Opposing effects of competitive exclusion determine invasion outcomes and structure ant assemblages

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Abstract

Insight into how species' phenotypic differences shape coexistence is key to understanding the processes structuring, maintaining and threatening biodiversity. In line with modern coexistence theory, increasing studies on plants show that traits may not only confer niche differences fostering the coexistence of dissimilar species, but also competitive advantages promoting the exclusion of weaker competitors. These variable relationships between traits and the outcomes of interspecific competition, and particularly their effects on assemblage structure, are relatively unexplored for animals in natural settings. Here we show that the abundance of an invasive ant as well as the composition and structure of native ground-foraging ant assemblages in a tropical grassland are largely attributable to differences in the morphological, dietary, physiological and behavioural traits between invasive and native species. Crucially, we find that individual traits vary in association with niche differences and competitive advantages, which have opposing effects on competitive exclusion. Competition along these separate trait axes consequently generates contrasting patterns of overdispersion and clustering in the functional structure of invaded assemblages. Our results highlight that different traits across the phenotypic spectrum may distinctly influence stabilizing and equalizing mechanisms of coexistence, ultimately modulating the role and effect of competition in structuring biodiversity.

Keywords

Assembly, coexistence, competition, competitive ability, functional diversity, functional trait, limiting similarity, niche

Introduction

Interspecific competition has long been presumed (Darwin, 1859) and is increasingly shown to be a fundamental force structuring and maintaining biodiversity (Scheffer & Van Nes, 2006; Levine & HilleRisLambers, 2009; Kunstler et al., 2016). Competitive interactions may also modulate the consequences of climate change, habitat alteration and biological invasions on ecological communities (Sanders et al., 2003; Violle et al., 2010; Legault et al., 2020). An important goal in ecology is therefore to accurately predict the outcome of interspecific competition from general and measurable properties of species, such as their functional traits (McGill et al., 2006). However, this is seldom achieved empirically because the ways by which species' trait differences affect competitive outcomes are not straightforward.

Ecological theory suggests that trait differences can actually drive two contrasting effects on competitive outcomes between species (Chesson, 2000; HilleRisLambers et al., 2012; D'Andrea & Ostling, 2016). On the one hand, differences in species' trait values may facilitate the use of different ecological *niches*, such as different resources or climatic windows. These 'niche effects' of trait differences promote coexistence between species with dissimilar trait values and drive competitive exclusion among species with similar trait values (*sensu* 'limiting similarity'; MacArthur & Levins, 1967). On the other hand, differences in trait values may also distinguish species in terms of their *competitive abilities* in a competition hierarchy, such as in the exploitation of a shared resource or climatic window. Contrary to niche effects, these 'competitive effects' of trait differences promote coexistence among species with comparably strong competitive abilities and similar trait values, while driving the exclusion of other species with weaker competitive abilities and dissimilar trait values (Chesson, 2000; Mayfield & Levine, 2010). Importantly, niche and competitive effects of trait differences generate contrasting patterns in the trait structures of species assemblages; species tend to be overdispersed in assemblage trait space when niche effects are at play but clustered when competitive effects are involved (Mayfield & Levine, 2010; D'Andrea & Ostling, 2016).

Burgeoning studies on plant assemblages evidence the importance of trait-based niche and competitive effects as well as their interactions in determining coexistence outcomes and shaping community assembly. While the high diversity of Amazonian forests has been attributed to interspecific niche differentiation across multiple trait axes (Kraft, Valencia & Ackerly, 2009), globally consistent competitive effects linked to species' differences in traits such as wood density, specific leaf area and maximum height have also been identified (Kunstler et al., 2016). Notably, results from competition experiments suggest that both niche and competitive effects may operate simultaneously and associate variably with individual traits or across multidimensional trait space (Kraft, Godoy & Levine, 2015), and differently for morphological versus physiological traits (Pérez-Ramos et al., 2019). In empirical studies of most animals, however, limiting similarity is often presumed to be the sole basis for competitive exclusion. Interspecific trait variation – along and across morphological, dietary, physiological and behavioural axes – is taken to reflect niche differentiation; and attempts to detect the effects of competition in community assembly involve tests for trait overdispersion only (Münkemüller et al., 2020). The potential for trait differences across the phenotypic spectrum to confer competitive advantages as opposed to niche

differences may therefore be underestimated.

Invasions by non-native species represent novel settings for testing the mechanisms which underpin community assembly (Shea & Chesson, 2002). Many differences between the traits of invasive and native plant species, such as leaf-area allocation and growth rate, have been shown to correspond to competitive advantages instead of niche differences (Van Kleunen, Weber & Fischer, 2010). However, the extent to which trait differences between invasive and native animal species promote niche and competitive differences, as well as their consequences for the functional structure of animal assemblages – from overdispersion to clustering – are less understood.

We investigated if niche and competitive effects of trait differences in interspecific competition accounted for the outcomes of an invasion by the Red Imported Fire Ant, *Solenopsis invicta*, and their impacts on the functional structure of ground-foraging ant assemblages in a tropical grassland. To maximize the likelihood of studying ant assemblage structure that was competition-driven, our study design naturally minimized the effects of potential environmental and dispersal assembly processes by focusing on a simple grassland setting with relatively low environmental heterogeneity, and sampling at fine spatial scales well within ant species' dispersal ranges (see Materials and Methods).

