vector competence of Culex tarsalis and Culex quinquefasciatus from the Coachella, Imperial, and San Joaquin Valleys of California for St. Louis encephalitis virus. Am J Trop Med Hyg 2003, 32:305–311.
12. Hammond WM, Reeves WC: Laboratory transmission of western equine encephalomyelitis virus by mosquitoes of the genera Culex and Culiseta. J Exp Med 1943, 78:425–434.
13. Bohart RM, Washino RK: Mosquitoes of California. Berkeley: University of California Press; 1978.

14. Reisen WK: Effect of temperature on Culex tarsalis (Diptera: Culicidae) from the Coachella and San Joaquin Valleys of California. J Med Entomol 1995, 32:536–645.

15. Mather TH, DeChiara GR: Effect of host blood source on the gonotrophic cycle of Aedes triseriatus. Am J Trop Med Hyg 1983, 32:189–195.

16. Day JF, Van Handel E: Differences between the nutritional reserves of laboratory-manimated and field-collected adult mosquitoes. J Am Mosq Control Assoc 1986, 2:154–157.

17. Aguilar R, Simard F, Kamdem C, Shields T, Glass GE, Garver LS, Dimopoulos G: Genome-wide analysis of transcriptomic divergence between laboratory colony and field Anopheles gambiae mosquitoes of the M and s molecular forms. Insect Mol Biol 2010, 19:695–705.

18. Reisen WK, Hardy JL, Presser SB: Effects of water quality on the vector competence of Culex tarsalis (Diptera: Culicidae) for western equine encephalomyelitis (Togaviridae) and St. Louis encephalitis (Flaviviridae) viruses. J Med Entomol 1997, 34:631–643.

19. Brubaker JF, Turell MJ: Effect of environmental temperature on the susceptibility of Culex pipiens (Diptera: Culicidae) to Rift Valley fever virus. J Med Entomol 1998, 35:918–921.

20. Kay BH, Edman JD, Fanning ID, Mottram P: Larval diet and the vector competence of Culex annulirostris (Diptera: Culicidae) for Murray Valley encephalitis virus. J Med Entomol 1989, 26:487–488.

21. Turell MJ: Effect of environmental temperature on the vector competence of Aedes taeniorhynchus for Rift Valley fever and Venezuelan equine encephalitis viruses. Am J Trop Med Hyg 1993, 49:67–76.

22. Grimmstad PR, Haramis LD: Aedes triseriatus (Diptera: Culicidae) and LaCrosse Virus. III. Enhanced oral transmission by nutrition-deprived mosquitoes. J Med Entomol 1984, 21:249–256.

23. Alto BW, Lounibos LP, Moore CN, Reiskind MH: Larval competition alters susceptibility of Aedes mosquitoes to dengue infection. Proc Biol Sci 2008, 275:463–471.

24. Alto BW, Reiskind MH, Lounibos LP: Size alters susceptibility of vectors to dengue virus infection and dissemination. Am J Trop Med Hyg 2008, 79:688–695.

25. Paulson SL, Hawley WA: Effect of body size on the vector competence of field and laboratory populations of Aedes triseriatus for La Crosse virus. J Am Mosq Control Assoc 1991, 7:170–175.

26. Dodson BL, Kramer LD, Rasgon JL: Larval nutritional stress does not affect vector competence for West Nile virus (WNV) in Culex tarsalis. Vector Borne Zoonotic Dis 2001, 11:1493–1497.

27. Reisen WK, Fang Y, Martinez VM: Effects of temperature on the transmission of West Nile virus by Culex tarsalis (Diptera: Culicidae). J Med Entomol 2006, 43:309–317.

28. Reisen WK, Hardy JL, Presser SB, Chiles RE: Seasonal variation in the vector competence of Culex tarsalis (Diptera: Culicidae) from the Coachella Valley of California for western equine encephalomyelitis and St Louis encephalitis viruses. J Med Entomol 1996, 33:433–437.

29. Hardy JL, Meyer RP, Presser SB, Milby MM: Temporal variations in the susceptibility of a semi-isolated population of Culex tarsalis to peroral infection with western equine encephalomyelitis and St Louis encephalitis viruses. Am J Trop Med Hyg 1990, 42:500–511.

30. Payne AF, Binduga-Gajewoska I, Kauffman EB, Kramer LD: Quantitation of flavivirus by fluorescent focus assay. J Virol Methods 2006, 134:183–187.

31. Khasnis AA, Nettleman MD: Global warming and infectious disease. Arch Med Res 2005, 36:89–96.

32. Tabachnick WJ: Challenges in predicting climate and environmental effects on vector-borne disease epistemists in a changing world. J Exp Biol 2010, 213:946–954.

33. Gould EA, Higgs S: Impact of climate change and other factors on emerging arbovirus diseases. Trans R Soc Trop Med Hyg 2009, 103:109–121.

34. Epstein PR: Chikungunya fever resurgence and global warming. Amer J Trop Med Hyg 2007, 76:403–404.

35. Gubler DJ: The global threat of emergent/re-emergent vector-borne diseases. In Vector Biology, Ecology and Control. Edited by Atkinson PW. New York: Springer; 2010:39–62.

36. Goddard LB, Roth AE, Reisen WK, Scott TW: Vector competence of California mosquitoes for West Nile virus. Emerg Infect Dis 2002, 8:1385–1391.

37. Reisen WK, Milby MM, Bock ME: The effects of immature stress on selected events in the life history of Culex tarsalis. Mosq News 1984, 44:385–395.

38. Agnew P, Haussy C, Malachias Y: Effects of density and larval competition on selected life history traits of Culex pipiens quinquefasciatus (Diptera: Culicidae). J Med Entomol 2000, 37:733–735.

39. Nasri RN, Mitchell C: Larval diet, adult size, and susceptibility of Aedes aegypti (Diptera: Culicidae) to infection with Ross River virus. J Med Entomol 1994, 31:123–126.

40. Pajaikas KP, Blanford S, Bell AS, Blanford JI, Read AF, Thomas MB: Influence of climate on malaria transmission depends on daily temperature variation. Proc Natl Acad Sci USA 2010, 107:15135–15139.

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