Population genetics and invasion history of the European Starling across Aotearoa New Zealand

Bryan Thompson[1], Kamolphat Atsawawaranunt[1], Melissa C. Nehmens[1], William S. Pearman[1], E Owen Perkins[1], Pavel Pipek[2,3], Lee A. Rollins[4], Hui Zhen Tan[1], Annabel Whibley[1,5], Anna W. Santure[1], Katarina C. Stuart[1,4 *]

[1] School of Biological Sciences, University of Auckland, Auckland, Aotearoa/New Zealand
[2] Department of Invasion Ecology, Institute of Botany, Czech Academy of Sciences, Průhonice, Czech Republic.
[3] Department of Ecology, Faculty of Science, Charles University, Prague, Czech Republic.
[4] Evolution & Ecology Research Centre, University of New South Wales, Sydney, Australia
[5] Grapevine Improvement, Bragato Research Institute, Lincoln, New Zealand / Aotearoa

* Corresponding author email: katarina.stuart@auckland.ac.nz

Abstract

The expansion of human settlements over the past few centuries is responsible for an unprecedented number of invasive species introductions globally. An important component of biological invasion management is understanding how introduction history and post-introduction processes have jointly shaped present-day distributions and patterns of population structure, diversity, and adaptation. One example of a successful invader is the European starling (*Sturnus vulgaris*), which was intentionally introduced to numerous countries in the 19th century, including Aotearoa New Zealand, where it has become firmly established. We used reduced-representation sequencing to characterise the genetic population structure of the European starling in New Zealand, and compare the population structure to that present in sampling locations in the native range and invasive Australian range. We found a relatively high level of genetic differentiation for samples taken from across the north of New Zealand, compared to other invasive and native populations, congruent with documented introductions from multiple localities while also implying restricted gene flow. Other New Zealand locations presented more homogenous genetic population structure, suggesting potential connectivity between southern regions. We also profiled genetic bottlenecks and shared outlier genomic regions as a means of corroborating translocation records between invasive populations. Using these results as well as historic demographic patterns, we demonstrate how genomic analysis

complements even well-documented invasion histories to better understand invasion processes, with direct implication for understanding contemporary gene flow and informing invasion management.

Keywords: *Sturnus vulgaris*, invasion history, population structure, reduced representation sequencing, invasive species, historical records

1 | INTRODUCTION

Invasive alien species are a threat to biodiversity, primary industries, and human health, costing a global estimate in excess of US$400 billion annually (Roy et al., 2024). With globalisation facilitating an increasing rate of accidental and deliberate introductions (Hulme, 2009; Seebens et al., 2017, 2021), and human-mediated environmental disturbance providing ideal environments for invasive species to thrive (Essl et al., 2020), the impact of invasive species will undoubtedly continue to grow. Ongoing efforts to mitigate the costs of invasive species often require single-species-targeted approaches (Roy et al., 2024). To this effect, new genomic tools have played a vital role in describing properties of invasive species during their transportation, introduction, establishment, and spread (McGaughran et al., 2024). Population genomics is one such tool that allows for the profiling of genetic patterns across an invasive species' range, giving insight into likely source populations, species movement, and patterns of adaptive change (McGaughran et al., 2024). In particular, comparing a species' documented introduction history to present-day population genetics may reveal the underlying mechanisms that drive population expansion and contribute to invasion success (Colautti & Lau, 2015). However, despite risks associated with invasive species continuing to escalate at an alarming rate, population genomic data is notably lacking for many of the worst invasive species globally (Matheson & McGaughran, 2022).

The Common or European Starling, *Sturnus vulgaris*, is considered one of the most successful invasive avian species worldwide (Lowe et al., 2000), with their presence negatively impacting agricultural, conservation, and societal interests (Campbell et al., 2016; Evans et al., 2020). Worldwide, the starlings' native Eurasian range extends across North Africa, the Middle East, and Central Asia (Cabe, 2020). The starling has now established invasive populations on all other human-populated continents (Fig. 1, Stuart, Hofmeister, et al., 2023). Most of the starling introductions were deliberate attempts by colonial acclimatisation societies to introduce the species to newly colonised countries (Feare, 1984; McDowall, 1994, pp. 1861–1990; Stuart, Hofmeister, et al., 2023). In addition to human-mediated introductions, the starling has expanded its native range, most likely aided by both direct and indirect human-facilitated factors such as climate change and ecosystem disturbance (Ferrer et al., 1991; G. Harris, 1964; Webster, 1975). Paradoxically, although the starling's native range has expanded, population numbers have declined in more recent times across multiple regions within the native range, likely as a result of ongoing changes in land use practices (Heldbjerg et al., 2016; Rintala et al., 2003; Wretenberg et al., 2006). There is a pressing need to understand patterns of invasion

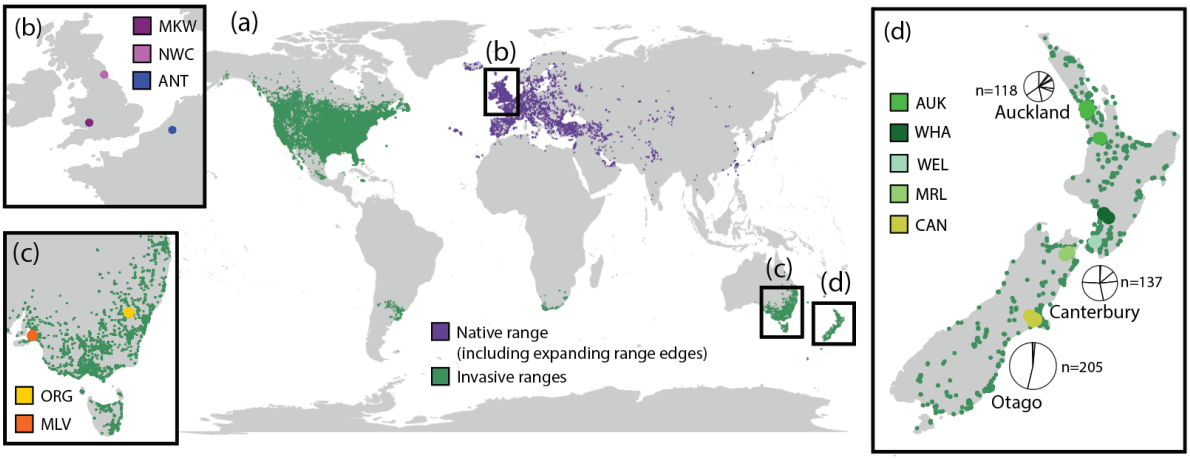
success and adaptation within this species, to inform both invasive species management as well as conservation efforts within native ranges of concern.

Several invasive starling ranges have been characterised by high-resolution genomic datasets revealing underlying genetic population structure associated with demography, spatial and temporal patterns of dispersal, and adaptive potential in this highly invasive species (Bodt et al., 2020; Fiorini et al., 2022; Hofmeister et al., 2021; Stuart et al., 2021). Amongst these invasive populations there exists a diversity of demographic and genetic patterns, with some populations displaying migration and panmixia (North America; Cabe, 1999), while others show signals of population substructure and spatial sorting (South Africa; Phair et al., 2018). In particular, the Australian starling range contains two major genetic subpopulations, that are thought to be a result of genetic differences at introduction sites, range edge effects, and landscape barriers (Rollins et al., 2011). However, until now, only low-resolution allozyme markers have been used to characterise the genetic structure of starlings in the New Zealand range, providing a limited view of population structure which reported fairly high levels of genetic differentiation and some loss of genetic diversity within New Zealand (Ross, 1983).

In Aotearoa New Zealand, repeated introductions to multiple locations - totalling more than 600 individuals (2,200 when including intra-country translocations) over a twenty-year period in the mid-1800s - have contributed to the present-day population (Pipek et al., 2019). Detailed historical accounts track the complicated introduction history of the New Zealand starlings, which included the importation of starlings from their native range, reciprocal translocations to and from Australia, and many within-New Zealand translocation events (Jenkins, 1977; Pipek et al., 2019; Stuart, Hofmeister, et al., 2023; Thomson, 1922). Starlings are now spread over the a vast majority of mainland New Zealand (Fig. 1). Within New Zealand, understanding how the introduction history has interacted with invasion processes to lead to the current population structure can form the basis for proactive management strategies.

This study aims to examine genetic patterns within the invasive starlings across New Zealand on a backdrop of historical information about introductions and translocations. To do this we used reduced-representation sequencing data to compare multiple populations to an existing and characterised dataset (Stuart, Sherwin, et al., 2022) of both the invasive Australian and native Eurasian ranges. We examine the population structure and genetic bottlenecks within New Zealand and contrast these patterns to the other invasive and native sampling locations. In particular we aim to understand if the standing genetic diversity is more reflective of documented introduction histories identified within this study and previous studies (Pipek et

al., 2019) or post-introduction events such as geographical isolation. We then examine the independent invasive lineages for signals of shared outlier regions, noting that these shared regions across independent introductions may indicate parallel (ongoing) selection, but could also be indicative of post-introduction translocations between geographically separate locations. Finally, we examine the ancient and more recent demographic history of the species, to contextualize recent patterns of genetic diversity loss and bottlenecks across the native and invasive ranges.



113

114 **Figure 1 | Global distribution of the starling (*Sturnus vulgaris*) and study sampling**
115 **locations.** Global distribution **(a)** is plotted based on bird sighting data (Sullivan et al., 2009),
116 with native range (including native range expansions) plotted in purple and invasive ranges in
117 green. Invasive bird sighting data is re-plotted within subpanels. Subpanels include sampling
118 locations from **(b)** the native range, **(c)** Australia (Stuart, Sherwin, et al., 2022), and **(d)** New
119 Zealand (samples from this study). Within New Zealand, pie charts indicate sites of major
120 introduction (>100 individuals) from overseas sources, with each sector representing an
121 independent introduction event as based on Table 1, Pipek et al. (2019). Otago is presented as
122 an empty circle as this was a major introduction site that was not sampled as part of this study.
123 Native range samples: Monks Wood, United Kingdom (MKW), Newcastle, United Kingdom
124 (NWC), Antwerp, Belgium (ANT); invasive range samples: Orange, Australia (ORG), McLaren
125 Vale, Australia (MLV), and in New Zealand: Auckland (AUK), Manawatū-Whanganui (WHA),
126 Wellington (WEL), Marlborough (MRL) and Canterbury (CAN).

127

2 | METHODS

2.1 | Collection of historical translocation records

For this study, we sought to identify evidence of translocations of starlings between Australia and New Zealand in the Papers Past (https://paperspast.natlib.govt.nz) and Trove (https://trove.nla.gov.au) newspaper archives and historical documents of the acclimatisation societies of New Zealand. In case of newspapers, we looked for articles published between years 1875 to 1900 (i.e., a period in which starlings were already acclimatised and redistributed in New Zealand) that contained not only "starling" or "starlings", but also words like "shipped", "liberated", "received", "acclimatisation", "distributed", "few", "several", "pairs" or "dozen" in reasonable proximity (both Papers Past and Trove allow to search for co-occurring strings separated by set maximum number of words). We also explored the annual reports of the largest acclimatisation societies of Australia in 1880s, when available on Trove. The list might still not be exhaustive. Furthermore, some birds might have escaped from bird fanciers. Part of the search was done using Trove API, accessed through R.

2.2 | Sample Collection

A total of 106 starling specimen samples were obtained from various contributors within New Zealand from five geographically distinct locations between May 2022 and October 2023 (Table S1). Sampling covered three locations in the North Island, specifically in the Auckland region (AUK: n=18), the Manawatū-Whanganui region (WHA: n=12), the Wellington region (WEL: n=40) and two in the South Island in the Marlborough region (MRL: n=15) and Canterbury region (CAN: n=21). Sampling locations were recorded, and individuals were stored on ice and transported to the University of Auckland. Tissue subsampling was performed using a biopsy punch of breast muscle tissue, which was then stored in 90% ethanol at -30°C until DNA extractions could be performed.