The study design facilitated the use of a novel comparative approach. We investigated the extent to which the abundance of the invader *S. invicta* across 61 plots (including multiple plots where it was absent) was explained by its trait differences with the 'observed assemblage' of ant species at each plot, and also with the 'missing assemblage' of ant species that were absent from each plot (but occurring in the species pool). To detect potential niche and competitive effects, trait differences were measured in two ways: as absolute dissimilarities to approximate niche differences, and as hierarchical differences to approximate competitive differences (after Kraft et al., 2015; Kunstler et al., 2016). Confirming weak environmental effects on invasion outcomes as well as assemblage structure, and assuming low barriers to dispersal across the landscape, we then scrutinized relationships between the abundance of *S. invicta* and its trait differences with missing assemblages to understand competitive mechanisms driving the potential exclusion of particular ant species in the presence of *S. invicta*. We likewise scrutinized trait differences between *S. invicta* and observed assemblages to understand mechanisms promoting the abundance of *S. invicta* and the potential coexistence of particular ant species with *S. invicta* (or at the least their persistence in the presence of *S. invicta*).

We further investigated how interspecific competition with the invader *S. invicta* structured ant assemblages by comparing uninvaded and invaded assemblages in terms of the degree of overdispersion or clustering along individual trait axes as well as in multidimensional trait space. Besides being among the first to assess both niche and competitive effects of trait differences in shaping an animal assemblage, our work goes further than previous studies by incorporating a diverse suite of traits spanning species' morphology (seven traits), diet (Trophic Position, proxied by stable isotope ratios of Nitrogen, δ15N), physiology (Critical Thermal Maximum), and behaviour (Interference Ability, the ability of individuals of a species to usurp and defend resources). All

traits were measured as continuous variables directly from multiple individuals of each species captured in field. This work advances our understanding of how interspecific differences across the phenotypic spectrum drive competitive outcomes among animal species, and more broadly shows that trait differences can deterministically shape the responses of biodiversity to biotic disturbances.

Results

As expected from the study design, there was little evidence to suggest that environmental effects influenced invasion outcomes across the 61 plots (Table 1) as well as the species richness and functional structure of the ant assemblages (Supporting information). Instead, the abundance of *S. invicta* was well explained by its average trait differences with ant species in the missing assemblage at each plot (Fig. 1a-c) and those in the observed assemblage at each plot (Fig. 1d-f). Both niche and competitive effects were detected and associated with separate traits (Table 1; Fig. 1).

Only differences in Body Size were broadly associated with niche effects (Table 1). Plots with higher abundances of *S. invicta* contained ant species of dissimilar Body Size from *S. invicta* (Fig. 1f) but were missing species of similar Body Size to *S. invicta*. In contrast, differences in Interference Ability, Trophic Position and Head Width were associated with strong competitive effects. Plots with higher abundances of *S. invicta* contained species of comparable Interference Ability to *S. invicta* and species which had less carnivorous diets and narrower heads than *S. invicta* (Table 1; Fig. 1b,c). Species missing from these plots had far lower (weaker) Interference Ability, more carnivorous diets and wider heads than *S. invicta* (Table 1; Fig. 1e).

Interestingly, differences in Critical Thermal Maximum (CT_{max}) were associated with both niche and competitive effects. Higher abundances of *S. invicta* were significantly associated with the positive effects of *S. invicta*'s competitive advantages and the negative effects of their niche differences with missing assemblages in CT_{max}, with the former driving a stronger effect (Table 1; Fig. 1a). Higher abundances of *S. invcita* were also significantly associated with the negative effects of *S. invicta*'s competitive inferiority and the positive effects of their niche differences with the observed assemblages in CT_{max}, with the former driving a stronger effect (Table 1; Fig. 1d).

Invaded assemblages were marginally lower in species richness ($M_{Uninvaded}$=6.84±1.76, $M_{Invaded}$=5.67±2.01, P=0.02) and significantly more clustered in multidimensional trait space overall (Fig. 2a). Importantly, niche and competitive effects also structured the invaded assemblages along individual trait axes, with contrasting results (Fig. 2). Consistent with competitive effects, invaded assemblages were significantly clustered in Interference Ability and Head Width relative to uninvaded assemblages (Fig. 2b,d). However, consistent with niche effects, invaded assemblages were more overdispersed in CT_{max} than uninvaded assemblages (Fig. 2c).

