



Sex-specific shifts in morphology and colour pattern polymorphism during range expansion of an invasive lizard

Kimberly A. Miller^{1,2} | Andressa Duran¹ | Jane Melville³ | Michael B. Thompson⁴ | David G. Chapple^{1,3,5} 

¹School of Biological Sciences, Monash University, Clayton, Vic., Australia

²Conservation and Research, Healesville Sanctuary, Zoos Victoria, Vic., Australia

³Division of Sciences, Museum Victoria, Melbourne, Vic., Australia

⁴School of Biological Sciences (A08), University of Sydney, Sydney, NSW, Australia

⁵Allan Wilson Centre for Molecular Ecology and Evolution, School of Biological Sciences, Victoria University of Wellington, Wellington, New Zealand

Correspondence

David G. Chapple, School of Biological Sciences, Monash University, Clayton, Vic, Australia.
Email: david.chapple@monash.edu

Funding information

Australian Research Council, Grant/Award Number: DP0771913; National Geographic Society, Grant/Award Number: 8085-06, 8952-11; the Hermon Slade Foundation, Grant/Award Number: HSF 09-02; Allan Wilson Centre for Molecular Ecology and Evolution; Monash University

Editor: Krystal Tolley

Abstract

Aim: Human-assisted range expansion of animals to new environments can lead to phenotypic shifts over ecological time-scales. We investigated whether phenotypic changes are sex-specific using an invasive lizard (*Lampropholis delicata*).

Location: Pacific region (Hawaiian Islands, Lord Howe Island, New Zealand, eastern Australia).

Methods: Using our knowledge of the introduction history of *L. delicata*, we examined museum specimens of individuals collected across the native and introduced range to determine whether shifts in morphology or colour pattern polymorphism had occurred during its range expansion, and if so, whether they differed between the sexes.

Results: Sexual dimorphism in both size and shape was documented within the native range of the delicate skink. However, during range expansion, phenotypic shifts were observed in shape, but not size. In two of the three invasive populations, these phenotypic shifts were sex-specific. In the Hawaiian Islands, changes in shape were driven by males, whereas in New Zealand it was due to shifts in females. Similarly, changes in the frequency of a colour pattern polymorphism, a mid-lateral stripe shown to have sex-specific impacts on fitness (positive in females, negative in males), occurred following colonization of the Hawaiian Islands and Lord Howe Island. In Hawaii, the incidence of the polymorphism increased over time in females, and decreased in males.

Main conclusions: Phenotypic shifts during the range expansion of invasive species may be sex-specific, and are potentially related to the degree of realized niche shift that has occurred between the source and introduced range.

KEYWORDS

colour pattern polymorphism, genetic admixture, Hawaiian Islands, invasive species, *Lampropholis delicata*, Lord Howe Island, morphology, New Zealand, rapid evolution, sexual dimorphism

1 | INTRODUCTION

Widespread species generally display geographic variation in their life history and phenotype across their range (Morrison & Hero,

2003; Stillwell, Morse, & Fox, 2007). This spatial variation may reflect the differing habitats and climates that they inhabit, and/or local adaptations for these environments (Hassall, Keat, Thompson, & Watts, 2014; Meiri & Dayan, 2003). Alternatively, the phenotypic



divergence of populations might be due to neutral processes during range expansion, for instance, founder effects/genetic drift (e.g. Wares, Hughes, & Grosberg, 2005) or spatial sorting (e.g. Shine, Brown, & Phillips, 2011). Similarly, populations that spread via human-mediated dispersal may experience founder events (Clegg et al., 2002; Kolbe, Leal, Schoener, Spiller, & Losos, 2012; Sax et al., 2007) and rapid environmental shifts (e.g. climate, habitat, prey or food availability, competitors and predators), resulting in changes in phenotype or life history (Campbell & Echternacht, 2003; Herrel et al., 2008). Morphological shifts in the introduced range may also result from intraspecific hybridization among individuals from different native-range source regions (Rius & Darling, 2014).

Invasive species provide illustrative examples of how these phenotypic shifts can occur over ecological time-scales (Sax et al., 2007; Suarez & Tsutsui, 2008). For instance, the formation of geographic variation in phenotype can be documented in "real time" (e.g. long-versus short-winged forms in bush crickets, *Conocephalus discolor* and *Metroptera roeselii*, Thomas et al., 2001; leg length and body size in cane toads, *Rhinella marina*, Phillips & Shine, 2005, 2006; Shine, 2012). In particular, one of the best demonstrations for the potential adaptive basis of these phenotypic shifts is the rapid evolution of latitudinal clines in phenotypic traits in the introduced range that mirror those evident in the native range (e.g. house sparrow, *Passer domesticus*, Johnston & Selander, 1964; fruit flies, *Drosophila subobscura*, Huey, Gilchrist, Carlson, Berrigan, & Serra, 2000).

Surprisingly, few studies have investigated whether the sexes exhibit divergent phenotypic responses during range expansions to non-native regions (Campbell & Echternacht, 2003; Simberloff, Dayan, Jones, & Ogura, 2000). In lizards, sexual dimorphism is widespread in both size and shape (e.g. male body size or head width; female body size and interlimb length; Cox, Skelly, & John-Alder, 2003; Scharf & Meiri, 2013). This dimorphism is driven by selection, such as increased reproductive output in females (body size and interlimb length), and male–male competition (e.g. head width) (Scharf & Meiri, 2013). Environmental factors such as food availability and habitat can also drive sexual dimorphism in lizards (Bonneaud et al., 2016; Herrel, Spithoven, Van Damme, & De Vree, 1999; Herrel, Van Damme, & De Vree, 1996). Thus, intuitively, range expansion during species invasions may alter sexual dimorphism of a species due to: (1) reduced population densities in the invasive range decreasing the level of intraspecific competition (e.g. relaxing male–male competition and decreasing dimorphism in head size) and (2) exposure to novel environmental conditions. Depending on the specific circumstances, these factors have the potential to lead to either reduced, or accentuated, levels of sexual dimorphism in invasive populations (Campbell & Echternacht, 2003).

Here we investigate sex-specific shifts in morphology and colour pattern in the invasive delicate skink (*Lampropholis delicata* De Vis) during its range expansion throughout the Pacific region. The delicate skink is a widespread lizard species with a native distribution spanning 26° latitude across a range of natural and urban environments in eastern Australia (Figure 1; Chapple, Hoskin, Chapple, & Thompson, 2011). It exhibits a range of behaviours that increase its

propensity for human-assisted dispersal (e.g. exploratory behaviour, hiding within freight and cargo; Chapple, Simmonds, & Wong, 2011; Cromie & Chapple, 2012), and consequently, it is a frequent stow-away in freight and cargo (Chapple, Kneegtmans, Kikillus, & van Winkel, 2016; Chapple, Whitaker, Chapple, Miller, & Thompson, 2013). The delicate skink is oviparous, and has successfully established populations in the Hawaiian Islands, New Zealand and Lord Howe Island (Table 1; Chapple, Miller, Kraus, & Thompson, 2013; Chapple, Reardon, & Peace, 2016).

