

Malleefowl and anthropogenic change:
an integrated analysis of population trends, landscape
genetics and movement ecology



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Figure on previous page: Yalanda, August 2018. This male (DA1, left) was tracked for five seasons. The pair still lives in the same 107 ha patch of native vegetation in an agricultural matrix. Credit: Peri Stenhouse

Contents

CONTENTS	3
LIST OF FIGURES	5
LIST OF TABLES	7
LIST OF SUPPLEMENTARY FIGURES	9
LIST OF SUPPLEMENTARY TABLES	11
PUBLICATIONS ARISING FROM THIS PHD	15
ABSTRACT	17
THESIS DECLARATION	19
ACKNOWLEDGEMENTS	21
CHAPTER 1: GENERAL INTRODUCTION	25
1.1 BACKGROUND	27
1.2 SPECIES BACKGROUND	28
1.3 HISTORICAL IMPACTS.....	29
1.4 NEW EMERGING THREATS	31
1.5 KNOWLEDGE GAPS & AIMS.....	31
1.6 THESIS STRUCTURE	34
1.7 REFERENCES	36
CHAPTER 2: TRENDS IN BREEDING ACTIVITY OF THE THREATENED MALLEEFOWL (<i>LEIPOA OCELLATA</i>): WHAT CAN WE EXPECT UNDER A CHANGING CLIMATE?	45
2.1 STATEMENT OF AUTHORSHIP	47
2.2 ABSTRACT.....	49
2.3 KEYWORDS	49
2.4 INTRODUCTION	51
2.5 METHODS.....	52
2.6 RESULTS	56
2.7 DISCUSSION	59
2.8 MISCELLANEOUS	62
2.9 REFERENCES	64
2.10 APPENDIX	69
CHAPTER 3: SPATIAL GENETIC STRUCTURE AND LIMITED GENE FLOW IN FRAGMENTED POPULATIONS OF THE THREATENED MALLEEFOWL (<i>LEIPOA OCELLATA</i>)	79
3.1 STATEMENT OF AUTHORSHIP	81
3.2 ABSTRACT.....	83
3.3 KEYWORDS	83
3.4 INTRODUCTION	85
3.5 METHODS.....	87

3.6 RESULTS	92
3.7 DISCUSSION	96
3.8 MISCELLANEOUS	101
3.9 REFERENCES	103
3.10 APPENDIX	108
CHAPTER 4: PATCH SIZE AND BREEDING STATUS INFLUENCE MOVEMENT PATTERNS IN THE THREATENED MALLEEFOWL (<i>LEIPOA OCELLATA</i>)	119
4.1 STATEMENT OF AUTHORSHIP	121
4.2 ABSTRACT	123
4.3 KEYWORDS	123
4.4 INTRODUCTION	125
4.5 METHODS	127
4.6 RESULTS	130
4.7 DISCUSSION	141
4.8 MISCELLANEOUS	146
4.9 REFERENCES	148
4.10 APPENDIX	154
CHAPTER 5: MALLEEFOWL MOVEMENT AND FINE-SCALE HABITAT SELECTION DRIVEN BY CANOPY	167
5.1 STATEMENT OF AUTHORSHIP	169
5.2 INTRODUCTION	171
5.3 METHODS	173
5.4 RESULTS	177
5.5 DISCUSSION	184
5.6 REFERENCES	188
5.7 APPENDIX	192
CHAPTER 6: GENERAL DISCUSSION	213
6.1 TRENDS	215
6.2 LANDSCAPE GENETICS	215
6.3 MOVEMENT	216
6.4 HABITAT USE	217
6.5 LIMITATIONS	217
6.6 MANAGEMENT IMPLICATIONS	219
6.7 FUTURE DIRECTIONS	220
6.8 WIDER APPLICATION OF RESULTS	221
6.9 CONCLUSIONS	222
6.10 REFERENCES	224

