

Specialist by preference, generalist by need: availability of quality hosts drives parasite choice in a natural multihost-parasite system

Darío Manzoli¹, María Saravia-Pietropaolo¹, Sofia Arce¹, Alejandro Percara¹, Leandro Antoniazzi¹, and Pablo Beldomenico²

¹Laboratorio de Ecología de Enfermedades

²Universidad Nacional del Litoral

May 5, 2020

Abstract

Encountering suitable hosts is key for parasite success. In a natural system involving a parasitic fly and its multiple bird hosts there are profound differences in host quality. The Great Kiskadee tolerates and does not invest in resisting the infection, which makes it an optimal host. Alternative hosts are frequently used, but whilst some of them may be good options, others are bad alternatives (they resist efficiently or die). Here we examined the host selection processes that drive parasite dynamics in this system with a thorough longitudinal study under natural conditions. We found that host selection is strongly driven by availability of quality hosts: the parasite chooses suboptimal hosts only when better alternatives are not sufficiently available. This adds evidence from a natural system that hosts are chosen as a function of their profitability, and shows that host selection by a parasite may be plastic and context-dependent.

INTRODUCTION:

Host-parasite interactions are inter-specific relationships in which the parasite is engaged in exploiting the host, while the host tries to counter the infection by minimising parasite burden (resistance) and its damage (tolerance) (Raberg et al. 2009). The outcome of this battle will determine consequences for host and parasite: the fitness cost paid by the host (virulence) and the success of an infection (transmission) (Leggett et al. 2013). The majority of parasites use more than one host species (Woolhouse et al., 2001; Schmid-Hempel, 2011). Using different host species implies exposure to different host competences and defence strategies, resulting in differential parasite transmission (Gervasi et al. 2015). From a parasite's perspective, an optimal host is one that survives the infection, so that the cycle of the parasite can be completed; and one that makes little effort to reduce the development, reproduction or replication of the parasite. Hence, to maximise parasite transmission, an ideal host should be little resistant and highly tolerant (Martin et al. 2016).

In nature, parasites encounter potential hosts that may differ in their quality (host competence) or quantity (abundance) (Fenton et al. 2015; Gervasi et al. 2015). Some parasites are considered generalists because they use a wide range of hosts, while others specialise in one or a few host species. Under these opposing strategies, generalist parasites benefit from broad host availability (quantity) whereas specialists prioritise host quality. Favouring quantity over quality may result in suboptimal exploitation of the host, maladaptive virulence and poor transmission (Rigaud et al. 2010; Lievens et al. 2018). On the other hand, specialists are less resilient during environmental disruptions that may affect the structure of the host community (Auld et al. 2017).

For an infection to occur a parasite has to encounter a compatible host. In this exposure process, the parasite may exhibit clear host preferences (Combes, 1991). Although encounter rates are often assumed to

be a function of host density or frequency, some parasites and vectors can also display non-random or even targeted host selection (Johnson et al. 2019). In those cases, host preference should reflect host competence, as selection pressures would favour choosing hosts with high tolerance and low resistance. Motile parasites, therefore, would not seek and infect hosts in a random manner, but rather their preference would be driven by host quality. Theoretical approaches explored this notion (Best et al. 2014; Forbes et al. 2017), but the existing empirical evidence is still very limited. One of the few studies that assessed the hypothesis that host quality drives parasite preference found that cercariae of an amphibian trematode discriminated among host species and preferred those that least limited the infection (Sears et al. 2012). Another study found that cercariae showed a consistent preference, but in this case host attractiveness was decoupled with host competence (Johnson et al. 2019). In other system, a parasitoid appears to contact its dinoflagellate hosts at random (Alacid et al. 2016).

Studying the complex dynamics of multihost-multiparasite communities in nature is a major outstanding challenge (Auld et al. 2017). This is because in natural systems causal relationships are embedded in a wider web of complex interactions, making extremely difficult dissecting cause-effect patterns from background noise. A singular system, comprising a parasitic fly and its multiple bird hosts, possesses characteristics that make it very amenable to detailed study of the ecology of host-parasite interactions in nature. The genus *Philornis* Meinert 1890 (Diptera: Muscidae) consists of a group of Neotropical flies that depend on bird broods (Arendt 1985; Couri 1999). While adults are free living, larval trophic behaviour is associated with bird nestlings. Most parasitic *Philornis* spp. have subcutaneous burrowing larvae that feed on blood, tissue and fluids at all instars (Dudaniec et al. 2006). The majority of subcutaneous *Philornis* parasitise exclusively bird nestlings. Because nestlings can be monitored daily from egg to fledging (i.e. the whole period they are susceptible to infection by *Philornis* spp.), collection of sequential infection data from the whole bird community is feasible (Manzoli et al. 2013). Moreover, subcutaneous larvae are relatively large and stay at the site where they penetrated the skin, so they can also be individually followed throughout this parasitic stage, enabling assessment of parasite success (Manzoli et al. 2018).