We use our knowledge of the native-range source populations and introduction history of the delicate skink (Table 1; Chapple, Miller, et al., 2013) to identify the phenotypic changes during its human-mediated range expansion. First, we will document the geographic variation in morphology that exists within the native range of the delicate skink. Specifically, we will (1) assess whether morphological variation reported among six native-range populations (Forsman & Shine, 1995a) is also evident among the nine genetic lineages of the delicate skink (Figure 1; Chapple, Hoskin, et al., 2011) and (2) determine the relative incidence of a colour pattern polymorphism (presence or absence of a mid-lateral stripe) shown to have sex-specific impacts on fitness (positive in females, negative in males; Forsman & Shine, 1995b), across the native range of the delicate skink. This will provide a valuable baseline from which to investigate our hypothesis that post-introduction shifts in phenotype have occurred in the invasive populations, relative to their native-range source population. Specifically, we will test three key predictions about the phenotypic shifts that have occurred during the range expansion of the delicate skink:

1. The magnitude of phenotypic change during range expansion will be related to the degree of realized niche shift between each source population(s) and invasive population. For instance, as niche conservatism is evident on Lord Howe Island (Tingley, Thompson, Hartley, & Chapple, 2016), the potential for phenotypic shifts is expected to be minor. Any changes that are present will most likely be the result of the genetic admixture among four native-range source populations that is present on the island (Chaplin, 2013). Moderate levels of phenotypic change are expected in New Zealand, where niche shifts are characterized by niche unfilling (i.e. when a species fails to colonize climates in the invaded range that are occupied in the native range; Tingley et al., 2016). The most substantial phenotypic shifts are expected in the Hawaiian Islands, where both niche expansion (i.e. when a species colonizes environmental conditions in its invaded range that are present, but unoccupied, in its native range) and unfilling have been demonstrated (Tingley et al., 2016).
2. As sexual dimorphism in lizards is primarily driven by selection related to environmental factors (Bonneaud et al., 2016; Herrel et al., 1996, 1999), the potential for sex-specific shifts in morphology will be highest in populations that experience the largest changes in their realized niche (i.e. most likely to occur in Hawaii and New Zealand).

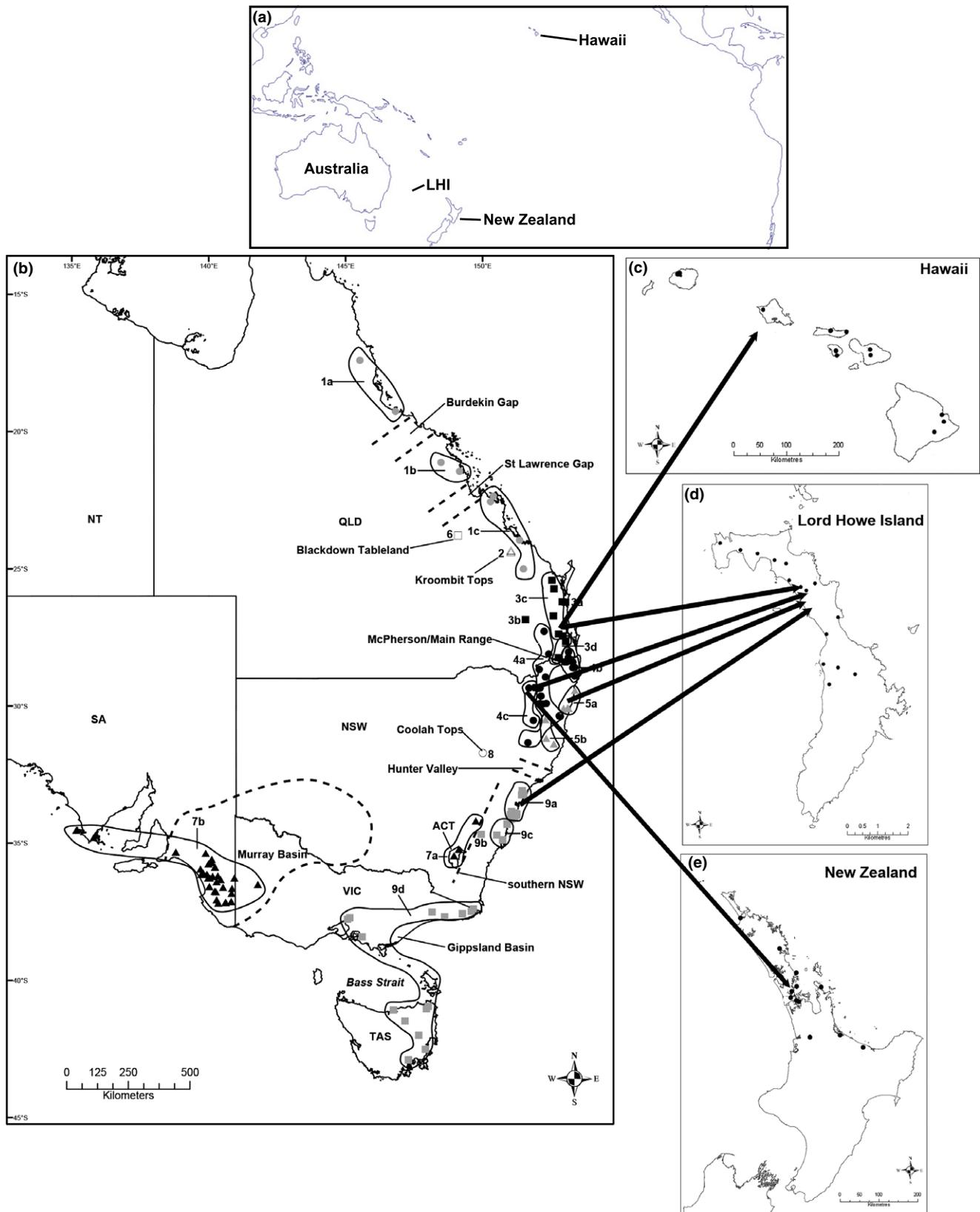


FIGURE 1 (a) Map of the Pacific region, indicating the source and introduced range of the delicate skink. (b) The distribution of delicate skink genetic clades and subclades in the native-Australian range (adapted from Chapple, Hoskin, et al., 2011). The location of recognized biogeographic barriers in eastern Australia is indicated. (c–e) The source regions for each introduced population (Hawaii, Lord Howe Island, New Zealand is indicated with arrows [see Table 1 for further information]). The sample codes for each introduced population follows that presented in Chapple, Miller, et al. (2013) [Colour figure can be viewed at wileyonlinelibrary.com]



TABLE 1 Introduction history of the invasive delicate skink (*Lampropholis delicata*) in the Pacific region. The native-range clades are indicated in Figure 1. Intraspecific hybridization (i.e. genetic admixture) has been shown to occur among the four source clades on Lord Howe Island (Chaplin, 2013). Abbreviations for Australian States: NSW = New South Wales, QLD = Queensland. For the Lord Howe Island introduction, we indicate whether individuals from each of these source regions occur in the northern (N), central (C) or southern (S) regions of the island

Introduced region	Initial establishment	No. generations since establishment	Known sources		
			Region	Clade	References
Hawaiian Islands	~1900–1909	~110	Brisbane, QLD	3	Baker (1979), Chapple, Miller, et al. (2013)
New Zealand	1960s	~50	Inland northern NSW	4	Chapple, Miller, et al. (2013)
Lord Howe Island	1980s	~30	Brisbane, QLD	3 (C, S)	Chapple, Miller, et al. (2013); Chapple et al. (2014)
			Inland northern NSW	4 (C, S)	
			Yamba-Coffs Harbour, NSW	5 (N, C, S)	
			Sydney, NSW	9a (C, S)	

3. As the relative presence of the mid-lateral stripe appears to be closely linked to latitude (Forsman & Shine, 1995b), we predict that post-introduction shifts in this colour pattern polymorphism will only be evident where there are substantial differences in the latitude of the source and introduced population (i.e. most pronounced in Hawaii [27°S to 21°N] and New Zealand [29°S to 36°S], relative to Lord Howe Island [27–33°S to 31°S]). Thus, we predict that the incidence of the mid-lateral stripe will decrease in New Zealand, increase in Hawaii and remain unchanged on Lord Howe Island.

