**Discussion**

In this study we used a Bayesian hierarchical approach to estimate mosquito lifespan by analysing a published database of mark-release-recapture experiments (MRRs) and another of mosquito dissection studies which we assembled via a literature search. By applying a single framework, we combined information across experiments which individually estimate lifespan with high uncertainty. Due to the assumptions required to analyse the field data, our estimates represent lower bounds on lifespan (LBL). Across both meta-analyses, the majority of estimated LBLs were less than 10 days, hinting that relatively few mosquitoes survive to be able to transmit disease. We determined that LBL varies across species and genera with most variance explained by differences at the genus level. The MRR analysis includes experiments conducted on each sex individually, and we estimate that, on average, females outlive males. Pre-release feeding with sugar also lengthens lifespan across all three genera, although this eﬀect is less marked than the sex diﬀerences. In contrast to some laboratory experiments (Yang et al., 2009; Brady et al., 2013), temperature was not determined to impact lifespan. By fitting a range of survival models to the data in both meta-analyses, we assessed evidence for mosquito senescence which yielded mixed results: in the MRR analysis, 8 of 33 species the data was better fit by models incorporating a mortality hazard increasing with age, with only 2 out of 25 species for the dissection studies.

MRR experiments are thought to underestimate lifespan: laboratory experiments have shown that marking mosquitoes with dye can negatively impact survival (Verhulst, Loonen, and Takken, 2013; Dickens and Brant, 2014); additionally MRR studies typically cannot diﬀerentiate between a mosquito dying and its dispersal from the study area further inflating mortality estimates. There was insufficient variation in the dyes used to mark mosquitoes in the database meaning it was not possible to quantify their effect on our estimates. We determined a positive correlation between lifespan estimates and trapping density however, indicating better trapping coverage likely raises estimates towards their true value. We conducted an *in silico* Monte Carlo study to determine how accurately mosquito lifespan can be estimated in an ideal MRR experiment, where mosquitoes are not harmed by marking nor emigrate from the study area (see SOM). This indicated that the short study durations and small release sizes of most experiments results in high uncertainty in lifespan estimates (Fig. S12), meaning pooling data across studies offers considerable improvements in statistical power.

Polovodova was the first to suggest that dissection can determine the number of gonotrophic cycles a female mosquito has lived through (Polovodova, 1948), which is termed their ``physiological age’’. The key assumptions in using dissection data to estimate mosquito age are: (i) physiological age is metered one-for-one by ovariolar dilations (unlike MRR, this method can only be applied to females) and (ii) the relationship between physiological and chronological age is known. Dissection data likely understates physiological age because most ovarioles have fewer dilations than the number of gonotrophic cycles undergone (Fox and Brust, 1994; Hugo et al., 2008), an issue known to worsen with age (Fox and Brust, 1994), meaning a large number of ovarioles must be dissected to locate those representative few (Gillies and Wilkes, 1965). The exchange rate between physiological age and chronological age is the duration of gonotrophic cycles which is predominantly estimated by two methods: MRR studies (see, for example, Gillies and Wilkes, 1965), where marked mosquitoes are recaptured and dissected to determine the number of gonotrophic cycles occurring since release; and dissections of specimens from laboratory colonies (see, for example, Afrane et al., 2005). In our analysis, laboratory studies indicated the longer gonotrophic cycle of the two approaches, which was somewhat surprising given that, in the wild, mosquitoes must additionally locate hosts to blood-feed and aquatic habitats in which to oviposit (Fig. S9). The literature does not however favour one experimental method over another, and so to convert physiological age into calendar age we pooled data from both sources. It is possible, however, that aggregating data across both methods induces biases and an approach more entrenched in experimental knowledge would fare better. The dissection of numbers of wild-caught specimens yields estimates of the age structure of the population at snapshots in time, which can be used to determine mean lifespans. This requires additional assumptions however and here we suppose that the population is at equilibrium (adult recruitment matches mortality) and that collected individuals are random samples from that population. If a population is shrinking, there are relatively few young mosquitoes and the survival curve is flatter than at equilibrium, meaning we would overestimate lifespan. To balance those samples collected whilst the population density was declining with those taken during expansion, we aggregated capture data across collections occurring at different timepoints for each site. Field entomologists have argued that trapping methods do not yield unbiased samples of the population, although there is not consensus on whether this results in a relative paucity (Gillies and Wilkes, 1965) or inflation (Clements and Paterson, 1981) of actual nulliparous numbers. Though after excluding those data with a large deficit of nulliparous individuals, posterior predictive checks indicated that our models fit the data well across all age classes (see SOM) hinting the assumption of random sampling may be reasonable. Overall, the assumptions underpinning our analysis of dissection data suggest these estimates, like those from MRR, represent lower bounds on lifespan. The alternative dissection-based approach of Detinova (Detinova, 1962), based on dichotomous categorisation of female mosquito specimens as ‘parous’ or ‘unparous’ relies on fewer assumptions, and is also widely used. Further work examining parity rates in field specimens may be fruitful although, in principle, it oﬀers less information on the age structure of a population than Polovodova’s.