The parasitic fly identified in central Argentina was designated ‘*Philornis torquans* complex genotype central Argentina’ (hereafter, ‘*P. torquans* c. A.’) (Monje et al. 2013; Quiroga et al. 2016). *Philornis torquans* is considered a generalist (Löwenberg-Neto 2008), and in central Argentina it has been found to parasitise around half of the passerine birds that breed there (Antoniazzi et al. 2011; Manzoli et al. 2013). However, it is closely associated with Great Kiskadees, *Pitangus sulphuratus* Linnaeus, 1766 (Passeriformes: Tyrannidae) (de la Peña et al. 2004; Antoniazzi et al. 2011; Manzoli et al. 2013). Given that prevalences and burdens are highest in Great Kiskadees, and that the abundance of their broods determines the occurrence of *P. torquans* in the whole bird community (Antoniazzi et al. 2011; Manzoli et al. 2013), Great Kiskadees are arguably the main host of *P. torquans* c. A. Many other bird species can be alternative hosts, of which Thornbirds (The Little Thornbird - *Phacellodomus sibilatrix* Sclater, 1879; and the Greater Thornbird - *Ph. ruber* Vieillot, 1817) are among the ones most frequently used (Antoniazzi et al. 2011; Manzoli et al. 2013; de la Peña et al. 2004). A recent study tested the hypothesis that, in this system, the outcome of host-parasite interactions is different when comparing main (Kiskadees) and alternative hosts (Thornbirds), and that this is related to contrasts in host defence strategies (Manzoli et al. 2018). It was found that main hosts have a strategy of high tolerance and negligible resistance, ensuring great larval success (>90%). In Thornbirds, an inflammatory response to infection is mounted, indicating a strategy of resistance. This response is efficient in Little Thornbirds (parasite success is reduced in >80%), but much less so in Greater Thornbirds (~20% reduction). These results highlighted the importance of defence strategies and its efficacies in determining virulence and infection dynamics, and also provided support to the hypothesis posited above which claimed that host selection is driven by host quality. It is noteworthy that despite the large difference in host competence between both Thornbird species, the parasite does not appear to prefer the better quality alternative host over the other: 38% of Little Thornbird broods parasitised compared to 26% Greater Thornbird broods (Manzoli et al. 2013).

The above prompted some questions, such as, why would a gravid female fly select a bad host instead of the optimal host or good alternative ones? A preliminary analysis suggested that ‘*P. torquans* c. A.’ selects

Thornbirds when the availability of Kiskadee nestlings has been low (Manzoli et al. 2013). Here we offer data from a thorough longitudinal study conducted along 8 breeding seasons under natural conditions to test the hypothesis that *Philornis* chooses suboptimal hosts when better alternatives are not sufficiently available.

Materiales y Métodos

Community studied

The data were collected from a 40-ha patch of native forest located in the centre of Santa Fe Province (60° 55' W, 31° 23' S), Argentina, representing relicts of the biogeographic province 'El Espinal'. The climate in the region is Pampean Temperate, with an average annual temperature of 18 °C (mean minimum= 12 °C; mean maximum= 23 °C) (www.climayagua.inta.gov.ar). These forests are breeding grounds for around 100 bird species, mostly Passeriformes (de la Peña, 2018). Because trees of El Espinal are not high, accessing the vast majority of bird nests of the community becomes feasible. At the study area, more than 20 species have been found to be parasitized by '*P. torquans* c. A.' larvae (Antoniazzi et al. 2011; Manzoli et al. 2013), but the Great Kiskadee is by far the preferred host, with prevalences over 3 times higher than those of the second most used host (Manzoli et al, 2013). Among the most frequently parasitized alternative hosts are the Greater Thornbird and the Little Thornbird (Antoniazzi et al, 2011). These three species were selected for this study, due to their abundance, the prevalence of infection by the parasite, and because there are prior data on resistance, tolerance and parasite success (Manzoli et al. 2018). Nonetheless, data on parasite occurrence was weekly obtained from all hosts present in the area. Given the established differences in host competence for the three focal host species, they may be categorised as the 'optimal host' (the Great Kiskadee; it survives and does not resist), the 'good alternative' (the Greater Thornbird; resists poorly and high burdens cause death) and the 'bad alternative' (the Little Thornbird; resists efficiently and moderate to high burdens cause death).

The breeding season of these host species ranges from October to March. In this study we analysed data from 8 breeding seasons, corresponding to the dates 2006/2007, 2007/2008, 2008/2009, 2009/2010, 2012/2013, 2013/2014, 2014/2015, 2015/2016.

Data collection

Once a week, the entire 40-ha forest patch was thoroughly surveyed searching for active nests (of all bird species). This was consistently done during the 8 breeding seasons. When eggs were identified, the nest was marked and systematically followed weekly until all nestlings left or died. Every week, the nestlings of each brood (of all bird species) were examined in search of external parasites following procedures described in detail by Manzoli et al. (2013). Larvae of '*P. torquans* c. A.' detected were counted and their stage recorded as L1 (<4mm), L2 (4 – 7mm) and L3 (>7mm). For this study, we only used the data from the period of the breeding season in which the three species of interest are expected to be breeding (weeks 5 to 25 from the September [spring] equinox; Figure 1).

Climatic data were obtained from the closest meteorological station. The climatic variables used in the analysis were weekly precipitation (mm) and weekly temperature (minimum, maximum and mean), as they were previously found to be significantly associated with '*P. torquans* c. A.' occurrence and abundance (Antoniazzi et al. 2011; Manzoli et al. 2013).

Statistical approach

The analyses were done using Generalized Linear Mixed Models (with Poisson distribution). The software used was R 3.2.3 (the R-Project for Statistical Computing; <http://www.R-project.org>) and the specific packages used were *glmmTMB*, function *glmmTMB* (Brook et al., 2017).

