Molecular Ecology (2008) **17**, 3668–3683 doi: 10.1111/j.1365-294X.2008.03864.x

A SINE of restricted gene flow across the Alpine Fault: phylogeography of the New Zealand common skink (*Oligosoma nigriplantare polychroma*)

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Abstract

New Zealand has experienced a complex climatic and geological history since the Pliocene. Thus, identifying the processes most important in having driven the evolution of New Zealand's biota has proven difficult. Here we examine the phylogeography of the New Zealand common skink (*Oligosoma nigriplantare polychroma*) which is distributed throughout much of New Zealand and crosses many putative biogeographical boundaries. Using mitochondrial DNA sequence data, we revealed five geographically distinct lineages that are highly differentiated (pairwise Φ_{ST} 0.54–0.80). The phylogeographical pattern and inferred age of the lineages suggests Pliocene mountain building along active fault lines promoted their divergence 3.98–5.45 million years ago. A short interspersed nuclear element (SINE) polymorphism in the myosin gene intron (*MYH-2*) confirmed a pattern of restricted gene flow between lineages on either side of the mountain ranges associated with the Alpine Fault that runs southwest to northeast across the South Island of New Zealand. An analysis of molecular variance confirmed that ~40% of the genetic differentiation in *O. n. polychroma* is distributed across this major fault line. The straits between the main islands of New Zealand accounted for much less of the variation found within *O. n. polychroma*, most likely due to the repeated existence of landbridges between islands during periods of the Pleistocene that allowed migration. Overall, our findings reveal the relative roles of different climatic and geological processes, and in particular, demonstrate the importance of the Alpine Fault in the evolution of New Zealand's biota.

Keywords: glacial cycles, mitochondrial DNA, mountain building, Pleistocene, Pliocene, transposable element

Received 22 March 2008; revision accepted 18 June 2008

Introduction

Phylogeographical studies worldwide have recognized that patterns in genetic structuring and speciation are intimately related to events during the Pliocene and Pleistocene. The specific geological and climatic events that have driven phylogeographical patterns within these periods vary depending on the focal region. In continental regions of the Northern Hemisphere, such as Europe and North America, studies of genetic variation across many taxa have led to the development of well-supported hypotheses implicating the glacial cycles of the Pleistocene (Hewitt 1996, 2000). However, in New Zealand tectonic events have been suggested as being more important in determining general patterns (McGlone 1985).

Many biogeographical hypotheses surround the Pliocene and Pleistocene history of New Zealand. During this period, the New Zealand landscape experienced partial marine inundation, volcanic activity, glacial cycles, and, most significantly, tectonism associated with its position at the boundary of the Australasian and Pacific plates (Suggate 1985; Worthy & Holdaway 2002). Tectonic activity has been concentrated along the Alpine Fault (Fig. 1) that stretches diagonally across the South Island from the southwest to the

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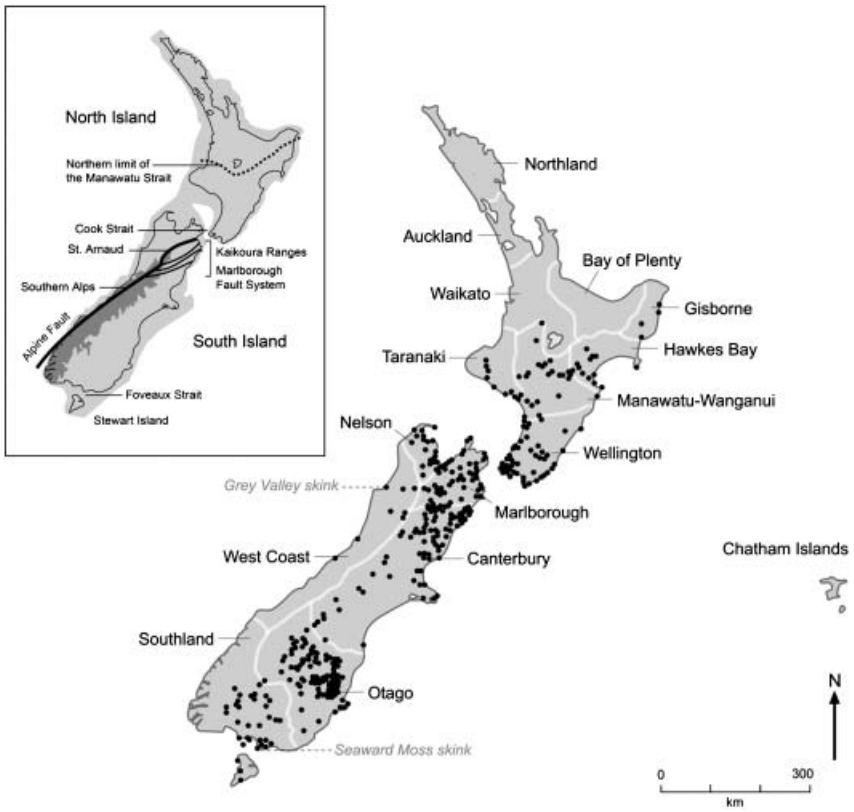

Fig. 1 The distribution of *Oligosoma nigriplantare polychroma* (BioWeb Herpetofauna Database 2006) and regions of New Zealand. The locations of the Grey Valley and Seaward Moss skinks are indicated. Inset: the biogeographical features of New Zealand. The dashed line approximates the Pliocene northern boundary of the Manawatu Strait in the North Island. The solid lines represent the approximate location of the Alpine Fault (heaviest) and Marlborough Fault System in the South Island. The land area (light shaded) and extent of glacial ice (dark shaded) during the Pleistocene are shown.

northeast, bisecting the ranges of many taxa. Fault activity has generated > 460 km of lateral motion over the past 25 million years (Myr) and 20 km of uplift in just 10 Myr (Whitehouse & Pearce 1992), likely to have induced both allopatric divergence and adaptive divergence through the creation of alpine habitat. Mountain building was greatest during the Pliocene 2–5 million years ago (Ma) (Whitehouse & Pearce 1992; Batt *et al*. 2000), forming the Southern Alps that run almost the length of the South Island and reach over 3000 m above sea level (a.s.l.) (Suggate 1990). This uplift drained the Manawatu Strait that existed (2–6 Ma) across the present-day Cook Strait (between the North and South Islands, Fig. 1) and southern North Island, now considered to be a biologically depauperate region (Rogers 1989). Furthermore, the creation of high-elevation areas accentuated the impact of the climate related glacial cycles of the Pleistocene that followed (Wallis & Trewick 2001).

Glacial periods (< 20 cycles) during the Pleistocene (0.01–1.8 Ma) are thought to have been less severe in New Zealand than in the continental Northern Hemisphere due to New Zealand's maritime setting (Wallis & Trewick 2001; Gibbs 2006). However, glaciation was still extensive in areas of high altitude (> 30% of land area; Wallis & Trewick 2001) and in some regions ice reached sea level (Newnham *et al*. 1999). The combined effects of glaciation during interstadials, and glacial outwash during interglacials, are thought to have extirpated populations from the central 'waist' of the South Island (Canterbury), resulting in a biologically depauperate region called the 'Beech gap' (Gibbs 2006). In contrast, the lowered sea levels (~130 m) during interstadials, are also hypothesized to have facilitated gene flow among island populations of terrestrial species through the formation of landbridges across Cook Strait and Foveaux Strait (between the South Island and Stewart Island, Fig. 1; Suggate *et al*. 1978; Fleming 1979; McGlone 1988).

Phylogeographical investigation in New Zealand has generally focused on taxa restricted to specific habitats (e.g. coastal: pohutukawa, *Metrosideros* spp., Gardner *et al*. 2004; forest dwelling: brown kiwi, *Apteryx australis*, Baker *et al*. 1995; Burbidge *et al*. 2003; frogs, *Leiopelma* spp., Holyoake *et al*. 2001; bats, *Mystacina tuberculata*, Lloyd 2003; freshwater: galaxiid fish, Waters & Wallis 2000; gobiid fish, Smith *et al*. 2005; crayfish *Paranephrops* spp., Apte *et al*. 2007; snails, *Potamopyrgus antipodarum*, Neiman & Lively 2004; alpine: weta, *Deinacrida connectens*, Trewick 2001; cicada, *Maoricicada* spp., Buckley *et al*. 2001; Buckley & Simon 2007; plants, *Pachycladon* spp., Heenan & Mitchell 2003) or have been geographically limited (e.g. Northland: skinks, *Oligosoma suteri*, *O. smithi*, *O. moco*, Hare *et al*. 2008; southern South Island: skinks, *O. grande*, Berry & Gleeson 2005). Few studies have considered common and widespread species. Studying ubiquitous species may help exclude the confounding

influences of habitat specialization on the identification of general phylogeographical patterns (Trewick 2001; Whitely *et al*. 2006) and examining the phylogeography of widespread species enables multiple hypothesized biogeographical barriers/regions to be considered simultaneously (Berry & Gleeson 2005).

The New Zealand common skink (*Oligosoma nigriplantare polychroma*) is one such ubiquitous species. It is an ideal model species to examine the evolutionary history of the New Zealand biota since the Pliocene. Terrestrial reptiles are generally poor dispersers, and thus, maintenance of gene flow is sensitive to the formation of landscape barriers making them particularly informative in phylogeographical studies (Avise 2000). *O. n. polychroma* is the most widespread skink species in New Zealand and one of the most extensively and continuously distributed endemic organisms. Its range throughout the southern North Island, much of the South Island and Stewart Island (Fig. 1, Patterson & Daugherty 1990; Gill & Whitaker 2001) has been less affected by human landscape modification and introduced predators than most New Zealand endemics (Towns & Ferriera 2001). *O. n. polychroma* remains abundant in coastal areas, shingle riverbeds, tussock grassland, farmland and urban areas, up to an altitude of 1700 m a.s.l. (Patterson 1992; Towns & Elliot 1996; Gill & Whitaker 2001; Towns *et al*. 2002).

The substantial geographical variation in morphology (*polychroma* = 'many colours') and ecology of *O. n. polychroma* might be indicative of phylogeographical structure and potentially unrecognized taxa (Daugherty *et al*. 1990; Patterson & Daugherty 1990; Towns *et al*. 2002). For instance, an *O. n. polychroma* population (named the 'Grey Valley' skinks) found on the West Coast of the South Island (Fig. 1) has been proposed to be distinct on the basis of morphology and allozymes (Miller 1999). At Tiwai Point in Southland (Fig. 1), there exist two distinct morphotypes of *O. n. polychroma*, one of which has been referred to as the 'Seaward Moss' skink (previously called the 'Tiwai Point Brown') due to its highly divergent brown colour and unstriped pattern (R. Hitchmough, personal communication). Allozyme evidence suggests that the Seaward Moss skink might represent a colour morph of *O. n. polychroma* (C.H.D., unpublished data), but this warrants further investigation.