It is widely believed mosquitoes live artificially long under the benign conditions of the laboratory and their lifespans likely constitute an upper bound on wild populations. A large cage experiment of *Ae. aegypti* determined that females lived on average for nearly 32 days days (Styer et al., 2007), and a similar study with female *A. stephensi* determined a median lifespan between 31 and 42 days (Dawes et al., 2009). These estimates are multiples of the modal estimates from either of our analyses but without a gold standard method to measure mosquito lifespan in the field, it is diﬃcult to quantify the gap that exists between field and laboratory populations. The development of additional methods to estimate mosquito age such as ‘Near-Infrared Spectroscopy’ (Mayagaya et al., 2009; Sikulu et al., 2011; Lambert et al., 2018) if they are proven to work in the field, may be of considerable worth here.

In our analysis of the MRR data, we considered the effect of air temperature, sex and pre-release feeding status on lifespan. There is evidence mainly from laboratory studies that temperature modulates mosquito ecology and behaviour (Yang et al., 2009; Brady et al., 2013; Murdock et al., 2012; Beck-Johnson et al., 2013). The locations and times of year when the MRR studies were conducted encompassed a large range of average air temperatures, from approximately 10 oC to 35 oC yet we found no relationship between lifespan and temperature when pooling all series (Fig. S5) or for those individual species with the most data (Fig. S6). It is possible that by considering a raw average of air temperatures across the month and ignoring rainfall (whose historical data is likely less reliable for a given location), this masked more complex interactions between temperature and lifespan. The observed laboratory relationship between lifespan and temperature, however, may not be as robust in the field if mosquitoes adjust their behaviour (such as, by seeking shade) in reaction to changes in temperature. As in laboratory studies (Styer et al., 2007; Dawes et al., 2009) our analysis indicates that females outlive male mosquitoes, although the magnitude of this difference is not as large in absolute or percentage terms. Ethical concerns mean it is now more common than historically for MRR experiments to release males opposed to females (Fig. SM2). Since differences in lifespan exist between the sexes, it is possible that other ecological parameters that can be determined by MRRs also differ, highlighting the need for field methods that directly measure these characteristics of wild females. Our estimates of lifespan indicate that mosquitoes sugar-fed prior to release lived longer than unfed individuals (Fig. S4) which may partly explain recent successes of vector control methods reducing access of the insects to sugar or using toxic sugar baits (Müller, Kravchenko, and Schlein, 2008; Müller, Junnila, and Schlein, 2010; Müller et al., 2010a; Müller et al., 2010b; Beier et al., 2012).