Three models were constructed. The first assessed whether the use of bad alternative hosts (Little Thornbirds) depended on the availability of optimal hosts (Great Kiskadees). The second model evaluated if the use of bad hosts was a function of the availability of good alternatives (Greater thornbirds). Finally, a third model assessed how availability of optimal hosts influenced the preference of good alternative hosts.

The study unit was the whole bird community present at the study area at a given week (w_i). The response variables were the mean burdens of first instar larvae on nestlings of bad alternative hosts at a given week (first and second model), or of good alternatives (third model). This was estimated for each week by dividing the total number of L1 on all nestlings of the focal species by the total number of nestlings of that species present at that moment (i.e. $baL1/ baN_{w_i}$ or $gaL1/ gaN_{w_i}$). We used the count of L1 and not all instars because L1 represent recent infections (complete larval development takes approximately 4 days), and also because the success of larval development differs across species (e.g. most L1 fail to progress to subsequent instars in Little Thornbirds).

The independent variable of interest was the availability of optimal hosts (first and third models) or good alternatives (second model). This was estimated by counting the number of broods present at the study site (40-ha forest patch) during a given week ($ohBroods_{w_i}$ or $gaBroods_{w_i}$). To take into account that the effect of host availability may depend on the parasite abundance in the area (host demand), the interaction between $oh/gaBroods_{w_i}$ and the total count of L1 in the whole bird community at a given week ($tL1_{w_i}$) was included, where $tL1_{w_i}$ is used as a proxy of parasite abundance (a large number of $tL1_{w_i}$ means that many gravid female flies were seeking hosts recently). This proxy is more precise and informative than using prevalence of infected nestlings or broods, taking into account that a gravid female of '*P. torquans* c. A.' may lay from 1 to 8 clutches of eggs, therefore being able of parasitising a single brood or several ones (Saravia-Pietropaolo et al. 2018). Additional independent variables were included to adjust for potential confounding. These were weekly precipitation (1, 2 or 1+2 weeks previously); minimum, maximum and mean weekly temperatures (same lags); count of broods of other bird species (potential hosts or non-hosts that are present simultaneously); and week of the breeding season (continuous variable, ranging from 1 to 40). All models included the random intercept 'breeding season ID', to account for the lack of independence of observations of the same breeding season.

Model selection and comparison was carried out in a stepwise manner using Akaike information criteria (AIC) (Burnham et al. 2011). All models with AIC values no greater than 5 units compared to the best model were considered. Using information about the AIC values, the selected models were weighed, and then the multimodal inference was done using the weighed mean of the β coefficient and its standard error. The terms that were considered significant were those with a coefficient's 95% confidence interval that did not include 0.

Results

In the 8 breeding seasons, the species of interest were recorded in 191 community-weeks, yielding data from 167 broods of Little Thornbird, 122 of Great Kiskadee and 79 of Greater Thornbird. Along the breeding season, the three species of interest occurred simultaneously from week 5 (mid October) to week 25 (early March) (Figure 1). Observations collected before week 5 or after week 25 were not used for the analyses.

None of the potential confounders assessed remained in the final models. The final first model (Table 1) showed that the burdens in bad alternative hosts (Little Thornbird) were negatively associated with the availability of optimal hosts (Great Kiskadee), and the magnitude of this association depended on the parasite abundance in the community (Figure 2a). For example, when the host demand in the bird community was low (i.e. low parasite abundance, $<10 tL1_{w_i}$), bad alternative hosts were parasitised only in the absence of optimal hosts, and at very low burdens. At higher levels of host demand, burdens in the bad alternative host were moderate to high only if optimal hosts were absent.

The same pattern was observed in the second model (Table 2). Availability of good alternative hosts (Greater Thornbird) reduced substantially the probability of infection in bad alternatives, and the strength of this association was related to host demand (Figure 2b). Regarding infection of good alternative hosts as a function of optimal host occurrence, the effect of availability of optimal hosts was less marked, when compared with the magnitude of the associations observed in the other two models, although at high host demand levels ($\sim 40 tL1_{w_i}$), each brood of optimal host reduces the probability of parasitism in good alternatives by 20% (Table 3, Figure 2c).

Discussion

Host selection (= choice or preference) is a trait of high relevance for the study of infection dynamics, which has drawn much attention for parasitoids and vectors (e.g. Henry et al., 2009; Campbell et al., 2013), but which has been often neglected for other parasites. Parasites differ in their ability to drive the encounter of hosts. While some parasites rely on contact between hosts or passive encounter of a free-living stage to reach compatible hosts, motile and vector-borne parasites are able to find hosts in a less random manner. Discriminating among hosts provides the opportunity of maximising fitness by selecting the most suitable options. Little is known about how parasites select hosts or the degree to which such choices are linked to their success, but some empirical evidence supports the notion that host preference is adaptive. Ectoparasitic mites (*Spinturnix* spp.) are more attracted by female and subadult bats than by male bats, and they survive longer in the former, indicating an adaptive selection of the more beneficial type of host (Christe et al. 2007). Similar evidence of host preference in relation with host competence was observed in multi-host systems. Cercariae of trematodes in the family Plagiorchiidae were put in selection chambers to choose among tadpoles of four host species, and the choice pattern observed was negatively correlated with the level of resistance of each host species. The preferred host species was that with the greatest proportion of metacercariae encysted, whereas the least chosen host was the one in which most cercariae failed (Sears et al. 2012). In the *Philornis*- multihost system, larvae that feed on the main host have higher chances of surviving than those that parasitise alternative hosts (Manzoli et al. 2018).