2.3 | DNA Extraction and Sequencing

Extracted DNA samples were sent to Diversity Arrays Technology Pty Ltd company (DArT P/L) for processing and sequencing (Kilian et al., 2012). Briefly, DArTseq is a reduced representation sequencing methodology, which uses double restriction enzyme digest (here *PstI-SphI*) to randomly subsample and then sequence a subset of the genome. DNA extraction for the MRL tissue samples was conducted using the New England Biolabs (NEB) Monarch Genomic DNA Purification Kit following standard manufacturer's protocols, and these were sequenced in January 2023. All other tissue samples from New Zealand were extracted using the DNeasy

Blood & Tissue Kit (Qiagen), also following the manufacturer's protocols, and were sequenced in November 2023. Sequencing was performed on an Illumina Hiseq2500/Novaseq6000, and the raw fastq data was obtained for all samples, including DArT-produced technical replicates.

In addition to reduced representation sequencing, whole genome resequencing (WGR) data of 12 individuals from four locations were used (Table S2), including three newly sequenced individuals from New Zealand, and three individuals each from the three native range sites and the two Australian genetic groups with data obtained from previous studies (Hofmeister et al., 2024; Stuart, Edwards, et al., 2023). For the three newly sequenced New Zealand individuals, we used the gDNA extracted for DArTseq sequencing, and individuals were resequenced using a short-read whole genome resequencing approach, with a coverage aim of approximately 20x. Sequencing was done on the Illumina NovaSeq platform (150 bp paired end reads) and was completed by Custom Science, Australasia. These individuals were taken from Marlborough (MRL), New Zealand.

2.4 | Raw Sequence Processing

In addition to the newly generated DArTseq sequence data, we also incorporated a previously published DArTseq dataset (Stuart, Sherwin, et al., 2022), which contains samples from the native European range (Antwerp, Belgium; ANT: n=15, Newcastle, United Kingdom; NWC: n=15, Monks Wood, United Kingdom; MKW: n=15), as well as two sampling locations from within the invasive Australian range (Orange; ORG: n=15, McLaren Vale; MLV: n=15). The two sampling locations in Australia were chosen to represent the two major genetic subpopulations (Rollins et al., 2011). These existing raw sequence data files, along with the MRL samples (January 2023 sequencing batch) were demultiplexed using STACKS v2.2 (Catchen et al., 2013) *process_radtags*, while also discarding low quality reads (-q), reads with uncalled bases (-c), and rescuing barcodes and RAD-Tag cut sites (-r). It was not necessary to perform this step on the remainder of the new sequence data because DArT performing in-house demultiplexing using a proprietary bioinformatic pipeline (Kilian et al., 2012).

For all the data, we used FASTP v0.23.2 (Chen et al., 2018) to remove adapter sequences and in the same step filtered reads for a minimum phred quality score of 22 (-q 22) and a minimum length of 40 (-l 40). Both batches of sequence data produced as part of this study were additionally length trimmed to reduce the read length of the newer sequence data to match the base length of the older sequence data (-b 69) from Stuart *et al.* (2022).

2.5 | Mapping, Variant Calling, and Filtering

We used the program BWA v0.7.17 (Li & Durbin, 2009) to index the reference genome *S. vulgaris* vAU1.0 (Stuart, Edwards, et al., 2022), and align the trimmed reads using the BWA *aln* function (-B 5 to trim the first 5 base pairs of each read), which is optimised for single-end short reads, followed by the BWA *samse* function for producing the SAM formatted output files containing the alignments and their respective base qualities. Alignments were then sorted and indexed using SAMTOOLS v1.16.1 (Li et al., 2009), and single nucleotide polymorphisms (SNPs) were subsequently called and annotated using BCFTOOLS v1.16 (Danecek et al., 2021) with the *mpileup* (-a "DP,AD,SP", --ignore-RG) and *call* (-mv, -f GQ) functions.

Next, we performed several filtering steps. We removed known technical replicates and identified relatives from the data (for full methods, see Appendix 1: Filtering replicates and relatives and Table S3 for original and final sample sizes), which resulted in a final individual count of 141. VCFTOOLS v0.1.15 (Danecek et al., 2011) was used to remove indels (--remove-indels), and quality filter for a minimum site quality score of 30 (--minQ30), minimum genotype quality score of 20 (--minGQ 20), and minimum and maximum depth of coverage of 5 (--minDP 5) and 100 (--maxDP 100). Then, to account for batch effects that may impact the sequenced loci, we kept only SNPs present in at least 50% of the individuals in each sampling location. We ran one final filtering step to ensure appropriate levels of missingness and rare alleles using the following parameters: maximum missingness per site of 30% (--max-missing 0.7), minor allele count of 5 (--mac 5), and a minimum and maximum allele per locus of 2 (--min-alleles 2 --max-alleles 2), resulting in a dataset containing 19,174 SNPs.

2.6 | Genetic diversity and bottlenecks across invasive lineages

First, we assessed genetic diversity metrics within sampling location using the DARTR v2.9.7 (Mijangos et al., 2022) package in R v4.2.1 (R Core Team, 2022) to run the *gl.report.heterozygosity* function, which calculated observed heterozygosity (Ho), sample size corrected unbiased expected heterozygosity (uHe), and inbreeding coefficient (F_{IS}). We identified SNPs that were private within sampling locations using the *populations* function in STACKS with the '--phylip' flag.

In addition to other genetic diversity metrics, the folded site frequency spectrum (SFS) of each sampling location was constructed to visualise the genetic bottlenecks experienced by the different populations. SNPs were filtered using DARTR to retain only SNPs that were genotyped in all individuals (1,451 SNPs), with this level of SNP filtering being used only for this analysis. This stringent filter was used as SNP missingness interacts with the binning of the SFS histogram and introduces irregular distributions into the SFS (Fig. S1). To produce comparable

SFS from populations with unequal numbers of samples, each population was subsampled to 10 individuals, and the *gl.percentage.freq* function in DARTR was used to calculate the minor allele frequency. This process was repeated 100 times and the distribution plotted.

2.7 | Genetic structure and differentiation

Next, we sought to profile the population structure within the newly sequenced invasive starling samples from New Zealand and compare this to previously sequenced locations from the native range and the invasive Australian range. For this, we used the DARTR package in R to run principal components analysis (PCA) using the *gl.pcoa* function on the full SNP data set.

The program ADMIXTURE v1.3.0 (Alexander et al., 2009) was used to infer ancestry proportions, using the default 200 bootstraps (-B), with cross-validation enabled (--cv). We tested a range of K values (1-10) and plotted the K value with the lowest cross-validation error above K=1 (Fig. S2). We assessed but did not detect contemporary gene flow among the New Zealand populations using the program BA3-SNPs version 3.0.4 (Mussmann et al., 2019; Wilson & Rannala, 2003), but we note that our dataset did not meet a number of assumptions for gene flow analyses (for full methods, see Appendix 2: Assessing gene flow).

We also assessed pairwise population genetic differentiation using two methods. First, we assessed pairwise F_{ST} values between sampling locations using the DARTR *gl.fst.pop* function in R. Secondly, we used Jaccard dissimilarity, a metric borrowed from numerical ecology, to quantify dissimilarity in minor alleles between locations (Legendre & Legendre, 2012). Here, Jaccard similarity was first calculated between a pair of individuals across all jointly genotyped loci as the number of loci where both individuals had at least one minor allele in their genotype, divided by the total number of loci where at least one individual has at least one minor allele (Prokopenko et al., 2016). Jaccard dissimilarity values were then calculated as 1 – Jaccard similarity, the number of loci where both individuals have a copy of the minor allele over the number of loci where either individual has a copy of the minor allele, for each pair of individuals; and pairwise Jaccard dissimilarity values between sampling locations were calculated by averaging over individuals. This approach ignores both the joint presence of major alleles and the joint absence of minor alleles and is interpretable as differentiation based upon the uniqueness of minor variants. This was executed using a custom Python script leveraging the PANDAS v2.2.2 (The pandas development team, 2020) and NUMPY v1.26.4 (Harris et al., 2020) libraries.

2.8 | Isolation by distance and isolation by environment within New Zealand

To understand the patterns of genetic structure observed across New Zealand in the context of landscape heterogeneity, we performed multiple matrix regression with randomisation (MMRR; (Wang, 2013b). MMRR takes a list of distance matrices and calculates the regression coefficients for each explanatory variable, with random permutations of the response variable then performed to estimate significance values. Specifically, we were interested in the effects of isolation by distance (IBD) and isolation by environment (IBE), which are major forces that shape genetic structure in natural populations (Nanninga et al., 2014). We first imported the file containing our SNPs into R and converted it into a genlight object using the function *genomic_converter* in RADIATOR (Gosselin et al., 2020). SNPs were filtered to a dataset of only the New Zealand samples (refiltered to --mac 5 --max-missing 0.7, leaving 14,890 markers). We then calculated pairwise Nei's genetic distance, our response variable, for all samples using the *stamppNeisD* function in STAMPP (Pembleton et al., 2013). Geodesic distances in metres were calculated from the sample coordinates using GEODIST (Padgham, 2021). To calculate environmental distances, we extracted climatic data from WorldClim data (30s resolution) (Fick & Hijmans, 2017) corresponding to the sampling coordinates for each sample. We performed MMRR using available R scripts (Wang, 2013a). We ran MMRR on two datasets, the first including all samples across New Zealand (n = 75), while the second excluded samples from AUK (n = 57) based on population structure analysis indicating that Auckland may be a separate introduction lineage and present-day population. We performed 9,999 permutations of MMRR, and visualised our data using GGPLOT2 (Wickham, 2016).

2.9 | Genetic outlier analysis within and across invasion lineages

We sought to identify outlier regions within each of the four distinct invasive lineages; two in New Zealand (identified here; see results) and the two major Australian genetic subpopulations. For this we used an analytical approach that identifies loci that have statistically diverged allele frequencies between population comparisons. We followed a similar approach used in Parvizi et al. (2024), which involved using genetic outlier analysis through the command line program BAYPASS v2.31 (Gautier, 2015). BAYPASS accounts for confounding effects of population structure when identifying outlier signals, and specifically we used the C2-contrast statistic which allows for the testing of binary covariates such as invasion status (native or invasive) and is more robust when computing statistic for even a small number of populations (Olazcuaga et al., 2020). We separately contrasted the three native samples sites (MKW, NWC, ANT) with the four invasive lineages of ORG, MLV, AUK, NZrest (comprising MRL, CAN, WHA and WEL). A top 1 percentile C2-contrast statistic threshold was chosen for each of the four separate invasion lineages based on a neutral simulated dataset, above which SNPs were considered an outlier.

This was done in R by using the BAYPASS function *simulate.baypass* to generate a neutral dataset of 5,000 SNPs based on each comparisons beta distribution of the ancestral reference allele frequency, which captures the neutral population structure present in the data. Outlier SNP IDs were retained and compared across invasion lineages and then visualized using GGVENNDIAGRAM (Gao et al., 2021) in order to examine unique and shared SNP outliers across lineages. Additionally, a combined C2-contrast statistic was run comparing all native with all invasive lineages together, and the overlap with the C2-contrast comparisons with each invasive lineage were considered. Genes overlapping outlier regions (+/- 10 kb) were identified using BEDTOOLS v2.30 (Quinlan & Hall, 2010), and over-represented genes examined with PANTHER (Mi et al., 2019) via the Gene Ontology enrichment webtool (release 2024-04-24).

2.10 | Historical demography of *Sturnus vulgaris*

For this, we used the programs PSMC (Pairwise Sequentially Markovian Coalescent) and STAIRWAY PLOT, at ancient- and recent- historical timescales, respectively, to examine fluctuations in effective population size (N_e). To examine the ancient timescale of fluctuations in N_e we used PSMC v0.6.5 (Li & Durbin, 2011). WGR sequence data files were prepared for PSMC using trimmed sequences mapped to the *S. vulgaris* vAU1.0 genome (Stuart, Edwards, et al., 2022). Variant calling was then performed with the BCFTOOLS *mpileup* function (-C 50 -q 20 -Q 25), *call*, *filter*, and *sort* functions. The dataset was filtered by removing indels and selecting SNPs for a minimum depth of 5 and a maximum depth of 50. Each individual's VCF file was then converted into fastq format using BCFTOOLS *vcfutils.pl,* which were then converted to PSMCFA files. We ran PSMC for 30 iterations (-N30), an upper limit of time to the most recent common ancestor set to 5 (-t5), an initial h:q value of 5 (-r5), and free atomic time intervals set to (4 + 30*2 + 4 + 6 + 10) based on recommendations from previous work where these intervals were successful for avian species (Nadachowska-Brzyska et al., 2015). We performed bootstrapping (100 iterations) for each individual to check for variation in N_e estimates using the same parameters used for the original PSMC analysis. The PSMC and bootstrap results were then scaled using an estimated generation time of 2 years, the approximate age of first breeding (Fear & Craig, 1999), and a yearly mutation rate of 2.3 × 10-9 based on related avian species estimates (Nadachowska-Brzyska et al., 2015; Smeds et al., 2016) and plotted using *psmc_plot.pl*.