2 | MATERIALS AND METHODS

2.1 | Specimens and data collection

A total of 1,188 delicate skink specimens (native range: 505, Hawaiian Islands: 389, New Zealand: 99, Lord Howe Island: 195) were examined from museum collections in Australia (Queensland Museum, Australian Museum, Museum Victoria), New Zealand (Te Papa Tongarewa: National Museum of New Zealand) and the United States (Bishop Museum-Hawaii, California Academy of Sciences, University of California Berkeley Museum of Vertebrate Zoology, Smithsonian National Museum of Natural History). There are nine major genetic clades (some with subclades) within the native range of the delicate skink (Chapple, Hoskin, et al., 2011; Figure 1). Based on previously published genetic work (Chapple, Hoskin, et al., 2011; Chapple, Miller, et al., 2013), and the collection location of the individuals, skink specimens were assigned to either one of the previously established clades, or to one of the invasive populations (i.e. Hawaiian Islands, New Zealand and Lord Howe Island). The three invasive populations were linked back to the Clades 3, 4, 5 and 9a from the native range based on their colonization history. These clades are hereafter named source clades. The Hawaiian Island population was linked to the Clade 3, and the New Zealand population was linked to the Clade 4 (Table 1; Chapple, Miller, et al., 2013). The Lord Howe Island population was treated as three separate invasive populations because each region of the island has different

source clades. The northern portion of the island was colonized by the source Clade 5; the central region by Clades 3, 4, 5 and 9a; and the southern region by source Clades 4 and 9a (Chapple, Miller, et al., 2013; Chapple et al., 2014; Moule et al., 2015; Table 1). Additionally, the density of historic sampling in Hawaii also allowed an examination of phenotypic changes over time. For these temporal analyses, the Hawaiian data were divided into three time periods: 1909–1939 (initial establishment until World War II [WWII]); 1940–1979 (WWII and post-war spread phase); and 1980–2008 (post-spread phase). However, due to low sample size for the initial time period, most analyses only compared the latter two time periods.

The collection year, location, sex and presence/absence of a mid-lateral stripe (Forsman & Shine, 1995b) were recorded for each specimen. For each specimen, we recorded seven morphometric traits that have been shown to capture size and shape variation in skinks (Wu, Alton, Clemente, Kearney, & White, 2015). Measurements (± 0.01 mm) were taken of snout-vent length (SVL; measured from the tip of the snout to the cloacal opening), snout-axilla length (SAL; from the tip of the snout to the axilla), interlimb length (ILL; measured from the axilla to the groin), hindlimb length (HLL; from the groin to the palm of the hindleg), length of 4th toe on hindleg (ToeL; from the base of the toe to the tip, excluding the claw), head width (HW; measured at the level of the ear apertures) and head length (HL; from the ear aperture to the tip of the snout; See Table S1 for descriptive statistics of morphological measurements for each clade and invasive population). The reproductive status and clutch size were also recorded for each female specimen.

2.2 | Statistical analyses

The seven morphometric measurements were first evaluated for their normality using Shapiro–Wilk normality tests, and as they did not deviate from normal distributions (data not shown), no data transformation was required. A principal component analysis (PCA) was performed on the covariance matrix of the measurements to

detect sources of variation in morphology among the delicate skink specimens, and the PC scores were used in subsequent analysis. PCAs were performed using the package `PCAMETHODS` 1.67.0 (Stacklies, Redestig, Scholz, Walther, & Selbig, 2007), and the statistical analyses were performed in R 3.1.3 (R Development Core Team 2015). Initially, the genetic clades within the native range were analysed separately to assess the degree of sexual dimorphism in the PC scores, and to evaluate if the morphological variation was consistent among the native range clades. Each of the invasive populations was then analysed individually and compared to their source clades. All the comparisons among native clades, and among source and invasive populations were performed using two-way analyses of variance (ANOVAs), followed by post hoc tests conducted using Tukey's honest significant difference (HSD). All ANOVAs included sex and the different populations (source and invasive) as independent variables, as well as their interaction. For the historic sampling in Hawaii, one-way ANOVAs were performed including the different time periods against the source clade as independent variables. Because multiple comparisons were performed for Lord Howe Island, the *p*-values were adjusted using a Bonferroni correction: $\alpha = 0.016$. A series of chi-square tests were used to identify whether prevalence of the mid-lateral stripe differed among samples, except where the expected values were <5 . In such cases, a Fisher's exact test was used. Prevalence of the mid-lateral stripe in females and males was compared. Finally, a one-way ANOVA was used to determine whether mean clutch size of females, relative to body size, differed between groups. Because clutch size is correlated with SVL, the studentized residuals of clutch size over SVL among groups were compared. Post hoc tests were conducted using Tukey's HSD where required. Small sample sizes of reproductive females precluded any statistical comparison of minimum size at maturity among groups (see Table S1 and S2 in Appendix S1).

3 | RESULTS

The PCA efficiently summarized the morphological variation in the delicate skink, with the first two PC axes accounting for 80% of the total variance in the dataset (Table 2). The remaining PC axes explained $<7\%$ each of the total variation, thus the subsequent analyses were based primarily on PC1 and PC2. Loadings of all variables on the first PC were positive and of similar magnitude, reflecting overall variation in size, especially SVL, SAL and HL. The second PC corresponded to the most pronounced variation in body shape, particularly related to ILL and HLL. In particular, positive scores on PC2 indicate specimens with more robust bodies (low ILL) with longer hindlimbs (high HLL) and longer toes (ToeL) (Figure 2).

3.1 | Sexual dimorphism among clades in the native range

There were significant differences in size (PC1; $F_{1,483} = 8.70$, $p = .003$) and shape (PC2; $F_{1,483} = 288.89$, $p < .001$) among genetic

TABLE 2 Loadings of a principal components analysis of morphological measurements of delicate skink

Variables	PC1	PC2
Snout-vent length (SVL)	0.42	-0.38
Snout-axilla length (SAL)	0.41	-0.04
Interlimb length (ILL)	0.37	-0.51
Hindlimb length (HLL)	0.30	0.62
Length of fourth toe on hindleg (ToeL)	0.34	0.38
Head width (HW)	0.39	-0.10
Head length (HL)	0.40	0.22
Proportion of variance	0.68	0.12
Cumulative proportion	0.68	0.80

clades in the native range (Figure 3). However, variation in size involved exclusively differences between Clades 7b+9a and Clades 2+3+4+5 (see Table S3 in Appendix S1), whereas no significant difference in PC1 scores were detected between males and females of the same clade (Figure 3; Table S3). On the other hand, there were significant differences in shape between Clade 1 and several other Clades (3, 4, 5, 7a, 7b, 9a) and between Clade 2 and Clades 4+7a (Table S3). Interestingly, there were significant differences in shape between males and females in all clades except 7a and 8 (Table S3). The prevalence of the mid-lateral stripe differed among clades, and was greater at more northern latitudes (Table 3). In the six clades where present, the mid-lateral stripe was also more prevalent in females than males ($\chi^2 = 50.283$, $df = 1$, $p < .001$). These differences indicate not only that identifying the source clade is crucial to understanding morphological variation in an introduced population but also that the variation in the native range is much stronger in terms of shape than in size.