It is encouraging that our pooled estimates of lifespan from the MRR (6.0 days) and dissection (5.5 days) analyses are comparable and that there was a positive (although insignificant) correlation between corresponding species-level estimates. Across genera, the greatest discrepancy in estimates was for *Aedes*, with 8.1 days estimated from the MRR studies compared to 3.5 days from the dissection analysis. This was followed by *Culex* (2.9 days from the MRRs versus 4.9 days fromdissection) with the smallest discrepancy for *Anopheles* (6.8 versus 6.4 days). Some of the diﬀerences in estimates between the two approaches are likely due to environmental and genetic diﬀerences between mosquitoes included in this analysis. However, we believe that part of the discrepancy can be explained by the assumptions required to analyse each field method and speculate that diﬀerences in dispersal rate may be responsible. Both *Anopheles* and *Culex* mosquitoes are generally thought to fly farther than *Aedes* [Charles, do you have a reference here?], meaning that the estimates from MRR-based approaches will be most downwardly-biased for these genera. This is supported by our results since the dissection-based estimates (themselves not reliant on assumptions about dispersal) are similar or exceed the MRR estimates for *Anopheles* and *Culex* mosquitoes, but not for *Aedes*.

In our study, we determined that mosquito senescence could better explain patterns of mortality in only a minority of cases across both experimental approaches, contrasting with evidence from some laboratory studies (Styer et al., 2007; Dawes et al., 2009) and field experiments (Harrington et al., 2014). To determine if experimental characteristics were responsible for our inability to detect senescence, we conducted a power analysis of MRR experiments (see SOM). This work indicated that power to detect senescence senescence strongly depends on study length (Fig. S11B) but is insensitive to release size (Fig. S11C). Clements and Patterson (1981) conducted a meta-analysis of MRR and dissection field experiments and determined that mortality increased with age at a rate comparable to the ‘mild’ senescence case population we consider in the power analysis (see SOM). In this case, detecting senescence with a power of 80% required a study length of at least 18 days. Since the median study duration for experiments included in our analysis was 10 days (Table SM2) this could partly explain our failure to detect senescence at the species level. It is possible however that laboratory studies overstate the magnitude of senescence since colony mosquitoes may survive long enough to experience physiological decline not felt in the wild. To our knowledge the MRR study of Harrington et al., (2014) on *Aedes* aegypti in Thailand has been the sole field experiment aiming to detect senescence and we argue that other similar studies would be worthwhile.

In this work, we applied modern statistical methods to combine precious field data collected by entomologists past and present to produce lower bound estimates of mosquito lifespan. Although our approach to estimating mosquito lifespan is novel, its importance for disease transmission has long been recognized since even before 1957, when George Macdonald formulated his mathematical model of malaria transmission. Indeed, the recent declines in malaria prevalence in Sub-Saharan Africa were mainly due to interventions (insecticide-treated bednets and indoor residual spraying) that aim to reduce mosquito lifespan (Bhatt et al., 2015). Yet, there is ample evidence that mosquito resistance to pyrethroid insecticides has spread throughout the continent (World Health Organization, 2018) which may erode or reverse recent gains. A direct way to assess the performance of existing vector control methods is to estimate how mosquito lifespan responds to changes in measured resistance. The diﬀerent nature of the assumptions underpinning analysis of MRR and dissection studies means they oﬀer complimentary information on mosquito survival and lacking a gold standard method to estimate this quantity, we foresee continued reliance on these longstanding field entomological methods.

**References**

1. Y. A. Afrane et al. “Eﬀects of microclimatic changes caused by land use and land cover on duration of gonotrophic cycles of *Anopheles gambiae* (Diptera: Culicidae) in western Kenya highlands”. *Journal of Medical* *Entomology* 42.6 (2005), pp. 974–980.
2. L. M. Beck-Johnson et al. “The eﬀect of temperature on Anopheles mosquito population dynamics and the potential for malaria transmission”. *PLOS one* 8.11 (2013), e79276.