In this study, we have investigated the phylogeography of *O. n. polychroma* using mitochondrial DNA (mtDNA) sequence data (ND2, ND4) and a SINE polymorphism in the nuclear myosin gene intron (*MYH-2*). We have examined how genetic variation is partitioned across geological features and from our observations inferred the extent to which these features have been important in influencing the phylogeographical patterns within *O. n. polychroma*. We have also tested hypotheses concerning the taxonomic status of Grey Valley and Seaward Moss skinks.

Materials and methods

Sampling

We obtained tissue samples from across the entire distribution of *Oligosoma nigriplantare polychroma* including eight samples from the Grey Valley skink and one sample from the Seaward Moss skink morphotype (Fig. 1, see Table S1, Supplementary material). Samples were obtained from field collections, a frozen tissue collection (–80 °C) housed at Victoria University of Wellington and ethanol-preserved specimens stored at the Museum of New Zealand, Te Papa Tongarewa. Based on a broader phylogenetic study of New Zealand Scincidae (D.G.C., C.H.D. and P.A.R., unpublished data), samples were also taken from the endemic small-scaled skink (*O. microlepis*), robust skink (*Cyclodina alani*), and the Chatham Islands skink (*O. n. nigriplantare*), as outgroups for our analyses (see Table S1). (We examine the phylogeography of *O. n. nigriplantare* in more detail elsewhere; Liggins *et al*. 2008).

Molecular methods

We extracted whole genomic DNA from heart, muscle or liver tissues using a standard phenol/chloroform protocol (Sambrook *et al*. 1989) followed by ethanol precipitation.

Using the polymerase chain reaction (PCR), we chose to amplify the mitochondrial coding regions ND2 (~600 bp) and ND4 (~850 bp: incorporating most of the flanking-3′ tRNA cluster, including the histidine and serine tRNA genes) based on their relatively high rate of evolution and proven utility in both intraspecific and interspecific studies of skinks (Chapple & Keogh 2004; Chapple *et al*. 2004; Fuerst & Austin 2004). The primers used to amplify and sequence these regions are listed in Table 1. PCRs were 25 μL in total volume, containing: 1 μL of DNA template, 0.4 μM of each primer, 0.4 mg/mL BSA, 200 μM of each dNTP and either: (i) 1× reaction buffer (67 mM Tris-HCl (pH 8.8), 16 mM $(NH_4)_2SO_4$, 1.5 mM $MgCl_2$) and 1 U of BioTherm DNA Polymerase (GeneCraft), or (ii) 1× reaction buffer (20 mM Tris-HCl (pH 8.4), 50 mM (KCl), 1.5 mM $MgCl_2$ and 1 U of Platinum *Taq* DNA Polymerase (Invitrogen). Amplification of target DNA was conducted on an Eppendorf Mastercycler ep gradient thermal cycler, using the following conditions: denaturation at 94 °C for 3 min, 35 cycles of 30 s at 94 °C, 20 s at 50 °C (annealing) and 50 s at 72 °C, followed by extension at 72 °C for 5 min. For samples that failed to amplify using these conditions, a gradient PCR with annealing temperatures ranging from 40 to 60 °C was conducted. PCR products were purified either using the ExoSAP-IT enzyme method (USB Corporation) or a High Pure PCR Product Purification Kit (Roche Diagnostics). The purified products were directly sequenced using BigDye Terminator version 3.1 and analysed on an ABI 3730 Capillary Sequencer

Table 1 Oligonucleotide primers used in this study. Values in '5′ position' refer to the position of the 5′ base of the primer in the complete *Eumeces egregius* mtDNA sequence (Kumaza & Nishida 1999)

Gene region	Primer	5′–3′ sequence	5′ position	Source
ND2	L4437	AAGCTTTCGGGCCCATACC	3833	Macey *et al*. 1997
	ND2r102	CAGCCTAGGTGGGCGATTG	4432	Sadlier *et al*. 2004
	ND2F-infrapunctatum	GCATGATTYACCGGAAYATGAGACAT	4141	Greaves *et al*. 2007
	ND2R-infrapunctatum	GGGGCAAGKCCTAGTTTTATGG	4192	Greaves *et al*. 2007
ND4	ND4I	TGACTACCAAAAGCTCATGTAGAAGC	10 771	Forstner *et al*. 1995
	tRNA-Leu	TACTTTTACTTGGATTTGCACCA	11 691	Forstner *et al*. 1995
	ND4F-infrapunctatum	GCATGATTYACCGGAAYATGAGACAT	11 153	Greaves *et al*. 2007
	ND4R-infrapuctatum	GGGGGATCAGTTAAAYAAYGAGGTG	11 234	Greaves *et al*. 2007
Myosin heavy chain intron 2	Myh2-F	GAACACCAGCCTCATCAACC	.	Lyons *et al*. 1997
	G315 (F)	TCAGAGTCACAATAAGGACAC	.	Smith 2001

(Applied BioSystems). Sequence data were edited manually using ContigExpress in Vector NTI Advance 9.1.0 (Invitrogen) and then aligned using CLUSTAL W (Thompson *et al*. 1994) executed in MEGA 3.1 (Kumar *et al*. 2004).

Transposon screening

A SINE was identified in the nuclear myosin gene intron (heavy chain myosin 2, *MYH-2*) in *O. n. polychroma* by Smith (2001). Our subsequent screening of all New Zealand skink taxa revealed that the SINE was only present in *O. n. polychroma* and its presence/absence was polymorphic in this subspecies. These attributes made it an ideal nuclear marker to incorporate in our phylogeographical study of *O. n. polychroma*. The *MYH-2* intron was ~488 bp in individuals without the SINE, and ~864 bp in individuals with the SINE. This substantial size difference enabled us to determine its presence/absence in each *O. n. polychroma* sample using gel electrophoresis to size-separate the PCR products. *MYH-2* was amplified using the primers listed in Table 1 as per the PCR conditions listed above.

Molecular diversity and phylogenetic analyses of O. n. polychroma

ND2 and ND4 data sets were concatenated and estimates of genetic diversity within *O. n. polychroma* were performed in ARLEQUIN version 3.0 (number of haplotypes *na*, nucleotide diversity π; Excoffier *et al*. 2005) and MEGA (model-corrected sequence divergence *d*, segregating sites *S*, parsimony-informative sites Pi). For all taxa (*O. n. polychroma* plus outgroups), we used PAUP* to estimate the transition/transversion ratio (ti/tv), determine base frequencies, and implement χ^2 tests to test for equal base frequencies across sequences, using variable sites only. Tajima's *D* test (Tajima 1989, 1996) performed in ARLEQUIN tested the hypothesis that all the sequences represent neutral markers. To identify the model of evolution that best fit our sequence data, we performed hierarchical likelihood-ratio tests (hRLT) in MODELTEST 3.7 (Posada & Crandall 1998) from the log-likelihood scores generated in PAUP*. The Tamura–Nei model with invariable sites and gamma-distributed rate variation (TrN + *I* + *G*, *I* = 0.558, α parameter = 1.394, $-\ln L$ = 7987.2388) was selected as most appropriate.

Bayesian, maximum parsimony (MP) and maximum likelihood (ML) methods of tree-building were implemented using the parameters estimated by MODELTEST executed in PAUP*. MP and ML heuristic searches were conducted in PAUP* using random stepwise addition and the tree-bisection–reconnection (TBR) branch-swapping algorithm. For Bayesian analyses, we used the general time reversible model (Rodriguez *et al*. 1990) with invariable sites and gamma-distributed rate variation (GTR + *I* + *G*), because the TrN model was not an option in MRBAYES version 3.1 (Ronquist & Huelsenbeck 2003). We performed four Metropolis-coupled Markov chain Monte Carlo (MCMCMC) runs, started from a random tree and ran the analyses for 1 million generations in MRBAYES. The chains were sampled every 100 generations to obtain 10 000 trees, of which the first 2500 were discarded as the burn-in phase. To assess the statistical support for the final topology, we used both Bayesian posterior probabilities and ML bootstrap analyses (100 replicates).

To test the taxonomic status of proposed undescribed taxa in *O. n. polychroma*, we conducted Shimodaira–Hasegawa tests (SH tests; Shimodaira & Hasegawa 1999) of different phylogenetic tree topologies in PAUP* (1000 bootstraps). The optimal ML tree derived from our concatenated molecular data was compared with two alternate topologies: the first tree constrained the Grey Valley skink (Miller 1999) as monophyletic and sister to *O. n. polychroma*. The second positioned the Seaward Moss skink (ONP4) as a phylogenetically distinct taxon.

Spatial genetic patterns within O. n. polychroma

The association between genetic distance and geographical distance in *O. n. polychroma* was examined for isolation

by distance (Wright 1943). Genetic distance (pairwise TrN) among individuals was plotted against straight line geographical distance between individuals using reduced major axis (RMA) regression (Sokal & Rohlf 1981) as implemented by the Isolation By Distance Web Service (IBDWS; Bohonak *et al*. 2005). All zero values for both genetic and geographical distance were adjusted to 0.0001. The significance of the association was determined by applying Mantel's permutation test (Mantel 1967) with 10 000 matrix randomizations. Isolation-by-distance analyses were carried out at two levels: over the entire country incorporating all samples, and in posteriorly identified geographically and genetically isolated groups.

Multidimensional scaling (MDS) of pairwise TrN corrected genetic distances projected genetic differentiation between samples on a two-dimensional plane. Visualization of the relative genetic distances between samples allows identification of abrupt genetic breaks between sites, or clustering of sampling sites, that would suggest significant genetic structuring rather than simply isolation by distance. All zero values (representing no genetic distance) were adjusted to 0.0001 for projection.

Genetic differentiation across the range of *O. n. polychroma* was estimated in ARLEQUIN. Pairwise Φ_{ST} values (incorporating a measure of the molecular distance between haplotypes) were derived among groups that were identified before observing the genetic data to test specific hypotheses (i.e. the Alpine Fault, Cook Strait and Foveaux Strait), as well as groups that were allocated a posteriori based on observed phylogenetic relationships. We used analyses of molecular variance (AMOVA; Excoffier *et al*. 1992) as implemented by ARLEQUIN, to examine the significance of hypothesized genetic breaks, and to assess whether the groupings based on tree topology represented the distribution of genetic variation across the range of *O. n. polychroma.* Significance levels of all the estimated values were calculated by 10 000 permutations, and adjusted according to the Bonferroni correction procedure (Rice 1989) for multiple pairwise comparisons as described by Holm (1979).