List of Figures

- Figure 2-1 Locations of Malleefowl mound activity grids on the Eyre Peninsula. Green areas depict native vegetation cover. Spatial vegetation data (most likely landcover 2010-2015) were obtained from the South Australian Government Data Directory (Government of South Australia 2022). Map was made with QGIS v3.16 (QGIS Development Team 2021). 53
- Figure 2-2 Proportional mound activity of Malleefowl in the Eyre Peninsula grids between 1995 and 2018. The proportional mound activity is the number of active mounds divided by the number of monitored mounds..... 57
- Figure 2-3 Best model: Predicted probabilities of Activity ratio in a grid and, on the left, cumulative rainfall of the 2 years (24 months) prior to breeding, and, on the right, average maximum temperature in the winter months (June, July, August), 1-3 months prior to breeding. The shaded areas are the 95% confidence intervals for the predicted values..... 58
- Figure 2-4 Second (equivalent) model: Predicted probabilities of activity ratio in a grid and, on the left, the average SOI in the winter months (June, July, August) 2 years prior to breeding (i.e. 25-30 months before breeding), and, on the right: bare surface (no vegetation) in the winter, 1-3 months before breeding. The shaded areas are the 95% confidence intervals for the predicted values..... 58
- Figure 3-1 Malleefowl sample collection sites on and near the Eyre Peninsula, South Australia. The predominant colour of the pie-chart matches that of the clusters identified by the PCA. The proportions within each pie chart reflect the LEA plot ancestry composition of samples ($K = 2$). Pie-chart line and label colours indicate the sex of the individual. The light green areas are native vegetation, the darker green highlights are nature protection and wilderness areas under South Australian Government management. 87
- Figure 3-2 Principal Component Analysis of population genetic structure of 55 Malleefowl samples from the Eyre Peninsula, South Australia. The fill colours depict the genetic clusters identified in the PCA. Different outer colours show two samples (CO04 and CO09) where the collection location did not match the PCA cluster assignments. Shapes show the gender of the individual. Also, see PC3 and PC4 in Figure S 3-1..... 93
- Figure 3-3 Bar plots show individual genetic cluster assignment from LEA results from $K = 2$ and $K = 3$. Samples are grouped horizontally according to the clusters observed in the PCA (separated by black vertical lines). 94
- Figure 3-4 Correlograms showing spatial genetic structure in male and female Malleefowl on the Eyre Peninsula. Genetic correlation coefficient (r) is displayed with 95% confidence intervals (U = upper, L = lower) and error bars determined by bootstrapping..... 95
- Figure 4-1 Overview map of the Eyre Peninsula study sites. Darker green shows government conservation areas and light green native vegetation cover. Dashed lines indicate outer boundaries of Malleefowl movement ranges and correspond to sections in Figure 4-5. P.A. = Port Augusta, A. = Adelaide. 128
- Figure 4-2 Predicted home range (ha) values by patch size (ha) for Malleefowl on the Eyre Peninsula ($n = 7$) while breeding and non-breeding. The lines are the fitted multiple linear regression and shaded areas are the 95% confidence intervals. 134
- Figure 4-3 Daily movement statistics of Malleefowl on the Eyre Peninsula by sex and breeding status. A) daily distance (sum of distances between all consecutive fixes during a day), B) daily displacement

(distance between first and last fix of a day) and C) maximum distance away from the mound recorded per day..... 135

Figure 4-4 Hourly movement statistics of Malleefowl on the Eyre Peninsula by sex. A) hourly distance travelled by breeding status, B) distance away from the mound only while breeding. Note the different y-axis scales. 136

Figure 4-5 Examples of movement patterns recorded in the Eyre Peninsula Malleefowl (clockwise from top left): Dispersal of LOC7 in fragmented habitat (initially NB, then B); Dispersal of SR60 in continuous habitat (NB); Excursion outside of the home range in SR159 in continuous habitat (NB); home range in SR260 (NB); Sedentarism in DA1 over five years in fragmented habitat (B). *Symbology*: Light blue contours: NB 95% UD; green dashed contours: NB 95% MCP; orange triangles: mound where bird was caught; white arrows: dispersal distance and direction. LOC7 - Magenta contours & triangle: 95% UD and mound of the breeding season after dispersal; yellow dashed contours: B 95% MCP. SR159 - Dark blue dashed line: pest-proof enclosure. Edges of each map correspond to the dashed lines in Figure 1. 139

Figure 4-6 Linear mixed-effects model predictions of daily distance (m) travelled by Malleefowl on the Eyre Peninsula. The left panel shows the modelled estimates for the breeding period vs maximum temperature (°C). The right three panels show the estimates while non-breeding for maximum and minimum temperature and the presence or absence of rain. Data were analysed on a transformed scale (SI table 5), but effects are displayed on the scale of the response (m) for easier interpretation. The shaded areas are the 95% confidence intervals for the predicted values. The rug plot at the bottom shows the location of the explanatory values. 140

Figure 5-1 Overview map of the Eyre Peninsula Malleefowl study sites. Darker green shows government conservation areas and light green native vegetation cover. Dashed lines indicate the outer boundaries of Malleefowl movement ranges..... 173

Figure 5-2 Optimal classification trees for the habitat selection of three non-breeding Malleefowl on the Eyre Peninsula. The left plots show the best trees for all three strata combined, the middle plots for the combination of ground cover and midstorey, and the right plots for the canopy. The first row shows the data for all three birds combined, the remaining rows for each bird, as labelled. Each node shows the predicted (most likely) class (High/Low/Control) in the top row; the predicted probability of each of these three classes in the middle row, and the percentage of observations in that node in the bottom row. Blue colours indicate high use sites and green colours low use sites. Controls were never predicted. Stronger colours indicate higher probabilities of prediction. G = Ground cover, M = Midstorey, C = Canopy. Understorey is the combined data of ground cover and midstorey. 181