For analysis of N_e demographic changes from the recent historical timescale, we used the tool STAIRWAY PLOT v2 which infers detailed population demographic history using the site frequency spectrum (SFS). We used native range individuals from the DArT-seq SNP dataset, choosing

MKW and NWC (n=22, 25,531 SNPs) based on their genetic similarity indicated by earlier analysis. We then ran stairway plot again for invasive New Zealand individuals, specifically choosing WHA and WEL (n=22, 25,283 SNPs), which were the most equivalent sample site pair for comparison due to equivalent sample sizes and genetic similarity to the native range sample site pair. Both SNP subsets were refiltered using VCFTOOLS to the same filtering criteria as used in initial SNP filtering (--max-missingness 0.7 --max-alleles 2 --min-alleles 2 --max-meanDP 100 --thin 1000), but with no minor allele count filtering because this would alter the site frequency spectrum. The tool VCF2SFS was used to generate SFS data (Liu et al., 2018), and we ran STAIRWAY PLOT with the same mutation rate and generation time from using in PSMC analysis.

3 | RESULTS

3.1 | Evidence of translocations between Australia and New Zealand

We found evidence from historical newspaper articles and the original records kept by acclimatisation societies of translocations between different regions of Australia and New Zealand (Table 1, Appendix 3: Historical records of starlings). We found that many of the recorded introductions occurred at or around 1880, and that all found translocation events between the two regions were from New Zealand to Australia, and on a larger scale than several shipments in the opposite direction in 1860s and 1870s (Pipek et al., 2019). Otago was the most common source population of birds, though two translocations were recorded as occurring from AUK to Australia and Canterbury Acclimatisation Society was contacted multiple times by Acclimatisation Society of South Australia with request of transporting starlings ("ACCLIMATISATION SOCIETY," 1880a; "ACCLIMATISATION SOCIETY," 1880b; "ACCLIMATISATION SOCIETY," 1882), but we have not found any direct evidence that the birds were shipped there in the end. Late shipment of 48 starlings in 1887 to Victoria is from unknown New Zealand source region ("CITY COUNCIL.," 1887), while a shipment to Tasmania a year earlier arrived from London ("SHIPPING," 1886).

3.2 | Genetic diversity and bottlenecks across invasive lineages

Genetic diversity metrics indicate all ten sampling locations have a minor deficit of heterozygosity compared to what would be expected under Hardy-Weinberg equilibrium, with all recorded values of observed heterozygosity (Ho) being less than unbiased expected heterozygosity (uHe) and further supported by marginally positive F_{IS} values (Table 2). However, it is worth noting that the three native range sample locations all have slightly higher heterozygosity deficits compared to the invasive sample sites. A notable exception to the otherwise similar genetic indices across the ten sampling locations is the relatively high number of private alleles found in Auckland (AUK; 20 private alleles), being four times greater than the next highest sampling locations (Table 2). AUK and the Australian McLaren Vale (MLV) sample display the lowest levels of unbiased expected heterozygosity.

The shape of the folded site frequency spectrum plots suggests that New Zealand populations from Canterbury (CAN), Marlborough (MRL), Wellington (WEL), and Manawatū-Whanganui (WHA) have comparable genetic diversity to native populations (Fig. 2), as indicated by a similar and higher median number of very rare SNPs. In comparison, sampled Australian and AUK populations display signals of genetic bottlenecks often observed in invasive populations, with

366 Orange (ORG; Australia) and Auckland populations displaying similar levels of genetic
367 bottlenecks and MLV showing the strongest genetic bottlenecks signature.

368 **3.3 | Genetic structure and differentiation of New Zealand starlings**

369 Analysis of the five New Zealand invasive sampling locations (AUK, WHA, WEL, MRL and CAN),
370 the two invasive Australian sampling locations (MLV, ORG), and the three native sampling
371 locations (ANT = Antwerp Belgium, NWC = Newcastle UK, and MKW = Monks Wood UK) reveals
372 strong population genetic structure within New Zealand (Fig. 3).

373 The two sampling locations of WHA and WEL form a single cluster on the two-dimensional PCA
374 (Fig. 3a), though F_{ST} analysis does still indicate some genetic differentiation between these two
375 sites (Fig. 3c). There is strong differentiation between AUK and the rest of the New Zealand
376 sampling locations, indicated by both PC distances and F_{ST} (Fig. 3a), with admixture analysis
377 also reporting different patterns of historical ancestry (Fig. 3b). This indicates that genetic
378 differentiation across New Zealand is more pronounced than the genetic differentiation seen in
379 the two Australian sample locations, which each are representative of the two main genetic
380 subpopulations within Australia (Stuart et al., 2021). AUK also has high F_{ST} values in comparison
381 to both invasive Australian sampling locations (Fig. 3c).

382 Our comparative analysis of the New Zealand sampling locations to both native and invasive
383 Australian ranges reveal CAN as the most similar New Zealand sampling location to the native
384 range, and MRL as the most similar New Zealand sampling location to Australia, with MRL
385 having higher genetic similarity to ORG compared to MLV (Fig. 3).

386 We explore Jaccard dissimilarity as a second diversity metric that emphasises counting minor
387 (rarer) allele dissimilarity alongside the traditional F_{ST} metric that incorporates information from
388 both alleles. We find that both these two approaches produce similar results (Fig. 3c), for
389 example using both metrics the Australian sample location of McLaren Vale (MLV) seems to be
390 quite differentiated from the native range, and Auckland (AUK) is quite distinct from the rest of
391 the New Zealand sampling sites. However, we also note some key differences that indicate
392 more nuanced patterns of genetic differentiation only visible when considering only minor
393 alleles, in particular higher levels of minor allele dissimilarity in the UK sample sites (NWC,
394 MKW), as well as lowest dissimilarity being seen in Marlborough (MRL) meaning a high level of
395 shared minor alleles with other locations.

396 **3.4 | Drivers of patterns of genetic differentiation within New Zealand**

Results of the isolation by distance (IBD) and isolation by environment (IBE) multiple matrix regression with randomization (MMRR) analyses revealed contrasting determinants of genetic variation across New Zealand. In the first dataset including all samples, geographical and environmental distances combined explained 27.9% of genetic variability (Table 3). The regression coefficient for geographical distance is almost five-times greater than that for environmental distance (β_{IBD} = 0.476, β_{IBE} = 0. 0878; Table 3), suggesting that IBD was more important in explaining the observed genetic distances, although contributions by IBE were also significant (Fig. S3). In the second dataset excluding Auckland samples, geographical and environmental distances combined explained 7.4% of genetic variability (Table 3). Unlike MMRR performed on the whole dataset, IBD was less important than IBE in explaining the observed genetic distances (β_{IBD} = 0.136, β_{IBE} = 0.180; Table 3, Fig. S3).

3.5 | Genetic outlier analysis within and across invasion lineages

The BAYPASS C2-contrast statistics identified the largest number of outlier SNPs (214) in comparisons between the native range and ORG (Fig. 4b), followed closely by AUK with 211 outliers (Fig. 4c). This was followed by 165 outliers when comparing the native range to MLV (Fig. 4a), while NZrest had the smallest number of outliers at 122 (Fig. 4d). Only two loci were identified across all comparisons (Fig. 4e), and these loci were also identified in the combined C2-comparison statistic analysis comparing all native with all invasive populations (Fig. S4). The largest number (24) of non-lineage specific outlier SNPs were shared across all lineages except AUK, and similarly AUK also reported the largest number of unique, lineage specific outliers (Fig. 4e). Many but not all overlapped SNPs were observed as outliers in the combined C2-comparison statistical analysis (Fig. S4).

In total 110 outlier SNPs were identified in at least two independent lineage comparisons. All genes falling within 10 kb of these outliers were assessed for GO term enrichment using the genome annotation from Stuart, Edwards *et al.,* (2022), but no GO terms were returned as significantly differentiated from the background dataset.

3.6 | Ancient and recent demographic changes in *Sturnus vulgaris*

Patterns of genomic structure, diversity, and adaptation within invasive populations may often be hard to interpret, due to the complex assortment of neutral and adaptive processes during establishment and spread (North et al., 2021). Additionally, as previously mentioned, starling numbers have been declining within native ranges and the species is becoming of increasing conservation concern (Robinson et al., 2005). Despite this, no genomic based historical

demographic estimates for this species exist, and thus, we sought to contextualise patterns of diversity and bottlenecks across the native and invasive lineages by examining the historical demography of the species.

The PSMC plots for all twelve individuals, spanning both native and invasive populations, showed complementary patterns with peak N_e estimated at around 150kya, followed by a steady decline that preceded even the last glacial period and continued until it's resolution at roughly 20kya. This pattern was fairly consistent across all individuals examined, though some variation existed in the bootstrapping values (Fig. S5). The results of STAIRWAY PLOT on native range samples reported a similar decrease between the timeframes of 100kya and 10kya, with the estimated population size holding steady since then (Fig. 5b). The invasive range samples had a similar trend, though reported a steeper decrease in N_e and a smaller present day estimate by roughly a factor of 10. This is not unexpected because previous analysis on another invasive avian species has demonstrated that invasive populations may display exaggerated changes in N_e using these methods, possibly due to recent bottlenecks and other demographic features present in invasive populations (Hilgers et al., 2024; Stuart et al., 2024).

Table 1 | Historical records of translocation of starlings between the invasive populations in Australia and New Zealand. AUK – Auckland (New Zealand), CAN – Canterbury (New Zealand), OTA – Otago (New Zealand), NSW – New South Wales (Australia), SA – South Australia, VIC – Victoria, TAS – Tasmania. In the reduced representation genetic dataset included in this study, NSW is represented by the sampling location ORG, while VIC and TAS are from the same genetic cluster represented by MLV. OTA is not directly represented in this study, with CAN being the closest sample site, while AUK is sampled in this study.

YEAR	SOURCE	TARGET	HOW MANY	REFERENCE
1878 - 1879 [a]	AUK	NSW	Two shipments, one reaching 50 birds. Birds were sent to 5 different localities, one received 28 starlings.	("INTERCOLONIAL NEWS.," 1879; "NEW SOUTH WALES MEMS.," 1878; "NEWS IN BRIEF," 1879; "NEWS OF THE DAY.," 1878; "Privy Council Selections.," 1878)
1880 [b]	OTA	VIC	9, one died on the way	("THE ACCLIMATISATION SOCIETY," 1880)
1881	AUK	VIC	Large consignment	("ACCLIMATISATION SOCIETY," 1881a; Auckland Acclimatisation Society, 1881)
1881 [c]	OTA	TAS	100, one died on the way, about one fourth short after the arrival, 50 liberated at once, rest given to some members to aviaries	("ACCLIMATISATION SOCIETY," 1881b; "OTAGO ACCLIMATISATION SOCIETY," 1881; "STARLINGS.," 1881)
1882 [c]	OTA	TAS	50	(Otago Acclimatisation Society, 1882)
1882	OTA	VIC	39	(Otago Acclimatisation Society, 1882)
1887		VIC	48, 36 were liberated in the gardens	("CITY COUNCIL.," 1887; "No Title," 1887)
1898	OTA	VIC (Gippsland)	78, maybe just around 50 survived?	("BRIEF MENTION," 1898; "THE Warragul Guardian, WITH WHICH IS INCORPORATED The Warragul News.," 1898; Otago Acclimatisation Society, 1900)

[a] Higgins et al. (2006) mentions the introduction of 'two small batches' of starlings into NSW from either VIC or NZ in 1880. In reality, there were two batches in 1878 and 1879, from Auckland.

