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Short Communication

Leaf inputs from invasive and native plants drive differential mosquito abundances

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ABSTRACT

Ecological impacts of invasive alien species can be unpredictable and simultaneously span multiple habitat types and taxonomic groups. Invasive alien plants can have particularly severe impacts, and plant inputs into aquatic environments can profoundly alter community composition of invertebrates, such as mosquitoes. Here, we examine larval mosquito colonisation of aquatic containers treated with leaves from four different terrestrial plants: the invasive tickberry *Lantana camara*, invasive guava *Psidium guajava*, native sycamore fig *Ficus sycomorus* and native silver cluster-leaf *Terminalia sericea*. Larval mosquito abundances differed significantly accordingly to leaf treatment, whilst no mosquitoes colonised leaf-free controls. Leaf litter from the invasive *L. camara*, invasive *P. guajava* and native *F. sycomorus* drove significant increases in mosquito abundances relative to native *T. sericea*. We demonstrate the importance of plant identity for larval mosquito proliferations in aquatic habitats, with changes in plant community composition following terrestrial plant invasions potentially resulting in increased mosquito abundances. In turn, this may have implications for the vectoring of mosquito-borne disease.

Key words

invasive alien plant; plant-vector interactions; container-breeding mosquitoes; leaf litter; vector-borne disease; oviposition

1. Introduction

The ecosystem-level impacts of invasive alien species can be multifaceted, with introductions often driving unexpected consequences in novel ranges. For example, invasive alien species can alter recipient ecosystems through greater provisioning of habitat, increased resource availability and direct interference with consumers (e.g. Green et al., 2011; Kobak et al., 2016; Sheppard et al., 2018). As plants are often the most important energy sources in terrestrial ecosystems, invasive alien plants can have especially marked ecological consequences (Sax et al., 2005), and these effects can span multiple habitat types. In particular, aquatic ecosystems may be heavily impacted by changes to terrestrial vegetation composition due to differences in detrital nutrient properties (Yanoviak, 1999). Indeed, alterations to species-level plant inputs can significantly change invertebrate community dynamics in aquatic habitats, where mosquitoes are often numerically dominant and benefit directly from leaf litter as a resource (Leonard and Juliano, 1995; Yanoviak, 1999; Reiskind et al., 2009). Furthermore, invasive terrestrial plants can provide increased shelter or breeding sites for adult mosquitoes, with potential benefits for population-level fitness and the transmission of mosquito-borne disease (Rajnikant et al., 1992; Webb et al., 2012; Stone et al., 2018). Mosquitoes are known to be highly responsive to plant cues when ovipositing (Shaan and Canyon, 2018), and an increasing body of evidence suggests that some invasive alien plant species may enhance mosquito demographics and exacerbate disease risk (Reiskind et al., 2010; Mack and Smith, 2011). However, understanding differential colonisation responses of mosquitoes among plant species has remained elusive.

In the present study, we examine the effects of leaf litter inputs from two invasive and two native terrestrial plant species on larval mosquito abundances (*Culex* spp. and *Aedes* spp. combined): the invasive tickberry *Lantana camara*, invasive guava *Psidium guajava*, native sycamore fig *Ficus sycomorus* and native silver cluster-leaf *Terminalia sericea*. The *L.*

camara species complex is native to tropical central and southern America, and is known to impact ecosystems through excessive resource additions, alterations to fire regimes and increased erosion (Richardson and van Wilgen, 2004; Vardien et al., 2012). *Psidium guajava* is also native to central and southern America and has substantial negative impacts on native species through competition and replacement (Urquía et al., 2019). Both species are recognised as invasive in South Africa and known to colonise close to waterbodies, whilst the two native plants serve as representative resident species.

2. Materials and Methods

Plant leaves of invasive i.e. *L. camara* and *P. guajava* and native i.e. *F. sycomorus*, and *T. sericea* and were collected in November 2018 by hand from trees along Muvudi River riparian zone (22° 58.967'S 30° 26.840'E) in Thohoyandou, Limpopo province, South Africa, before being air dried at room temperature. Only fresh plant leaves were selected, given many plants reabsorb nutrients prior to senescence (e.g. Staaf 1982). After drying, approximately 3 g of the material for each species was weighed and added into 5 L white polyethylene buckets, filled with 3.9 L of filtered (filter size 63 µm) borehole + river water (70:30 ratio). The initial mean water temperature was 27.2 ± 0.2 °C (SD), conductivity was 169.3 ± 4.0 µS cm⁻¹, total dissolved solids were 85.5 ± 2.6 mg L⁻¹ and pH was 6.6 ± 0.1 . Weekly, the buckets were topped up to 3.9 L with filtered borehole water. The twenty-five 5 L buckets (i.e. 5 replicates × 4 species, + 5 controls) were placed in a shaded area on the University of Venda campus in a randomised array, and mosquitoes could freely colonise any bucket. Mosquito larvae counting was initiated after three weeks to allow for colonisation, with first instar stages omitted from counts due to low visual detectability. During each sampling event, container contents were strained, and larval mosquitoes counted before being returned to their respective containers. Container-breeding mosquito larvae (*Culex* spp. and *Aedes* spp.) were subsequently enumerated weekly over 4 weeks.