List of Tables

Table 2-1 Attributes of the Malleefowl nest activity monitoring grids on the Eyre Peninsula. Dominant canopy species based on Matthews (2002). Rain = Mean annual rainfall across all years in which monitoring took place [mm]; Species = Predominant canopy species; Size = Total grid size (Patch size) [km ²]; Mounds = Number of mounds known in 2018; Fires: Years in which part of the grid was burnt (number of mounds) Note: No mounds were burnt twice.	54
Table 3-1 Cluster name, size after filtering (n), observed (H _o) and expected (H _E) heterozygosity (both values range from 0 to 1, with 0 meaning no diversity) and the inbreeding coefficient (F _{is} , range 0 to 1 with 0 meaning no inbreeding). Results are presented with one standard deviation in brackets.	94
Table 4-1 Overview of tracked Malleefowl on the Eyre Peninsula. We present the trapping location, bird-ID, tracking period and duration, cause of death (fate) and number (percentage) of successful day and night-time fixes for each bird. F = Female. ¹ Cat DNA present. ² DNA Failed to amplify. ³ Chewed feather ends. ⁴ Cut feather ends. ⁵ Tooth mark on transmitter. ⁶ Feathers plucked. ⁷ Kill cached. ⁸ Feathers scattered	131
Table 4-2 Summary of movement metrics of Malleefowl on the Eyre Peninsula. We present sex (F = Female, M = Male); breeding stage (B = Breeding, NB = Non-breeding (number of stages used for analyses); home range (95% UD, ha); total range length (km); and mean ± 1 s.d. and maximum of daily distance, daily displacement, distance to mound and hourly movement (m). Maxima for each sex are in bold. Details in Table S 4-3.	133
Table 5-1 Overview of species and genera found in each visit frequency category	177
Table 5-2 Top 30% genera and species by cover (average cover per bird, height and visit frequency in %) found in the high use day sites and how much the average cover of the genera/species was in the low day and control sites. H = Height, G = Ground cover, M = Midstorey, C = Canopy. Genera that were included in the top models are highlighted in bold font.	178
Table 5-3 Variables that predicted high site use among Malleefowl on the Eyre Peninsula, using rpart classification tree models. G = Ground cover, M = Midstorey, C = Canopy. Understorey is the combined data of ground cover and midstorey. We also present variance explained, prediction accuracy and misclassification rate in %. CI = 95% Confidence intervals for hundred repetitions.	180

List of Supplementary Figures

- Figure S 2-1 Estimated marginal means, i.e. probabilities of year-to-year mound activity and grid-to-grid differences shown for the period from 2003 – 2018 for Malleefowl monitoring grids in Cowell, Hincks, Munyaroo and Lock. Left: Year to year variation. Right: Grid to grid variation. Error bars show the 95% confidence intervals. 2018 had to be removed for scale reasons. 75
- Figure S 2-2 Estimated marginal means, i.e. probabilities of year-to-year and grid-to-grid differences shown for the period from 2010 – 2018 for Malleefowl monitoring grids in Cowell, Hincks, Munyaroo, Lock and Secret Rocks. Left: Year to year variation. Right: Grid to grid variation. Error bars show the 95% confidence intervals. 2018 had to be removed for scale reasons. 75
- Figure S 3-1 Principal Component Analysis (Axes PC3 and PC4) of population genetic structure of 55 Malleefowl samples from the Eyre Peninsula, South Australia. The fill colours depict the genetic clusters identified in the PCA. Different outer colours show two samples (CO04 and CO09) where collection location did not match the PCA cluster assignments. Shapes show the gender of the individual. 111
- Figure S 3-2 Isolation-by-distance scatterplots for all birds, $K = 2$ and $K = 3$. The x-axis shows the natural logarithm of the geographic distance after Rousset (2000). The y-axis shows the pairwise genetic distance calculated as a percentage. 112
- Figure S 3-3 LEA cross-entropy results for Malleefowl on the Eyre Peninsula, South Australia. The cross-entropy validation errors are plotted for K values of 1 to 10. The lowest error indicates the greatest support at $K = 2$ 113
- Figure S 3-4 Malleefowl sample collection sites on and near the Eyre Peninsula, South Australia. The predominant colour of the pie-chart matches that of the clusters identified by the PCA. The proportions within each pie chart reflect the LEA plot ancestry composition of samples ($K = 3$). Pie-chart line and label colours indicate the sex of the individual. The light green areas are native vegetation, the darker green highlights are nature protection and wilderness areas under South Australian Government management. 114
- Figure S 3-5 Visualisation of the relationship between ancestry proportions and native vegetation cover of Malleefowl on the Eyre Peninsula, South Australia. Each plot shows the proportion of the named ancestry proportion on the y-axis and the % native vegetation cover on the x-axis. The colours indicate the cluster assignments as calculated by the PCA. Three samples (SR15, CO04 and CO09) where collection location did not match the PCA cluster assignments were excluded. 115
- Figure S 3-6 Probabilities of three sample type groups being successfully genotyped for the Eyre Peninsula Malleefowl. Pink bars are the confidence intervals. 117
- Figure S 4-1 Predicted total range (km) values by patch size (ha) for Malleefowl on the Eyre Peninsula ($n = 7$) while breeding (blue) and non-breeding (red). The shaded areas are the 95% confidence intervals for the fitted values. 159
- Figure S 4-2 Timeline of GPS tracked Malleefowl on the Eyre Peninsula. The coloured vertical bars show the daily total distance moved (i.e. sum of all distances between fixes per day) and the black bars in front show the daily displacement (i.e. distance between the first and last fix of a bird day). The horizontal line shows the long-distance movement threshold of $4 \times$ mean daily displacement of all birds (4×502 m). 160
- Figure S 4-3 Long-range movements of SR60 between April and November 2020 (NB). The white arrow depicts the length and direction of resettlement. The arrows are colour coded by season, with

autumn brown to purple, winter in blues and spring in greens. The orange triangle shows the mound at which the bird was captured, the blue contours the 95% UD, the grey contours the 50% UD and the green dashed line the 95% MCP. Compare excursion numbers to Table S 4-6. 163