Most previous work on host selection has been conducted in experimental settings, under unnatural circumstances and providing limited opportunities. These experiments can establish differential attractiveness among hosts, but are unable to evaluate the degree to which parasites can reliably differentiate among an entire assemblage of alternative hosts in natural contexts. The *Philornis*- multihost system provides the rare opportunity to evaluate host attractiveness in the real world (for a detailed description of the advantages of this system see Suppl. Mat. in Manzoli et al. 2018). It is noteworthy that the longitudinal studies conducted here not only provide correlational evidence (very strong associations between putative cause and alleged effect), but also demonstrate robust temporal coherence, and the analyses controlled for potential confounders that may cause spurious associations (Höfler 2005).

Gravid *Philornis* females can fly relatively long distances seeking bird broods that could be hosts of their larvae. Being the encounter the result of such an active search, there is opportunity for evolving capacity to discriminate among hosts and select the ones which maximise larval fitness (Forbes et al. 2017). Our results showed that, in this system, the parasite has an enormous ability to discriminate between hosts of differing quality, and that host selection depends on the structure of the host assemblage and on the size of the parasite population (host demand). The probability and intensity of parasitism in alternative hosts increased as better hosts were less available, especially with growing host demand. It is worth highlighting that the results observed were drastic: bad hosts were virtually not used at all when better alternatives were sufficiently available, even at high host demand levels.

Another study in a different system did not find such context-dependency of host selection. Under experimental conditions, motile cercariae of the trematode *Ribeiroia ondatrae* were attracted differentially by five alternative hosts (Johnson et al. 2019). Unlike the experiment by Sears et al. (2012), the attractiveness was not correlated with host competence. The most preferred host was the bullfrog (*Rana catesbiana*), a non-native and large-bodied species in which most cercariae failed to encyst. The attractiveness of the most competent host in that experiment, *Peudacris regila*, was not altered by variations in the assemblage composition, including presence/absence of bullfrogs. However, bullfrog presence decreased infection for the other 3 alternative host species, which is in coincidence with the effect of Great Kiskadees on Thornbirds observed in this work.

In response to environmental variability, parasites can evolve from generalism to specialism or vice versa (Lievens et al. 2018). Dietary specialisation is predicted to evolve when the fitness obtained from feeding on different resources (i.e. hosts) is lower than that obtained by consuming (i.e. parasitising) a limited subset, whereas generalism should evolve when differences in energetic gains between resources are not large (Lyimo

& Ferguson, 2009). The robust dataset analysed here shows that specificity may also be plastic within individuals, swinging conveniently between generalism and specialism depending on the structure of the host community. In presence of sufficient broods of optimal hosts, '*P. torquans* c. A.' behaves as a specialist, being found almost exclusively on Great Kiskadees. When the optimal host is absent or very little available, however, the parasite can broaden its host range, beginning by choosing the most suitable options, leaving bad hosts for moments when good alternatives cannot be found. This sort of facultative generalism is highly advantageous for the parasite, as it may accommodate its choice so it uses suboptimal resources only at times when it is profitable. In the study area, Great Kiskadees are present only during a limited window of time, while Little Thornbirds (bad alternative hosts) breed actively for much longer (Fig. 1). Hence, being able to decide using bad hosts only when more competent ones are missing may prove essential for the parasite to be able to persist and thrive in this ecosystem.

From the host's perspective, the degree of host selection observed in this system may favour the evolution of resistance. The strategy of tolerance of Great Kiskadees appears to be very successful (Manzoli et al. 2018), but it implies that this host is targeted and therefore highly exposed to the parasite. Resisting is highly costly, but it may be beneficial not only because it would reduce the parasite burden, but also because of its potential to dissuade host selection, as parasites would be driven to select hosts in which their fitness is maximised.

The level of host discrimination observed for *Philornis* flies rivals the cognitive decisions made by vertebrates when selecting their food items, their refuge or their partners. Further, it is in line with the optimal foraging theory (Pyke et al. 1977) which predicts that foragers will be selective when a high-quality food type is abundant, but less selective when that high-quality item is scarce (Emlen 1966, Stephens and Krebs 1986). Underlying these selection processes there are supply-demand dynamics. For example, avian parents should include lower quality foods in the nestling diet only when brood demand is higher than expected or when there is a shortage of food availability (Wright et al. 1998; Sauter et al. 2006). Similarly, in our system, *Philornis* flies select low quality hosts when host demand is high and better hosts are lacking.