Mismatch frequency histograms were plotted in DNASP version 3 (Rozas & Rozas 1998) to determine whether the number of pairwise differences within a posteriori-identified groups reflected spatial range expansion or a stationary population history (Harpending 1994). A smooth bell shape signifies either population expansion or spatial range expansion, whereas a multimodal distribution represents a long history *in situ* (Rogers & Harpending 1992; Ray *et al*. 2003; Excoffier 2004). To distinguish between these two types of distribution, a raggedness index (*r*, sum of the squared difference between neighbouring peaks) and the sum of squared deviations (SSD) between the observed and the expected mismatch were calculated using the methods of Schneider & Excoffier (1999) in ARLEQUIN. The validity of the spatial expansion hypothesis (both *r* and SSD) was tested using a parametric bootstrap approach (200 replicates).

Estimating divergence dates within O. n. polychroma

To estimate the divergence time of lineages, we calibrated the evolutionary rate of ND2 by re-analysing the data from Macey *et al*. (1998) for the agamid lizards of the *Laudakia caucasia* species group. Specifically, we recalculated the evolutionary rate for *L. caucasia* using only the 550 bp fragment of ND2 used in the present study (e.g. Greaves *et al*. 2007, 2008; Smith *et al*. 2007; Hare *et al*. 2008). We calculated average between-group nucleotide differences across each of the calibrated nodes (1.5 Ma, 2.5 Ma, 3.5 Ma) from Macey *et al*. (1998), plotted them against time and then used the slope of the linear regression to calculate a rate of evolution for our 550 bp fragment of ND2. This resulted in an evolutionary rate of 1.4% per Myr (0.7% per lineage, per Myr) and is slightly faster than the rate of 1.3% per Myr found by Macey *et al*. (1998). All divergence time estimates presented are based on mean uncorrected divergences derived from ND2 (550 bp).

Results

Molecular diversity and phylogenetic analyses of O. n. polychroma

The ND2 (550 bp) and ND4 (773 bp) gene regions were successfully amplified in 84 *Oligosoma nigriplantare polychroma* samples (representing 74 localities); however, only ~535 bp of ND2 was obtained for ONP41 and ONP76. The full data set comprised the concatenated ND2 and ND4 sequences of *O. n. polychroma*, three *O. n. nigriplantare* individuals and two outgroups (no insertions or deletions; concatenated length = 1323 bp). The aligned sequences were translated into amino acid sequences using the vertebrate mitochondrial genetic code. As no premature stop codons were observed (apart from within the tRNAs flanking the ND4 sequence), we conclude that all sequences obtained are true mitochondrial copies. Sequences are available on GenBank Accession nos EF033052, EF033068, EF043106, EF043107, EF043162–EF043170, EF043225–EF043229, EU074693–EU074769 and EU077420–EU077496 (see Table S1).

The ingroup (*O. n. polychroma*) had 951 conserved sites and 372 variable sites (per site, pS = 0.2812), of which 273 were parsimony informative. At the first position, 81 sites were polymorphic, compared with 116 and 171 polymorphisms at the second and third positions, respectively. Across all sites and all sequences (*O. n. polychroma* plus outgroups) the relative rates of each substitution type estimated under a TrN + I + G model revealed a predominance of transitions (ti/tv = 5.4: A↔C = 1.000, A↔G = 25.143, A↔T = 1.000, C↔G = 1.000, C↔T = 14.781, relative to

Clade	*n*	π	*S*	Φ	Tajima's *D*	%TrN *d*
1a	20	0.024 ± 0.012	133	0.682	–0.638	2.485 (0–4.568)
1b	20	0.011 ± 0.006	77	0.698	–1.311	1.104 (0–2.631)
2	5	0.022 ± 0.014	103	0.689	–0.729	2.335 (0.384–4.560)
3	7	0.031 ± 0.018	92	0.678	–0.151	3.206 (1.070–5.918)
4	6	0.029 ± 0.017	2	0.680	–0.261	3.023 (0.456–4.334)
5	26	0.018 ± 0.009	119	0.689	–0.867	1.881 (0.076–3.516)
Σ =	84	0.054 ± 0.026	372	0.491	–0.125	5.186 (0–10.549)

Table 2 Nucleotide diversity (π ± SD), polymorphic sites (*S*), Φ_{ST} values within, Tajima's *D*, mean and range of Tamura–Nei corrected genetic distances (%TrN *d*) of *Oligosoma nigriplantare polychroma* clades. Number of individuals per clade is given as *n*

G↔T = 1.000), but a χ^2 test indicated no significant base composition heterogeneity for variable sites among sequences (χ^2 = 101.0853, d.f. = 264, *P* = 1.000, *P* > 0.05). For all variable sites, the mean base frequencies for A, C, G, and T were 0.340, 0.319, 0.107 and 0.234, respectively.

We found considerable genetic diversity within *O. n. polychroma* (Table 2). There were 77 haplotypes across our 84 samples with an overall nucleotide diversity (π ± SD) of 0.054 ± 0.026. Pairwise sequence divergence (%TrN *d* ± SE) in *O. n. polychroma* was up to 10.549 ± 1.039 (ONP93, Wairau Valley and ONP25, Lake Hawea), with a mean of 5.186.

All phylogenetic analyses recovered the same five clades (designated 1, 2, 3, 4 and 5; Fig. 2). Mean divergences among the five clades ranged from 5.740 (Clades 1 and 2) to 8.509%TrN *d* (Clades 3 and 5), placing divergence dates around 3.98–5.45 Ma. Clade 1 contained two subclades, designated 1a and 1b (2.946%TrN *d*, estimated divergence 1.93 Ma). In our ML tree (–ln*L* = 7978.498; Fig. 2), Clades 1, 4 and 5 were well supported with posterior probabilities and bootstrap values of 1.0 and 100, respectively, but Clades 2 and 3 had much lower support (posterior probabilities and bootstrap values of 0.5 and 55, respectively; Fig. 2). The basal relationships between Clades 1, 2, 3 and 4 were not consistent across the different phylogenetic methods used. Bayesian analysis presented Clades 1, 2 and 3 as sister groups, whereas MP analysis found Clades 1 and 3 as sister groups, as well as Clades 2 and 4.

Analysis of the mtDNA variation within clades using Tajima's *D* indicated that all clades had negative *D* values (Table 2) and Clade 1b had a *D* value near significance (*P* = 0.097). However, the null hypothesis of neutral evolution could not be rejected for any clade or for the entire ingroup (*P* > 0.05). Nucleotide sequence diversity (π ± SD; Table 1) within each of the clades ranged from 0.011 ± 0.006 (Clade 1b) to 0.031 ± 0.018 (Clade 3).

Transposon screening

At least two individuals from each of the mtDNA clades within *O. n. polychroma* were successfully screened for the SINE (63 total; see Table S1). The insertion was always present in samples from Clades 1 and 2 (including samples from the North Island and the north of the South Island) and absent from all others (Fig. 3). Where *MYH-2* was not successfully amplified, there was also difficulty in the amplification of the mtDNA gene regions, most likely due to poor quality of the DNA template (predominantly those sourced from museum collections without information on method of preservation; see Table S1). The mean divergence (%TrN *d* ± SE) between those individuals that contained the transposon and those that did not was 7.516 ± 0.550 (estimated divergence time 5.11 Ma).

Spatial genetic patterns within O. n. polychroma

The five clades are geographically distinct (Fig. 3). Clade 1 is widespread throughout the North Island and across Cook Strait, in the northwest of the South Island and down the West Coast. Clade 1a is confined to the North Island and Clade 1b is largely confined to the South Island, except for ONP71 from Castlepoint on the east coast of the Wellington region. Clade 1b is mostly restricted to the west of the major mountain ranges of the Alpine Fault, although it occupies lower altitude mountain passes (Fig. 1). There is one shared haplotype in Clade 1a (ONP63, Ward Island and ONP50, Matiu-Somes Island; both in Wellington Harbour), and two shared haplotypes in Clade 1b (GVS1 and GVS3, Kangaroo Creek; and GVS4-GVS7, Blaketown Beach and GVS2, Mawheraiti Railway). Clade 1b contains the proposed divergent populations from the Grey Valley on the West Coast. These Grey Valley skinks did not form a monophyletic group within the ML tree, and the topology representing the hypothesized phylogenetic relationship between the Grey Valley skinks and *O. n. polychroma* (–ln*L* = 8141.172) was significantly less likely (SH test; *P* < 0.05) than that of the optimal ML tree (–ln*L* = 7978.498, Fig. 2).

Clades 2, 3 and 4 are found in the northeast and central South Island (mean 5.880–7.670%TrN *d* among, estimated divergence times 3.98–4.63 Ma). Clade 2 is found around the coastal areas of the Marlborough region and the Wairau River that runs alongside the Alpine Fault (Figs 1 and 3). Clade 3 is found farther south in the mountainous Canterbury region, including the Seaward and Inland Kaikoura ranges (Fig. 1) and those directly associated with the Alpine Fault. This clade is the most range-restricted, but