Figure S 4-4 Long-range movements of SR260 between December 2017 and February 2019 (NB). The first three excursions occurred when this male was tending the mound by himself. The excursion arrows are colour coded by season, with summer in yellow to red, winter in blues and spring in greens. The orange triangle shows the mound at which the bird was captured, the blue contours the 95% UD, the grey contours the 50% UD and the green dashed line the 95% MCP. Compare excursion numbers to Table S 4-6..... 164

List of Supplementary Tables

Table S 2-1 An overview of grid activity of all Eyre Peninsula grids. 'Total' is the total number of mounds; 'Checked' is the number of monitored mounds per grid; 'Not checked' is the number of mounds not monitored that year; 'Active' is the number of active mounds per grid; and 'Ratio' is the proportional mound activity which is calculated as number of active mounds divided by number of monitored mounds.....	69
Table S 2-2 An overview of Malleefowl mound numbers and densities in all Eyre Peninsula grids. 'Total' is the total number of mounds per grid; 'Active' is the number of active mounds per grid. The next two columns show the number of mounds and active mounds per square km.	70
Table S 2-3 All environmental and habitat variables tested in the models and how they were calculated. Description: times are in relation to September of that year when breeding (egg laying) has commenced. Calc: calculation, cu = cumulative, ma = monthly average; Trans: transformation, lc = log-centred, sc = scaled and centred, snc = scaled not centred; na: not applicable; All rainfall was measured in mm, temperatures in °C and distances in km.	73
Table S 2-4 Estimated marginal means, i.e. probabilities of year-to-year and grid-to-grid mound activity differences. The first two rows show the model formulas and remaining rows the modelling results for the 2003-2018 period for 4 grids on the left and the 2010-2018 period for 5 grids on the right. Year-to-year models are on the left and grid-to-grid on the right within each period. Odds: odds ratio, CI: confidence intervals, p: p-value (significant results in bold font).	74
Table S 2-5 Model outputs with $\Delta AICc \leq 4$ (5 grids, 2010-2018, n = 45). AICc = corrected Akaike Information Criterion; R^2_m / R^2_c = Marginal / Conditional r-squared (%); SE = Standard Error; LCI/UCI = Lower/Upper confidence intervals; P = P-value.....	75
Table S 3-1 Sequenced samples Eyre Peninsula Malleefowl. We present sample ID, collection date, location (Hamb = Hambidge CP, Mara = Maralinga, Muny = Munyaroo CP, SR/Iron = Secret Rocks/Ironstone NP, Yala = Yalanda, Yella = Yellabinna), sex (S = sex, F = female, M = male, U = unknown), sample type (Toe = toepad, Mus = muscle, Liv = liver, Yolk = yolk sac, Fea = feather, Egg= egg membrane, Blo = blood), storage method (Sto = Storage, ta = taxidermied, fr = frozen (-80°C if from museum, -20°C for others), ro = room temperature, bu = lysis buffer, et = 100% EtOH), origin of sample (Sou = Source, SAM = South Australia Museum, PS = Author, Gov = DEW or NRM, Misc = Miscellaneous, AM = Australian Museum), collection event (Field = found during field work or monitoring, Trap = from captured bird at active mound, Chick = chick hatched while author at mound and feather sample collected, Active = from active mound), whether sample was from immediate proximity of the mound (M = Mound), and reason why sample wasn't used for analysis (Rem = Removed, Kin = kinship >0.3, Trans = four Malleefowl from Lock were translocated to Lincoln NP in 2001, making the heritage of this bird unclear), DNA = DNA absent or contaminated, Data = >50% missing data). Dup = Duplicate sample(s).	108
Table S 3-2 Overview of sample type and storage methods for Eyre Peninsula Malleefowl samples. We present DNA quality (good (bad)= sufficient (insufficient) for SNP genotyping), sample types (Toe = toepad, Yolk = yolk sac, Egg m.= egg membrane), storage method (Storage, bu = lysis buffer, fr = frozen (-80°C if from museum, -20°C for others), ro = room temperature, ta = taxidermy, et = 100% EtOH), age classes (1 = under 5, 2 = 5 - 10; 3 = 11 -20; 4 = over 20 years) and number of samples of that type (Count). The table is sorted by most successful sample type.....	116
Table S 3-3 Parameter estimates of the generalised linear model investigating the relationship between sample type, storage and age on DNA usability. SE = Standard Error; z = z-value; LCI/UCI	

= Lower/Upper confidence intervals; p = P-value. We also present the Hosmer and Lemeshow goodness of fit test results (p <0.05 means data is not a good fit)..... 117

Table S 4-1 Attributes of the study sites on the Eyre Peninsula. Predominant vegetation types (Berkinshaw and McGregor 2010; Matthews 2002; Wilderness Advisory Committee 2014). Rain = Mean annual rainfall from the nearest BOM weather station for 2016 to 2020 (mm); Columns 3 and 4 show the predominant plant species in the upper and lower vegetation levels, respectively. 154

Table S 4-2 Movement after capture of the Eyre Peninsula Malleefowl in the initial season of capture. We present the trapping date; the date on which the tagged bird returned to the mound (d: number of days between trapping and return); date until which the mate continued to maintain the mound in the absence of the tagged bird (d: days); date on which breeding activities ceased; whether the breeding season was completed. ¹Tagged bird killed. ²Tagged bird tending mound without mate. 157