The remarkable capability of '*P. torquans* c. A.' of discriminating among hosts should be enabled by a substantial amount of time and energy dedicated for host searching, coupled by cues that allow reliable differentiation among hosts. It is noteworthy that the parasite can definitely distinguish between Little Thornbird (bad alternative host) and Greater Thornbird (good alternative host), which is outstanding considering that both *Phacellodomus* spp. are phylogenetically very close to each other, and both build nests of identical materials and similar shape. The mechanisms of host selection by dipteran parasites have been studied mainly for haematophagous species. The cues involved in them are thermal, visual and chemical (Takken and Knols, 2010). Nonetheless, chemical cues seem to prevail in highly selective species (Jossart et al., 2019). *Anopheles gambiae* is highly anthropophilic, and this preference is driven by specific components found in human sweat (Braks et al., 2001; Zwiebel & Takken, 2004) and skin microbiota (Verhulst et al., 2009). Variability in the chemical composition of the sweat determines differential attractiveness among humans (Smallegange et al., 2011). Likewise, species of *Glossinia* (tse-tse fly) show variable attraction to different odoriferous compounds (Torr & Solano, 2010). Oviposition site selection by *Glossina brevipalpis* was found to be independent of the salinity and pH of the substrate, and instead was driven by presence of con-specific or hetero-specific puppae (Renda et al., 2016), which would be mediated by odoriferous compounds (Saini et al., 1996). Chemical cues have also been shown to drive host selection in other arthropods, such as ticks (Ferreira et al., 2019).

Our findings have important implications for the epidemiology of multi-host parasites, specifically for community scale patterns of disease transmission. The concept of facultative generalism adds pertinent elements to the notion of a 'dilution effect' and the relevance of biodiversity on infection dynamics (Schmidt & Ostfeld 2001). Shifts in biodiversity and community composition can influence the likelihood of a parasite or vector encountering a highly competent host species rather than a low-quality host. The 'dilution effect' implies that greater biodiversity results in less encounters with highly competent hosts and, therefore, in less parasite success and transmission (Schmidt & Ostfeld 2001). However, if parasites or vectors selectively infect the

most competent hosts as observed in this study, the presence of alternative ones (which increases with biodiversity) may have little effect on transmission (Levine et al., 2017). Nevertheless, when the alternative hosts are humans, domestic animals, or an endangered wildlife species, the plastic host selection observed here, influenced by biodiversity, can be of high importance for public health, animal husbandry, and biodiversity conservation, respectively. For example, humans might not be targeted by a given parasite as long as more suitable hosts are available. This kind of 'deflection effect' is related to the 'dilution effect' because they are both a function of biodiversity. More biodiversity may dilute the infection of highly competent hosts, or it can also divert it from a host of interest. Reductions in biodiversity might therefore cause that humans (or domestic animals, or endangered species) become increasingly chosen by parasites. Thus, although the abundance of a given parasite is highly dependent on the abundance of its 'reservoir' host (e.g. mean abundance of '*P. torquans* c. A.' is low in the community in years with few Great Kiskadee broods; Manzoli et al. 2013), lack of main hosts would paradoxically increase infection risk in an alternative host of interest.

Acknowledgements

All procedures conducted in this study comply with the current National and Provincial laws, and were approved by the University Bioethics Committee. This study was funded by Wildlife Conservation Society (Wildlife Health Fund, www.wcs.org), Morris Animal Foundation (www.morrisanimalfoundation.org, Grant No. D08ZO-304), and CONICET (www.conicet.gov.ar, Grant No. PIP11220100100261). We would like to thank Dr. Martín de la Peña for the ornithological. The authors declare no competing interests.

References

- Antoniazzi, L.R., Manzoli, D.E., Rohrmann, D., Saravia, M.J., Silvestri, L. & Beldomenico, P.M. (2011). Climate variability affects the impact of parasitic flies on Argentinean forest birds. *J. Zool.* , 283, 126–134.
- Arendt, W. (1985). Philornis ectoparasitism of pearly-eyed thrashers. I. Impact on growth and development of nestlings. *Auk* , 102, 270–280.
- Auld, S.K.J.R., Searle, C.L. & Duffy, M.A. (2017). Parasite transmission in a natural multihost–multiparasite community. *Philos. Trans. R. Soc. Lond., B Biol. Sci.* , 372, 20160097.
- Best, A., White, A. & Boots, M. (2014). The coevolutionary implications of host tolerance: coevolution of tolerance. *Evolution* , 68, 1426–1435.
- Braks, M.A.H., Meijerink, J. & Takken, W. (2001). The response of the malaria mosquito, *Anopheles gambiae* , to two components of human sweat, ammonia and l-lactic acid, in an olfactometer. *Physiol. Entomol.* , 26, 142–148.
- Campbell, R., Thiemann, T.C., Lemenager, D. & Reisen, W.K. (2013). Host-Selection patterns of *Culex tarsalis* (Diptera: Culicidae) determine the spatial heterogeneity of West Nile Virus enzootic activity in Northern California. *J. Med. Entomol.* , 50, 1303–1309.
- Christe, P., Glaizot, O., Evanno, G., Bruyndonckx, N., Devevey, G., Yannic, G., et al. (2007). Host sex and ectoparasites choice: preference for, and higher survival on female hosts. *J. Anim. Ecol.* , 76, 703–710.
- Combes, C. (1991). Ethological Aspects of Parasite Transmission. *Am. Nat.* , 138, 866–880.
- Couri, M.S. (1999). Myiasis caused by obligatoty parasite. Ia Philornis Meinert (Muscidae). In: *Myiasis in Man and Animals in the Neotropical Region* (eds. Guimarães, J.H. & Papavero, N.). Editora Plêiade, São Paulo, pp. 51–70.
- Dudaniec, R.Y., Kleindorfer, S. & Fessl, B. (2006). Effects of the introduced ectoparasite *Philornis downsi* on haemoglobin level and nestling survival in Darwin's Small Ground Finch (*Geospiza fuliginosa*). *Austral Ecol.* , 31, 88–94.
- Emlen, J.M. (1966). The role of time and energy in food preference. *Am. Nat.* , 100, 611–617.