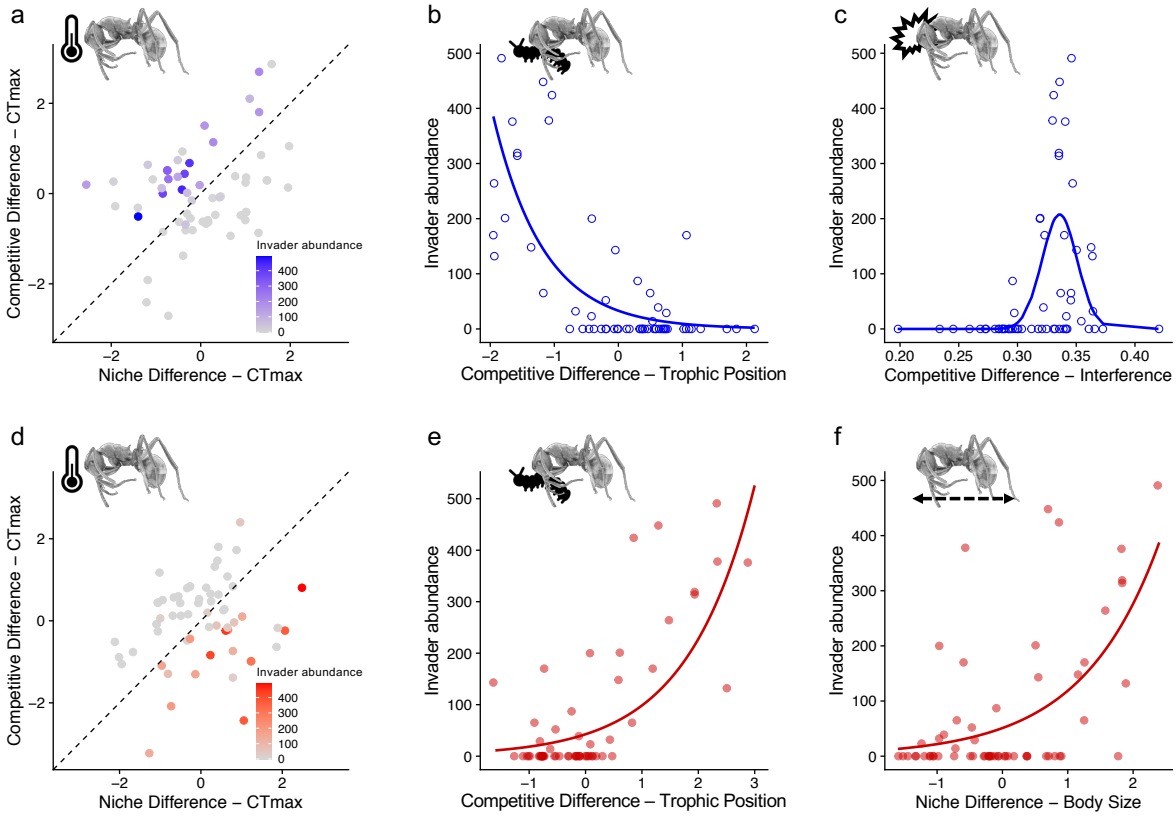

Figure 1. Abundance of the invasive species *S. invicta* as a function of its average trait differences with the 'missing assemblage' of ant species at each plot (**a-c**), and with the 'observed assemblage' of ant species at each plot (**d-f**). Against the missing assemblages, the model for Critical Thermal Maximum (CT_{max}) detected significant effects of both Niche Difference and Competitive Difference, and predicted higher densities of *S. invicta* when Competitive Difference exceeded Niche Difference (**a**). The abundance of *S. invicta* was also significantly higher across plots where missing species tended to be more carnivorous than *S. invicta* (**b**), and where missing species had far poorer Interference Ability than *S. invicta* (**c**). Against the observed assemblages, the model for CT_{max} detected significant effects of both Niche Difference and Competitive Difference, and predicted higher abundances of *S. invicta* when Niche Difference exceeded Competitive Difference (**d**). The abundance of *S. invicta* was also significantly higher across plots where the species present were less carnivorous than *S. invicta* (**e**), and where the species present tended to be more dissimilar in Body Size (i.e., smaller or larger) from *S. invicta* (**f**).

Table 1. Results of trait and environment models for the abundance of the invasive ant *S. invicta* across 61 plots, with standardized coefficients. Trait models explain *S. invicta* abundance as a function of its average trait differences with either the ant species observed at each plot, or the ant species missing from each plot (but occurring in the species pool). Two types of trait differences between *S. invicta* and ant species were measured: Niche Difference and Competitive Difference. Environment models were built for the percentage ground cover (Ground Cover) and mean annual temperature (Temperature) at each plot. These variables were also included as covariates in trait models if they improved model performance.

Model Type	**Model Name**	**AICc**	**Terms**	**β**	***P***
Trait Difference: Invader vs. Observed					
	Head Width	401.6	Competitive Difference	4.5	<0.001***
			Ground Cover	1.77	<0.05*
	CT_{max}	398.1	Competitive Difference	-3.32	<0.001***
			Niche Difference	3.02	<0.001***
	Trophic Position	409.6	Competitive Difference	0.84	<0.05*
	Body Size	411.7	Niche Difference	0.85	<0.05*
Trait Difference: Invader vs. Missing					
	Head Width	409.1	Competitive Difference	-5.01	<0.001***
			Ground Cover	2.09	<0.05*
	CT_{max}	401	Competitive Difference	4.18	<0.001***
			Niche Difference	-3.44	<0.001***
	Interference	396.9	Competitive Difference	3.1	<0.001***
			Competitive Difference^2	-3.17	<0.001***
	Body Size	424.2	Niche Difference	-2.46	<0.05*
	Trophic Position	405.9	Competitive Difference	-1.29	<0.001***
			Temperature	0.62	0.11
Environment					
	Ground Cover	428.5	Ground Cover	0.2	0.79
	Temperature	428.2	Temperature	0.51	0.54