3.2 | Shifts in morphology and colour pattern during the introduction process

Although there were no significant shifts in body size in any of the invasive populations (see Table S4 in Appendix S1; see Figure S1 in Appendix S2), there were intriguing differences in the degree of shape dimorphism following the establishment of invasive populations (Table 4; Figure S1). However, the way in which these shifts took place was largely idiosyncratic. For instance, there were significant accentuations in shape dimorphism in invasive populations of Hawaii and New Zealand, yet they were driven by shifts only in males in the former and females in the latter (Table 4). The historical sampling from the Hawaiian Islands was evaluated in detail only for males, given that it was the only comparison of specimens of the same sex that showed body shape differences (Table 4). There were significant body shape differences between the source range and each of the time periods (Table 5; Figure 4). Interestingly, there was no significant difference between the time periods compared to each other, suggesting that the detected shape changes occurred early during their invasion of the Hawaiian island. Finally, the

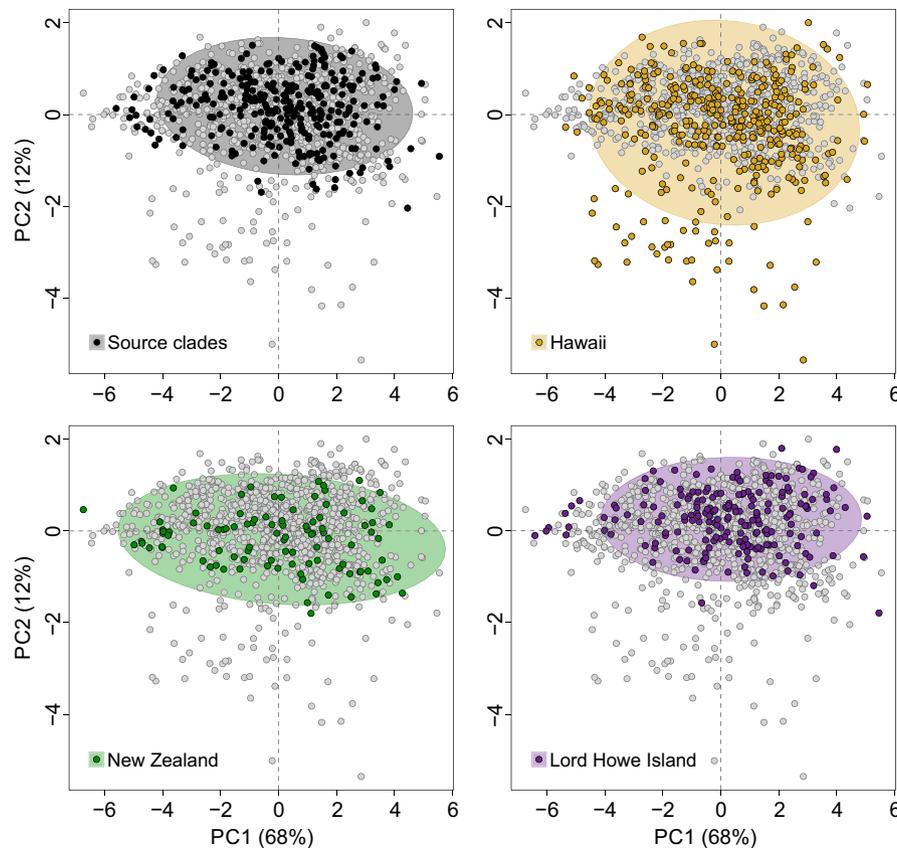


FIGURE 2 Morphological space of skink lizards represented by the scores of the first two PC axes. Each point represents one of the 1,188 specimens. Points highlighted in each plot represents the position of each population in the morphological space, with their respective 95% confidence ellipses. High scores on PC1 indicate larger specimens (high SVL, SAL and HL), whereas lower scores on PC2 indicate specimens with more elongated bodies (high ILL) [Colour figure can be viewed at wileyonlinelibrary.com]

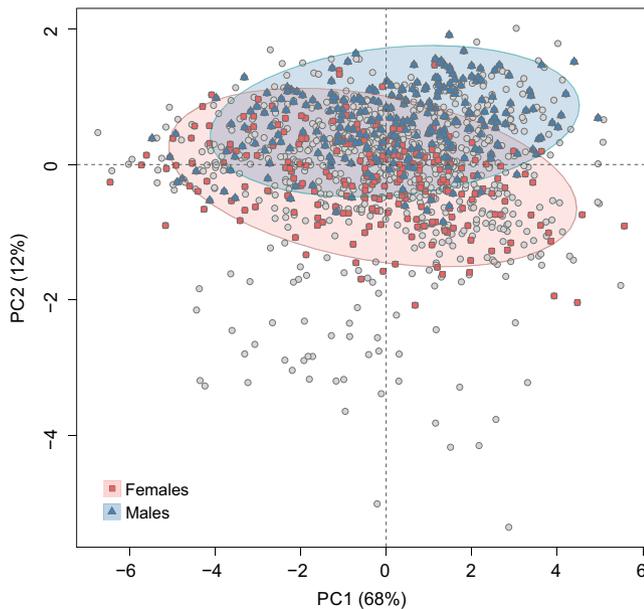


FIGURE 3 Sexual dimorphism in clades within the native range along the first two PC axes. Background points in grey represent each one of the 1,188 specimens. 95% confidence ellipses are provided [Colour figure can be viewed at wileyonlinelibrary.com]

degree of shape dimorphism remained unchanged in all three populations of Lord Howe when compared to their source clades (Table 4).

The mean clutch size of females in Hawaii (4.05, $N = 105$) was larger than females in the source population (Clade 3: 2.40, $N = 5$), but the difference was not statistically significant after correcting for SVL ($F_{1,108} = 3.621$, $p = .060$). The small number of gravid females in the source population ($N = 5$) and the earliest time period on Hawaii (1909–1939) precluded comparison of clutch sizes over time. The mean clutch size of females in New Zealand (3.76, $N = 7$) was not significantly different to that of females in Clade 4 (4.14, $N = 10$; $F_{1,15} = 0.449$, $p = .513$). The mean clutch size of females on Lord Howe Island (3.32, $N = 56$) was not significantly different from that of females in the source regions (Clade 3: 3.50, $N = 5$; Clade 4: 3.67, $N = 10$, Clade 5: 2.40, $N = 3$; Clade 9a: 3.43, $N = 5$) after correcting for SVL ($F_{4,74} = 1.568$, $p = .192$, $p > .05$ for all post hoc comparisons). Clutch sizes of females in any region on Lord Howe Island (northern, central and southern) did not differ from any other population or any source clade after correcting for SVL ($F_{6,72} = 1.243$, $p = .295$, $p > .05$ for all post hoc comparisons).