- Fenton, A., Streicker, D.G., Petchey, O.L. & Pedersen, A.B. (2015). Are all hosts created equal? Partitioning host species contributions to parasite persistence in multihost communities. *Am. Nat.* , 186, 610–622.
- Ferreira, L.L., Sarria, A.L.F., de Oliveira Filho, J.G., de Silva, F. de O., Powers, S.J., Caulfield, J.C., *et al.* (2019). Identification of a non-host semiochemical from tick-resistant donkeys (*Equus asinus*) against *Amblyomma sculptum* ticks. *Ticks Tick Borne Dis.* , 10, 621–627.
- Forbes, M.R., Morrill, A. & Schellinck, J. (2017). Host species exploitation and discrimination by animal parasites. *Philos. Trans. R. Soc. Lond., B Biol. Sci.* , 372, 20160090.
- Gervasi, S.S., Civitello, D.J., Kilvitis, H.J. & Martin, L.B. (2015). The context of host competence: a role for plasticity in host–parasite dynamics. *Trends Parasitol.* , 31, 419–425.
- Henry, L.M., Ma, B.O. & Roitberg, B.D. (2009). Size-mediated adaptive foraging: a host-selection strategy for insect parasitoids. *Oecologia* , 161, 433–445.
- Höfler, M. (2005). The Bradford Hill considerations on causality: a counterfactual perspective. *Emerg. Themes Epidemiol.* , 2, 1–9.
- Johnson, P.T.J., Calhoun, D.M., Riepe, T.B. & Koprivnikar, J. (2019). Chance or choice? Understanding parasite selection and infection in multi-host communities. *Int. J. Parasitol.* , 49, 407–415.
- Jossart, Q., Terrana, L., De Ridder, C., Eeckhaut, I., Monteyne, D. & Caulier, G. (2019). To see or to smell: the role of vision in host-recognition by an ectoparasitic crab. *Symbiosis* .
- Leggett, H.C., Buckling, A., Long, G.H. & Boots, M. (2013). Generalism and the evolution of parasite virulence. *Trends Ecol. Evol.* , 28, 592–596.
- Lievens, E.J.P., Perreau, J., Agnew, P., Michalakis, Y. & Lenormand, T. (2018). Decomposing parasite fitness reveals the basis of specialization in a two-host, two-parasite system. *Evol. Lett.* , 2, 390–405.
- Löwenberg-Neto, P. (2008). The structure of the parasite–host interactions between *Philornis* (Diptera: Muscidae) and neotropical birds. *J. Trop. Ecol.* , 24, 575–580.
- Lyimo, I.N. & Ferguson, H.M. (2009). Ecological and evolutionary determinants of host species choice in mosquito vectors. *Trends Parasitol.* , 25, 189–196.
- Magnusson, A., Skaug, H., Nielsen, A., Berg, C., Kristensen, K., Maechler, M., *et al.* (2017). Package ‘glmmTMB’. R Package Version 0.2. 0.
- Manzoli, D.E., Antoniazzi, L.R., Saravia, M.J., Silvestri, L., Rorhmann, D. & Beldomenico, P.M. (2013). Multi-level determinants of parasitic fly infection in forest passerines. *PLoS ONE* , 8, e67104.
- Manzoli, D.E., Saravia-Pietropaolo, M.J., Antoniazzi, L.R., Barengo, E., Arce, S.I., Quiroga, M.A., *et al.* (2018). Contrasting consequences of different defence strategies in a natural multihost–parasite system. *Int. J. Parasitol.*, 48, 445 - 455 .
- Martin, L.B., Burgan, S.C., Adelman, J.S. & Gervasi, S.S. (2016). Host competence: an organismal trait to integrate immunology and epidemiology. *Integr. Comp. Biol.* , 56, 1225–1237.
- Monje, L.D., Quiroga, M., Manzoli, D., Couri, M.S., Silvestri, L., Venzal, J.M., *et al.* (2013). Sequence analysis of the internal transcribed spacer 2 (ITS2) from *Philornis seguysi* (García, 1952) and *Philornis torquans* (Nielsen, 1913) (Diptera: Muscidae). *Syst. Parasitol.* , 86, 43–51.
- de la Peña, M.R. (2016). *Aves y características de la reserva natural Martín R. de la Peña* . 1st edn. Editorial de la Universidad Nacional del Litoral, Santa Fe, Argentina. pp: 411
- de la Peña, M.R., Beldomenico, P.M. & Antoniazzi, L.R. (2003). Pichones de aves parasitados por larvas de *Philornis sp.* (Diptera: Muscidae) en un sector de la Provincia biogeográfica del Espinal de Santa Fe, Argentina. *FAVE- Cs.Vet.* , 2, 141–146.