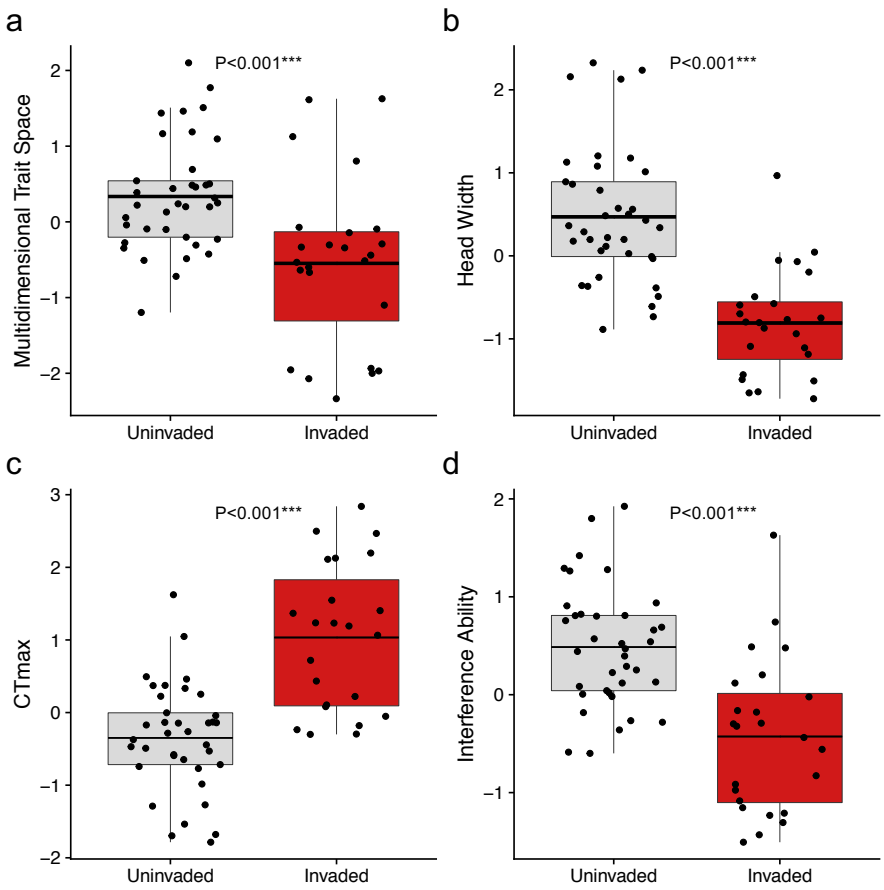

Figure 2. Standardized Effect Sizes (SES) for functional dispersion in Multidimensional Trait Space (**a**) and three separate traits – Head Width (**b**), Critical Thermal Maximum (CT_{max}) (**c**), and Interference Ability (**d**) – across uninvaded and invaded ground-foraging ant assemblages. In comparison with the uninvaded assemblages, ants in the invaded assemblages were significantly more overdispersed in CT_{max} but significantly more clustered in Multidimensional Trait Space, Head Width and Interference Ability. P-values correspond to results from t-tests (**b,c**) and Wilcoxon Mann-Whitney tests (**a,d**); full details in Supporting information.

Discussion

A sound empirical understanding of how trait differences shape interspecific competition is key to explaining community assembly and predicting biodiversity responses. Here, we found that interspecific trait differences largely determined the outcomes of an ant invasion through opposing mechanisms of competitive exclusion (*sensu* Mayfield & Levine, 2010). Specifically, differences between the invader *S. invicta* and resident ant species in separate morphological, dietary, physiological and behavioural traits facilitated two distinct mechanisms: limiting similarity driven by niche effects (MacArthur & Levins, 1967) and competitive hierarchies shaped by the competitive effects (Keddy & Shipley, 1989; Chesson, 2000; Kunstler et al., 2016). These mechanisms well explained the plausible exclusion of some species and the apparent persistence of the invader and other species. Crucially, the two mechanisms acted distinctly on separate traits, and their opposing effects generated contrasting structures among those traits in the invaded assemblages. Our findings cast light on the thus far underestimated role of traits in conferring competitive advantages (as opposed to niche opportunities) to animal species, underscoring their importance to interspecific competition in natural settings. The findings also illustrate how multiple, even opposing assembly mechanisms can simultaneously structure functional diversity.

That both niche and competitive effects played key roles in determining invasion outcomes among ant species suggests that a wider view of competition and coexistence – one extending beyond classical niche theory, incorporating stabilizing and equalizing mechanisms (Chesson, 2000), and which is increasingly well-established for plant assemblages (Kraft et al., 2015; Kunstler et al., 2016) – is likewise applicable to animals. Consistent with limiting similarity, niche differences led to the exclusion, in a symmetrical manner, of species with similar body sizes as the invader *S. invicta,* while promoting coexistence between the invader and species that were sufficiently larger or smaller than itself, potentially via resource partitioning. Nonetheless, niche differences alone did not determine competitive outcomes in the invasion; their stabilizing effects more likely counteracted the demographic effects of species' differences in competitive abilities that promoted exclusion (Chesson, 2000; HilleRisLambers et al., 2012). Furthermore, contrary to the commonly held assumption that trait differences mainly reflect niche partitioning (Mayfield & Levine, 2010), we detected competitive effects associated with multiple traits of ant species. For instance, species with relatively wider heads and more carnivorous diets than *S. invicta* appeared to be excluded where *S. invicta* occurred, while species with relatively narrower heads and more carbohydrate-rich diets than *S. invicta* were observed in its presence. This may have occurred if *S. invicta* depleted arthropod populations and more strongly impacted more carnivorous ant species (as observed by Porter & Savignano, 1990), driving a competition hierarchy in omnivory among native species. Another trait associated with strong competitive effects, as revealed from asymmetrical patterns of exclusion (Fig. 1c) and consequent clustering in assemblage trait structure (Fig. 2d), was interference ability. It has been proposed that interspecific trade-offs in stress tolerance and competitive dominance akin to those in plants (e.g. Grime, 1979) may likewise facilitate coexistence in ant assemblages; such as between species with abilities to monopolize resources and those which can forage over wider thermal ranges (Bestelmeyer, 2000). Given that *S. invicta* possessed the strongest interference ability, and that other ant species in invaded assemblages had dissimilar or higher thermal tolerances than *S. invicta* (Fig. 1d), such dominance-tolerance trade-offs may have been at