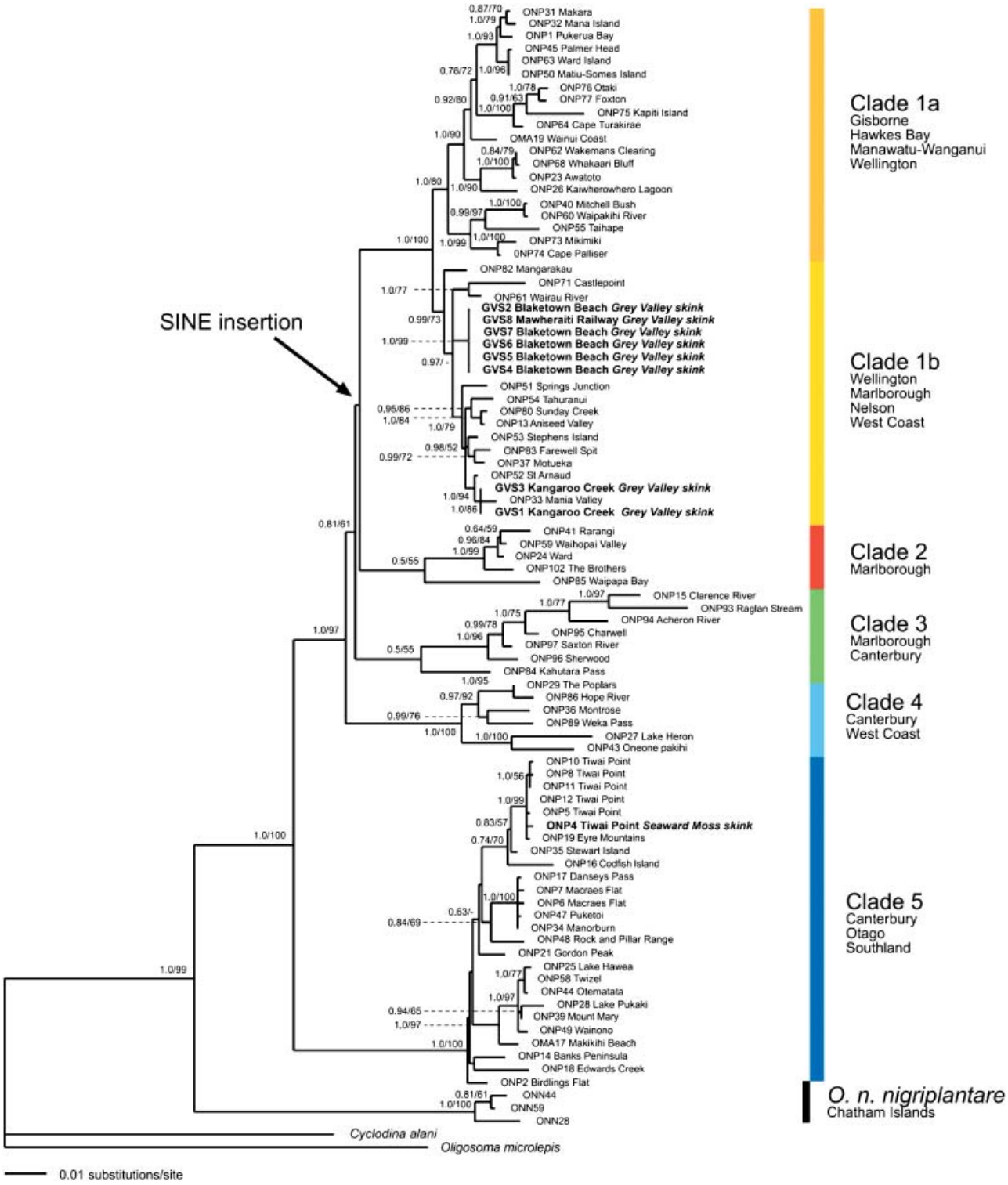

Fig. 2 Maximum likelihood (ML) phylogenetic tree based on the combined mtDNA gene regions (ND2 and ND4, TrN + *I* + *G* model of substitution), showing the relationships within *Oligosoma nigriplantare polychroma* (–ln*L* = 7978.498). The short interspersed nuclear element (SINE) insertion event in the nuclear gene intron *MYH-2* is mapped onto the phylogeny (presence/absence). Branch support is shown as Bayesian posterior probability/ML bootstrap support. The Grey Valley and Seaward Moss skink samples are indicated in bold text. The tree is rooted with *O. n. nigriplantare*, *O. microlepis* and *Cyclodina alani*.

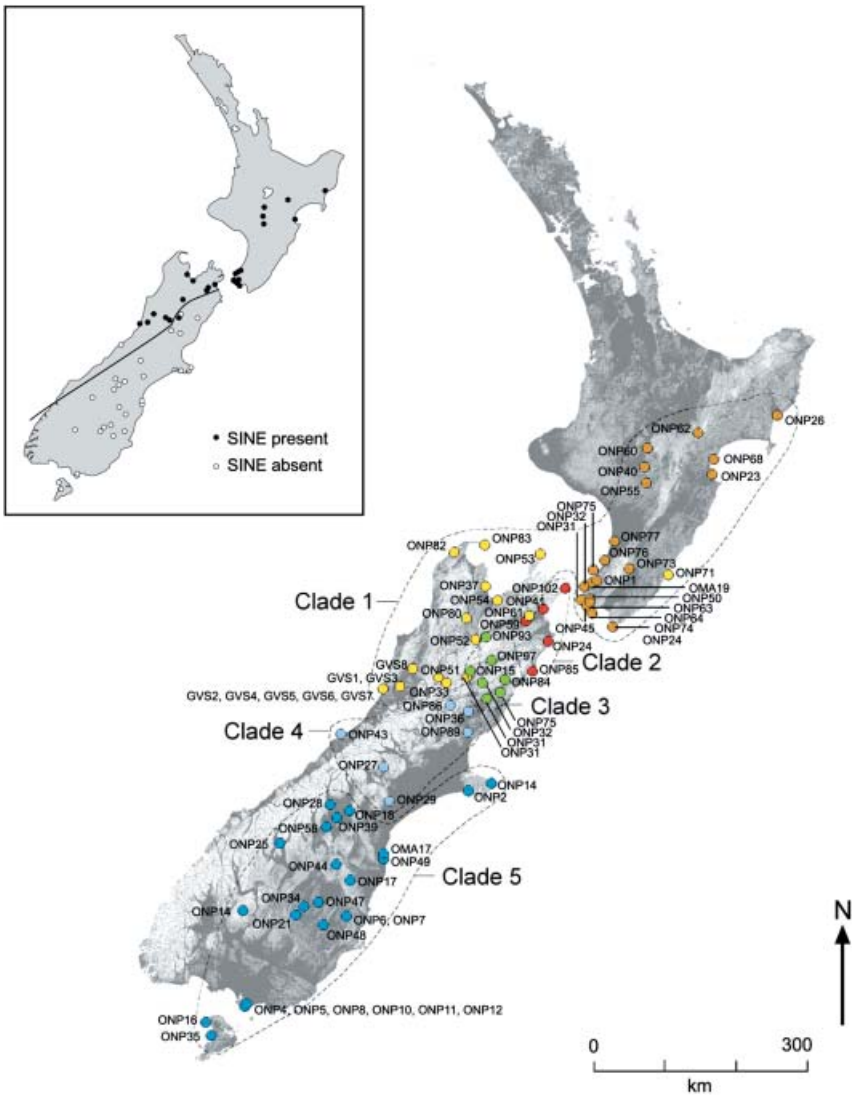

Fig. 3 Distribution of *Oligosoma nigriplantare polychroma* samples. Colours denote the phylogenetic affinity of the samples: orange, Clade 1a; yellow, Clade 1b; red, Clade 2; green, Clade 3; light blue, Clade 4; dark blue, Clade 5. Inset: distribution of samples successfully screened for the presence (filled circles) and absence (empty circles) of the short interspersed nuclear element (SINE) in the nuclear gene intron *MYH-2*. The solid line represents the approximate position of the Alpine Fault.

contained the greatest divergences (%TrN $d \pm$ SE; Table 2), up to 5.918 ± 0.720 (ONP93, Raglan Stream and ONP84, Kahutara Pass) with a mean of 3.206. In the central South Island, Clade 4 occupies the Canterbury coast south of the Waiau River, and crossed the central South Island to the West Coast.

Clade 5 represents the southernmost range of the common skink and is widespread in Canterbury, Otago and across Foveaux Strait in Southland (Figs 1 and 3). However, divergences in Clade 5 are lower than those of the other clades (%TrN $d \pm$ SE; Table 2). The morphologically divergent Seaward Moss skink sample (ONP4) from Tiwai Point in Southland was not found to be genetically distinct. An SH test confirmed that the hypothesized phylogenetic relationship between the Seaward Moss skink and *O. n. polychroma* presented a topology that was significantly less likely ($-\ln L = 8090.784$, $P < 0.05$) than the optimal ML topology ($-\ln L = 7978.498$, Fig. 2).

Although we found a significant positive relationship between genetic differentiation and geographical distance across all of our samples ($r^2 = 0.321$, $r = 0.567$, one-sided $P = 0.0001$), this was likely an artefact of more abrupt genetic breaks across the range of *O. n. polychroma*. Multidimensional scaling of the TrN genetic distances revealed three distinct clusters representing separate geographical regions of New Zealand (Fig. 4). The most isolated cluster of samples corresponded to Clade 5 representing samples from the Canterbury, Otago and Southland regions. Likewise, Clade 1 forms a relatively tight cluster representing those samples from the North Island and the northwest of the South Island. Clades 2, 3 and 4 do not separate clearly in the two-dimensional space, possibly indicating close genetic affinity to one another and/or not with Clades 1 and 5. The cluster of Clades 2, 3 + 4 has a larger spread, indicative of the greater genetic diversity found among these samples from the northeast of the South Island. This cluster was most

Table 3 Hierarchical analyses of molecular variance (AMOVA) employing uncorrected genetic distances for *Oligosoma nigriplantare polychroma* over a priori and a posteriori hypothesized barriers. All samples were included in each analysis unless stated otherwise. SI indicates samples form the South Island; SINE refers to the short interspersed nuclear element polymorphism in the nuclear gene intron *MYH-2*. Statistical significance (*P*) was tested with 10 000 permutations

	Observed partition					
Source of variation	Among groups percentage	Within groups percentage	Total variance components	*P*	Total d.f.	Total sum of squares
a priori						
Cook Strait (Clade 1 only)	39.3	60.7	19.121	< 0.01	39	603.000
Foveaux Strait (Clade 5 only)	7.3	92.7	15.844	0237	26	386.222
Alpine Fault (SI samples only)	40.3	59.7	46.311	< 0.01	62	2248.429
a posteriori						
Clades 1a, 1b, 2, 3, 4, 5	68.8	31.2	42.066	< 0.01	83	2973.929
SINE-delineated break	44.8	55.2	46.070	< 0.01	83	2973.929

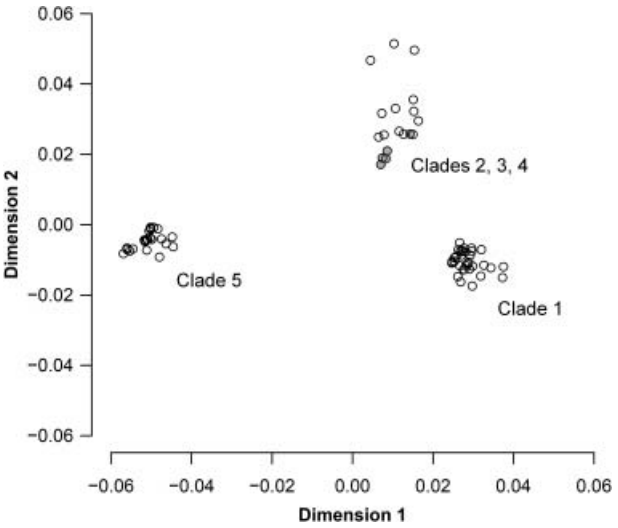

Fig. 4 Multidimensional scaling of Tamura–Nei (TrN) corrected genetic distances among the *Oligosoma nigriplantare polychroma* individuals revealed three clusters that represent separate geographical regions of New Zealand, labelled: Clade 1, Clades 2, 3 + 4, and Clade 5. Shaded circles represent individuals from Clade 2.

Table 4 Pairwise Φ_{ST} estimates (below diagonal) and significance of differentiation (above diagonal) among clades of *Oligosoma nigriplantare polychoma*. Asterisks denote statistical significance following sequential Bonferroni correction ($\alpha = 0.05$)

Clade	1a	1b	2	3	4	5
1a	—	0.000*	0.000*	0.000*	0.000*	0.000*
1b	0.393	—	0.000*	0.000*	0.000*	0.000*
2	0.590	0.733	—	0.003*	0.003*	0.000*
3	0.575	0.707	0.540	—	0.001*	0.000*
4	0.590	0.728	0.564	0.559	—	0.000*
5	0.730	0.795	0.736	0.720	0.728	—

closely related to that of Clade 1, particularly the samples that corresponded to Clade 2 (containing the SINE).