- Pyke, G.H., Pulliam, H.R. & Charnov, E.L. (1977). Optimal Foraging: a selective review of theory and tests. *Q. Rev. Biol.* , 52, 137–154.
- Quiroga, M.A., Monje, L.D., Arrabal, J.P. & Beldomenico, P.M. (2016). New molecular data on subcutaneous *Philornis* (Diptera: Muscidae) from southern South America suggests the existence of a species complex. *Rev. Mex. Biodivers.* , 87, 1383–1386.
- Råberg, L., Graham, A.L. & Read, A.F. (2009). Decomposing health: tolerance and resistance to parasites in animals. *Philos. Trans. R. Soc. Lond., B Biol. Sci.* , 364, 37–49.
- Renda, S., De Beer, C.J., Venter, G.J. & Thekisoe, O.M.M. (2016). Evaluation of larviposition site selection of *Glossina brevipalpis* . *Vet. Parasitol.* , 215, 92–95.
- Rigaud, T., Perrot-Minnot, M.-J. & Brown, M.J.F. (2010). Parasite and host assemblages: embracing the reality will improve our knowledge of parasite transmission and virulence. *Proc. Biol. Sci.* , 277, 3693–3702.
- Saini, R.K., Hassanali, A., Andoke, J., Ahuya, P. & Ouma, W.P. (1996). Identification of major components of larviposition pheromone from larvae of tsetse flies *Glossina morsitans morsitans* Westwood and *Glossina morsitans centralis* Machado. *J. Chem. Ecol.* , 22, 1211–1220.
- Saravia-Pietropaolo, M.J., Arce, S.I., Manzoli, D.E., Quiroga, M. & Beldomenico, P.M. (2018). Aspects of the life cycle of the avian parasite *Philornis torquans* (Diptera: Muscidae) under laboratory rearing conditions. *Can. Entomol.* , 150, 317–325.
- Sauter, A., Bowman, R., Schoech, S.J. & Pasinelli, G. (2006). Does optimal foraging theory explain why suburban Florida scrub-jays (*Aphelocoma coerulescens*) feed their young human-provided food? *Behav. Ecol. Sociobiol.* , 60, 465–474.
- Schmid-Hempel, P. (2011). *Evolutionary parasitology: the integrated study of infections, immunology, ecology, and genetics* . Oxford University Press, Oxford.
- Schmidt, K. & Ostfeld, R.S. (2001). Biodiversity and the dilution effect in disease ecology. *Ecology* , 82, 609–619.
- Sears, B.F., Schlunk, A.D. & Rohr, J.R. (2012). Do parasitic trematode cercariae demonstrate a preference for susceptible host species? *PLoS ONE* , 7, e51012.
- Smallegange, R.C., Verhulst, N.O. & Takken, W. (2011). Sweaty skin: an invitation to bite? *Trends Parasitol.* , 27, 143–148.
- Stephens, D. & Krebs, J. (1986). *Foraging theory* . Princeton University Press, New Jersey.
- Takken, W. & Knols, B. (Eds.). (2010). *Olfaction in vector-host interactions* . Ecology and control of vector-borne diseases. 1st edn. Wageningen Academic Publishers, Netherlands. pp 437
- Torr, S. & Solano, P. (2010). Olfaction in *Glossina* - host interactions: a tale of two testse. In: *Olfaction in vector-host interaction* , Ecology and control of vector-borne diseases (eds. Takken, W. & Knols, B.). Wageningen Academic Publishers, Netherlands, pp. 265–289.
- Verhulst, N.O., Beijleveld, H., Knols, B.G., Takken, W., Schraa, G., Bouwmeester, H.J., *et al.* (2009). Cultured skin microbiota attracts malaria mosquitoes. *Malar. J.* , 8, 302.
- Woolhouse, M.E.J. (2001). Population biology of multihost pathogens. *Science* , 292, 1109–1112.
- Wright, J., Both, C., Cotton, P.A. & Bryant, D. (1998). Quality vs. Quantity: energetic and nutritional trade-offs in parental provisioning strategies. *J. Anim. Ecol.* , 67, 620–634.
- Zwiebel, L.J. & Takken, W. (2004). Olfactory regulation of mosquito–host interactions. *Insect Biochem. Mol. Biol.* , 34, 645–652.

Table 1 : Mean burden of first instar larvae of *Philornis*(L1) in bad alternative hosts (Little Thornbird) as a function of the number of co-existing optimal host (Great Kiskadee) broods and total L1 in the community.

Parameter	Coefficient
<i>Intercept</i>	-0.4274
$tL1_{w0}$	0.0385
$gaBroods_{wi}$	-0.6443
$tL1_{w0} * gaBroods_{wi}$	-0.0169

$baL1/baN_{wi}$: mean L1 burden in bad alternative hosts at a given week

$tL1_{wi}$: total count of L1 in all nestlings present in the studied community at a given week

$ohBroods_{wi}$: total count of broods of optimal hosts in the studied community at a given week

Table 2 : Mean burden of first instar larvae of *Philornis*(L1) in bad alternative hosts (Little Thornbird) as a function of the number of co-existing good alternative host (Greater Thornbird) broods and total L1 in the community.

$baL1/baN_{wi}$: mean L1 burden in bad alternative hosts at a given week

$tL1_{wi}$: total count of L1 in all nestlings present in the studied community at a given week

$gaBroods_{wi}$: total count of broods of good alternative hosts in the studied community at a given week

Table 3 : Mean burden of first instar larvae of *Philornis*(L1) in good alternative hosts (Greater Thornbird) as a function of the number of co-existing optimal hosts (Great Kiskadee) broods and total L1 in the community.