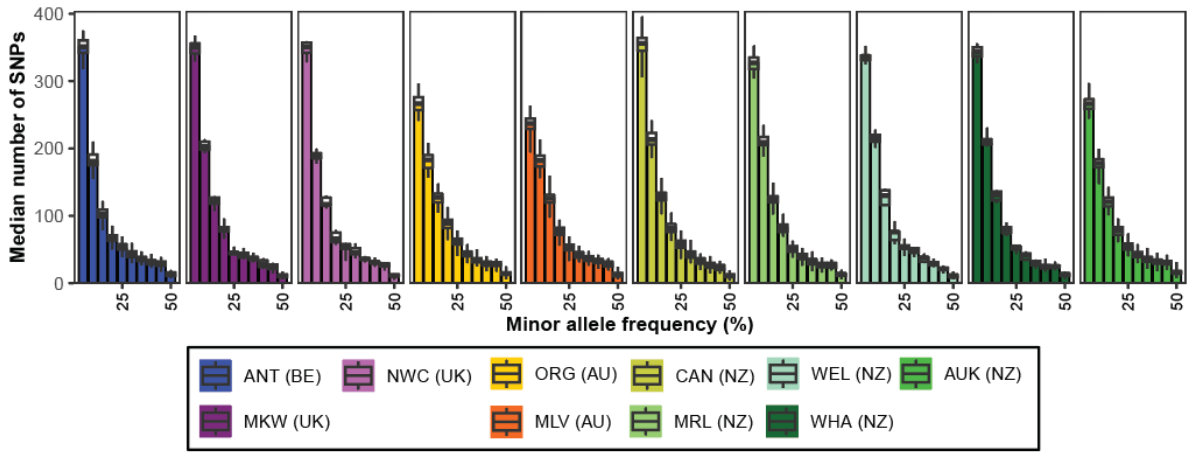
[b] Higgins et al. (2006) mentions the introduction of an unknown number of starlings into VIC in 1880 from New Zealand, and doesn't mention source location.

[c] Higgins et al. (2006) mentions the introduction of 75 starlings into TAS during 1880 (though

uncertainty exists with this date as alternate dates of 1800 and 1860 are also mentioned). Most likely this number is just a rough estimate – Crowther reported that 99 birds arrived and soon after arrival about one fourth died. 50 were released at once, the rest (unknown number, as others could die) to different places.

Table 2 | Global genetic population diversity of the starling, showing both genetic diversity indices (observed heterozygosity Ho, sample size corrected unbiased expected heterozygosity uHe, and inbreeding coefficient F_{IS}) and the number of private alleles of the ten sampling locations, including their respective sample sizes (n). F_{IS} ranges from -1 to +1, where positive results indicate a deficit of heterozygotes (excess of homozygotes), and conversely, negative results indicate an excess of heterozygotes (deficit of homozygotes). Metrics are derived from the full SNP dataset, which is comprised of a total of 19,174 SNPs. Population abbreviations are provided in the Fig. 1 caption.

Range	**Sample Location**	**n**	**Private alleles**	**Ho**	**uHe**	**F_{IS}**
Native	MKW	11	0	0.178	0.192	0.072
	NWC	11	0	0.174	0.192	0.092
	ANT	15	5	0.176	0.191	0.078
Australia	ORG	14	4	0.180	0.190	0.057
	MLV	15	0	0.175	0.186	0.063
New Zealand	AUK	18	20	0.181	0.187	0.031
	WEL	11	0	0.185	0.193	0.038
	WHA	11	1	0.190	0.194	0.021
	MRL	14	4	0.185	0.194	0.047
	CAN	21	3	0.187	0.195	0.041



476

477 **Figure 2 | Folded site frequency spectrum (SFS) for native and invasive starling** sampling
478 locations used as part of this study. The plot is generated from 1,451 SNPs (0% missingness),
479 each population was subset 100 times to 10 individuals to generate the error distribution.
480 Population abbreviations are provided in the Fig. 1 caption.

481

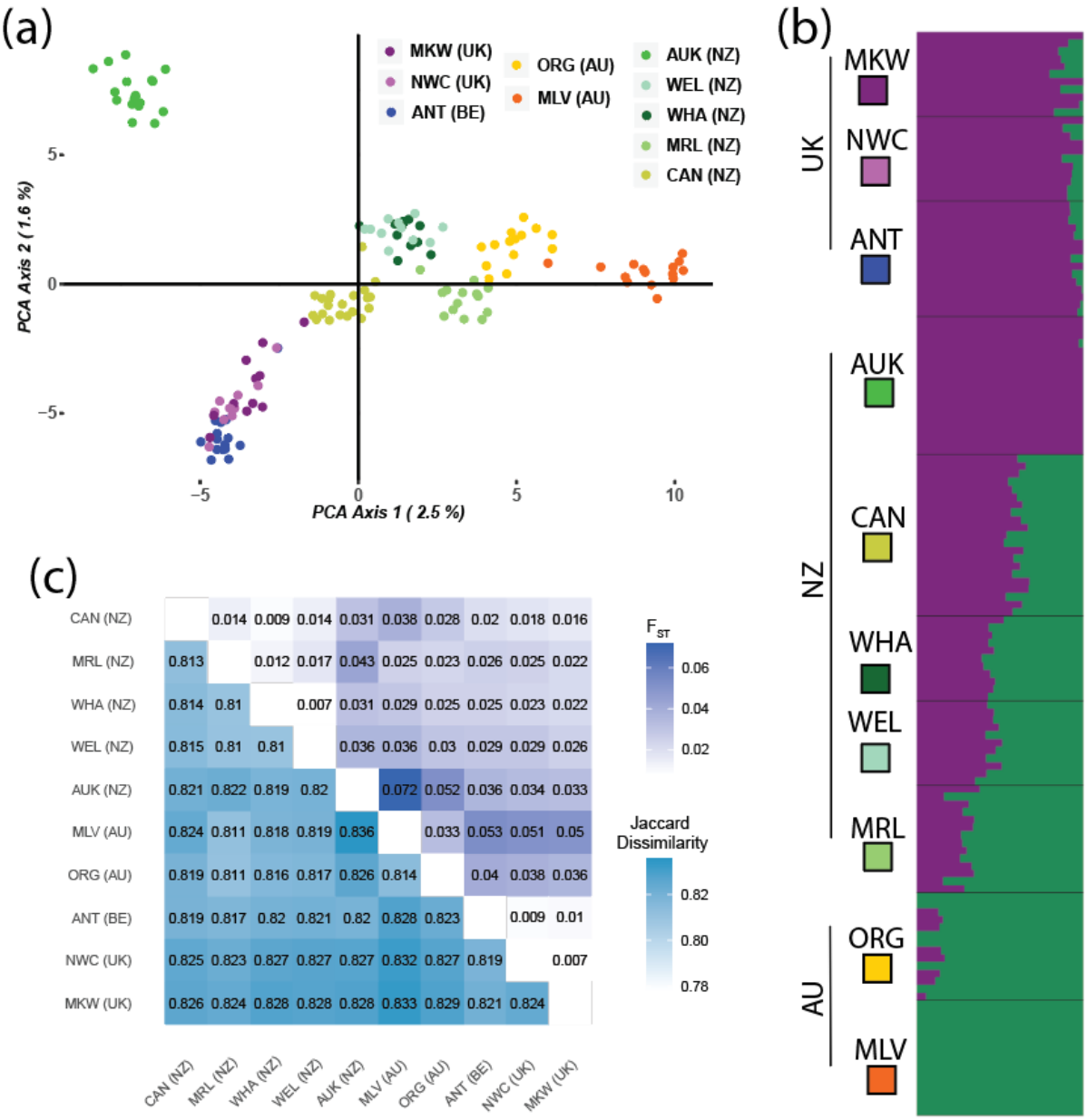


Figure 3 | Global genetic population structure of the starling. Panel **(a)** depicts a PCA of the ten sampled locations from across European native and Australian and New Zealand invasive ranges displaying PCA axis 1 (2.5% variance explained) and PCA axis 2 (1.6% variance explained). Panel **(b)** depicts the ADMIXTURE ancestry Q profile of the SNP dataset at K=2 calculated over 200 bootstrap resamplings. Panel **(c)** depicts a heatmap of pairwise genetic differentiation analysis between each of the ten sampled locations. Above the diagonal is pairwise F_{ST} values, with darker colour indicating a higher F_{ST}, which indicates more genetic differentiation. Below the diagonal is a heatmap of pairwise Jaccard dissimilarity values, with darker colour indicating a higher dissimilarity which is interpretable as fewer shared minor alleles. Population abbreviations are provided in the Fig. 1 caption.

Table 3 | Results of multiple matrix regression with randomization testing for isolation by distance (β_{IBD}) and isolation by environment (β_{IBE}), where β indicates the respective regression coefficient and *P* the associated p-value of the regression.

Samples	R^2	β_{IBD} (*P*)	β_{IBE} (*P*)
All samples (n = 75)	0.279	0.476 (0.0001)	0.0878 (0.0356)
Without Auckland (n = 57)	0.074	0.136 (0.0057)	0.180 (0.0108)

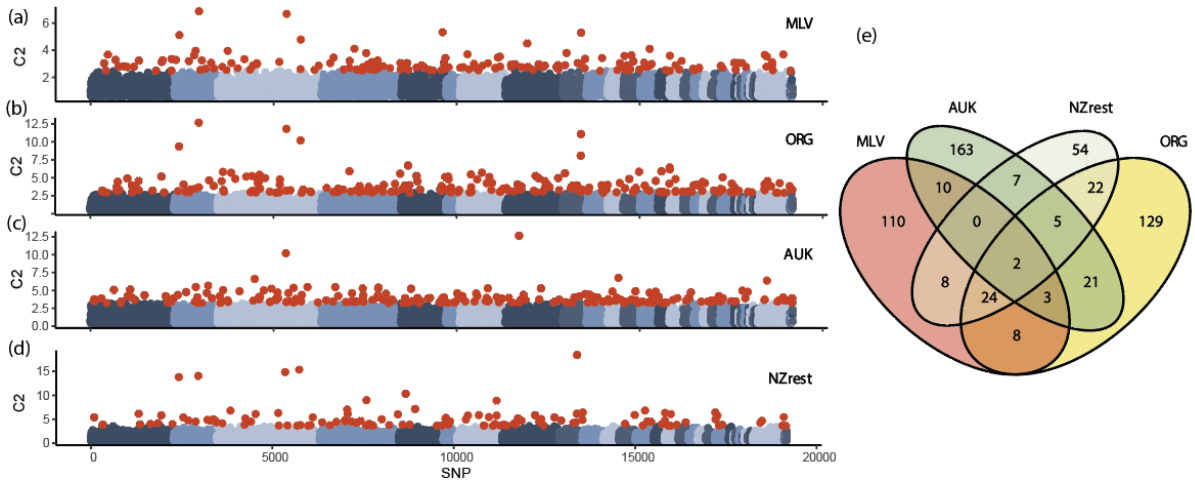
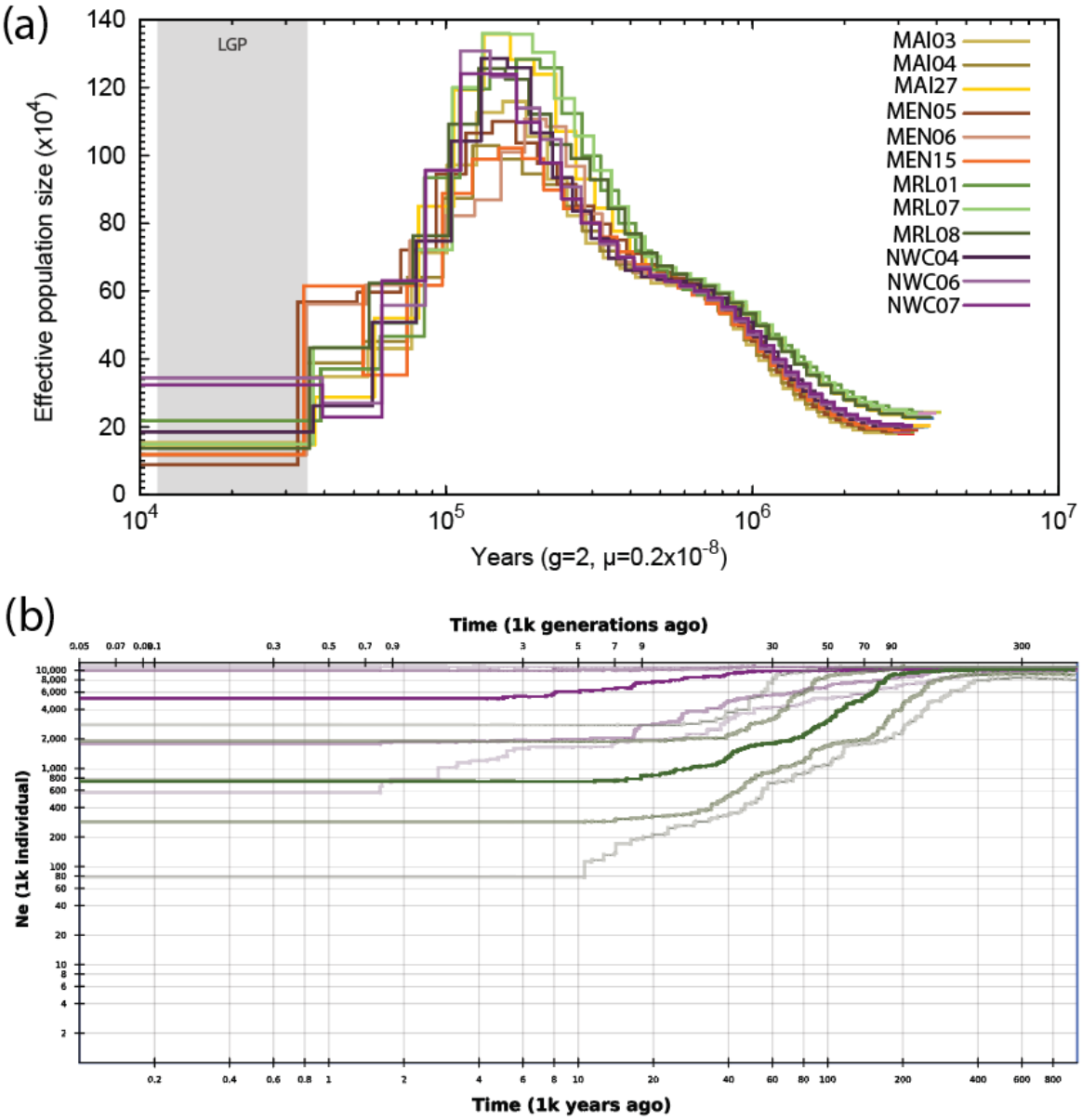