Hierarchical AMOVA revealed that among-clade (Clades 1a, 1b, 2, 3, 4 and 5) differences accounted for 68.8% of the overall variation in *O. n. polychroma* (Table 3). Pairwise Φ_{ST} comparisons for the clades were all significant following Bonferroni correction and Clade 5 was found the most differentiated (Table 4). By comparison, genetic differentiation across the Alpine Fault accounted for 40.3% ($P < 0.01$, Table 3) of the variation found within South Island samples of *O. n. polychroma* (mean 6.806% uncorrected ND2 *d* between South Island individuals either side of the fault, 4.86 Ma). The SINE-delineated break (between Clades 1 + 2 and Clades 3, 4 + 5) largely coincided with individuals distributed on either side of the Alpine Fault and explained 44.8% ($P < 0.01$, Table 3) of the variation found across the range of *O. n. polychroma*. Within Clade 1, 39.3% ($P < 0.01$) of the variation was accounted for by Cook Strait (mean 2.658% uncorrected ND2 *d* between Clade 1 individuals on either side of the strait, 1.90 Ma). In contrast, only 7.3% ($P = 0.237$) of the variation found within Clade 5 was accounted for by Foveaux Strait.

Individuals within Clade 5 have the strongest relationship between genetic distance and geographical distance ($r^2 = 0.342$, $r = 0.585$, one-sided $P = 0.0001$), but there was also a relationship found within the individuals of Clade 1 ($r^2 = 0.176$, $r = 0.420$, one-sided $P = 0.0001$) and the individuals of Clades 2, 3 + 4 (pooled in order to represent a region; $r^2 = 0.202$, $r = 0.450$, one-sided $P = 0.0001$).

There was significant support for the model of spatial expansion in Clade 1 (sum of squared deviation, SSD = 0.008, $P = 0.260$; raggedness index = 0.006, $P = 0.905$). Analysis of mismatch distributions for Clade 5 and the combined individuals of Clades 2, 3 + 4 (pooled to represent a region) yielded no significant support for the model

of spatial expansion (SSD = 0.010, $P = 0.040$; and SSD = 0.010, $P = 0.045$, respectively). However, the raggedness index indicated the distribution for both was not significantly different to what would be expected from an expanding population (Clades 2, 3 + 4: $r = 0.013$, $P = 0.715$; Clade 5: $r = 0.008$, $P = 0.530$). None of the mismatch distributions were strictly unimodal although Clades 1 and 5 had a relatively smooth frequency histogram indicative of spatial population expansion. In contrast, the mismatch distribution of Clades 2, 3 + 4 was very ragged, suggesting a stationary population of constant population size.

Discussion

In Daugherty *et al*.'s (1990) original designation of *Oligosoma nigriplantare polychroma* based on an analysis of allozyme variation, two subclades were defined across the range of the subspecies. The authors identified that despite some regional differentiation, the relationships between the populations generally followed a north-to-south transect, consistent with that of a geographically widespread polytypic species. However, they did not exclude the possibility of further differentiation in populations not yet studied and suggested that a taxonomically significant boundary may exist between the two subclades they identified (one restricted to the north of St Arnaud, one to the south). Although we also see a largely north-to-south arrangement of populations within our mtDNA data, we can now identify several distinct regions of genetic disjunction. Five clades (Clades 1–5) were identified to be geographically distinct (Figs 2 and 3). Our MDS plot indicated the isolation by distance trend was likely an artefact of more abrupt genetic breaks being incorporated into the overall signal (Fig. 4). Accordingly, AMOVA suggested much of the genetic differentiation within *O. n. polychroma* was explained by among clade differences rather than within clade differences (Table 3). The phylogeographical pattern found across the range of *O. n. polychroma* enables us to build on previous phylogeographical studies on New Zealand taxa, and discuss many aspects of New Zealand biogeography.

Pliocene mountain building

All the major phylogeographical breaks across the range of *O. n. polychroma* were in the South Island. The basal relationships among the five major clades in *O. n. polychroma* were not well resolved (low Bayesian posterior probabilities and ML bootstrap values, Fig. 2), indicating either an absence of genetic signal at this level, or rapid and simultaneous divergence. Divergence of the clades was estimated to be 3.98–5.45 Ma, suggesting the principal stimulus for the pattern of genetic structuring was mountain emergence in the South Island. This date coincides with prior knowledge that many other South Island taxa diverged in response to the mountain building of the Pliocene (alpine buttercups, *Ranunculus* spp., Lockhart *et al*. 2001; peripatus, Peripatopsidae; weevils, *Lyperobius* spp.; carabid beetles, *Mecodema* spp.; grasshoppers, Acrididae; Trewick & Wallis 2001; cockroaches, *Celattoblata* spp., Trewick & Wallis 2001; Chinn & Gemmell 2004; *Maoricicada campbelli*, Buckley *et al*. 2001; freshwater crayfish, *Paranephrops* spp., Apte *et al*. 2007; moa, *Megalapteryx* spp., *Dinornis* spp., *Pachyornis* spp., Baker *et al*. 2005; skinks, *O. lineoocellatum* and *O. chloronoton* spp. complex, Greaves *et al*. 2007).

Both the mitochondrial markers and SINE in the nuclear gene intron *MYH-2* indicated a significant genetic disjunction south of St Arnaud as suggested by Daugherty *et al*. (1990; based on allozyme variation) across the Alpine Fault (Fig. 3). AMOVA indicated 40.3% of the differentiation found across the South Island range of *O. n. polychroma* could be attributed to this break (Table 3). The Alpine Fault has previously been implicated in numerous biogeographical patterns in New Zealand. The hypothesized processes responsible for biological disjunction across the fault line include lateral displacement (e.g. Heads 1989), Pliocene mountain building (e.g. McGlone 1985) and Pleistocene glaciation (e.g. Wardle 1963). Recent evidence connects lateral movement of the fault with the distribution of congeneric lineages (hydrobiid gastropods, Haase *et al*. 2007), but for intraspecific and species level patterns Pleistocene glaciation or Pliocene mountain building is generally implicated (Waters & Wallis 2000; Trewick & Wallis 2001). Our findings support the latter hypothesis, dating the divergence of lineages over the Alpine Fault 4.07–4.61 Ma.

A hypothesis consistent with our observations that most genetic variation is partitioned across the Alpine Fault, and that estimates of genetic divergence predate Pleistocene glaciations, is that the Alpine Fault initiated allopatric divergence in *O. n. polychroma* by providing a sustained barrier to gene flow. We found the transposable element in only Clades 1 and 2 (see Table S1), providing evidence for restricted gene flow across the Alpine Fault where it intersects the distributions of Clades 1b and 3, and Clades 1b and 4 (Fig. 3). Being biparentally inherited, SINEs provide a more sensitive representation of gene flow than mtDNA markers (Shedlock & Okada 2000; Ray 2006). Despite this, SINEs are rarely used at the population level or for phylogeographical study, because they are most often fixed within a species, thus offering no meaningful indication of gene flow. The SINE polymorphism within *O. n. polychroma* provides a valuable indication of restricted gene flow over the mountain ranges associated with the Alpine Fault. The consequence of this biogeographical barrier in the evolution of New Zealand taxa has been controversial. Our study provides strong evidence indicating its importance for understanding some New Zealand biotic distributions.

Despite sharing the SINE, the genetic divergence found between Clades 1 and 2 (6.222% TrN *d*) is sufficient to

suggest that the transposon insertion predates the vicariance of these clades via mountain building (4.61 Ma). In contrast, the occurrence of Clade 4 on either side of the fault (Fig. 3) is likely due to an anomalous dispersal event through a lowland mountain pass of the Southern Alps, rather than vicariance. Likewise, recent movement following glacial retreat (< 14 000 years ago; McGlone 1995) has been inferred for closely related lineages distributed on either side of the Southern Alps in *Celatoblatta* cockroaches (Chinn & Gemmell 2004) and a freshwater fish (*Galaxias vulgaris* complex) despite being limited to the east of the fault throughout the rest of its range (Wallis *et al*. 2001).

Further genetic breaks within *O. n. polychroma* were revealed by mtDNA in the vicinity of the Marlborough Fault System (Fig. 1). The Marlborough/north Canterbury area is extremely mountainous due to secondary faulting associated with the Alpine Fault. Just like the Alpine Fault, the Marlborough fault system formed during the Miocene and was most active in the Pliocene (Suggate *et al*. 1978). The resultant topography is inferred to have driven diversification in several plant taxa and formed two divergent lineages of *G. vulgaris* on either side of the Kaikoura Ranges (Waters & Wallis 2000). Although the breaks we identified among clades are not consistent with the Kaikoura ranges (Figs 1 and 3), their uplift may have instigated the diversification of lineages (3.98–4.63 Ma) that have now shifted in range.

Pleistocene glacial processes

All of the *O. n. polychroma* clades converge in the Canterbury and Marlborough regions of the South Island (Figs 1 and 3). Clades 2, 3, and 4 are only found in this region and are range-restricted in comparison to Clades 1 and 5. Despite this finding, these clades each harbour disproportionately high levels of genetic diversity (π of each ranges from 0.022 to 0.031, Table 2). There are two possible explanations for this pattern that are not mutually exclusive: genetic divergence has been promoted by Pliocene mountain building, and/or it represents a refugial region during the Pleistocene.

High genetic diversity in the Canterbury and Marlborough regions could be argued as evidence for a glacial refugium. The north of the South Island has been suggested as an important centre of Pleistocene survival (Cockayne 1926; Willet 1950; Wardle 1963; McGlone *et al*. 2001). Pronounced endemism and the discontinuous distribution of a number of taxa (grasses, Connor 2002; *Nothofagus* & Wardle 1963) suggest that the climate was more favourable in both Marlborough and Nelson than in adjacent regions to the north and south (Willet 1950; Wardle 1963). While the Nelson region probably supported plants adapted to moist, mild conditions, the importance of Marlborough/north Canterbury as a refugium is indicated by endemic lowland and montane plants (Wardle 1963; Wagstaff *et al*. 1999). This climate may have provided suitable habitat for the persistence of a disproportionately high number of *O. n. polychroma* lineages.