Parameter	Coefficient
<i>Intercept</i>	-1.3771
$tL1_{w0}$	0.0635
$ohBroods_{wi}$	0.0328
$tL1_{w0} * ohBroods_{wi}$	-0.0073

$gaL1/gaN_{wi}$: mean L1 burden in good alternative hosts at a given week

$tL1_{wi}$: total count of L1 in all nestlings present in the studied community at a given week

$ohBroods_{wi}$: total count of broods of optimal hosts in the studied community at a given week

Figure 1

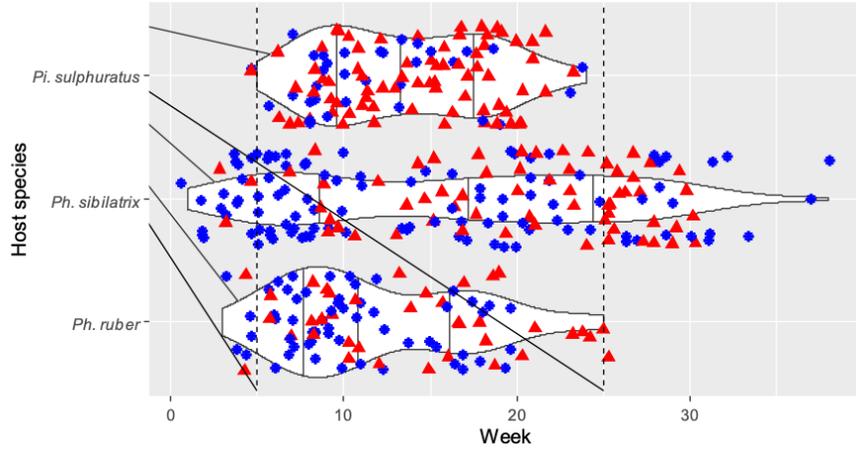
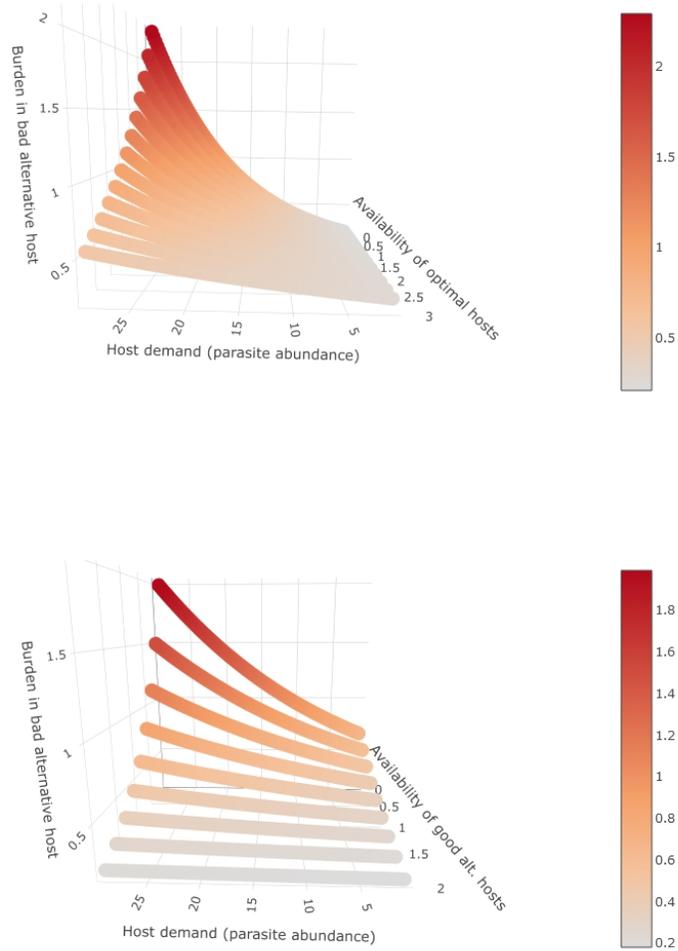
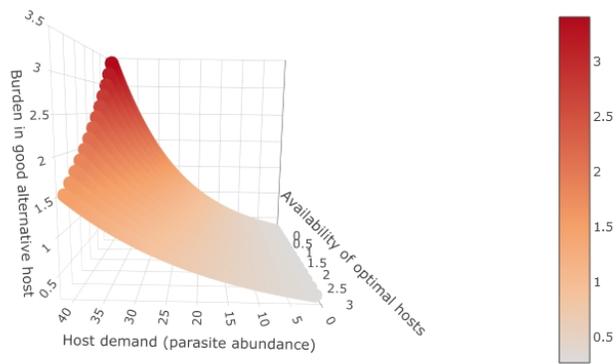


Figure 2





Legend to figures:

Figure 1: Temporal distribution of broods of each focal host species along the breeding season. Data from 8 consecutive breeding seasons. Blue dots and red triangles represent non-infected and infected broods, respectively. Week 0 refers to the week of the spring equinox (September in the southern hemisphere). *Pi. sulphuratus* (Great Kiskadee) is the optimal host, *Ph. sibilatrix* (Little Thornbird) the bad alternative host and *Ph. ruber* (Greater Thornbird) the good alternative host.

Figure 2: Predicted burdens of first instar larvae (L1) on bad alternative hosts (Little Thornbird) (**a** and **b**) or good alternative hosts (Greater Thornbird) as a function of host demand (parasite abundance) and the co-existing number of broods of optimal hosts (Greater Kiskadee) (**a** and **c**) and good alternative hosts (**b**).