Table S 4-3 Basic movement statistics of Malleefowl on the Eyre Peninsula. We present the years for the bird observed over multiple years and the different patches used by the birds that resettled (Period), the breeding status (B), the days tracked (Days), the patch size (Patch; ha), the home range (MCP = 95% MCP, HR = 95% UD; ha), the total range length (TRL; km), and the mean ± 1 s.d. and the maximum of the daily total distance, the daily displacement, the distance to mound and the hourly movement in meters. B = Breeding, NB = Non-Breeding; F = Female; S = Single Malleefowl tending mound without its mate; ° = were not used for analyses due to short tracking time; * = MCP bootstraps did not converge; DA has 5 periods because of missing fixes in winter, Lock has 3 patches with a total range length of 7.8 km over the whole tracking period, SR60 has 2 patches with a total range length of 23.1 km for the whole tracking period..... 158

Table S 4-4 Model summary showing the effect of breeding status and patch size on home range (UD95) and total range (km) for Malleefowl on the Eyre Peninsula. We present model formulas, marginal R² values (in%), terms used, model estimates, lower and upper confidence levels, standard errors (SE) and p-values. Significance is indicated in bold. 159

Table S 4-5 Comparative statistics of distance metrics. Comparisons are between breeding (B) and non-breeding (NB) periods; between the female and the males while breeding (Sex - B) and non-breeding (Sex - NB); between seasons (spring, summer, autumn and winter) and between the times of day (dawn, day, dusk and night). We present the effect size (Sullivan and Feinn 2012) with lower and upper confidence interval limits (r_{tb} = rank biserial correlation, W_k = Kendall's coefficient of concordance). Significant p-values in bold. n = number of records. Please note there was only one female tracked! 161

Table S 4-6 Long-range movement with daily displacement greater than four times the mean displacement of all birds (4*502 m). We present the breeding status (B = Breeding, NB = Non-breeding, S = Single), the date, the type of movement (Type, O = outside of home range, I = inside of home range, R = resettlement), the excursion number (Also see Fig. 3 and 4 for SR260 and SR60) and their length in days. Bold font indicates the maxima for each bird. 162

Table S 4-7 Model summaries showing the effect of environmental features on daily and hourly movement. R²m = Marginal r-squared (in %); R²c = Conditional r-squared (in %); Est= Estimate, LCI/UCI = Upper/Lower Confidence Interval; SE = Standard Error; DF = degrees of freedom, All models (lme) have random = ~1|idSeason, correlation = corCAR1(form = ~ timeSeq | idSeason). Significant values in bold. 165

Table S 5-1 Plants consumed by Malleefowl (modified after Reichelt and Jones 2008). Header row: O = observation, C & G = crop and gizzard content, F = faecal matter, P = palatability study. Plant

parts consumed: F = flowers, f = foliage, b = buds, p = pods, berries, other fruit, s = seeds, t = tubers.
..... 192

Table S 5-2 Predominant plant communities in the home ranges three tracked Malleefowl on the Eyre Peninsula. Table adapted from Brandl, McDonald *et al.* (2009). Rainfall is the cumulative rainfall for the years the birds were tracked (BOM 2022). We used QGIS (QGIS Development Team 2021) to plot sites and report the intersecting Native Vegetation Floristic Areas sourced from the Government of South Australia (2022)..... 194

Table S 5-3 List of all plant species recorded at all sites independently of visit frequency. Species are listed by height class. Count is the number of locations at which a species was found, and n is the total number of day sites surveyed (including control sites). S = Stratum. 195

Table S 5-4 Top 30% most common genera and species recorded as present at high use day sites (i.e. number of sites found at) and their percent occurrence at high, low and control sites. S = Stratum, G = Ground cover, M = Midstorey, C = Canopy..... 199

Table S 5-5 Summary statistics of explanatory variables (Structure, diversity and visits). We present the percentage cover mean with 1 standard deviation (mean), the median (med), the interquartile range (IQR) and the percentage cover range (range). Rows with *All* visits show statistics of the complete dataset with all visits taken into account. Rows with *High/Low/Ctrl* visits show the statistics of the reduced subset of the data with high (top 25% visited) and low use (bottom 25% visited) sites as well as controls (not visited) which were used for the decision tree models. V = Visits, A = All, H/L = High/Low use, C = Control, G = Ground cover, M = Midstorey, C = Canopy 201

Table S 5-6 Summary statistics of explanatory variables (Ground cover). We present the percentage cover mean with 1 standard deviation (mean), the median (med), the interquartile range (IQR) and the percentage cover range (range). Rows with *All* visits show statistics of the complete dataset with all visits taken into account. Rows with *High/Low/Ctrl* visits show the statistics of the reduced subset of the data with high (top 25% visited) and low use (bottom 25% visited) sites as well as controls (not visited) which were used for the decision tree models. V = Visits, A = All, H/L = High/Low use, C = Control, G = Ground cover, M = Midstorey, C = Canopy 203