During the last Pleistocene glaciation, suitable lizard habitat is thought to have been restricted to the north of the North Island and narrow coastal strips of the South Island (Bull & Whitaker 1975). Refugia have been suggested to exist in coastal Fiordland, Southland, Stewart Island, Otago, Banks Peninsula, and the Nelson/Marlborough Sounds region. Specifically for *Leiolopisma nigriplantare* (now *O. nigriplantare*, following taxonomic revision, which removed six species from within the species complex; Daugherty *et al*. 1990; Patterson & Daugherty 1990, 1994; Patterson 1997), refugia were also suggested to exist in Te Anau and in Stewart Island, from which the central South Island was recolonized (based on heam banding patterns of populations across the species distribution, Hardy 1977).

From our study, it appears *O. n. polychroma* survived the Pleistocene in a number of regions over its entire range, but it is possible there were originally more lineages that did not survive. Clade 5 of the southern South Island is the most divergent of the *O. n. polychroma* lineages, indicating that there may have been intermediary lineages (both genetically and geographically) that went extinct (Figs 2 and 4, Table 4). Studies of other taxa have likewise identified divergent lineages in this region (cicada, *M. campbelli*, Buckley *et al*. 2001; alpine scree weta, *D. connectens*, Trewick *et al*. 2000; alpine plant, *Pachycladon* spp., Heenan & Mitchell 2003; brown kiwi, *A. australis*, Baker *et al*. 1995), and it is renowned for its high proportion of endemic taxa (Climo 1975; Craw 1988; Connor 2002). Following the last glacial maximum, it is likely Clade 5 extended its range north to occupy Otago and Canterbury as suggested by Hardy (1977) and supported by our significant isolation-by-distance measures, mismatch distribution and raggedness index.

Many organisms were locally extirpated by the direct action of glaciers and/or harsher climates during the glacial periods (Cockayne 1926; Willet 1950; Wardle 1963). This activity was the greatest in the central South Island and is hypothesized to have formed the 'Beech gap' where beech and many other organisms are absent (Willet 1950; Wardle 1963, 1988; McGlone *et al*. 2001; Wallis & Trewick 2001). *O. n. polychroma* is present in the 'Beech gap' and even has a lineage endemic to this region (Clade 4, Canterbury), suggesting the skink was either not extirpated or that it was successful in recolonizing after the Pleistocene.

Past and present straits

Two distinct faunal breaks exist in the distributional patterns of New Zealand skinks: the Cook Strait limits the distribution of the *Cyclodina* genus to the north (Chapple *et al*. 2008a); and the historical northern boundary of the

Manawatu Strait affects *Oligosoma* skinks (Bull & Whitaker 1975). The northern limit of *O. n. polychroma* largely coincides with this Pliocene boundary (39° S latitude, Wardle 1963; Fig. 1). Our mismatch analyses and isolation-by-distance measures lend significant support for range expansion in Clade 1 (Fig. 4), suggesting *O. n. polychroma* recolonized much of the North Island from pockets of suitable habitat following remission of the Manawatu Strait during the Pleistocene (Lewis *et al*. 1994).

The spread of Clade 1 over the north and south of Cook Strait and its absence from the area east of the Alpine Fault (Fig. 3) is similar to patterns also seen in three galaxiid and gobiid freshwater fishes (McDowall 1996, 2005; Smith *et al*. 2005), extant and extinct species of the frog genus *Leiopelma* (Holyoake *et al*. 2001), large carnivorous landsnails of the subgenus *Powelliphanta* (Powell 1979), and brown kiwi (*A. australis*, Burbidge *et al*. 2003). Collectively, these organisms indicate that Cook Strait (i.e. Late Pleistocene) is a more recent barrier to dispersal than the Alpine Fault (i.e. Pliocene).

It is accepted that a landbridge existed between the North and South Island during glacial periods of the late Pleistocene (Fig. 1). However, the timing of the last land bridge over Cook Strait and whether terrestrial organisms used it, has been the subject of some debate (Worthy & Holdaway 2002). Conventional wisdom dictates that land extended between the islands during each glacial period of the late Pleistocene (e.g. Fleming 1979; McGlone 1988). Some have used geological inference to detail that this was the case, dating the most recent land connection across Cook Strait only 15–16 Ka (Lewis *et al*. 1994; Stevens *et al*. 1995). Others have observed terrestrial fossil faunas and propose there was no faunal exchange between the North and South Islands during the latest, and most severe, glacial period (Worthy & Holdaway 1994). Under the latter scenario, the differentiation of North Island and South Island taxa should date from the last interglacial highstand ~130 000 years ago (Worthy & Holdaway 1994).

Hardy (1977) suggested that *L. nigriplantare* moved freely across Cook Strait prior to its final formation. We found Cook Strait accounted for 39.3% of the variation found across the range of Clade 1 (Table 3). Within the phylogenetic tree, there was an evident break between samples of the North Island and South Island (bar ONP71) delineating Clade 1a and Clade 1b (Fig. 2). Divergence between samples of the North Island and South Island was dated at 1.90 Ma, suggesting gene flow across Cook Strait in this taxon predates any landbridge that may have been formed during the last glacial period of the Pleistocene. The existence of one sample from Clade 1b (ONP71, Castlepoint; Fig. 3) in the North Island is likely a result of incomplete lineage sorting or ancient dispersal, not recent dispersal. Although molecular evidence for gene flow across Cook Strait since formation has recently been found in *O. zelandicum* (O'Neill *et al*. 2008), other molecular studies in a variety of animal taxa provide no support for the use of a landbridge by terrestrial animals during the Late Pleistocene (brown kiwi, *A. australis*, Baker *et al*. 1995; bats, *M. tuberculata*, Lloyd 2003; skinks, *O. lineoocellatum* and *O. chloronoton* spp. complex, Greaves *et al*. 2007).

During the lowered sea levels of at least the last two glacial periods, a landbridge is also expected to have facilitated gene flow across Foveaux Strait (Suggate *et al*. 1978). Accordingly, we found little differentiation across Foveaux Strait (7.3% of the differentiation found across the range of Clade 5 could be attributed to the strait, Table 3), despite the strait being a known region of species disjunction in plants (Wardle 1963, 1988) and in the skinks *O. stenotis* and *O. notosaurus* (Gill & Whitaker 2001).

Taxonomic and conservation implications

Many new skink species have recently been described in New Zealand based on molecular studies (Chapple & Patterson 2007; Chapple *et al*. 2008b, c). We found no mtDNA support for the previously proposed distinct groupings within *O. n. polychroma*, suggesting they represent only distinct colour morphs of *O. n. polychroma* (as has been suggested by allozyme data for the Seaward Moss skink; C.H.D., unpublished data). The Seaward Moss skink was nested among *O. n. polychroma* samples in Clade 5 (Fig. 2). Samples of the Grey Valley skink fell within Clade 1b, and the presence of the SINE in these samples confirmed their affinity with both Clades 1 and 2 (Fig. 2, see Table S1). The presence of five divergent clades within *O. n. polychroma* (Fig. 2), highlights the need for future taxonomic and morphological work. The level of genetic differentiation among the clades is substantial (pairwise Φ_{ST} ranges from 0.540 to 0.795, Table 4) and worth consideration both for the taxonomy and conservation of this species. This need is reinforced by the presence of the SINE in only Clades 1 and 2 (Fig. 3, see Table S1). The marker indicates there has been no gene flow between these clades (Clades 1 + 2) and Clades 3, 4 + 5 for 4–5 Myr. Given this evidence, it is also likely there is restricted gene flow among all of the clades.

Conclusions

Overall, our findings provide considerable support for some suggested biogeographical patterns in New Zealand. In line with previous molecular and fossil evidence, we find it unlikely that *Oligosoma nigriplantare polychroma* used any landbridge formed across Cook Strait during the last glacial period of the Pleistocene. Tectonism during the Pliocene and associated mountain building is implicated as more important than glacial cycles and periods of marine inundation in driving the phylogeographical pattern of *O. n. polychroma*. Our study provides novel evidence for restricted gene flow across the Alpine Fault, possibly

the most significant phylogeographical break in New Zealand.

Some relatively unexplored biogeographical hypotheses have also emerged from our study. We nominate two regions of glacial refugium for *O. n. polychroma*. The first in the southern South Island, concordant with previous findings, and the second in the northeast of the South Island previously only proposed for plant taxa. We suggest that the milder and drier climate in the northeast South Island may have acted as a faunal refugium, like that identified for forest-dwelling fauna in Nelson. Further studies on species of similar ecological characteristics will facilitate greater understanding of the role of these glacial refugia in New Zealand.

Acknowledgements

We wish to thank the National Museum Te Papa Tongarewa for making their collections available and T. Bell for field collections. This research was conducted with the approval of the Victoria University of Wellington (VUW) Animal Ethics Committee 2003R16: Conservation of New Zealand reptiles, and with Department of Conservation collection permits (National Permit Nos: WA-17797-FAU and TT-17773-FAU). We are grateful to S. Smith and S. Greaves for information on *MYH-2* primers and the transposable element in New Zealand skinks. Thanks to the VUW herpetology group for valuable comments on the manuscript. L. Liggins thanks Heritage Expeditions and the Enderby Trust for their generosity and the New Zealand Herpetological Society for their support. This research was supported by the Allan Wilson Centre for Molecular Ecology and Evolution and grants to L. Liggins from the Miss E. L. Hellaby Indigenous Grasslands Research Trust, the George Mason Charitable Trust, the Royal Forest & Bird Society of New Zealand, the Society for Research on Amphibians and Reptiles in New Zealand, the Helen Stewart Royle Charitable Trust, the Tongariro Natural History Society, the Scientific Research Society (Sigma Xi G-I-A-R), the Royal Zoological Society of New South Wales and a VUW Masters Scholarship.

References

Apte S, Smith PJ, Wallis GP (2007) Mitochondrial phylogeography of New Zealand freshwater crayfishes, *Paranephrops* spp. *Molecular Ecology*, **16**, 1897–1908.

Avise JC (2000) *Phylogeography: The History and Formation of Species*. Harvard University Press, Cambridge, Massachusetts.

Baker AJ, Daugherty CH, Colbourne R, McLennan JL (1995) Flightless brown kiwis of New Zealand possess extremely subdivided population structure and cryptic species like small mammals. *Proceedings of the National Academy of Sciences, USA*, **92**, 8254–8258.

Baker AJ, Huynen LJ, Haddrath O, Millar CD, Lambert DM (2005) Reconstructing the tempo and mode of evolution in an extinct clade of birds with ancient DNA: the giant moas of New Zealand. *Proceedings of the National Academy of Sciences, USA*, **102**, 8257–8262.

Batt GE, Braun J, Kohn BP, McDougall I (2000) Thermochronological analysis of the dynamics of the Southern Alps, New Zealand. *Geological Society of America Bulletin*, **112**, 250–266.

Berry O, Gleeson DM (2005) Distinguishing historical fragmentation from a recent population decline — shrinking or pre-shrunk skink from New Zealand? *Biological Conservation*, **123**, 197–210.

BioWeb Herpetofauna Database (2006) New Zealand Department of Conservation, Wellington, New Zealand.