On the other hand, several differences in coloration were detected between source and invasive populations. In Hawaii, the

TABLE 3 Prevalence of mid-lateral stripe in the delicate skink across the native range and the invasive populations on Lord Howe Island (LH), New Zealand (NZ) and Hawaii (HI). Significant differences between source clades and the introduced populations are indicated by superscript in the latter

Clade	Source(s)	All		Females		Males	
		Prevalence	N	Prevalence	N	Prevalence	N
1		35.19	54	52.17	23	22.58	31
2		63.16	19	88.89	9	40.00	10
3		41.07	56	80.95	21	17.14	35
4		32.98	94	65.79	38	10.71	56
5		21.28	47	33.33	27	5.00	20
7a		0.00	20	0.00	5	0.00	15
7b		0.00	38	0.00	20	0.00	16
8		0.00	2	0.00	1	0.00	1
9a		4.72	106	8.16	49	1.79	56
9d1		0.00	39	0.00	20	0.00	18
9d2		0.00	26	0.00	14	0.00	11
LH (overall)	3, 4, 5, 9a	29.74 ^{9a}	195	55.45 ^{3,5,9a}	101	2.13 ³	94
LH (Northern)	5	25.00	60	46.88	32	0.00	28
LH (Central)	3, 4, 5	20.45 ³	44	40.91 ³	22	0.00	22
LH (Southern)	4, 9a	37.36 ^{9a}	91	68.09 ^{9a}	47	4.55	44
NZ	4	38.38	99	73.33	45	9.26	54
HI (overall)	3	47.26	402	95.03 ³	181	6.57 ³	213
HI (1909–1939)	3	37.50	16	66.67	6	20.00	10
HI (1940–1979)	3	44.09	127	90.74	54	6.06	66
HI (1980–2009)	3	49.42	259	98.33 ³	120	5.88 ³	136

female mid-lateral stripe was more prevalent in the invasive population than in the source region (Clade 3; $p = 0.033$, Fisher's exact test). Conversely, in males, the mid-lateral stripe was more prevalent in the source region than in the Hawaiian population ($p = 0.045$, Fisher's exact test). The prevalence of the mid-lateral stripe in females increased over time in the Hawaiian population (Table 3), and was significantly greater than Clade 3 only in the most recent time period (1909–1939: $p = .588$, 1940–1979: $p = .256$; 1980–2009: $p = .004$, Fisher's exact tests). In males, the prevalence of the mid-lateral stripe decreased over time in the Hawaiian population (Table 3), and was significantly smaller than Clade 3 only in the most recent time period (1909–1939: $p = 1.0$; 1940–1979: $p = .091$, 1980–2009: $p = .041$, Fisher's exact tests). On the other hand, the mid-lateral stripe was equally prevalent in the Clade 4 and New Zealand populations for both males and females (Table 3; $p > .05$ for all comparisons). Finally, several differences were detected in Lord Howe Island populations. In females, the mid-lateral stripe was less prevalent on Lord Howe Island than Clade 3 ($\chi^2 = 4.706$, $df = 1$, $p = .03$), but more prevalent than Clades 5 ($\chi^2 = 4.168$, $df = 1$, $p = .041$) and 9a ($\chi^2 = 30.733$, $df = 1$, $p < .001$) (Table 3). In males, the mid-lateral stripe was less prevalent on Lord Howe Island than in Clade 3 ($p = .0051$, Fisher's exact test), but equal in prevalence to Clades 4, 5 and 9a (Table 3; $p > .05$ for all comparisons). The mid-lateral stripe was equally prevalent in the northern LH region and its sole source population (Clade 5) for males and females (Table 3;

$p > .05$ for all comparisons). The prevalence of the mid-lateral stripe was greater in source Clade 3 than in the central LH region for females ($\chi^2 = 7.207$, $df = 1$, $p = .007$). No other comparisons of the central LH region and its source populations were significant. In the southern LH region, the prevalence of the mid-lateral stripe was greater than in Clade 9a for females (Table 3; $\chi^2 = 36.752$, $df = 1$, $p < .001$). No other comparisons of the southern LH region and its source populations were significant.

4 | DISCUSSION

The range expansion of the delicate skink throughout the Pacific region has been associated with shifts in both its morphology and colour pattern, but the extent and nature of these shifts vary between the three invasive regions (Hawaiian Islands, New Zealand and Lord Howe Island). Importantly, the post-establishment morphological shifts involved shape (PC2), rather than size (PC1). Differences were also evident in the underlying nature of these morphological shifts, with changes driven by males in Hawaii, and females in New Zealand. Shifts in the relative incidence of the mid-lateral stripe were pervasive in Hawaii, relatively minor on Lord Howe Island, and absent in New Zealand. No shifts in clutch size were observed in the introduced range of the delicate skink. Our study is unable to determine whether the phenotypic shifts observed



TABLE 4 Comparisons in body shape variation (PC2 scores) between source clades and invasive populations of the delicate skink with the respective post hoc tests. Significant differences are in bold

	Sex	Two-way ANOVA			Post hoc test			
			Population (source/invasive)	Sex:population	M-F (source)	M-F (invasive)	M (invasive)—M (source)	F (invasive)—F (source)
Hawaii	F	43.889	8.380	1.398	1.022	0.652	-0.653	-0.283
	<i>p</i>	<.001	.004	.238	.003	<.001	.025	.538
New Zealand	F	119.716	10.883	0.894	0.720	0.857	-0.171	-0.308
	<i>p</i>	<.001	.001	.346	<.001	<.001	.325	.017
Lord Howe North	F	55.538	0.738	3.935	1.028	0.602	-0.125	0.301
	<i>p</i>	<.001	.392	.050	<.001	<.001	.847	.194
Lord Howe Central	F	212.422	1.606	1.268	3	1.022	-0.156	0.125
	<i>p</i>	<.001	.172	.282	<i>p</i>	<.001	.992	.998
					4	0.720	0.080	0.059
					<i>p</i>	<.001	1.000	1.000
					5	1.028	-0.141	0.145
					<i>p</i>	<.001	.997	.995
					9a	0.767	-0.133	-0.107
					<i>p</i>	<.001	.992	.999
Lord Howe South	F	132.094	3.918	0.373	4	0.720	0.150	0.239
	<i>p</i>	<.001	.021	.689	<i>p</i>	<.001	.693	.390
					9a	0.767	-0.063	0.072
				<i>p</i>	<.001	.988	.992	