play. Further supporting this hypothesis, species with higher CT_{max} but lower interference ability than *S. invicta* were observed recruiting to baits abandoned by *S. invicta* when those baits were heated by sun exposure with changes in cloud cover (Supporting information). Such trade-offs would represent equalizing mechanisms which, in addition to stabilizing mechanisms, promoted coexistence among the invader and other ant species by mitigating interspecific differences in competitive abilities (Chesson, 2000).

The above findings shed light on potential trait-based stabilizing and equalizing mechanisms that may determine the outcomes of competition among ant species and assemblage structure. Rigorous tests for these coexistence mechanisms would require competition experiments measuring demographic parameters such as invasion growth rates (e.g., Kraft et al., 2015); yet such approaches are not readily transferrable to animals (see Terry et al., 2020). Nonetheless, we suggest that the potential for trait differences to reflect niche or competitive effects warrants explicit consideration of both in the structuring of empirical assemblages. We therefore agree with recent calls for community assembly studies to test for patterns of clustering in addition to overdispersion as signatures of competition (D'Andrea & Ostling, 2016; Münkemüller et al., 2020). However, our findings also highlight important limitations of inferring assembly processes exclusively from patterns in multidimensional trait space.

Of direct relevance to observational studies, our results show that inferences about underlying ecological processes from patterns in the multidimensional trait structure of assemblages may be misguided if distinct processes with opposing effects act on different traits simultaneously. Here, potentially important effects of niche partitioning in body size (Fig. 1f) and thermal tolerances (Fig. 2c) in structuring the invaded assemblages were not well reflected in their strongly clustered multidimensional trait space (Fig. 2a). This was possibly due to the overwhelming, opposing effects of hierarchical competition along other trait axes such as head width (Fig. 2b) and interference ability (Fig. 2d). Such effects from multiple interactive and simultaneous assembly processes on functional structure are likely underestimated, especially where biotic interactions are concerned (but see Fortunel et al., 2016). Yet these distinct competitive mechanisms have significant implications that reach beyond the fundamental understanding of community assembly. For instance, they can strongly determine biodiversity-ecosystem function relationships. While niche differences and resource partitioning drive complementarity effects (Godoy et al., 2020), competitive hierarchies drive selection effects; each has a vastly different outcome for ecosystem functions (see Loreau & Hector, 2001).

Overall, our study uncovered new insights into the drivers and consequences of an ant invasion, which were consistent with the predictions of modern coexistence theory (Chesson, 2000; Mayfield & Levine, 2010). Distinguishing between the contributions of traits to niche and competitive differences among species revealed alternative paths to competitive exclusion by invasion (MacDougall et al., 2009), and potential trait-based stabilizing and equalizing mechanisms structuring invaded assemblages. These specific mechanisms are useful for understanding invasion responses and assembly processes at fine spatial scales. The broader implication for empirical work is to embrace the potential for different traits across the phenotypic spectrum to reflect the

varying role and effect of competition in structuring biodiversity.

Materials and Methods

Sampling ant assemblages and environmental variables

The study was conducted in two reserves of open grassland in Hong Kong which have been protected for >35 years, and which contain networks of exposed grass bunds (width ≤5 m) separating individual ponds (Wong et al., 2020). Pilot surveys from 2015 to 2017 recorded colonies of *S. invicta* present at high densities at multiple locations (Wong et al., 2020). From April to September 2018, we systematically characterized the ground-foraging ant assemblages at 61 plots (each a 4 x 4 m quadrat; ≥25 m between adjacent plots) using pitfall traps followed by observations at baits. We intentionally sampled at this fine spatial scale to enhance the detection of patterns driven by biotic interactions, since S*. invicta* and most ant species in the region forage within 5 m of their nests (Eguchi, Bui & Yamane, 2004; Weeks, Wilson & Vinson, 2004). For the same reasons, the minimum distance of 25 m between adjacent plots facilitated independent observations. The maximum distance between any two plots was 4 km; at this spatial scale, the effects of dispersal limitation were very likely minimized as all species disperse via flying alates (Helms, 2018).

In each plot, six pitfall traps (diameter: 5.5 cm) were installed to sample the ants over a period of 48 hrs. Baits were then installed within 72 hrs from the retrieval of the pitfall traps. Between 1000-1500 hrs, five bait stations were installed in each plot, each comprising a slice of chicken sausage (diameter: 20 mm; height: 2 mm) on a white plastic disc (diameter: 5 cm) flushed with the ground. The sausage bait contained trophic resources required by most ants: proteins, lipids, carbohydrates and sodium (Calcaterra et al., 2010). Each baiting session lasted 40 min, during which photographs were taken with a digital camera in 5 min intervals; these were subsequently used to determine the species recruiting, their abundances, and interspecific interactions (see 'Behavioural trait: Interference Ability' below). The mean ground surface temperature around each bait during the 40 min session was recorded with a thermogun. In pilot trials, the 40-min duration allowed for competitive interactions to reach unequivocal outcomes and for baits to be monopolized by single species. After each bating session, live workers of all species encountered were collected and used for dietary and physiological trait measurements in the laboratory (below). Specimens collected from pitfall traps were used for morphological measurements, confirming species identities, and determining the occurrences of species in plots. While several species collected in pitfall traps were not recorded at baits (but not vice versa), many of these were hypogaeic. Given that our study aimed to investigate competition, we focused the analyses on the pool of species recorded at baits as these best represented the ground-foraging ant assemblage limited by common trophic (and other) resources.