Bohonak AJ, Jensen JL, Kelley ST, Ngan EC (2005) Isolation by distance, web service. *BMC Genetics*, **6**, 13.

Buckley TR, Simon C (2007) Evolutionary radiation of the cicada genus *Maoricicada Dugdale* (Hemiptera: Cicadoidea) and the origins of the New Zealand alpine biota. *Biological Journal of the Linnean Society*, **91**, 419–435.

Buckley TR, Simon C, Chambers GK (2001) Phylogeography of the New Zealand cicada *Maoricicada campbelli* based on mitochondrial DNA sequences: ancient clades associated with Cenozoic environmental change. *Evolution*, **55**, 1395–1407.

Bull PC, Whitaker AH (1975) The amphibians, reptiles, birds and mammals. In: *Biogeography and Ecology in New Zealand* (ed. Kuschel G), pp. 231–276. Dr. Dr W. Junk, The Hague.

Burbidge ML, Colbourne RM, Robertson HA, Baker AJ (2003) Molecular and other biological evidence supports the recognition of at least three species of brown kiwi. *Conservation Genetics*, **4**, 167–177.

Chapple DG, Daugherty CH, Ritchie PA (2008a) Comparative phylogeography reveals pre-decline population structure of New Zealand *Cyclodina* (Reptilia: Scincidae) species. *Biological Journal of the Linnean Society*, in press.

Chapple DG, Keogh JS (2004) Parallel adaptive radiations in arid and temperate Australia: molecular phylogeography and systematics of the *Egernia whitii* (Lacertilia: Scincidae) species group. *Biological Journal of the Linnean Society*, **83**, 157–173.

Chapple DG, Keogh JS, Hutchinson MN (2004) Molecular phylogeography and systematics of the arid-zone members of the *Egernia whitii* (Lacertilia: Scincidae) species group. *Molecular Phylogenetics and Evolution*, **33**, 549–561.

Chapple DG, Patterson GB (2007) A new skink species (*Oligosoma taumakae* sp. nov., Reptilia: Scincidae) from the Open Bay Islands, New Zealand. *New Zealand Journal of Zoology*, **34**, 347–357.

Chapple DG, Patterson GB, Bell T, Daugherty CH (2008b) Taxonomic revision of the New Zealand copper skink (*Cyclodina aenea*: Squamata: Scincidae) species complex, with description of two new species. *Journal of Herpetology*, **42**, 437–452.

Chapple DG, Patterson GB, Gleeson DM, Daugherty CH, Ritchie PA (2008c) Taxonomic revision of the marbled skink (*Cyclodina oliveri*, Reptilia: Scincidae) species complex, with a description of a new species. *New Zealand Journal of Zoology*, **35**, 129–146.

Chinn WG, Gemmell NJ (2004) Adaptive radiation within New Zealand endemic species of the cockroach genus *Celatoblatta* Johns (Blattidae): a response to Plio–Pleistocene mountain building and climate change. *Molecular Ecology*, **13**, 1507–1518.

Climo F (1975) The landsnail fauna. In: *Biogeography and Ecology in New Zealand* (ed. Kuschel G), pp. 459–489. Junk, W., The Hague, The Netherlands.

Cockayne L (1926) Monograph on the New Zealand beech forests. Part 1. The ecology of the forests and taxonomy of the beeches. *Bulletin of the New Zealand Forest Service*, **4**, 1–71.

Connor HE (2002) Regional endemism in New Zealand grasses. *New Zealand Journal of Botany*, **40**, 189–200.

Craw R (1988) Continuing the synthesis between panbiogeography, phylogenetic systematics and geology as illustrated by empirical studies on the biogeography of New Zealand and the Chatham Islands. *Systematic Zoology*, **37**, 291–310.

Daugherty CH, Patterson GB, Thorn CJ, French DC (1990) Differentiation of the members of the New Zealand *Leiolopisma nigriplantare* species complex. *Herpetological Monographs*, **4**, 61–76.

Excoffier LG (2004) Patterns of DNA sequence diversity and genetic structure after a range expansion: lessons from the infinite-island model. *Molecular Ecology*, **13**, 853–864.

Excoffier LG, Laval G, Schneider S (2005) ARLEQUIN, Version 3.0: an integrated software package for population genetics data analysis. *Evolutionary Bioinformatics Online*, **1**, 47–50.

Excoffier LG, Smouse PE, Quattro JM (1992) Analysis of molecular variance inferred from metric distances among DNA haplotypes: application to human mitochondrial DNA restriction data. *Genetics*, **131**, 479–491.

Fleming CA (1979) *The Geological History of New Zealand and its Life*. Auckland University Press, Auckland.

Forstner MRJ, Davis SK, Arevalo E (1995) Support for the hypothesis of Anguimorph ancestry for the suborder Serpentes from phylogenetic analysis of mitochondrial DNA sequences. *Molecular Phylogenetics and Evolution*, **4**, 93–102.

Fuerst GS, Austin CC (2004) Population genetic structure of the prairie skink (*Eumeces septentrionalis*): nested clade analysis of post Pleistocene populations. *Journal of Herpetology*, **38**, 257–268.

Gardner RC, De Lange PJ, Keeling DJ, Bowala T, Brown HA, Wright SD (2004) A late Quaternary phylogeography for *Metrosideros* (Myrtaceae) in New Zealand inferred from chloroplast DNA haplotypes. *Biological Journal of the Linnean Society*, **83**, 399–412.

Gibbs G (2006) *Ghosts of Gondwana: The History of Life in New Zealand*. Craig Potton Publishing, Nelson, New Zealand.

Gill BJ, Whitaker AH (2001) *New Zealand Frogs and Reptiles*. David Bateman Limited, Albany, Auckland, New Zealand.

Greaves SNJ, Chapple DG, Daugherty CH, Gleeson DM, Ritchie PA (2008) Genetic divergences pre-date Pleistocene glacial cycles in the New Zealand speckled skink, *Oligosoma infrapunctatum*. *Journal of Biogeography*, **35**, 853–864.

Greaves SNJ, Chapple DG, Gleeson DM, Daugherty CH, Ritchie PA (2007) Phylogeography of the spotted skink (*Oligosoma lineoocellatum*) and green skink (*O. chloronoton*) species complex (Lacertilia: Scincidae) in New Zealand reveals pre-Pleistocene divergence. *Molecular Phylogenetics and Evolution*, **45**, 729–739.

Haase P, Marshall JC, Hogg ID (2007) Disentangling causes of disjunction on the South Island of New Zealand: the Alpine fault hypothesis of vicariance revisited. *Biological Journal of the Linnean Society*, **91**, 361–374.

Hardy GS (1977) The New Zealand Scincidae (Reptilia: Lacertilia); a taxonomic and zoogeographic study. *New Zealand Journal of Zoology*, **4**, 221–325.

Hare KM, Daugherty CH, Chapple DG (2008) Comparative phylogeography of three skink species (*Oligosoma moco*, *O. smithi*, *O. suteri*; Reptilia: Scincidae) in northeastern New Zealand. *Molecular Phylogenetics and Evolution*, **46**, 303–315.

Harpending HC (1994) Signature of ancient population growth in a low-resolution mitochondrial DNA mismatch distribution. *Human Biology*, **66**, 591–600.

Heads M (1989) Integrating earth and life sciences in New Zealand natural history: the parallel arcs model. *New Zealand Journal of Zoology*, **16**, 549–585.

Heenan PB, Mitchell AD (2003) Phylogeny, biogeography and adaptive radiation of *Pachycladon* (Brassicaceae) in the mountains of South Island, New Zealand. *Journal of Biogeography*, **30**, 1737–1749.

Hewitt GM (1996) Some genetic consequences of ice ages, and their role in divergence and speciation. *Biological Journal of the Linnean Society*, **58**, 247–276.

Hewitt G (2000) The genetic legacy of the Quarternary ice ages. *Nature*, **405**, 907–913.

Holm S (1979) A simple sequentially rejective multiple test procedure. *Scandinavian Journal of Statistics*, **6**, 65–70.

Holyoake A, Waldman B, Gemmell NJ (2001) Determining the species status of one of the world's rarest frogs: a conservation dilemma. *Animal Conservation*, **4**, 29–35.

Kumar S, Tamura K, Nei M (2004) MEGA 3.1: integrated software for molecular evolutionary genetics analysis and sequence alignment. *Briefings in Bioinformatics*, **5**, 150–163.

Kumaza Y, Nishida M (1999) Complete mitochondrial DNA sequences of the green turtle and blue-tailed mole skink: statistical evidence for archosaurian affinity of turtles. *Molecular Biology and Evolution*, **16**, 784–792.

Lewis KB, Carter L, Davey FJ (1994) The opening of Cook Strait: interglacial tidal scour and aligning basins at a subduction to transform plate edge. *Marine Geology*, **116**, 293–312.

Liggins L, Chapple DG, Daugherty CH, Ritchie PA (2008) Origin and post-colonization evolution of the Chatham Islands skink (*Oligosoma nigriplantare nigriplantare*). *Molecular Ecology*, **17**, 3290–3305.

Lloyd BD (2003) Intraspecific phylogeny of the New Zealand short-tailed bat *Mystacina tuberculata* inferred from multiple mitochondrial gene sequences. *Systematic Biology*, **52**, 460–476.

Lockhart PJ, McLenachan PA, Havell D, Glenny D, Huson D, Jensen U (2001) Phylogeny, radiation, and transoceanic dispersal of New Zealand alpine buttercups: molecular evidence under split decomposition. *Annals of the Missouri Botanical Garden*, **88**, 458–477.

Lyons LA, Laughlin TF, Copeland NG, Jenkins NA, Womack JE, O'Brien SJ (1997) Comparative anchor tagged sequences (CATS) for integrative mapping of mammalian genomes. *Nature*, **15**, 47–56.

Macey JR, Larson A, Ananjeva NB, Fang Z, Papenfuss TJ (1997) Two novel gene orders and the role of light-strand replication in rearrangement of the vertebrate mitochondrial genome. *Molecular Biology and Evolution*, **14**, 91–104.

Macey JR, Shulte JA, Ananjeva NB, Larson A, Rastegar-Pouyani N, Shammakov SM, Papenfuss TJ (1998) Phylogenetic relationships among agamid lizards of the *Laudakia caucasia* species group: testing hypotheses of biogeographic fragmentation and an area cladogram for the Iranian Plateau. *Molecular Phylogenetics and Evolution*, **10**, 118–131.

Mantel N (1967) The detection of disease clustering and a generalized regression approach. *Cancer Research*, **27**, 209–220.

McDowall RM (1996) Volcanism and freshwater fish biogeography in the northeastern North Island of New Zealand. *Journal of Biogeography*, **23**, 139–148.