Generalised linear mixed models (GLMMs) assuming a Poisson error distribution and log link were used to analyse larval mosquito counts over the experimental period according to the *treatment* and *time* terms, and their interaction (Brooks et al., 2017). Bucket *ID* was included as a random effect to account for repeated measures of each bucket over time, with *time* included as a within-subject variable. Resulting models were tested for zero inflation and overdispersion *via* simulation comparisons (Hartig, 2018). Analysis of deviance with Type II sums of squares was used to infer significance levels of main effects (Fox and Weisberg, 2011), with estimated marginal means used *post-hoc* for Tukey comparisons (Lenth, 2018). In all analyses, significance was inferred at $p < 0.05$. All statistical analyses were performed using the R environment (v3.5.1; R Development Core Team, 2018).

3. Results

No larval mosquitoes were recovered from control groups, and thus this treatment level was removed from further analyses. Contrastingly, mosquitoes colonised all containers treated with leaf litter, however abundances in native *T. sericea* were considerably reduced overall (Fig. 1). The interaction term between *treatment* and *time* was not statistically clear (likelihood ratio test: $\chi^2 = 6.66$, $df = 9$, $p = 0.67$), and was thus removed from the final model. Accordingly, differences among leaf treatments in larval abundances were consistent across the monitoring period. Leaf treatments significantly affected larval mosquito abundances (GLMM: $\chi^2 = 61.66$, $df = 3$, $p < 0.001$). Significantly fewer mosquitoes were present in containers treated with native *T. sericea* as compared to invasive tickberry *L. camara*, invasive guava *P. guajava* and native *F. sycomorus* (all $p < 0.001$); there were no significant differences among the latter three plant species (all $p > 0.05$). Larval mosquito abundances also differed significantly over time overall (GLMM: $\chi^2 = 20.23$, $df = 3$, $p < 0.001$), with abundances falling significantly between the first and fourth week of the monitoring period ($p < 0.001$) (Fig. 2).

4. Discussion

The establishment and spread of invasive alien species can often have unexpected ecosystem-level impacts across multiple habitat types. In the present study, we demonstrate that leaf litter inputs among invasive and native terrestrial plants can differ markedly in their effects on larval mosquito abundances. Specifically, two highly invasive terrestrial plant species, *L. camara* and *P. guajava*, and the native *F. sycomorus*, drove significantly elevated mosquito abundances compared to native *T. sericea*. Mosquitoes were entirely absent from leaf-free controls, owing to a lack of organic content. Accordingly, alterations to plant stands following invasions could worsen mosquito problems by driving increases in mosquito abundances, with possible implications for public health. Yet, this effect is likely species-specific. Furthermore, plant incursions into previously unvegetated areas in proximity to water bodies could further promote mosquito proliferations.

The complete lack of colonisation of leaf-free controls in the present study is not surprising, given the strong general attractant effect of organic material on gravid mosquitoes (Hazard et al., 1967). Indeed, given the capacity for mosquitoes to exploit small, container-style habitats which are often present in high densities across terrestrial landscapes, there is frequently a high degree of selectivity for ‘rewarding’ habitats among available patches. Whilst we observed negligible colonisation of native *T. sericea* treated waters, a much stronger attractant effect was found towards native *F. sycomorus*. Accordingly, botanical effects appear to be tightly linked to plant identity, with effects of invasion potentially dependent on background vegetative composition.

Nevertheless, although not assessed in the present study, natural compounds from plants can have lethal effects on larval mosquitoes (Shalan and Canyon, 2018), and so it cannot be ruled out that these effects altered abundances in specific treatments here, rather than *via*

selectivity. Indeed, whilst previous research has demonstrated attraction to *P. guajava* fruits by adult mosquitoes (Müller et al., 2010), compounds from *P. guajava* and *L. camara* have demonstrated mosquitocidal properties through lipophilic extracts (Mendes et al. 2017; Hari and Mathew, 2018). Yet, the specific compounds that drive colonisation differences require further investigation. In particular, to our knowledge, no studies have examined effects of compounds from *T. sericea* on mosquito abundances. Given the importance of the identity of plant inputs for the productivity of mosquito populations in aquatic habitats (Reiskind et al., 2009, 2010), quantifications of differential responses among invasive and native plant species is of high importance in the context of public health. Further research should therefore examine the direct effects of these and other plant inputs on additional mosquito life history traits, such as size, sex and fecundity, as well as direct lethal effects. In turn, this may aid in the identification of specific compounds that deter colonisation.

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Fig. 1. Mean ($\pm$ SE) larval mosquito abundances in 3.9 L containers under four plant leaf treatments (Fs, native *Ficus sycomorus*; Ts, native *Terminalia sericea*; Lc, invasive *Lantana camara*; Pg, invasive *Psidium guajava*) over four week monitoring period. Letters denote significant differences.

Fig. 2. Mean ($\pm$ SE) weekly larval mosquito abundances in 3.9 L containers under four plant leaf treatments (Fs, native *Ficus sycomorus*; Ts, native *Terminalia sericea*; Lc, invasive *Lantana camara*; Pg, invasive *Psidium guajava*) over four week monitoring period. Locally-weighted scatterplot smoothing lines are presented (9/10 smoother span) to illustrate trends.

Highlights

- Mosquito colonisation under invasive and native leaf litter treatments examined.
- No mosquitoes colonised leaf-free control aquatic habitats.
- Significant differences were found among invasive and native plant species.
- Mosquitoes significantly avoided native *Terminalia sericea*.
- Terrestrial plant invasion impacts can span multiple habitat types.

ACCEPTED MANUSCRIPT

Figure 1

Figure 2