At each plot, we also estimated the percentage of ground cover (Ground Cover) by applying colour thresholding techniques in ImageJ (Abramoff, 2004) to digital photographs, and obtained high resolution (30 x 30 m) estimates of mean annual temperature (Temperature) from local climate models (developed by Morgan &

Guénard, 2019). As these environmental factors were shown to influence ant diversity in other invaded systems (e.g. King & Tschinkel, 2008), we used our data to test for environmental effects on invasion outcomes (the abundance of *S. invicta* across the plots), species richness and the trait structures of the assemblages.

Morphological traits

We measured seven morphological traits (Body Size, Head Width, Eye Width, Mandible Length, Scape Length, Pronotum Width, Leg Length) on ≥10 individual workers of every species (N=319). These traits regulate ant physiology and behaviour and are hypothesized to impact performance and fitness (Supporting information).

Dietary trait: Trophic Position

We measured the relative Trophic Position of each ant species using stable isotope ratios of Nitrogen (δ15N) (Tillberg et al., 2006). Live ants collected from the field were killed in a -20 °C freezer. We then rinsed the ants with distilled water, removed their abdomens to avoid contamination by undigested material in the gut (Tillberg et al., 2006), and dried the samples in an oven at 40 °C until a constant mass was reached. Dried samples with ≥5 workers were transferred into an aluminium capsule weighing 0.3–1 mg (workers of larger species were first ground and homogenized using a mortar and pestle following Hyodo et al., 2011). We measured the δ15N values of each sample using a Nu Perspective Isotope Ratio Mass Spectrometer coupled to an Eurovector Elemental and reported in ‰ (Gebauer & Schulze, 1991). Mineral soil collected from the field was used for baseline calibration. The δ15N value of every species was determined using 1–3 samples.

Physiological trait: Critical Thermal Maximum

We measured the Critical Thermal Maximum (CT_{max}) of individual ants following established protocols for CT_{max} assays (Diamond et al., 2017). The ants were first acclimated at 25 °C for ≥2 hrs in the laboratory. Individual ants were then placed in 1.5 mL Eppendorf tubes. The entrance of each tube was plugged with dry cotton wool, ensuring that each ant was confined to an area of even temperature distribution. The tubes were then placed in a digital dry bath (BSH1004) connected to an additional thermometer (UEi Test Instruments DT302 Dual Input IP67) for ensuring temperature accuracy. The assay began at a starting temperature of 36 °C, from which the temperature was increased at a constant rate of 1 °C min-1 (Diamond et al., 2017). Every 1 min, each tube was rotated and visually inspected to determine whether the ant had lost muscle coordination (Diamond et al., 2017); the temperature at which this occurred was recorded as the individual's CT_{max}. We measured the CT_{max} of ≥10 individual workers of each species (N=193).

Behavioural trait: Interference Ability

We assessed the Interference Ability of ant species from observations of interspecific interactions at baits after Le Brun and Feener (2007) (therein termed 'behavioural dominance'). Here, Interference Ability describes a species' relative success in two types of antagonistic interactions: usurping a resource from heterospecifics (expulsion) and defending an occupied resource from usurping heterospecifics (retention) (Le Brun & Feener, 2007). We recorded the outcomes (win/loss) in expulsion and retention incidents between different ant species

at baits. Each species' Interference Ability was then scored using the Colley rating method (after Le Brun & Feener, 2007), which adjusted the value of each win and loss by the Interference Ability of the competitors that a species interacted with.

Data analysis

Trait selection

For all morphological traits except Body Size, we corrected for the effects of body size by regressing each trait against Body Size and using the residuals as the new values for that trait. We used Principal Components Analysis (PCA) and correlation analysis to select a suite of traits that captured most interspecific variation in multidimensional trait space while minimizing redundancy from trait correlations (Supporting information). The first principal component, capturing 30% of the variation, was strongly positively associated with Head Width and Mandible Length and strongly negatively associated with Leg Length and Scape Length. The second (28% of the variation) was strongly positively and negatively associated with Interference Ability and Eye Width, respectively. The third (19% of the variation) was strongly positively and negatively associated with Body Size and Trophic Position, respectively. The fourth (13% of the variation) was strongly negatively associated with CT_{max}. All subsequent principal components had eigenvalues lesser than unity. Among traits showing strong positive correlations (Supporting information), we selected those with stronger loadings on principal components. Our final set of traits comprised four morphological, one dietary, one physiological and one behavioural trait: Body Size, Head Width, Eye Width, Leg Length, Trophic Position, CT_{max}, and Interference Ability.