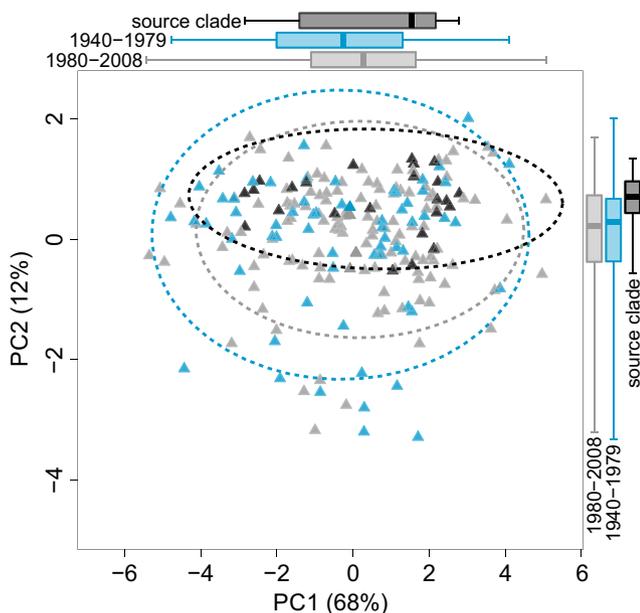


FIGURE 4 Principal component analysis of morphological variation in Hawaiian male delicate skinks in its native and invasive environment. Symbols in black, blue and grey correspond to specimens from the source clade, in the invasive range collected between 1940 and 1979, and between 1980 and 2008, respectively. Box and whisker plots summarize the variation in each of the PCs (medians and quartiles are shown) [Colour figure can be viewed at wileyonlinelibrary.com]

TABLE 5 Post hoc tests from the ANOVA comparing males from source population and the two different time periods from the historical sampling of Hawaiian delicate skinks ($F_{2,214} = 4.76$, $p = .009$). Significant differences are in bold

Comparisons	Difference	<i>p</i> adjusted
Source clade—(1940–1979)	0.711	.007
Source clade—(1980–2008)	0.539	.033
(1980–2008)—(1940–1979)	0.172	.476

in the introduced range are the result of neutral (e.g. founder effects, genetic drift) or selective processes (e.g. local adaptation), but provides a framework for future studies investigating the mechanisms responsible for these patterns.

4.1 | Delicate skinks exhibit sexual dimorphism among clades in the native range

We confirmed the presence of sexual dimorphism in morphology across the native range of the delicate skink. While there was no difference in size (PC1) within clades, the sexes differed in shape (PC2). Our results are largely concordant with a previous study of the delicate skink (Forsman & Shine, 1995a), which compared populations from across six different latitudes (one each from Clades 1, 3, 5 and 9a, and two from Clade 9d), with a smaller subset of morphological

traits (SVL, ILL). But contrary to our study, they also found females to have larger body sizes than males (except for Melbourne [our Clade 9d]). Importantly, both studies found clear evidence for geographic variation in morphology in the delicate skink. This geographic variation in delicate skink body proportions has been attributed to latitudinal clines in climate (rainfall, air temperatures and seasonality) in eastern Australia (Forsman & Shine, 1995a).

Our finding that the relative incidence of the mid-lateral stripe decreases in both sexes with increasing latitude is consistent with previous reports by Forsman and Shine (1995b), who attributed it to latitudinal clines in climate and predation rate in eastern Australia. Hence, the mid-lateral stripe may play a role in both thermoregulation (e.g. thermal reflectance) and antipredator behaviour (e.g. crypsis). Indeed, latitudinal clines in colour pattern (or the relative incidence of colour morphs) in species in the same region of eastern Australia (e.g. White's skink, *Liopholis whitii*, Chapple et al., 2008), North America and Europe (e.g. house sparrow, *Passer domesticus*, Johnston & Selander, 1964) have been associated with climatic variation (reviewed in McLean & Stuart-Fox, 2014). However, as the incidence of the mid-lateral stripe is higher in females than males for each delicate skink clade (where the mid-lateral stripe is present; Table 3), there may be sexual dimorphism in both ecology and the relative selective benefit of the colour pattern polymorphism. A field study where the sexes differed in their microhabitat selection and body temperatures provides evidence for both of these possibilities: striped females are larger (SVL), have larger clutch sizes and higher survival than non-striped females, whereas the presence of the mid-lateral stripe has a detrimental impact on males, reducing survival (Forsman & Shine, 1995b). However, while the mid-lateral stripe appears to have a sex-specific impact on fitness (Forsman & Shine, 1995b), the precise functional significance of the polymorphism is currently unknown, and the focus of our ongoing investigations.

4.2 | Sex-specific shifts in morphology and colour pattern during range expansion

While morphological shifts, relative to their native-range source populations, were evident in Hawaii and New Zealand, surprisingly these changes were restricted to shape (PC2), rather than size (PC1). Furthermore, these morphological changes were driven by a different sex in each invasive population (males in Hawaii, females in New Zealand). As predicted, shifts in morphology were limited to the invasive populations (Hawaii, New Zealand) that had experienced niche shifts, either via niche expansion or unfilling, relative to the native range (Tingley et al., 2016). This suggests that selective processes (e.g. adaptation to novel environments or environmental shifts) may have an important role in driving these morphological shifts in the invasive range of the delicate skink. Few other studies have investigated sex-specific shifts in morphology during range expansion to non-native regions. For instance, in the brown anole, body size increased over time due to an increase in male growth rate (Campbell & Echternacht, 2003). Similarly, an increase in sexual size dimorphism in the introduced range of the Indian Mongoose

(*Herpestes javanicus*) was driven by an increase in male body size (Simberloff et al., 2000).

The morphological shift in shape observed in Hawaii and New Zealand involved interlimb length and hindlimb length. Interlimb length is a trait that is most commonly associated with reproductive output in female lizards (as it allows females fit larger clutches; Cox et al., 2003; Scharf & Meiri, 2013), including the delicate skink (Forsman & Shine, 1995a). Limb length in the delicate skink has been shown to be a plastic trait that responds to habitat structure (Downes & Hoefer, 2007). Thus, while we found support for our hypothesis that changes in sexual dimorphism were most likely in Hawaii and New Zealand, only one of the traits involved in these shifts (ILL) has been shown to have a sex-specific function in lizards (Cox et al., 2003; Forsman & Shine, 1995a; Scharf & Meiri, 2013). Intriguingly, head width, which is linked with male–male competition and the ability to grasp females during mating (Cox et al., 2003; Scharf & Meiri, 2013), remained unchanged in the invasive populations. This is surprising, as changes in population density have been reported in both Hawaii and New Zealand, which could potentially influence the degree of male–male competition evident within populations. Baker (1979) reported very high densities of the delicate skink on Oahu, Hawaii. Similarly, the delicate skink occurs in high densities within the Auckland region of New Zealand (Chapple, Miller, et al., 2013; Chapple, Reardon, et al., 2016). However, it has a patchy distribution throughout the remainder of the country due to human-assisted ("spot-fire") dispersal (Chapple, Whitaker et al., 2013; Chapple, Knechtmans, et al., 2016; Chapple, Reardon, et al., 2016), and many populations are at lower densities (DGC, personal observation). In contrast, the increase in head size in an introduced population of the wall lizard (*Podarcis sicula*) was associated with a change in diet (towards a more plant-based diet) and gut morphology, and occurred in response to a higher population density in the introduced range (Herrel et al., 2008).

Despite the presence of intraspecific hybridization in the central and southern regions of Lord Howe Island among individuals from different native-range source regions (Chaplin, 2013; Chapple, Miller, et al., 2013; Chapple et al., 2014; Moule et al., 2015), no morphological shifts in shape were observed on the island. This result is in contrast to the invasive range of the brown anole (*Anolis sagrei*), where there was a positive relationship between the extent of morphological change and the degree of genetic admixture present within the invasive population (Kolbe, Larson, & Losos, 2007). Thus, genetic admixture on Lord Howe Island does not appear to have a fundamental impact on the aspects of delicate skink shape or size that we measured.