Figure 4 | Genetic outlier analysis conducted on the starling, using `BAYPASS` C2-contrast test on the native range sample sites (N = 3) against **(a)** McLaren Value, Australia (MLV, N = 1), **(b)** Orange, Australia (ORG; N = 1) **(c)** Auckland, New Zealand (AUK, N = 1), and **(d)** the rest of New Zealand (NZrest; N=4). Outlier SNPs are indicated in red. Panel **(e)** depicts the Venn diagram showing overlap between these 4 independent lineage genetic outlier analysis tests when compared to the invasive range sites.



509

510 **Figure 5 | Ancient and recent demographic history of the starling (*Sturnus vulgaris*).** Panel
511 (a) depicts N_e on an ancient timescale estimated by `PSMC` using whole genome resequencing
512 data of 12 samples. LGP = last glaciation period. MAI = New South Wales, Australia, MEN =
513 Victoria, Australia, MRL = Marlborough, New Zealand, NWC = Newcastle, UK. Panel **(b)** depicts
514 N_e during recent demographic history estimated by `STAIRWAY PLOT` using site frequency
515 spectrum data for native range individuals from Monks Wood (MKW) and Newcastle (NWC;
516 n=22) in purple, and invasive range individuals from Manawatū-Whanganui (WHA) and
517 Wellington (WEL; n=22) in green, using the DArT-seq SNP dataset. Lighter and darker lines
518 indicate 75% and 95% confidence intervals respectively.

519

4 | DISCUSSION

The invasive starlings within New Zealand have a complicated history of introductions involving both importation from the native range and many translocations between populations in the invasive ranges. In this study we have examined the genetic diversity and population structure present within this population and interpret the results alongside a backdrop of rich introduction history information. Additionally, we contrast these genetic patterns to those in the native range and invasive Australian range, to better understand how the invasion history and invasion processes have shaped the genetic structure of the present-day populations.

4.1 | Integrating present day population genomics with introduction history within New Zealand

According to historical records, the establishment of starlings across the New Zealand landscape involved the concerted introduction of over 2,000 birds, predominantly by local acclimatisation societies (Pipek et al., 2019). Over 600 starlings were imported from the native range to New Zealand between 1860 and 1873 in multiple shipments, most of which originated in London. These importations established populations in Auckland (AUK), Canterbury (CAN), and Otago (here unsampled), but also contributed to other locations in New Zealand. While most imports were of relatively small numbers of individuals (at most 41, but generally around a dozen), the Otago population was predominantly founded by two large imports of 97 and 104 birds from London (Fig. 1). Translocations of over 1,600 birds within New Zealand were then responsible for establishing or bolstering other populations, with over 1,400 of these translocated birds sourced from Otago. Auckland and Canterbury contributed a handful of birds to other locations, but there are no records of them receiving translocations from Otago or other regions (Pipek et al., 2019). In contrast, of the sampling locations obtained in this study, the Marlborough region received at least two batches of starlings from Otago: 50 in 1882, and an unknown number in 1883. The Wellington region received over 250 from Otago in five batches between 1877 and 1883. While the starling is partially migratory in its native range, there is no evidence for general migratory movement during winter in New Zealand (Ross, 1983), and it is likely that dispersal distances within New Zealand are fairly moderate, as documented in other invasive ranges (Cabe, 1999; Rollins et al., 2009; Waterman et al., 2008).

The primary feature of the population genomic profile of New Zealand starlings, and in strong alignment with the documented introduction history, is the genetic differentiation between Auckland and all other sampling locations. Several lines of evidence indicate that this is likely due to founder and admixture effects from multiple small, independent introductions along

553 with limited gene flow with other regions since establishment, and not late invasion processes
554 such as adaptation or a recent cryptic introduction. These lines of evidence include patterns
555 seen in population structuring and differentiation (Fig. 1), a distinct SFS signature but without
556 different inbreeding metrics (Fig. 2, Table 2, overlapping differences in outlier SNPs (Fig. 4,
557 discussed more below), and the fact that inclusion of Auckland into MMRR analysis drastically
558 increases the relative explanatory power of geographic distance over environmental similarity
559 (Table 3). Further, Auckland had the highest number of private alleles (Table 2), which in other
560 invasive species have been attributed to multiple introductions from source locations with
561 differing allele frequencies acting in concert with founder effects (Gonçalves da Silva et al.,
562 2010). The retention of these private alleles in Auckland suggests that there is likely restricted
563 gene flow between Auckland and the other New Zealand sampling locations. Thus, like with
564 Australia (Rollins et al., 2011), we may consider that within New Zealand there are at least two
565 distinct invasive lineages, with F_{ST} values across these different invasive lineages being
566 comparable (Fig. 3c). This result generally agrees with previous country wide patterns found in
567 allozyme data (Ross, 1983) which identified Auckland and Nelson, in the north of the South
568 Island (here unsampled) as clustering more closely with native range UK samples than to all
569 other New Zealand populations.

570 When considering the remainder of the New Zealand sampling locations, here termed 'NZrest',
571 comparing relative F_{ST} values within the invasive ranges to the native sample sites indicates
572 where gene flow may be occurring and where it is likely restricted. Pairwise F_{ST} values
573 comparable to native ranges (pairwise F_{ST}: 0.007-0.009) are seen between Wellington (WEL) and
574 Manawatū-Whanganui (WHA) (Fig. 3c), which due to geographical proximity and a lack of
575 separation by an elevational barrier, known to limit starling movement (Higgins et al., 2006), are
576 likely to be two readily interbreeding locations. There is indication of genetic similarities and
577 thus historic or ongoing gene flow across all these latitudinally southern locations despite no
578 evidence to date of migratory behaviour in New Zealand. Of particular note is the low genetic
579 differentiation between Canterbury (CAN) and the Manawatū-Whanganui (WHA) (Fig. 3c,
580 pairwise F_{ST} 0.009), two locations that are the most geographically separate of NZrest (Fig. 1)
581 and reportedly founded from different sources, with WHA most likely established predominantly
582 from Otago birds (Pipek et al., 2019). The similarity of Canterbury with the rest of New Zealand
583 therefore suggests that either Canterbury and Otago contained introductions with similar initial
584 genetic profiles, or that ongoing gene flow has reduced potential genetic differences. The former
585 explanation is supported by historical records that demonstrate that largescale bird shipments
586 to Canterbury and Otago were organised by one bird fancier family during the late 1800's (Pipek

et al., 2015). Meanwhile, the latter explanation is most supported by the Jaccard dissimilarity metrics (Fig. 3c), because Canterbury becomes more genetically distinct from NZrest when considering only minor alleles. This suggests that it is possible that higher immigration than emigration may have helped to maintain rare allelic differences, while also bolstering genetic diversity in this location (Fig 2) that only had a moderate number of individuals introduced compared to Otago (Fig. 1).

Within the NZrest sampling locations, patterns of genetic differentiation are explained by environment more so than geographic distance (Table 3), which supports the existence of some ongoing localised adaptation within this interbreeding genetic subpopulation. Nonetheless, only a small portion of genetic variation across these sampling sites is explained by geographic or environmental distance, indicating likely a strong role for processes related to introduction history and drift. These conclusions are in alignment with a study that found that starling morphometric variation was largely haphazard across the landscape and likely primarily driven by founding effects and drift (Ross & Baker, 1982).

The starling's northern invasive range within New Zealand overlaps that of a closely related second invasive species, the common myna (*Acridotheres tristis*). Interestingly, despite being phylogenetically close and having been introduced to similar localities around New Zealand during the late 1800's, present day population structure patterns of the two species are very different (Atsawawaranunt et al., 2023). While myna also show two genetic clusters, there is separation between myna populations in the far East coast and the rest of the North Island. This difference in genetic clustering compared to starlings is likely indicative of fundamental biological differences in, for example, dispersal between the two species shaping their present day gene flow, and likely also founder effects during introduction. This indicates that even population genomics of closely related, co-occurring species cannot accurately infer patterns in another species, emphasising the benefit of species-specific genomic resources.

4.2 | Comparison of New Zealand and Australian invasive lineages alongside historical translocation information

Within our study, comparative analysis between the New Zealand and Australian invasive starling populations revealed that both had similar levels of genetic differentiation across sampling locations (Fig. 3a) but with distinctive ancestry admixture signals (Fig. 3b). Due to the many repeated introductions from New Zealand to Australia over several decades (Table 1), the present-day invasive range patterns may be due to initial differences in introduced individuals, or a result of demographic effects caused by stochastic invasion processes during translocation

and establishment within founding lineages. Strong genetic differentiation has also been described between the invasive North American and Australian ranges, reiterating the strength of founder effects in invasive populations for this species globally (Hofmeister et al., 2024).

Genetic outlier analysis can be used to infer parallel signals of bottlenecks and/or adaptation within invasive lineages (e.g. Parvizi et al., 2024). Where the invasive history of a population is characterized by early translocation events between invasive ranges, as with the starling (Table 1), now-independent invasive lineages that share common outlier SNPs may be reflecting translocation history rather than parallel evolutionary biological processes. This is because outlier regions within invasive populations may be due to complicated signals generated from concurrent demographic processes such as bottlenecks, drift, and range expansion (Salloum et al., 2022; Stuart et al., 2021). If two geographically separated lineages with known historical translocations contain the same signal, it is possible that this occurs because of shared introduction history, though parallel adaptation is a plausible alternate theory (Hodgins et al., 2015; Zenni & Hoban, 2015).