Quantifying two measures of trait differences

For each trait, we quantified two measures of trait differences between the trait value of *S. invicta* ($T_{S.\ invicta}$) and the trait value of every other ant species (T_{Other}) recorded at baits in the study. We calculated Niche Difference as $|T_{S.\ invicta} - T_{Other}|$, a non-directional measure that can indicate the magnitude of niche differences between species. We calculated Competitive Difference as $T_{S.\ invicta} - T_{Other}$, a directional measure that detects for differences in competitive abilities along a competition hierarchy (Kraft et al., 2015; Kunstler et al., 2016).

Modelling invader abundance as a function of its trait differences with two assemblage types

The total number of *S. invicta* workers collected across the six pitfall traps at each of the 61 plots was used as the response term 'Invader Abundance'. For each trait, we modelled Invader Abundance as a function of the average trait differences (including Niche Difference and Competitive Difference as separate terms) between *S. invicta* and the observed assemblage of ant species at each plot. We repeated this process using the average trait differences between *S. invicta* and the 'missing assemblage' at each plot, which comprised any species missing from the plot but occurring in the species pool. We modelled the relationships using Poisson models with observation-level random effects as well as negative binomial models (prior to selecting the best model), as both of these could address the overdispersion in Invader Density (see Harrison, 2014). For each trait, we built a full model which included Niche Difference, Competitive Difference, the quadratic form of Competitive

Difference, and the environmental covariates Ground Cover and Temperature. We then selected the best model using a backward-step-wise variable selection procedure based on the Akaike information criterion corrected for small sample size (AICc).

Comparing patterns of trait dispersion between uninvaded and invaded assemblages

To determine whether invaded assemblages were more overdispersed or clustered relative to the uninvaded assemblages, we assessed the functional dispersion (FDisp) in each trait as well as in multidimensional trait space at the plot level (excluding *S. invicta*). We calculated FDisp using the 'fdisp' function of the FD package in R (R Core Team, 2017), and the ant species' frequencies of occurrence across the six pitfall traps at each plot. To control for potential effects of species richness on FDisp, we compared standardized effect sizes (SES) instead of the observed values (Swenson, 2014). We calculated SES by comparing the observed values to values generated from 999 constrained null models randomizing the matrix of species' frequencies of occurrence using the 'Independent Swap' algorithm. The formula for calculating SES is:

$$SES = \frac{Mean_{observed} - Mean_{null}}{Standard\ Deviation_{null}}$$

We then compared the SES values of FDisp in invaded plots to those in uninvaded plots using t-tests or Wilcoxon-Mann-Whitney tests (when sample variances were unequal).

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Author contributions

MKLW planned the study with inputs from BG and OTL. MKLW conducted the fieldwork. MKLW, RHL and CML performed trait measurements. MKLW conducted statistical analyses and wrote the first draft of the manuscript. MKLW, BG and OTL contributed substantially to manuscript revisions.

References

Abràmoff, M. D., Magalhães, P. J., & Ram, S. J. (2004). Image processing with ImageJ. Biophotonics international, 11(7), 36-42.

Bestelmeyer, B. T. (2000). The trade-off between thermal tolerance and behavioural dominance in a subtropical South American ant community. Journal of Animal Ecology, 69(6), 998-1009.

Calcaterra, L. A., Cuezzo, F., Cabrera, S. M., & Briano, J. A. (2010). Ground ant diversity (Hymenoptera: Formicidae) in the Iberá Nature Reserve, the largest wetland of Argentina. Annals of the Entomological Society of America, 103(1), 71-83.

Chesson, P. (2000). Mechanisms of maintenance of species diversity. Annual review of Ecology and Systematics, 31(1), 343-366.

D'Andrea, R., & Ostling, A. (2016). Challenges in linking trait patterns to niche differentiation. Oikos, 125(10), 1369-1385.

Darwin, C. (1859). On the origin of species. Murray.

Diamond, S. E., Chick, L., Perez, A. B. E., Strickler, S. A., & Martin, R. A. (2017). Rapid evolution of ant thermal tolerance across an urban-rural temperature cline. Biological Journal of the Linnean Society, 121(2), 248-257.

Eguchi, K., Bui, T. V., & Yamane, S. (2004). A preliminary study on foraging distance and nesting sites of ants in Indo-Chinese lowland vegetation. Sociobiology, 43(3).

Fortunel, C., Valencia, R., Wright, S. J., Garwood, N. C., & Kraft, N. J. (2016). Functional trait differences influence neighbourhood interactions in a hyperdiverse Amazonian forest. Ecology letters, 19(9), 1062-1070.

Godoy, O., Gómez-Aparicio, L., Matías, L., Pérez-Ramos, I. M., & Allan, E. (2020). An excess of niche differences maximizes ecosystem functioning. Nature communications, 11(1), 1-10.

Grime, J.P. (1979) Plant Strategies and Vegetation Processes. John Wiley & Sons, Chichester

Harrison, X. A. (2014). Using observation-level random effects to model overdispersion in count data in ecology and evolution. PeerJ, 2, e616.

Helms, J. A. (2018). The flight ecology of ants (Hymenoptera: Formicidae). Myrmecological News, 26(2), 19-30.

HilleRisLambers, J., Adler, P. B., Harpole, W. S., Levine, J. M., & Mayfield, M. M. (2012). Rethinking community assembly through the lens of coexistence theory. Annual review of ecology, evolution, and systematics, 43, 227-248.