As predicted, opposing temporal shifts in the relative incidence of the mid-lateral stripe (becoming more prevalent in females and less so in males) have occurred in the delicate skink in Hawaii since its initial introduction in the early 1900s. Our temporal analysis of colour morph frequency in the Hawaiian population indicates that this shift is not simply due to a founder effect, but rather the result of post-introduction evolution. The relative morph frequencies in the native range have been stable over the same period (Forsman & Shine, 1995b), and the sex-specific shifts in the prevalence of the



mid-lateral stripe in Hawaii are consistent with the reported selective advantage of the trait (beneficial in females, detrimental in males; Forsman & Shine, 1995b). Climate appears to be a key determinate of colour morph frequency in the delicate skink (the prevalence decreases with increasing latitude; Forsman & Shine, 1995b), and thus the climatic shift experienced during the transition from Brisbane (the native-range source region; latitude 27°S) to Hawaii (latitude 21°N) might have driven the change in morph frequency. The change in the prevalence of the colour morph in both males and females occurred over 70 years (i.e. ~70 generations). Rapid evolution of latitudinal clines in phenotypic traits (including colour pattern) in the introduced range, which reflect those in the native range, also occurred in the house sparrow (Johnston & Selander, 1964) and *Drosophila subobscura* (Huey et al., 2000). However, the temporal shift in colour pattern in the Hawaiian delicate skink population appears to be slightly different in that the relative morph frequencies currently observed (98% in females, 6% in males; this study) have over-shot those evident at an equivalent latitude in the native range (Townsville: 19°S, 83% in females, 34% in males; Forsman & Shine, 1995b). Thus, the divergent selection on colour pattern has intensified in the Hawaiian population, and might be a result of niche evolution in Hawaii (Tingley et al., 2016) or differences in microhabitat or predation pressure in the Hawaiian Islands. In contrast, contrary to our prediction, no shift in colour morph frequency was observed in New Zealand, and only minor shifts (presumably due to genetic admixture of source clades with differing colour morph frequencies) were observed on Lord Howe Island.

ACKNOWLEDGEMENTS

We thank D. Bray (Museum Victoria), R. Sadlier (Australian Museum), A. Amey, P. Couper (Queensland Museum), R. Coory, C. Miskelly (Te Papa Tongarewa, National Museum of New Zealand), A. Allison, K. Imada (Bishop Museum, Hawaii), the Californian Academy of Sciences, C. Moritz (Museum of Vertebrate Zoology, UC Berkeley), A. Wynn and G. Zug (Smithsonian, US National Museum of Natural History) for facilitating access to the delicate skink specimens in their collections. F. Kraus provided valuable information and advice. The research was funded by the Australian Research Council (grant to D.G.C.; DP0771913), the National Geographic Society (grants to D.G.C. and M.B.T.; 8085-06, 8952-11), the Hermon Slade Foundation (grant to D.G.C. and M.B.T.; HSF 09-02), the Allan Wilson Centre for Molecular Ecology and Evolution and a Monash University School of Biological Sciences Small Grant (to D.G.C.).

DATA ACCESSIBILITY

Data will be submitted to Dryad doi:10.5061/dryad.r0874

REFERENCES

Baker, J. K. (1979). The rainbow skink, *Lampropholis delicata*, in Hawaii. *Pacific Science*, 33, 207–212.

- Bonneaud, C., Marnocha, E., Herrel, A., Vanhooydonck, B., Irschick, D. J., & Smith, T. B. (2016). Developmental plasticity affects sexual size dimorphism in an anole lizard. *Functional Ecology*, 30, 235–243.
- Campbell, T. S., & Echternacht, A. C. (2003). Introduced species as moving targets: Changes in body sizes of introduced lizards following experimental introductions and historical invasions. *Biological Invasions*, 5, 193–212.
- Chaplin, K. (2013). *Intra-specific hybridisation of the delicate skink (Lampropholis delicata) on Lord Howe Island*. Honours thesis. Melbourne, Australia: Monash University.
- Chapple, D. G., Hoskin, C. J., Chapple, S. N. J., & Thompson, M. B. (2011). Phylogeographic divergence in the widespread delicate skink (*Lampropholis delicata*) corresponds to dry habitat barriers in eastern Australia. *BMC Evolutionary Biology*, 11, 191.
- Chapple, D. G., Hutchinson, M. N., Maryan, B., Pivlich, M., Moore, J. A., & Keogh, J. S. (2008). Evolution and maintenance of colour pattern polymorphism in *Liopholis* (Squamata: Scincidae). *Australian Journal of Zoology*, 56, 103–115.
- Chapple, D. G., Knechtmans, J., Kikillus, H., & van Winkel, D. (2016). Biosecurity of exotic reptiles and amphibians in New Zealand: Building upon Tony Whitaker's legacy. *Journal of the Royal Society of New Zealand*, 46, 66–84.
- Chapple, D. G., Miller, K. A., Chaplin, K., Barnett, L., Thompson, M. B., & Bray, R. D. (2014). Biology of the invasive delicate skink (*Lampropholis delicata*) on Lord Howe Island. *Australian Journal of Zoology*, 62, 498–506.
- Chapple, D. G., Miller, K. A., Kraus, F., & Thompson, M. B. (2013). Divergent introduction histories among invasive populations of the delicate skink (*Lampropholis delicata*): Has the importance of genetic admixture in the success of biological invasions been overemphasized? *Diversity and Distributions*, 19, 134–146.
- Chapple, D. G., Reardon, J. T., & Peace, J. E. (2016). Origin, spread and biology of the invasive plague skink (*Lampropholis delicata*) in New Zealand. Chapter 13. In D. G. Chapple (Ed.), *New Zealand lizards* (pp. 341–359). Switzerland: Springer.
- Chapple, D. G., Simmonds, S. M., & Wong, B. B. M. (2011). Know when to run, know when to hide: Can behavioral differences explain the divergent invasion success of two sympatric lizards? *Ecology and Evolution*, 1, 278–289.
- Chapple, D. G., Whitaker, A. H., Chapple, S. N. J., Miller, K. A., & Thompson, M. B. (2013). Biosecurity interceptions of an invasive lizard: Origin of stowaways and human-assisted spread within New Zealand. *Evolutionary Applications*, 6, 324–339.
- Clegg, S. M., Degnan, S. M., Moritz, C., Estoup, A., Kikkawa, J., & Owens, I. P. F. (2002). Microevolution in island forms: The roles of drift and directional selection in morphological divergence in a passerine bird. *Evolution*, 56, 2090–2099.
- Cox, R. M., Skelly, S. L., & John-Alder, H. B. (2003). A comparative test of adaptive hypotheses for sexual size dimorphism in lizards. *Evolution*, 57, 1653–1669.
- Cromie, G. L., & Chapple, D. G. (2012). Impact of tail loss on the behaviour and locomotor performance of two sympatric *Lampropholis* skink species. *PLoS ONE*, 7, e34732.
- Downes, S., & Hoefer, A. M. (2007). An experimental study of the effects of weed invasion on lizard phenotypes. *Oecologia*, 153, 775–785.
- Forsman, A., & Shine, R. (1995a). Parallel geographic variation in body shape and reproductive life history within the Australian scincid lizard *Lampropholis delicata*. *Functional Ecology*, 9, 818–828.
- Forsman, A., & Shine, R. (1995b). The adaptive significance of colour pattern polymorphism in the Australian scincid lizard *Lampropholis delicata*. *Biological Journal of the Linnean Society*, 55, 273–291.
- Hassall, C., Keat, S., Thompson, D. J., & Watts, P. C. (2014). Bergmann's rule is maintained during a rapid range expansion in a damselfly. *Global Change Biology*, 20, 475–482.