The outlier SNPs across the four distinct invasive lineages indicate the largest number of shared outliers between the two Australian lineages and NZrest, with the later sampling group having the smallest number of unique outlier loci (Fig. 5). These results offer genetic support to historical records that claim the translocations were successful (Table 1, Appendix 3: Historical records of starlings), and we may interpret this as the unidirectional sharing of alleles reducing the number of unique outliers within NZrest. Intriguingly, despite AUK being genetically distinct in terms of genome wide patterns, there is a high proportion of outlier loci overlap between AUK and ORG (Fig. 5). This may be because of translocations (Table 1) or because both these locations are more temperate region and thus parallel selection within this region is a possibility (see below). Sequencing of historical samples from the Auckland region would enable the origin of this genetic signature to be established.

4.3 | Weak inbreeding but not bottlenecks are ubiquitous across invasive starling lineages

Across both invasive and native sampling locations, we observe a consistently weak pattern of inbreeding, with slightly elevated levels of F_{IS} in all ten sampling locations (Table 2), likely reflective of generally moderate dispersal distances and philopatry recorded in other invasive ranges (Cabe, 1999; Rollins et al., 2009; Waterman et al., 2008). Interestingly, the three native range populations all have higher levels of inbreeding than any of their invasive counterparts, though the difference is marginal (Table 2). These relative values between native and invasive range F_{IS} could be attributed to the recent dramatic native range declines (Heldbjerg et al., 2016;

653 Rintala et al., 2003; Robinson et al., 2005). This contrasts with historical demographic patterns
654 evident in this species (Fig. 5), which indicate stable N_e estimates post the last glacial period
655 and suggest recent drops in starling numbers are not part of a more long-term trend.

656 Patterns of genetic bottlenecks as indicated by site frequency spectrum analysis (Fig. 2) are
657 consistent with the expected levels of heterozygosity (Table 2). The native range and Canterbury
658 exhibit similar proportions of rare alleles, with the NZrest sampling locations having marginally
659 fewer. This is in contrast to previous results from allozyme based estimates, which reported a
660 loss of rare alleles within New Zealand in comparison to the native range (Ross, 1983). We
661 observe strong genetic bottlenecks in Auckland, despite historical records suggesting that the
662 population was founded by similar number of individuals as Canterbury (118 vs 137 individuals
663 with little evidence for translocation to each of these two locations; see Pipek et al. (2019) for
664 more details). The stronger bottlenecks seen in Auckland and the Australian lineages could be
665 due to the warmer climates of these locations, which are more dissimilar to the starlings' native
666 range (Higgins et al., 2006). However, it could also reflect the contrasting impacts of many small
667 introductions to Auckland and the large Canterbury introductions over a shorter timeframe
668 (Pipek et al., 2019), with the former having stronger bottlenecks and less adaptive potential.
669 Environmental dissimilarities may have also exerted a stronger selection regime (Royall, 1966)
670 to result in increased population bottlenecks within these location, and may explain why Otago
671 was such a popular source of starling translocation to Australia and elsewhere in New Zealand,
672 as the species may have had more success establishing in this cooler region.

673 **4.4 | Implications for management**

674 Interpreting these population structure results alongside complementary literature on the
675 environmental niche the starlings occupy within New Zealand allows us to make some
676 hypotheses around future patterns of population structure and the feasibility of management
677 for this species. While there is mixed evidence for negative impacts of starlings on the New
678 Zealand ecology (Flux, 2013), they are still routinely controlled across New Zealand because of
679 their agricultural impacts, which are of particular concern within the wine industry (Campbell et
680 al., 2016). The present-day strong genetic division between the north-west of New Zealand
681 (represented by the sampling location of Auckland, AUK) and the rest of the country may initially
682 present as two management units. Unfortunately, previous niche modelling work on this
683 species found increasing suitable habitats at higher elevation under future climate change
684 scenarios, meaning that mountain ranges which may help reinforce current population
685 structure may present less of a barrier in the future (Atsawawaranunt et al., 2024). Increasing

sampling in the centre of the North Island, as well as along the west coast of the South Island would help to confirm the nature of dispersal and admixture at the boundaries of these two genetic subpopulations. However, it is likely that local removal of starlings within any New Zealand region is not a feasible management solution for this species, as reinvasion would likely occur from within the country.

5 | Conclusion

In summary, while the starlings' range in New Zealand may initially appear to be continuous, there are multiple lines of evidence for strong population structure that is likely a result of founder effects that are being maintained under present day gene flow patterns. Further, historical accounts of translocations between both Australian subpopulations and New Zealand remain supported by the genetic data, though the populations display distinct signatures of bottlenecks. The unique population genomic patterns of the New Zealand starlings emphasise the need for species-specific genetic data for management and informing our understanding of invasion processes within an invasive range and more broadly.

Author contributions

Project conceived by LAR, AW, AWS, and KCS. Sample coordination and extractions performed by KA, HZT, AW, AWS, and KCS. The historical data explored done by PP. Analysis performed by BT, KA, MCN, WP, EOP, HZT, and KCS. Manuscript drafting was done by BT and KCS, with contributions from KA, MCN, CP, EOP, PP and HZT. All authors edited and contributed to the final version of this manuscript.

Ethics

All birds that were newly sequenced as part of this study were captured and culled by private landowners or euthanised by BirdCare Aotearoa under the International Wildlife Rehabilitation Council (IWRC) and the National Wildlife Rehabilitation Association (NWRA) licence Code of Professional Ethics, so ethics approval was not needed for this study.

Data availability

The raw sequencing data have been deposited under BioProject accession no. XXXX in the NCBI BioProject database (https://www.ncbi.nlm.nih.gov/bioproject/). Processed genetic data files and basic code is available on Dryad (XXXXX) and Zenodo (XXXXX). More fully annotated and

cleaned code, along with some project vignettes and any other relevant files or metadata for this project are available on GitHub (https://github.com/katarinastuart/Sv10_NZstarlings/).

Acknowledgements

We extend many thanks to the collectors who contributed starling samples from New Zealand. In particular, we would like to acknowledge John, Ian, and Merryl Flux, Bart Arnst, Andrew Veale, Aimee Hoeberigs, Kristal Cain, Jim Cook, and Paige Matheson for providing samples as part of control efforts in New Zealand and thank Ariel-Micaiah Heswall, Lynn Miller, and Dani Najera Archila from BirdCare Aotearoa for providing samples from injured birds with untreatable injuries. We acknowledge the use of New Zealand eScience Infrastructure (NeSI) high-performance computing facilities and thank the NeSI team, particularly Dinindu Senanayake, for their support and troubleshooting. Open access publishing is facilitated by The University of Auckland, as organised by the Council of Australian University Librarians and its Member Institutions.

Funding

A Marsden Grant (UOA1911) awarded to AWS from the New Zealand Royal Society Te Aparangi supported KA, AWS, LAR, AW, and KCS and funded sample sequencing. BT was supported by a University of Auckland Summer Research Scholarship.

REFERENCES

ACCLIMATISATION SOCIETY. (1880a, June 11). *LYTTELTON TIMES*. https://paperspast.natlib.govt.nz/newspapers/LT18800611.2.32

ACCLIMATISATION SOCIETY. (1880b, November 5). *LYTTELTON TIMES*. https://paperspast.natlib.govt.nz/newspapers/LT18801105.2.5

ACCLIMATISATION SOCIETY. (1881a). *New Zealand Herald*, 6. https://paperspast.natlib.govt.nz/newspapers/NZH18810315.2.42

ACCLIMATISATION SOCIETY. (1881b). *Otago Daily Times*. https://paperspast.natlib.govt.nz/newspapers/ODT18810307.2.26

ACCLIMATISATION SOCIETY. (1882, June 1). *PRESS*, PAGE 3. https://paperspast.natlib.govt.nz/newspapers/CHP18820601.2.21

Alexander, D. H., Novembre, J., & Lange, K. (2009). Fast model-based estimation of ancestry in unrelated individuals. *Genome Research*, *19*(9), 1655–1664. https://doi.org/10.1101/gr.094052.109

Atsawawaranunt, K., Ewart, K. M., Major, R. E., Johnson, R. N., Santure, A. W., & Whibley, A. (2023). Tracing the introduction of the invasive common myna using population genomics. *Heredity*, *131*, 56–67. https://doi.org/10.1038/s41437-023-00621-w

Atsawawaranunt, K., Whibley, A., Cain, K. E., Major, R. E., & Santure, A. W. (2024). Projecting the current and potential future distribution of New Zealand's invasive sturnids. *Biological Invasions*, *26*(5), 1345–1366. https://doi.org/10.1007/s10530-024-03246-0

Auckland Acclimatisation Society. (1881). *Annual report and financial statement of the Auckland Acclimatisation Society for 1880-1881*.

Bodt, L. H., Rollins, L. A., & Zichello, J. M. (2020). Contrasting mitochondrial diversity of European starlings (Sturnus vulgaris) across three invasive continental distributions. *Ecology and Evolution*, *10*(18), 10186–10195. https://doi.org/10.1002/ece3.6679

759 BRIEF MENTION. (1898). *Evening Star*, 3.
760 https://paperspast.natlib.govt.nz/newspapers/ESD18980217.2.54
761 Cabe, P. R. (1999). Dispersal and Population Structure in the European Starling. *The Condor*, *101*(2),
762 451–454. https://doi.org/10.2307/1370014
763 Cabe, P. R. (2020). *European Starling (Sturnus vulgaris), version 1.0. In Birds of the World (S. M.*
764 *Billerman, Editor)*. Cornell Lab of Ornithology, Ithaca, NY, USA.
765 https://doi.org/10.2173/bow.eursta.01
766 Campbell, S., Roberts, E. J., Craemer, R., Pacioni, C., Rollins, L., & Woolnough, A. P. (2016). Assessing
767 the economic benefits of starling detection and control to Western Australia. *Australasian*
768 *Journal of Environmental Management*, *23*(1), 81–99.
769 https://doi.org/10.1080/14486563.2015.1028486
770 Catchen, J., Hohenlohe, P. A., Bassham, S., Amores, A., & Cresko, W. A. (2013). Stacks: An analysis tool
771 set for population genomics. *Molecular Ecology*, *22*(11), 3124–3140.
772 https://doi.org/10.1111/mec.12354
773 Chen, S., Zhou, Y., Chen, Y., & Gu, J. (2018). fastp: An ultra-fast all-in-one FASTQ preprocessor.
774 *Bioinformatics*, *34*(17), i884–i890. https://doi.org/10.1093/bioinformatics/bty560
775 CITY COUNCIL. (1887). *The Ballarat Star*, 4. https://trove.nla.gov.au/newspaper/article/203951949
776 Colautti, R. I., & Lau, J. A. (2015). Contemporary evolution during invasion: Evidence for
777 differentiation, natural selection, and local adaptation. *Molecular Ecology*, *24*(9), 1999–2017.
778 https://doi.org/10.1111/mec.13162
779 Danecek, P., Auton, A., Abecasis, G., Albers, C. A., Banks, E., DePristo, M. A., Handsaker, R. E., Lunter,
780 G., Marth, G. T., Sherry, S. T., McVean, G., & Durbin, R. (2011). The variant call format and
781 VCFtools. *Bioinformatics*, *27*(15), 2156–2158. https://doi.org/10.1093/bioinformatics/btr330
782 Danecek, P., Bonfield, J. K., Liddle, J., Marshall, J., Ohan, V., Pollard, M. O., Whitwham, A., Keane, T.,
783 McCarthy, S. A., Davies, R. M., & Li, H. (2021). Twelve years of SAMtools and BCFtools.
784 *GigaScience*, *10*(2), giab008. https://doi.org/10.1093/gigascience/giab008

Essl, F., Lenzner, B., Bacher, S., Bailey, S., Capinha, C., Daehler, C., Dullinger, S., Genovesi, P., Hui, C., Hulme, P. E., Jeschke, J. M., Katsanevakis, S., Kühn, I., Leung, B., Liebhold, A., Liu, C., MacIsaac, H. J., Meyerson, L. A., Nuñez, M. A., … Roura-Pascual, N. (2020). Drivers of future alien species impacts: An expert-based assessment. *Global Change Biology*, *26*(9), 4880–4893. https://doi.org/10.1111/gcb.15199

Evans, T., Blackburn, T. M., Jeschke, J. M., Probert, A. F., & Bacher, S. (2020). Application of the Socio-Economic Impact Classification for Alien Taxa (SEICAT) to a global assessment of alien bird impacts. *NeoBiota*, *62*, 123–142. https://doi.org/10.3897/neobiota.62.51150

Feare, C. J. (1984). *The Starling*. Oxford University Press.