Table S 5-7 Summary statistics of explanatory variables (Midstorey). We present the percentage cover mean with 1 standard deviation (mean), the median (med), the interquartile range (IQR) and the percentage cover range (range). Rows with *All* visits show statistics of the complete dataset with all visits taken into account. Rows with *High/Low/Ctrl* visits show the statistics of the reduced subset of the data with high (top 25% visited) and low use (bottom 25% visited) sites as well as controls (not visited) which were used for the decision tree models. V = Visits, A = All, H/L = High/Low use, C = Control, G = Ground cover, M = Midstorey, C = Canopy 208

Table S 5-8 Summary statistics of explanatory variables (Canopy). We present the percentage cover mean with 1 standard deviation (mean), the median (med), the interquartile range (IQR) and the percentage cover range (range). Rows with *All* visits show statistics of the complete dataset with all visits taken into account. Rows with *High/Low/Ctrl* visits show the statistics of the reduced subset of the data with high (top 25% visited) and low use (bottom 25% visited) sites as well as controls (not visited) which were used for the decision tree models. V = Visits, A = All, H/L = High/Low use, C = Control, G = Ground cover, M = Midstorey, C = Canopy 210

Publications arising from this PhD

Journal articles

Stenhouse, P. ¹ & Moseby, K. E. ¹²³ (2022). Trends in breeding activity of the threatened Malleefowl (*Leipoa ocellata*): what can we expect under a changing climate? *Emu - Austral Ornithology*. <https://doi.org/10.1080/01584197.2022.2045870>

Stenhouse, P. ¹, Onley, I. R. ¹, Mitchell, K. J. ⁴⁵, Moseby, K. E. ¹²³ & Austin, J. J. ⁴ (2022) Spatial genetic structure and limited gene flow in fragmented populations of the threatened Malleefowl (*Leipoa ocellata*). *Ecological Genetics and Genomics*. <https://doi.org/10.1016/j.egg.2022.100127>

Stenhouse, P. ¹ & Moseby, K. E. ¹²³ (2022) Patch size and breeding status influence movement patterns in the threatened Malleefowl (*Leipoa ocellata*). Submitted to *Austral Ecology*.

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Conference proceedings

Stenhouse, P., & Moseby, K. E. (2018). Movement patterns of Malleefowl on the Eyre Peninsula. In National Malleefowl Forum Proceedings 2018

Conference posters

Stenhouse, P. Malleefowl (*Leipoa ocellata*) in a Changing Climate: Can we help a threatened species deal with a dryer and hotter environment? In International Ornithological Conference 2018, Vancouver, Canada.

Stenhouse, P. Long term population trends and short term movement patterns of Malleefowl (*Leipoa ocellata*). In Ecological Society of Australia Conference 2018, Brisbane, Australia.

Presentations

Moseby, K. E. & Stenhouse, P. (August 2018). Long term population trends and shorter term movement patterns of Malleefowl (*Leipoa ocellata*) on the Eyre Peninsula. In National Malleefowl Forum in Mildura, Victoria, Australia.

Stenhouse, P. (February 2021). Malleefowl in a Changing Climate. In Field Naturalists' Society of SA, General Meeting

Stenhouse, P. (July 2022). Malleefowl and anthropogenic change. Department of Agriculture, Water and Environment (Canberra) Community of Practice.

Abstract

Malleefowl are iconic Australian birds that build large mounds to incubate their eggs using external sources of heat. Malleefowl numbers have drastically decreased since European settlement and they are now a nationally threatened species. Their decline is caused by habitat loss and fragmentation; predation by introduced species; inappropriate fire regimes and competition and habitat degradation through overabundant native and introduced herbivores. Climate change is expected to exacerbate these pressures by leading to more frequent and longer periods of high temperatures, reduced precipitation and increased frequency and intensity of wildfires.

This thesis aimed to investigate how historic and recent habitat fragmentation and climatic variables influenced Malleefowl on the Eyre Peninsula and how climate change may exacerbate future impacts. The objectives were to assess population trends, investigate landscape genetics, study movement patterns and the influence of habitat variables.

These objectives were addressed through a multidisciplinary approach to study discrete Malleefowl populations at study sites located on the Eyre Peninsula in South Australia. To assess population trends and the influence of environmental factors, 24 years of mound-based breeding activity data were modelled. Results showed a widespread and significant decline in Malleefowl breeding activity during the study period, driven by changes in vegetation cover and reduction in soil moisture.

High throughput DNA sequencing was used to investigate past dispersal patterns and the effects of recent anthropogenic land cover changes. The results indicate that Malleefowl population structure on the Eyre Peninsula was not homogenous and that habitat fragmentation with subsequent isolation has caused the differentiation of at least two distinct populations. Further, there was preliminary genetic evidence of female dispersal.

Solar-powered GPS trackers were used to determine individual movement patterns and the effects of habitat fragmentation and environmental factors. Malleefowl movement was tightly associated with breeding status, with breeding birds staying close to the mound and non-breeding birds disassociating from the mound and moving long distances. Movement was also influenced by patch size and fragmentation, with non-breeding birds in large patches moving further than birds in smaller patches, but Malleefowl were able to persist in small patches and breed successfully for many years. Malleefowl moved less with increasing temperatures. Malleefowl did not use cropping land and they avoided crossing open paddocks. Malleefowl also had high mortality rates with over 66% of tracked individuals dying within a year of trapping, mostly from cat and fox predation.