McDowall RM (2005) Seasonal pulses in migrations of New Zealand diadromous fish and the potential impacts of river mouth closure. *New Zealand Journal of Marine and Freshwater Research*, **29**, 517–526.

McGlone M (1985) Plant biogeography and the late Cenozoic history of New Zealand. *New Zealand Journal of Botany*, **23**, 723–749.

McGlone MS (1988) New Zealand. In: *Vegetation History* (eds Huntley B, Webb T), pp. 557–599. Kluwer Academic Publishers, Dordrecht, The Netherlands.

McGlone M (1995) Lateglacial landscape and vegetation change and the Younger Dryas climatic oscillation in New Zealand. *Quarternary Science Reviews*, **14**, 867–881.

McGlone MS, Duncan RP, Heenan PB (2001) Endemism, species selection and the origin and distribution of the vascular plant flora of New Zealand. *Journal of Biogeography*, **28**, 199–216.

Miller C (1999) *Genetic diversity and management units for conservation of West Coast Oligosoma (Reptilia: Lacertilia: Scincidae)*. MSc Thesis, Victoria University of Wellington, Wellington, New Zealand.

Neiman M, Lively CM (2004) Pleistocene glaciation is implicated in the phylogeographical structure of *Potamopyrgus antipodarum*, a New Zealand snail. *Molecular Ecology*, **13**, 3085–3098.

Newnham RM, Lowe DJ, Williams PW (1999) Quaternary environmental change in New Zealand: a review. *Progress in Physical Geographyraphy*, **23**, 567–610.

O'Neill SB, Chapple DG, Daugherty CH, Ritchie PA (2008) Phylogeography of two New Zealand lizards: McCann's skink (*Oligosoma maccanni*) and the brown skink (*O. zelandicum*). *Molecular Phylogenetics and Evolution*, in press, doi: 10.1016/j.ympev.2008.05.008.

Patterson GB (1992) The ecology of a New Zealand grassland lizard guild. *Journal of the Royal Society of New Zealand*, **22**, 91–106.

Patterson GB (1997) South Island skinks of the genus *Oligosoma*: description of *O. longipes* n. sp. with redescription of *O. otagense* (McCann) and *O. waimatense* (McCann). *Journal of the Royal Society of New Zealand*, **27**, 439–450.

Patterson GB, Daugherty CH (1990) Four new species and one new subspecies of skinks, genus *Leiolopisma* (Reptilia: Lacertilia: Scincidae) from New Zealand. *Journal of the Royal Society of New Zealand*, **20**, 65–84.

Patterson GB, Daugherty CH (1994) *Leiolopisma stenotis*, n. sp., (Reptilia: Lacertilia: Scincidae) from Stewart Island, New Zealand. *Journal of the Royal Society of New Zealand*, **24**, 125–132.

Posada D, Crandall KA (1998) MODELTEST: testing the model of DNA substitution. *Bioinformatics*, **14**, 817–818.

Powell AWB (1979) *New Zealand Mollusca Marine Land and Freshwater Shells*. Collins, Auckland.

Ray DA (2006) SINEs of progress: mobile element applications to molecular ecology. *Molecular Ecology*, **16**, 19–33.

Ray N, Currat M, Excoffier LG (2003) Intra-deme molecular diversity in spatially expanding populations. *Molecular Biology and Evolution*, **20**, 76–86.

Rice WR (1989) Analyzing tables of statistical tests. *Evolution*, **43**, 223–225.

Rodriguez F, Oliver JF, Marín A, Medina JR (1990) The general stochastic model of nucleotide substitution. *Journal of Theoretical Biology*, **142**, 485–501.

Rogers GM (1989) The nature of the lower North Island floristic gap. *New Zealand Journal of Botany*, **27**, 221–241.

Rogers AR, Harpending HC (1992) Population growth makes waves in the distribution of pairwise genetic differences. *Molecular Biology and Evolution*, **9**, 552–569.

Ronquist F, Huelsenbeck JP (2003) MRBAYES 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics*, **19**, 1572–1574.

Rozas J, Rozas R (1998) DNASP, version 3: an integrated program for molecular population genetics and molecular evolution analysis. *Bioinformatics*, **15**, 174–175.

Sadlier RA, Smith SA, Bauer AM, Whitaker AH (2004) A new genus and species of live-bearing scincid lizard from New Caledonia. *Journal of Herpetology*, **38**, 320–330.

Sambrook J, Fritsch EF, Maniatus T (1989) *Molecular Cloning: A Laboratory Manual*. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York.

Schneider S, Excoffier LG (1999) Estimation of demographic parameters from the distribution of pairwise differences when the mutation rates vary among sites: application to human mitochondrial DNA. *Genetics*, **152**, 1079–1089.

Shedlock AM, Okada N (2000) SINE insertions: powerful tools for molecular systematics. *Bioessays*, **22**, 148–160.

Shimodaira H, Hasegawa M (1999) Multiple comparisons of log-likelihoods with applications to phylogenetic inference. *Molecular Biology and Evolution*, **16**, 1114–1116.

Smith SA (2001) *A molecular phylogenetic study of the Eugongylus group of skinks*. PhD Thesis, University of Adelaide, Adelaide, Australia.

Smith PJ, McVeagh SM, Allibone R (2005) Extensive genetic differentiation in *Gobiomorphus breviceps* from New Zealand. *Journal of Fish Biology*, **67**, 627–639.

Smith SA, Sadlier RA, Bauer AM, Austin CC, Jackman T (2007) Molecular phylogeny of the scincid lizards of New Caledonia and adjacent areas: evidence for a single origin of the endemic skinks of Tasmantis. *Molecular Phylogenetics and Evolution*, **43**, 1151–1166.

Sokal RR, Rohlf FJ (1981) *Biometry*, 2nd edn. W.H. Freeman, New York.

Stevens GR, McGlone M, McCulloch B (1995) *Prehistoric New Zealand*, 2nd edn. Reed Publishing (NZ) Ltd, Auckland, New Zealand.

Suggate RP (1985) The geological perspective. In: *Landforms of New Zealand* (eds Soons JM, Selby MJ), pp. 1–13. Longman Paul Ltd, Auckland, New Zealand.

Suggate R (1990) Late Pliocene and Quaternary glaciations of New Zealand. *Quaternary Science Reviews*, **9**, 175–197.

Suggate RP, Stevens GR, Te Punga MT (1978) *The Geology of New Zealand*. Government Printer, Wellington, New Zealand.

Tajima F (1989) Statistical method for testing the neutral mutation hypothesis. *Genetics*, **123**, 585–595.

Tajima F (1996) The amount of DNA polymorphism maintained in a finite population when the neutral mutation rate varies among sites. *Genetics*, **143**, 1457–1465.

Thompson JD, Higgins DG, Gibson TJ (1994) CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Research*, **22**, 4673–4680.

Towns DR, Elliot GP (1996) Effects of habitat structure on distribution and abundance of lizards at Pukerua Bay, Wellington. *New Zealand Journal of Ecology*, **20**, 191–206.

Towns DR, Ferriera R (2001) Conservation of New Zealand lizards (Lacertilia: Scincidae) by translocation of small populations. *Biological Conservation*, **98**, 211–222.

Towns DR, Neilson KA, Whitaker AH (2002) *North Island Oligosoma Spp. Skink Recovery Plan 2002–12*. Threatened Species Recovery Plan no. 48. Department of Conservation, Wellington, New Zealand.

Trewick SA (2001) Scree weta phylogeography: surviving glaciation and implications for Pleistocene biogeography in New Zealand. *New Zealand Journal of Zoology*, **28**, 291–298.

Trewick SA, Wallis GP (2001) Bridging the 'beech-gap': New Zealand invertebrate phylogeography implicates Pleistocene glaciation and Pliocene isolation. *Evolution*, **55**, 2170–2180.

Trewick SA, Wallis GP, Morgan-Richards M (2000) Phylogeographical pattern correlates with Pliocene mountain building in the alpine scree weta (Orthoptera, Anostostomatidae). *Molecular Ecology*, **9**, 657–666.

Wagstaff SJ, Heenan PB, Sanderson MJ (1999) Classification, origins, and patterns of diversification in New Zealand Carmichaelinae (Fabaceae). *American Journal of Botany*, **86**, 1346–1356.

Wallis GP, Judge KF, Bland J, Waters JM, Berra TM (2001) Genetic

diversity in New Zealand *Galaxias vulgaris sensu lato* (Teleostei: Osmeriformes: Galaxiidae): a test of a biogeographic hypothesis. *Journal of Biogeography*, **28**, 59–67.
Wallis GP, Trewick SA (2001) Finding fault with vicariance: a critique of Heads (1998). *Systematic Biology*, **50**, 602–609.
Wardle P (1963) Evolution and distribution of the New Zealand flora as affected by Quarternary climates. *New Zealand Journal of Botany*, **1**, 3–17.
Wardle P (1988) Effects of glacial climates on floristic distribution in New Zealand 1. A review of the evidence. *New Zealand Journal of Botany*, **26**, 541–555.
Waters JM, Wallis GP (2000) Across the Southern Alps by river capture? Freshwater fish phylogeography in South Island, New Zealand. *Molecular Ecology*, **9**, 1577–1582.
Whitehouse IE, Pearce AJ (1992) Shaping the mountains of New Zealand. In: *Landforms of New Zealand* (eds Soons LM, Selby MJ). Longman Paul, Hong Kong.
Whitely AR, Spruell P, Allendorf FW (2006) Can common species provide valuable information for conservation? *Molecular Ecology*, **15**, 2767–2786.
Willet RW (1950) The New Zealand Pleistocene snow line, climatic conditions, and suggested biological effects. *New Zealand Journal of Science and Technology*, **32**, 18–48.
Worthy TH, Holdaway RN (1994) Quaternary fossil faunas from caves in Takaka Valley and on Takaka Hill, northwest Nelson, South Island, New Zealand. *Journal of the Royal Society of New Zealand*, **24**, 297–391.
Worthy TH, Holdaway RN (2002) *The Lost World of the Moa: Prehistoric Life of New Zealand*. Indiana University Press, Bloomington, Indiana.
Wright S (1943) Isolation by distance. *Genetics*, **16**, 114–138.

This study formed part of the MSc research of Libby Liggins. Her research interest is in biogeography and the recent evolutionary history of New Zealand's terrestrial and marine life. David Chapple's research is focused on the phylogenetics, phylogeography and evolutionary ecology of squamate reptiles. Charles Daugherty has research interests on the evolutionary and population biology of vertebrates, conservation genetics and ecological restoration. Peter Ritchie's research focuses on the evolutionary genetics of species in New Zealand, the Southern Ocean and Antarctica.

Supplementary material

The following supplementary material is available for this article:

Table S1 GenBank Accession numbers, tissue codes, sources, location and phylogenetic affinity for *Oligosoma* and *Cyclodina* taxa used in this study

This material is available as part of the online article from:
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