Ferrer, X., Motis, A., & Peris, S. (1991). Changes in the Breeding Range of Starlings in the Iberian Peninsula During the Last 30 Years: Competition as a Limiting Factor. *Journal of Biogeography*, *18*(6).

Fick, S. E., & Hijmans, R. J. (2017). WorldClim 2: New 1-km spatial resolution climate surfaces for global land areas. *International Journal of Climatology*, *37*(12), 4302–4315. https://doi.org/10.1002/joc.5086

Fiorini, V. D., Domínguez, M., Reboreda, J. C., & Swaddle, J. P. (2022). A recent invasive population of the European starling sturnus vulgaris has lower genetic diversity and higher fluctuating asymmetry than primary invasive and native populations. *Biological Invasions*, *24*, 437–448. https://doi.org/10.1007/s10530-021-02653-x

Flux, J. E. C. (2013). *[updated 2022] Common starling | tāringi. In Miskelly, C.M. (ed.)*. New Zealand Birds Online. www.nzbirdsonline.org.nz

Gao, C.-H., Yu, G., & Cai, P. (2021). ggVennDiagram: An Intuitive, Easy-to-Use, and Highly Customizable R Package to Generate Venn Diagram. *Frontiers in Genetics*, *12*, 706907. https://doi.org/10.3389/fgene.2021.706907

Gautier, M. (2015). Genome-Wide Scan for Adaptive Divergence and Association with Population-Specific Covariates. *Genetics*, *201*(4), 1555–1579. https://doi.org/10.1534/genetics.115.181453

Gonçalves da Silva, A., Eberhard, J. R., Wright, T. F., Avery, M. L., & Russello, M. A. (2010). Genetic evidence for high propagule pressure and long-distance dispersal in monk parakeet (Myiopsitta monachus) invasive populations. *Molecular Ecology*, *19*(16), 3336–3350. https://doi.org/10.1111/j.1365-294X.2010.04749.x

Gosselin, T., Lamothe, M., Devloo-Delva, F., & Grewe, P. (2020). *radiator: RADseq Data Exploration, Manipulation and Visualization using R* (10.5281/zenodo.3687060) [Computer software]. https://thierrygosselin.github.io/radiator/

Harris, C. R., Millman, K. J., van der Walt, S. J., Gommers, R., Virtanen, P., Cournapeau, D., Wieser, E., Taylor, J., Berg, S., Smith, N. J., Kern, R., Picus, M., Hoyer, S., van Kerkwijk, M. H., Brett, M., Haldane, A., del Río, J. F., Wiebe, M., Peterson, P., … Oliphant, T. E. (2020). Array programming with NumPy. *Nature*, *585*(7825), 357–362. https://doi.org/10.1038/s41586-020-2649-2

Harris, G. (1964). Climatic Changes Since 1860 Affecting European Birds. *Weather*, *19*(3), 70–79. https://doi.org/10.1002/j.1477-8696.1964.tb02074.x

Heldbjerg, H., Fox, A. D., Levin, G., & Nyegaard, T. (2016). The decline of the Starling Sturnus vulgaris in Denmark is related to changes in grassland extent and intensity of cattle grazing. *Agriculture, Ecosystems & Environment*, *230*, 24–31. https://doi.org/10.1016/j.agee.2016.05.025

Higgins, P. J., Peter, J. M., & Cowling, S. J. (2006). *Handbook of Australian, New Zealand & Antarctic birds. Volume 7, Boatbill to starlings*. Melbourne : Oxford University Press. https://trove.nla.gov.au/version/24973684

Hilgers, L., Liu, S., Jensen, A., Brown, T., Cousins, T., Schweiger, R., Guschanski, K., & Hiller, M. (2024). *Avoidable false PSMC population size peaks occur across numerous studies* (p. 2024.06.17.599025). bioRxiv. https://doi.org/10.1101/2024.06.17.599025

Hodgins, K. A., Bock, D. G., Hahn, M. A., Heredia, S. M., Turner, K. G., & Rieseberg, L. H. (2015). Comparative genomics in the Asteraceae reveals little evidence for parallel evolutionary change in invasive taxa. *Molecular Ecology*, *24*(9), 2226–2240. https://doi.org/10.1111/mec.13026

Hofmeister, N. R., Stuart, K. C., Warren, W. C., Werner, S. J., Bateson, M., Ball, G. F., Buchanan, K. L., Burt, D. W., Cardilini, A. P. A., Cassey, P., De Meyer, T., George, J., Meddle, S. L., Rowland, H. M., Sherman, C. D. H., Sherwin, W. B., Vanden Berghe, W., Rollins, L. A., & Clayton, D. F. (2024). Concurrent invasions of European starlings in Australia and North America reveal population-specific differentiation in shared genomic regions. *Molecular Ecology*, *in press*. https://doi.org/10.1111/mec.17195

Hofmeister, N. R., Werner, S. J., & Lovette, I. J. (2021). Environmental correlates of genetic variation in the invasive European starling in North America. *Molecular Ecology*, *30*(5), 1251–1263. https://doi.org/10.1111/mec.15806

Hulme, P. E. (2009). Trade, transport and trouble: Managing invasive species pathways in an era of globalization. *Journal of Applied Ecology*, *46*(1), 10–18. https://doi.org/10.1111/j.1365-2664.2008.01600.x

INTERCOLONIAL NEWS. (1879). *Wagga Wagga Express*, 3. https://trove.nla.gov.au/newspaper/article/145215072

Jenkins, C. F. H. (with Western Australia). (1977). *The Noah's ark syndrome: One hundred years of acclimatization and zoo development in Australia*. Zoological Gardens Board of Western Australia.

Kilian, A., Wenzl, P., Huttner, E., Carling, J., Xia, L., Blois, H., Caig, V., Heller-Uszynska, K., Jaccoud, D., Hopper, C., Aschenbrenner-Kilian, M., Evers, M., Peng, K., Cayla, C., Hok, P., & Uszynski, G.

(2012). Diversity arrays technology: A generic genome profiling technology on open platforms. *Methods in Molecular Biology (Clifton, N.J.)*, *888*, 67–89. https://doi.org/10.1007/978-1-61779-870-2_5

Legendre, P., & Legendre, L. (2012). *Numerical Ecology* (3rd edition). Elsevier.

Li, H., & Durbin, R. (2009). Fast and accurate short read alignment with Burrows–Wheeler transform. *Bioinformatics*, *25*(14), 1754–1760. https://doi.org/10.1093/bioinformatics/btp324

Li, H., Handsaker, B., Wysoker, A., Fennell, T., Ruan, J., Homer, N., Marth, G., Abecasis, G., Durbin, R., & 1000 Genome Project Data Processing Subgroup. (2009). The Sequence Alignment/Map format and SAMtools. *Bioinformatics (Oxford, England)*, *25*(16), 2078–2079. https://doi.org/10.1093/bioinformatics/btp352

Liu, S., Ferchaud, A.-L., Grønkjaer, P., Nygaard, R., & Hansen, M. M. (2018). Genomic parallelism and lack thereof in contrasting systems of three-spined sticklebacks. *Molecular Ecology*, *27*(23), 4725–4743. https://doi.org/10.1111/mec.14782

Lowe, S., Browne, M., & Boudjelas, S. (2000). *100 of the world's worst invasive alien species. A selection from the global invasive species database*. Invasive Species Specialist Group.

Matheson, P., & McGaughran, A. (2022). Genomic data is missing for many highly invasive species, restricting our preparedness for escalating incursion rates. *Scientific Reports*, *12*(1), Article 1. https://doi.org/10.1038/s41598-022-17937-y

McDowall, R. M. (1994). *Gamekeepers for the Nation: The Story of New Zealand's Acclimatisation Societies, 1861-1990*. Canterbury University Press.

McGaughran, A., Dhami, M. K., Parvizi, E., Vaughan, A. L., Gleeson, D. M., Hodgins, K. A., Rollins, L. A., Tepolt, C. K., Turner, K. G., Atsawawaranunt, K., Battlay, P., Congrains, C., Crottini, A., Dennis, T. P. W., Lange, C., Liu, X. P., Matheson, P., North, H. L., Popovic, I., … Wilson, J. (2024). Genomic tools in biological invasions: Current state and future frontiers. *Genome Biology and Evolution*, evad230. https://doi.org/10.1093/gbe/evad230

884 Mi, H., Muruganujan, A., Huang, X., Ebert, D., Mills, C., Guo, X., & Thomas, P. D. (2019). Protocol
885 Update for large-scale genome and gene function analysis with the PANTHER classification
886 system (v.14.0). *Nature Protocols*, *14*(3), 703–721. https://doi.org/10.1038/s41596-019-
887 0128-8
888 Mijangos, J. L., Gruber, B., Berry, O., Pacioni, C., & Georges, A. (2022). dartR v2: An accessible genetic
889 analysis platform for conservation, ecology and agriculture. *Methods in Ecology and*
890 *Evolution*, *13*(10), 2150–2158. https://doi.org/10.1111/2041-210X.13918
891 Mussmann, S. M., Douglas, M. R., Chafin, T. K., & Douglas, M. E. (2019). BA3-SNPs: Contemporary
892 migration reconfigured in BayesAss for next-generation sequence data. *Methods in Ecology*
893 *and Evolution*, *10*(10), 1808–1813. https://doi.org/10.1111/2041-210X.13252
894 Nadachowska-Brzyska, K., Li, C., Smeds, L., Zhang, G., & Ellegren, H. (2015). Temporal Dynamics of
895 Avian Populations during Pleistocene Revealed by Whole-Genome Sequences. *Current*
896 *Biology*, *25*(10), 1375–1380. https://doi.org/10.1016/j.cub.2015.03.047
897 Nanninga, G. B., Saenz-Agudelo, P., Manica, A., & Berumen, M. L. (2014). Environmental gradients
898 predict the genetic population structure of a coral reef fish in the Red Sea. *Molecular*
899 *Ecology*, *23*(3), 591–602. https://doi.org/10.1111/mec.12623
900 NEW SOUTH WALES MEMS. (1878). *Albury Banner and Wodonga Express*, 7.
901 https://trove.nla.gov.au/newspaper/article/257962187
902 NEWS IN BRIEF. (1879). *New Zealand Herald*, 7.
903 https://paperspast.natlib.govt.nz/newspapers/NZH18790111.2.42
904 NEWS OF THE DAY. (1878). *Sydney Morning Herald*, 5.
905 https://trove.nla.gov.au/newspaper/article/13426649
906 No title. (1887). *The Ballarat Star*, 2. https://trove.nla.gov.au/newspaper/article/207773316
907 North, H. L., McGaughran, A., & Jiggins, C. (2021). Insights into invasive species from whole-genome
908 resequencing. *Molecular Ecology*, *n/a*(n/a). https://doi.org/10.1111/mec.15999

Olazcuaga, L., Loiseau, A., Parrinello, H., Paris, M., Fraimout, A., Guedot, C., Diepenbrock, L. M., Kenis, M., Zhang, J., Chen, X., Borowiec, N., Facon, B., Vogt, H., Price, D. K., Vogel, H., Prud'homme, B., Estoup, A., & Gautier, M. (2020). A Whole-Genome Scan for Association with Invasion Success in the Fruit Fly Drosophila suzukii Using Contrasts of Allele Frequencies Corrected for Population Structure. *Molecular Biology and Evolution*, *37*(8), 2369. https://doi.org/10.1093/molbev/msaa098

OTAGO ACCLIMATISATION SOCIETY. (1881). *Otago Daily Times*, 3. https://paperspast.natlib.govt.nz/newspapers/ODT18810208.2.18

Otago Acclimatisation Society. (1882). *Sixteenth annual report of the Otago Acclimatisation Society*.

Otago Acclimatisation Society. (1900). *Cashbook Proper 1864-1900*.