Vegetation surveys were combined with GPS tracking data to investigate whether Malleefowl movement was influenced by vegetation composition or cover. Results indicate that Malleefowl seek out micropatches of tall mallee stands within a mature *Eucalyptus* matrix – likely as refuges from heat and predators – and that movement is less driven by a preference for plant species, reflecting the fact that Malleefowl have a highly variable and opportunistic diet.

In summary, this thesis shows how anthropogenic habitat alteration and changing environmental conditions have reduced breeding activity, restricted movement, dispersal and gene flow of Malleefowl populations on the Eyre Peninsula, with noticeable long-term genetic effects. Further, we can expect climate change to exacerbate all existing pressures and possibly drive Malleefowl closer to extinction. This research provides valuable new information about Malleefowl movement ecology which supports increased efforts to protect even small patches of native vegetation in agricultural matrices and the creation of habitat corridors between patches, as well as the improvements of matrix habitat, to facilitate between-patch movement. This, combined with continued and improved predator-control efforts, may enhance the conditions for future survival of Malleefowl populations, and support biodiversity in general, which is vital when faced with increasing climate change pressures.

Thesis declaration

I certify that this work contains no material which has been accepted for the award of any other degree or diploma in my name, in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text. In addition, I certify that no part of this work will, in the future, be used in a submission in my name, for any other degree or diploma in any university or other tertiary institution without the prior approval of the University of Adelaide and where applicable, any partner institution responsible for the joint award of this degree.

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Adelaide, 4 April 2022

Peri Stenhouse

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Chapter 1:

General Introduction



Figure on previous page: Yalanda, August 2018. A Malleefowl pair greeting each other. The male (DA1, left) was tracked for over five seasons. The pair still lives in the same 107 ha patch of native vegetation in an agricultural matrix. Credit: Peri Stenhouse

1.1 Background

Global biodiversity is in crisis and nearly one million species worldwide are in danger of extinction (Bergstrom, Wienecke *et al.* 2021; Díaz, Settele *et al.* 2019). Australia has one of the highest extinction rates in the world (Woinarski, Burbidge *et al.* 2015; Woinarski, Garnett *et al.* 2017) and many more species are listed as threatened and expected to go extinct in the near future (Geyle, Woinarski *et al.* 2018). There are multiple, interacting reasons for this decline (Doherty, Dickman *et al.* 2015), such as predation by introduced predators (Woinarski, Burbidge *et al.* 2015), competition with overabundant herbivores (introduced and native) (Foster, Barton *et al.* 2014; Kuiper and Parker 2013), altered fire regimes (Santos, Hradsky *et al.*) and the degradation and clearing of native habitat that has continued since European colonisation (Haddad, Brudvig *et al.* 2015).

Many terrestrial vertebrates persist in human-modified landscapes where much of the original habitat has been cleared and fragments of habitat are scattered within an agricultural matrix (Tulloch, Barnes *et al.* 2016). While there is no consensus about whether habitat fragmentation 'per se' (i.e. the habitat amount remains the same with increased patchiness) is beneficial or detrimental to biodiversity (Fahrig 2017; Fahrig, Arroyo-Rodríguez *et al.* 2019; Fletcher, Didham *et al.* 2018; Haddad, Brudvig *et al.* 2015), it is generally agreed that habitat loss and degradation is detrimental to species survival (Fahrig 2003; Legge, Kennedy *et al.* 2011; Lindenmayer and Fischer 2006; McGregor, Legge *et al.* 2014). Negative effects of habitat loss on wildlife can be as simple as the lack of access to suitable habitat and associated resources, but there are indirect effects too which are influenced by species-dependent factors (e.g. mobility or dietary flexibility) and interactions with other threatening processes (e.g. predation or wildfires) that may play a role in the spatial and temporal extent of these effects (Lindenmayer and Fischer 2006). Some impacts, such as local extinctions caused by isolation, genetic drift and inbreeding may only become apparent after several generations (Stevens, Harrison *et al.* 2018).

Climate change is expected to exacerbate existing effects (Chambers, Hughes *et al.* 2005; Mac Nally, Bennett *et al.* 2009; Warren 2006) and widely impact the physiology, morphology, phenology and genetic health of species (Parmesan and Yohe 2003; Scheffers, De Meester *et al.* 2016; Thackeray, Henrys *et al.* 2016). Impacts include temporal shifts in breeding patterns (Visser, Both *et al.* 2004), range shifts (Chambers, Hughes *et al.* 2005; Hughes 2011) or range contractions (Runge, Tulloch *et al.* 2015). Range shifts can lead to a spatial (Schweiger, Heikkinen *et al.* 2012) or temporal mismatch (Pelini, Prior *et al.* 2009) between interacting species. Further, an increased need to thermoregulate in hotter conditions leads to decreased movement (Tatler, Currie *et al.* 2021) and worse body condition (du Plessis, Martin *et al.* 2012). Climate change has already caused global

extinctions and is expected to cause more in the future (Conradie, Woodborne *et al.* 2019; Yalcin and Leroux 2018).