1. J. C. Beier et al. “Attractive toxic sugar bait (ATSB) methods decimate populations of *Anopheles* malaria vectors in arid environments regardless of the local availability of favoured sugar-source blossoms”. *Malaria* *Journal* 11.1 (2012), p. 31.
2. S. Bhatt et al. “The eﬀect of malaria control on *Plasmodium falciparum* in Africa between 2000 and 2015”. *Nature* 526.7572 (2015), pp. 207–211.
3. O. J. Brady et al. “Modelling adult Aedes aegypti and Aedes albopictus survival at diﬀerent temperatures in laboratory and field settings”. *Parasites & vectors* 6.1 (2013), p. 351.
4. R. Carter and K. N. Mendis. “Evolutionary and historical aspects of the burden of malaria”. In: *Clinical Microbiology Reviews* 15.4 (2002), pp. 564– 594.
5. A. Clements and G. Paterson. “The analysis of mortality and survival rates in wild populations of mosquitoes”. *Journal of Applied Ecology* (1981), pp. 373–399.
6. E. J. Dawes et al. “*Anopheles* mortality is both age-and *Plasmodium*-density dependent: implications for malaria transmission”. *Malaria* *Journal* 8.1 (2009), p. 228.
7. T. S. Detinova et al. “Age grouping methods in Diptera of medical impor-tance with special reference to some vectors of malaria”. In: *Monograph* *series World Health Organisation* (1962).
8. B. L. Dickens and H. L. Brant. “Eﬀects of marking methods and fluorescent dusts on *Aedes aegypti* survival”. In: *Parasites & Vectors* 7.1 (2014), p. 1.
9. A. Fox and R. Brust. “How do dilatations form in mosquito ovarioles?” *Parasitology Today* 10.1 (1994), pp. 19–23.
10. B Gates. *The Deadliest Animal in the World*. https://www.gatesnotes.com/Health/Most-Lethal-Animal-Mosquito-Week. 2014.
11. A. Gelman and D. B. Rubin. “Inference from iterative simulation using multiple sequences”. In: *Statistical Science* (1992), pp. 457–472.
12. M. Gillies and T. Wilkes. “A study of the age-composition of populations of *Anopheles gambiae* Giles and A. funestus Giles in North-Eastern Tanzania”. *Bulletin of Entomological Research* 56.02 (1965), pp. 237–262.
13. C. A. Guerra et al. “A global assembly of adult female mosquito mark-release-recapture data to inform the control of mosquito-borne pathogens”. *Parasite & Vectors* 7.1 (2014), p. 276.
14. L. C. Harrington et al. “Age-dependent survival of the dengue vector *Aedes aegypti* (Diptera: Culicidae) demonstrated by simultaneous release–recapture of diﬀerent age cohorts”. *Journal of Medical Entomology* 45.2 (2008), pp. 307–313.
15. T. Hoc and T. Wilkes. “The ovariole structure of *Anopheles gambiae* (Diptera: Culicidae) and its use in determining physiological age”. *Bulletin of Entomological Research* 85.01 (1995), pp. 59–69.
16. L. E. Hugo et al. “Adult survivorship of the dengue mosquito *Aedes aegypti* varies seasonally in central Vietnam”. *PLoS Neglected Tropical Diseases* 8.2 (2014), e2669.
17. L. E. Hugo et al. “Evaluations of mosquito age grading techniques based on morphological changes”. *Journal of Medical Entomology* 45.3 (2008), 353–369.
18. B. Kay. “Age structure of populations of *Culex annulirostris* (Diptera: Culicidae) at Kowanyama and Charleville, Queensland”. *Journal of* *Medical Entomology* 16.4 (1979), pp. 309–316.
19. B. Lambert. *A Student’s Guide to Bayesian Statistics*. Sage, 2018.
20. B. Lambert et al. “Monitoring the Age of Mosquito Populations Using Near-Infrared Spectroscopy”. *Scientific reports* 8.1 (2018), p. 5274.
21. A. Lange and T. Hoc. *Abortive oogenesis and physiological age in blood-sucking mosquitoes (Diptera: Culicidae). Meditsinskaya Parazitologiya i Parasitarnye Bolezni 50, 48–56*. 1981.