- Herrel, A., Huyghe, K., Vanhooydonck, B., Backeljau, T., Breugelmans, K., Grbac, I., ... Irschick, D. J. (2008). Rapid large-scale evolutionary divergence in morphology and performance associated with exploitation of a different dietary resource. *PNAS*, *105*, 4792–4795.
- Herrel, A., Spithoven, L., Van Damme, R., & De Vree, F. (1999). Sexual dimorphism of head size in *Gallotia galloti*; testing the niche divergence hypothesis by functional analyses. *Functional Ecology*, *13*, 289–297.
- Herrel, A., Van Damme, R., & De Vree, F. (1996). Sexual dimorphism of head size in *Podarcis hispanica atrata*: Testing the dietary divergence hypothesis by bite force analysis. *Netherlands Journal of Zoology*, *46*, 253–262.
- Huey, R. B., Gilchrist, G. W., Carlson, M. L., Berrigan, D., & Serra, L. (2000). Rapid evolution of a geographic cline in size in an introduced fly. *Science*, *287*, 308–309.
- Johnston, R. F., & Selander, R. K. (1964). House sparrows: Rapid evolution of races in North America. *Science*, *144*, 548–550.
- Kolbe, J. J., Larson, A., & Losos, J. B. (2007). Differential admixture shapes morphological variation among invasive populations of the lizard. *Molecular Ecology*, *16*, 1579–1591.
- Kolbe, J. J., Leal, M., Schoener, T. W., Spiller, D. A., & Losos, J. B. (2012). Founder effects persist despite adaptive differentiation: A field experiment with lizards. *Science*, *335*, 1086–1089.
- McLean, C. A., & Stuart-Fox, D. (2014). Geographic variation in animal colour polymorphisms and its role in speciation. *Biological Reviews*, *89*, 860–873.
- Meiri, S., & Dayan, T. (2003). On the validity of Bergmann's rule. *Journal of Biogeography*, *30*, 331–351.
- Morrison, C., & Hero, J. M. (2003). Geographic variation in life-history characteristics of amphibians: A review. *Journal of Animal Ecology*, *72*, 270–279.
- Moule, H., Chaplin, K., Bray, R. D., Miller, K. A., Thompson, M. B., & Chapple, D. G. (2015). A matter of time: Temporal variation in the introduction history and population genetic structuring of an invasive lizard. *Current Zoology*, *61*, 456–464.
- Phillips, B. L., & Shine, R. (2005). The morphology, and hence impact, of an invasive species (the cane toad, *Bufo marinus*): Changes with time since colonisation. *Animal Conservation*, *8*, 407–413.
- Phillips, B. L., & Shine, R. (2006). Spatial and temporal variation in the morphology (and thus, predicted impact) of an invasive species in Australia. *Ecography*, *29*, 205–212.
- R Development Core Team. (2015). *R: A language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing. Retrieved from <http://www.R-project.org/>.
- Rius, M., & Darling, J. A. (2014). How important is intraspecific genetic admixture to the success of colonising populations? *Trends in Ecology and Evolution*, *29*, 233–242.
- Sax, D. F., Stachowicz, J. J., Brown, J. H., Bruno, J. F., Dawson, M. N., Gaines, S. D., ... Rice, W. R. (2007). Ecological and evolutionary insights from species invasions. *Trends in Ecology and Evolution*, *22*, 465–471.
- Scharf, I., & Meiri, S. (2013). Sexual dimorphism of heads and abdomens: Different approaches to 'being large' in female and male lizards. *Biological Journal of the Linnean Society*, *110*, 665–673.
- Shine, R. (2012). Invasive species as drivers of evolutionary change: Cane toads in tropical Australia. *Evolutionary Applications*, *5*, 107–116.
- Shine, R., Brown, G. P., & Phillips, B. L. (2011). An evolutionary process that assembles phenotypes through space rather than through time. *PNAS*, *108*, 5708–5711.
- Simberloff, D., Dayan, T., Jones, C., & Ogura, G. (2000). Character displacement and release in the small Indian mongoose, *Herpestes javanicus*. *Ecology*, *81*, 2086–2099.
- Stacklies, W., Redestig, H., Scholz, M., Walther, D., & Selbig, J. (2007). *pcaMethods* a bioconductor package providing PCA methods for incomplete data. *Bioinformatics*, *23*, 1164–1167.
- Stillwell, R. C., Morse, G. E., & Fox, C. W. (2007). Geographic variation in body size and sexual size dimorphism of a seed-feeding beetle. *American Naturalist*, *170*, 358–369.
- Suarez, A. V., & Tsutsui, N. D. (2008). The evolutionary consequences of biological invasions. *Molecular Ecology*, *17*, 351–360.
- Thomas, C. D., Bodsworth, E. J., Wilson, R. J., Simmons, A. D., Davies, Z. G., Musche, M., & Conradt, L. (2001). Ecological and evolutionary processes at expanding range margins. *Nature*, *411*, 577–581.
- Tingley, R., Thompson, M. B., Hartley, S., & Chapple, D. G. (2016). Patterns of niche filling and expansion across the invaded ranges of an Australian lizard. *Ecography*, *39*, 270–280.
- Wares, J. P., Hughes, A. R., & Grosberg, R. K. (2005). Mechanisms that drive evolutionary change: Insights from species introductions and invasions. In D. F. Sax, J. J. Stachowicz & S. D. Gaines (Eds.), *Species invasions. Insights into ecology, evolution and biogeography* (pp. 229–257). Sunderland, MA: Sinauer Associates.
- Wu, N. C., Alton, L. A., Clemente, C. J., Kearney, M. R., & White, C. R. (2015). Morphology and burrowing energetics of semi-fossorial skinks (*Liopholis*). *Journal of Experimental Biology*, *218*, 2416–2426.

BIOSKETCH

David Chapple's research group (<https://sites.google.com/site/chapplelab/>) investigates the evolutionary ecology of environmental change. We use field studies, field- and laboratory-based experiments, comparative analyses, morphological analyses and molecular approaches to examine the impact of past, current and future environmental change on phenotype, life history and distribution. In particular, we focus on species that display divergent responses to environmental changes, namely invasive species that expand their range, and threatened species that contract their range.

Author contributions: D.G.C. and M.B.T. conceived the project and obtained funding; K.A.M. and D.G.C. conducted the morphological analyses; K.A.M., J.M. and A.D. analysed the data; D.G.C. led the writing; and all authors commented on and edited the complete draft.

SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

How to cite this article: Miller KA, Duran A, Melville J, Thompson MB, Chapple DG. Sex-specific shifts in morphology and colour pattern polymorphism during range expansion of an invasive lizard. *J Biogeogr.* 2017;44:2778–2788. <https://doi.org/10.1111/jbi.13075>