Padgham, M. (2021). *geodist: Fast, Dependency-Free Geodesic Distance Calculations* (R package version 0.0.7) [Computer software]. https://github.com/hypertidy/geodist

Parvizi, E., Vaughan, A. L., Dhami, M. K., & McGaughran, A. (2024). Genomic signals of local adaptation across climatically heterogenous habitats in an invasive tropical fruit fly (Bactrocera tryoni). *Heredity*, *132*(1), 18–29. https://doi.org/10.1038/s41437-023-00657-y

Pembleton, L. W., Cogan, N. O. I., & Forster, J. W. (2013). StAMPP: An R package for calculation of genetic differentiation and structure of mixed-ploidy level populations. *Molecular Ecology Resources*, *13*(5), 946–952. https://doi.org/10.1111/1755-0998.12129

Phair, D. J., Roux, J. J. L., Berthouly-Salazar, C., Visser, V., Vuuren, B. J. van, Cardilini, A. P. A., & Hui, C. (2018). Context-dependent spatial sorting of dispersal-related traits in the invasive starlings (Sturnus vulgaris) of South Africa and Australia. *bioRxiv*, 342451. https://doi.org/10.1101/342451

Pipek, P., Blackburn, T. M., & Pyšek, P. (2019). The ins and outs of acclimatisation: Imports versus translocations of skylarks and starlings in 19th century New Zealand. *Biological Invasions*, *21*(4), 1395–1413. https://doi.org/10.1007/s10530-018-1905-y

Pipek, P., Pyšek, P., & Blackburn, T. M. (2015). How the Yellowhammer became a Kiwi: The history of an alien bird invasion revealed. *NeoBiota*, *24*, 1–31. https://doi.org/10.3897/neobiota.24.8611

Privy Council Selections. (1878). *Australian Town and Country Journal*, 23. https://trove.nla.gov.au/newspaper/article/70597849

Prokopenko, D., Hecker, J., Silverman, E. K., Pagano, M., Nöthen, M. M., Dina, C., Lange, C., & Fier, H. L. (2016). Utilizing the Jaccard index to reveal population stratification in sequencing data: A simulation study and an application to the 1000 Genomes Project. *Bioinformatics*, *32*(9), 1366–1372. https://doi.org/10.1093/bioinformatics/btv752

Quinlan, A. R., & Hall, I. M. (2010). BEDTools: A flexible suite of utilities for comparing genomic features. *Bioinformatics*, *26*(6), 841–842. https://doi.org/10.1093/bioinformatics/btq033

R Core Team. (2022). *R: A Language and Environment for Statistical Computing* [Computer software]. R Foundation for Statistical Computing. https://www.R-project.org/

Rintala, J., Tiainen, J., & Pakkala, T. (2003). Population trends of the Finnish starling Sturnus vulgaris, 1952—1998, as inferred from annual ringing totals. *Annales Zoologici Fennici*, *40*(4), 365–385. https://www.jstor.org/stable/23736554

Robinson, R. A., Siriwardena, G. M., & Crick, H. Q. P. (2005). Status and population trends of Starling Sturnus vulgaris in Great Britain. *Bird Study*, *52*(3), 252–260. https://doi.org/10.1080/00063650509461398

Rollins, L. A., Woolnough, A. P., Sinclair, R., Mooney, N. J., & Sherwin, W. B. (2011). Mitochondrial DNA offers unique insights into invasion history of the common starling. *Molecular Ecology*, *20*(11), 2307–2317. https://doi.org/10.1111/j.1365-294X.2011.05101.x

Rollins, L. A., Woolnough, A. P., Wilton, A. N., Sinclair, R., & Sherwin, W. B. (2009). Invasive species can't cover their tracks: Using microsatellites to assist management of starling (Sturnus vulgaris) populations in Western Australia. *Molecular Ecology*, *18*(8), 1560–1573. https://doi.org/10.1111/j.1365-294X.2009.04132.x

960 Ross, H. A. (1983). Genetic differentiation of starling (Sturnus vulgaris: Aves) populations in New
961 Zealand and Great Britain. *Journal of Zoology*, *201*(3), 351–362.
962 https://doi.org/10.1111/j.1469-7998.1983.tb04281.x
963 Ross, H. A., & Baker, A. J. (1982). Variation in the size and shape of introduced starlings, Sturnus-
964 vulgaris (Aves, Sturnidae), in New-Zealand. *Canadian Journal of Zoology*, *60*, 3316–3325.
965 Roy, H. E., Pauchard, A., Stoett, P., Renard Truong, T., Bacher, S., Galil, B. S., Hulme, P. E., Ikeda, T.,
966 Sankaran, K., McGeoch, M. A., Meyerson, L. A., Nuñez, M. A., Ordonez, A., Rahlao, S. J.,
967 Schwindt, E., Seebens, H., Sheppard, A. W., & Vandvik, V. (2024). *IPBES Invasive Alien Species*
968 *Assessment: Summary for Policymakers*. Zenodo. https://doi.org/10.5281/zenodo.10521002
969 Royall, W. C., Jr. (1966). Breeding of the Starling in Central Arizona. *The Condor*, *68*(2), 196–205.
970 https://doi.org/10.2307/1365718
971 Salloum, P. M., Santure, A. W., Lavery, S. D., & de Villemereuil, P. (2022). Finding the adaptive needles
972 in a population-structured haystack: A case study in a New Zealand mollusc. *Journal of*
973 *Animal Ecology*, *91*(6), 1209–1221. https://doi.org/10.1111/1365-2656.13692
974 Seebens, H., Bacher, S., Blackburn, T. M., Capinha, C., Dawson, W., Dullinger, S., Genovesi, P., Hulme,
975 P. E., van Kleunen, M., Kühn, I., Jeschke, J. M., Lenzner, B., Liebhold, A. M., Pattison, Z., Pergl,
976 J., Pyšek, P., Winter, M., & Essl, F. (2021). Projecting the continental accumulation of alien
977 species through to 2050. *Global Change Biology*, *27*(5), 970–982.
978 https://doi.org/10.1111/gcb.15333
979 Seebens, H., Blackburn, T. M., Dyer, E. E., Genovesi, P., Hulme, P. E., Jeschke, J. M., Pagad, S., Pyšek, P.,
980 Winter, M., Arianoutsou, M., Bacher, S., Blasius, B., Brundu, G., Capinha, C., Celesti-Grapow,
981 L., Dawson, W., Dullinger, S., Fuentes, N., Jäger, H., … Essl, F. (2017). No saturation in the
982 accumulation of alien species worldwide. *Nature Communications*, *8*(1), 14435.
983 https://doi.org/10.1038/ncomms14435
984 SHIPPING. (1886, September 27). *Daily Telegraph Launceston*, Page 2.
985 https://trove.nla.gov.au/newspaper/article/149532782

Smeds, L., Qvarnström, A., & Ellegren, H. (2016). Direct estimate of the rate of germline mutation in a bird. *Genome Research*, *26*(9), 1211–1218. https://doi.org/10.1101/gr.204669.116

STARLINGS. (1881, February 2). *Mercury*, 3.

Stuart, K. C., Cardilini, A. P. A., Cassey, P., Richardson, M. F., Sherwin, W. B., Rollins, L. A., & Sherman, C. D. H. (2021). Signatures of selection in a recent invasion reveal adaptive divergence in a highly vagile invasive species. *Molecular Ecology*, *30*(6), 1419–1434. https://doi.org/10.1111/mec.15601

Stuart, K. C., Edwards, R. J., Cheng, Y., Warren, W. C., Burt, D. W., Sherwin, W. B., Hofmeister, N. R., Werner, S. J., Ball, G. F., Bateson, M., Brandley, M. C., Buchanan, K. L., Cassey, P., Clayton, D. F., De Meyer, T., Meddle, S. L., & Rollins, L. A. (2022). Transcript- and annotation-guided genome assembly of the European starling. *Molecular Ecology Resources*, *22*(8), 3141–3160. https://doi.org/10.1111/1755-0998.13679

Stuart, K. C., Edwards, R. J., Sherwin, W. B., & Rollins, L. A. (2023). Contrasting Patterns of Single Nucleotide Polymorphisms and Structural Variation Across Multiple Invasions. *Molecular Biology and Evolution*, *40*(3), msad046. https://doi.org/10.1093/molbev/msad046

Stuart, K. C., Hofmeister, N. R., Zichello, J. M., & Rollins, L. A. (2023). Global invasion history and native decline of the common starling: Insights through genetics. *Biological Invasions*, *25*(5), 1291–1316. https://doi.org/10.1007/s10530-022-02982-5

Stuart, K. C., Johnson, R. N., Major, R. E., Atsawawaranunt, K., Ewart, K. M., Rollins, L. A., Santure, A. W., & Whibley, A. (2024). The genome of a globally invasive passerine, the common myna, Acridotheres tristis. *DNA Research*, *31*(2), dsae005. https://doi.org/10.1093/dnares/dsae005

Stuart, K. C., Sherwin, W. B., Austin, J. J., Bateson, M., Eens, M., Brandley, M. C., & Rollins, L. A. (2022). Historical museum samples enable the examination of divergent and parallel evolution during invasion. *Molecular Ecology*, *31*(6), 1836–1852. https://doi.org/10.1111/mec.16353

1011 Sullivan, B. L., Wood, C. L., Iliff, M. J., Bonney, R. E., Fink, D., & Kelling, S. (2009). eBird: A citizen-based
1012 bird observation network in the biological sciences. *Biological Conservation*, *142*(10), 2282–
1013 2292. https://doi.org/10.1016/j.biocon.2009.05.006
1014 THE ACCLIMATISATION SOCIETY. (1880). *Evening Star*, 2.
1015 https://paperspast.natlib.govt.nz/newspapers/ESD18800811.2.13
1016 The pandas development team. (2020). *pandas-dev/pandas: Pandas* [Computer software].
1017 https://doi.org/10.5281/zenodo.3509134
1018 THE Warragul Guardian, WITH WHICH IS INCORPORATED The Warragul News. (1898). *Warragul*
1019 *Guardian*, 2. https://trove.nla.gov.au/newspaper/article/67434906
1020 Thomson, G. M. (1922). *The naturalisation of animals & plants in New Zealand,* (pp. 1–628). The
1021 University Press,. https://doi.org/10.5962/bhl.title.28093
1022 Wang, I. J. (2013a). *Data from: Examining the full effects of landscape heterogeneity on spatial*
1023 *genetic variation: A multiple matrix regression approach for quantifying geographic and*
1024 *ecological isolation [Dataset]. Dryad*. https://doi.org/10.5061/dryad.kt71r
1025 Wang, I. J. (2013b). Examining the full effects of landscape heterogeneity on spatial genetic variation:
1026 A multiple matrix regression approach for quantifying geographic and ecological isolation.
1027 *Evolution*, *67*(12), 3403–3411. https://doi.org/10.1111/evo.12134
1028 Waterman, M., Fuller, C., & Murray, MD. (2008). Studies of roosting common starlings Sturnus
1029 vulgaris in South Australia. *Corella*, *32*, 25–29.
1030 Webster, MA. (1975). Hong Kong's trade in wildlife. *Biological Conservation*, *8*, 203–211.
1031 Wickham, H. (2016). *ggplot2: Elegant Graphics for Data Analysis*. Springer-Verlag New York.
1032 https://ggplot2.tidyverse.org
1033 Wilson, G. A., & Rannala, B. (2003). Bayesian inference of recent migration rates using multilocus
1034 genotypes. *Genetics*, *163*(3), 1177–1191. https://doi.org/10.1093/genetics/163.3.1177
1035 Wretenberg, J., Lindström, Å., Svensson, S., Thierfelder, T., & Pärt, T. (2006). Population trends of
1036 farmland birds in Sweden and England: Similar trends but different patterns of agricultural

intensification. *Journal of Applied Ecology*, *43*(6), 1110–1120. https://doi.org/10.1111/j.1365-2664.2006.01216.x

Zenni, R. D., & Hoban, S. M. (2015). Loci under selection during multiple range expansions of an invasive plant are mostly population specific, but patterns are associated with climate. *Molecular Ecology*, *24*(13), 3360–3371. https://doi.org/10.1111/mec.13234