22. G. Macdonald et al. *The epidemiology and control of malaria.* London, Oxford University Press, 1957.
23. V. S. Mayagaya et al. “Non-destructive determination of age and species of *Anopheles gambiae* sl using near-infrared spectroscopy”. *The American Journal of Tropical Medicine and Hygiene* 81.4 (2009), pp. 622–630.
24. G. Müller, A Junnila, and Y Schlein. “Eﬀective control of adult *Culex* *pipiens* by spraying an attractive toxic sugar bait solution in the vegetationnear larval habitats”. *Journal of Medical Entomology* 47.1 (2010), 63–66.
25. G. C. Müller, V. D. Kravchenko, and Y. Schlein. “Decline of Anopheles sergentii and Aedes caspius populations following presentation of attractive toxic (spinosad) sugar bait stations in an oasis”. *Journal of the American* *Mosquito Control Association* 24.1 (2008), pp. 147–149.
26. G. C. Müller et al. “Field experiments of Anopheles gambiae attraction to local fruits/seedpods and flowering plants in Mali to optimize strategies for malaria vector control in Africa using attractive toxic sugar bait methods”. *Malaria journal* 9.1 (2010), p. 262.
27. G. C. Müller et al. “Successful field trial of attractive toxic sugar bait (ATSB) plant-spraying methods against malaria vectors in the *Anopheles* *gambiae* complex in Mali, West Africa”. In: *Malaria Journal* 9.1 (2010), 210.
28. C. Murdock et al. “Complex eﬀects of temperature on mosquito immune function”. *Proc. R. Soc. B* (2012), rspb20120638.
29. J. Nedelman. “A negative binomial model for sampling mosquitoes in a malaria survey”. *Biometrics* (1983), pp. 1009–1020.
30. V. Polovodova. “The determination of the physiological age of female *Anopheles* by the number of gonotrophic cycles completed”. *Meditsin-skaia Parazitologiia Parazitar Bolezni* 18 (1949), pp. 352–355.
31. R. C. Russell. “Population age composition and female longevity of the arbovirus vector *Culex annulirostris skuse* near Echua, Victoria, in the Murray Valley of southeastern Austria 1979-1985”. *Australian Journal* *of Experimental Biology & Medical Science* 64.6 (1986).
32. M. Service. “Studies on sampling larval populations of the *Anopheles* *gambiae* complex”. In: *Bulletin of the World Health Organisation* 45.2(1971), p. 169.
33. M. Sikulu et al. “Evaluating RNAlater® as a preservative for using near-infrared spectroscopy to predict *Anopheles gambiae* age and species”. *Malaria Journal* 10.1 (2011), p. 186.
34. J. B. Silver. *Mosquito ecology: field sampling methods*. Springer Science & Business Media, 2007.
35. M. E. Sinka et al. “The dominant *Anopheles* vectors of human malaria in Africa, Europe and the Middle East: occurrence data, distribution maps and bionomic précis”. *Parasites & Vectors* 3.1 (2010), p. 117.
36. L. M. Styer et al. “Mosquitoes do senesce: departure from the paradigm of constant mortality”.*The American Journal of Tropical Medicine and* *Hygiene* 76.1 (2007), pp. 111–117.
37. N. O. Verhulst, J. A. Loonen, and W. Takken. “Advances in methods for colour marking of mosquitoes”. *Parasites & Vectors* 6.1 (2013), p. 1.
38. World Health Organisation et al. “WHO statement on the first meeting of the International Health Regulations (2005) - Emergency Committee on Zika virus and observed increase in neurological disorders and neonatal malformations”. In: 37.3 (2016), pp. 332–333.
39. World Health Organization et al. “Global report on insecticide resistance in malaria vectors: 2010–2016”. WHO (2018).
40. H. Yang et al. “Assessing the eﬀects of temperature on the population of *Aedes aegypti*, the vector of dengue”. *Epidemiology and Infection* 137.08 (2009), pp. 1188–1202.