Hyodo, F., Takematsu, Y., Matsumoto, T., Inui, Y., & Itioka, T. (2011). Feeding habits of Hymenoptera and Isoptera in a tropical rain forest as revealed by nitrogen and carbon isotope ratios. Insectes Sociaux, 58(3), 417.

Keddy, P. A., & Shipley, B. (1989). Competitive hierarchies in herbaceous plant communities. Oikos, 234-241.

King, J. R., & Tschinkel, W. R. (2008). Experimental evidence that human impacts drive fire ant invasions and ecological change. Proceedings of the National Academy of Sciences, 105(51), 20339-20343.

Kraft, N. J., Godoy, O., & Levine, J. M. (2015). Plant functional traits and the multidimensional nature of species coexistence. Proceedings of the National Academy of Sciences, 112(3), 797-802.

Kraft, N. J., Valencia, R., & Ackerly, D. D. (2008). Functional traits and niche-based tree community assembly in an Amazonian forest. Science, 322(5901), 580-582.

Kunstler, G., Falster, D., Coomes, D. A., Hui, F., Kooyman, R. M., Laughlin, D. C., ... & Aiba, M. (2016). Plant functional traits have globally consistent effects on competition. Nature, 529(7585), 204-207.

Lebrun, E. G., & Feener Jr, D. H. (2007). When trade-offs interact: balance of terror enforces dominance discovery trade-off in a local ant assemblage. Journal of Animal Ecology, 76(1), 58-64.

Legault, G., Bitters, M. E., Hastings, A., & Melbourne, B. A. (2020). Interspecific competition slows range

expansion and shapes range boundaries. Proceedings of the National Academy of Sciences, 117(43), 26854-26860.

Levine, J. M., & HilleRisLambers, J. (2009). The importance of niches for the maintenance of species diversity. Nature, 461(7261), 254-257.

Loreau, M., & Hector, A. (2001). Partitioning selection and complementarity in biodiversity experiments. Nature, 412(6842), 72-76.

MacArthur, R., & Levins, R. (1967). The limiting similarity, convergence, and divergence of coexisting species. The american naturalist, 101(921), 377-385.

MacDougall, A. S., Gilbert, B., & Levine, J. M. (2009). Plant invasions and the niche. Journal of Ecology, 97(4), 609-615.

Mayfield, M. M., & Levine, J. M. (2010). Opposing effects of competitive exclusion on the phylogenetic structure of communities. Ecology letters, 13(9), 1085-1093.

McGill, B. J., Enquist, B. J., Weiher, E., & Westoby, M. (2006). Rebuilding community ecology from functional traits. Trends in ecology & evolution, 21(4), 178-185.

Morgan, B., & Guénard, B. (2019). New 30 m resolution Hong Kong climate, vegetation, and topography rasters indicate greater spatial variation than global grids within an urban mosaic. Earth System Science Data, 11(3).

Münkemüller, T., Gallien, L., Pollock, L. J., Barros, C., Carboni, M., Chalmandrier, L., ... & Talluto, M. V. (2020). Dos and don'ts when inferring assembly rules from diversity patterns. Global Ecology and Biogeography.

Pérez-Ramos, I. M., Matías, L., Gómez-Aparicio, L., & Godoy, Ó. (2019). Functional traits and phenotypic plasticity modulate species coexistence across contrasting climatic conditions. Nature communications, 10(1), 1-11.

Porter, S. D., & Savignano, D. A. (1990). Invasion of polygyne fire ants decimates native ants and disrupts arthropod community. Ecology, 71(6), 2095-2106.

R Core Team (2017). R: A language and environment for statistical computing. R Found. Stat. Comput. Vienna, Austria.

Sanders, N. J., Gotelli, N. J., Heller, N. E., & Gordon, D. M. (2003). Community disassembly by an invasive species. Proceedings of the National Academy of Sciences, 100(5), 2474-2477.

Scheffer, M., & van Nes, E. H. (2006). Self-organized similarity, the evolutionary emergence of groups of similar species. Proceedings of the National Academy of Sciences, 103(16), 6230-6235.

Shea, K., & Chesson, P. (2002). Community ecology theory as a framework for biological invasions. Trends in Ecology & Evolution, 17(4), 170-176.

Swenson, N. G. 2014. Functional and phylogenetic ecology in R. Springer.

Terry, C., Chen, J., & Lewis, O. T. (2020). The effect of natural enemies on the coexistence of competing species-an empirical test using Bayesian modern coexistence theory. bioRxiv.

Tillberg, C. V., McCarthy, D. P., Dolezal, A. G., & Suarez, A. V. (2006). Measuring the trophic ecology of ants using stable isotopes. Insectes sociaux, 53(1), 65-69.

Van Kleunen, M., Weber, E., & Fischer, M. (2010). A meta-analysis of trait differences between invasive and non-

invasive plant species. Ecology letters, 13(2), 235-245.

Violle, C., Pu, Z., & Jiang, L. (2010). Experimental demonstration of the importance of competition under disturbance. Proceedings of the National Academy of Sciences, 107(29), 12925-12929.

Weeks, R. D., Wilson, L. T., & Vinson, S. B. (2004). Resource partitioning among colonies of polygyne red imported fire ants (Hymenoptera: Formicidae). Environmental entomology, 33(6), 1602-1608.

Wong, M. K., Guénard, B., & Lewis, O. T. (2020). The cryptic impacts of invasion: functional homogenization of tropical ant communities by invasive fire ants. Oikos, 129(4), 585-597.