1.2 Species Background

The family Megapodiidae consists of 21 extant species distributed throughout the Australasian region (www.iucnredlist.org). All megapodes use external sources of heat, rather than their body heat, to incubate their eggs. External heat sources vary from warm beach sand to specially built incubation chambers that make use of the heat produced by decaying vegetation. There are three Australian megapode species, and while all are mound builders, their habitats and distribution differ significantly. The Orange-footed Scrubfowl (*Megapodius reinwardt*) is found in the tropical north, while the Australian Brush-turkey (*Alectura lathamii*) can be found all along the east coast of Australia ranging from temperate to tropical climates. Both are classified by the IUCN as of least concern. My study species, the Malleefowl (*Leipoa ocellata*) lives in semi-arid to arid regions of southern Australia.

Malleefowl are large (c. 60 cm, 1.5 - 2.5 kg) ground-dwelling birds that have evolved the most complex breeding system within the megapode family. Breeding pairs build incubation mounds from sand and dirt that can be up to 6 m in diameter and up to 1.5 m high (Benshemesh 1992a; Booth 1985; Frith 1959). Breeding activities can continue up to 11 months a year (Frith 1959) and mound maintenance can take up to 7 hours each day (Neilly, Wells *et al.* 2021a; Weathers and Seymour 1998). Winter rain is required to compost the leaf litter and if rain is insufficient, breeding attempts are aborted for that season (Booth and Seymour 1983; Frith 1959). If rain is sufficient the birds build an incubation chamber within the mound and begin laying eggs around September. The Malleefowl can regulate the mound temperature to approximately $34 \pm 4^{\circ}\text{C}$ using the heat from the sun and composting litter, and superprecocial chicks hatch after about 62 days, with warmer temperatures in the incubation chamber resulting in shorter incubation periods (Booth 1987; Frith 1959). Females lay one large egg that is around 10% of their body weight approximately every 6 days (Marchant and Higgins 1993). Clutch size varies depending on conditions but is usually around 20 eggs (Marchant and Higgins 1993). Malleefowl were thought to be behaviourally monogamous (Frith 1959; Frith 1962b; Priddel and Wheeler 2003), however extra-pair paternity and polygyny (male breeding with two females at two different mounds simultaneously) have also been observed (Cope, Mulder *et al.* 2014; Weathers, Weathers *et al.* 1990). Further, Cope, Mulder *et al.* (2014) reported egg-dumping by unrelated females.

Malleefowl are an iconic Australian bird species that inhabit mallee woodlands and shrublands in semi-arid to arid regions, where the danger of wildfires is high. Because they clear leaf litter around their nesting mounds, they reduce the fuel load and thus the intensity and likelihood of fires in that area (Smith, Avitabile *et al.* 2017). They can turn over up to 3 tonnes of soil every day and improve soil quality through their composting activities (Weathers and Seymour 1998). Through these actions, they provide habitat for a variety of other fauna and are potentially important ecosystem engineers (Catterall 2018; Neilly, Wells *et al.* 2021b). Like other ecosystem engineers in semi-arid environments, such as fossorial marsupials - bilbies, wombats, bettongs - studying the ecology of Malleefowl in anthropogenically modified landscapes and under climate change impacts is critical to the conservation of Malleefowl and the habitats and ecosystems that they live in.

1.3 Historical impacts

Malleefowl numbers and their range have declined greatly, at least since European settlement, and their numbers are continuing to decline (Benshemesh 2007; Benshemesh, Southwell *et al.* 2020). They are now recognised as threatened in all the states in which they occur (South Australia: Vulnerable; Victoria & New South Wales: endangered; Northern Territory: Critically Endangered; Western Australia: Fauna that is rare or is likely to become extinct). Malleefowl are also listed as Vulnerable with a declining trend on the IUCN Red List of endangered animals with an estimated number of 100,000 mature individuals across Australia (IUCN 2012, criteria VU A1c,e and A2b,c,e). The main reason for the decline is habitat loss (Benshemesh 2007; Parsons, Short *et al.* 2008; Woinarski 1989), followed by predation, overabundant herbivores and road kills (Benshemesh 2004; Benshemesh, Barker *et al.* 2007; Priddel and Wheeler 1994; Priddel, Wheeler *et al.* 2007; Wheeler and Priddel 2009).

Since European settlement, approximately 57% of the Australian landscape has been modified for agricultural and pastoral use (Thackway 2018). The primary issues that arise from land clearing are habitat loss and fragmentation. Since 1750, up to 70% of many Australian vegetation communities that would have supported Malleefowl in the past (e.g., various mallee woodland communities and other woodlands and shrublands) have been cleared and have become more fragmented, leading to less suitable habitat split into smaller patches (Tulloch, Barnes *et al.* 2016). Habitat fragmentation also changes fire regimes, with smaller patches experiencing less frequent fires than larger patches of continuous vegetation, and senescing, long-unburnt vegetation in these small patches leading to declining habitat quality that is unsuitable for Malleefowl (Parsons and Gosper 2011).

Wildfires can destroy Malleefowl habitat, with the extent of the damage and speed of recovery depending on the intensity and scale of the fire (Benshemesh 1992b; Gill, Woinarski *et al.* 1999). Malleefowl breeding densities (Benshemesh 1992a) and the number of birds (Woinarski 1989) were greatest in habitats that were unburnt for over 40 years. Modelling revealed a positive relationship between breeding activity and time since the last fire and a negative relationship between breeding activity and the proportion of a site burnt (Benshemesh, Southwell *et al.* 2020), but also that Malleefowl strongly prefer areas not burnt in the last 20 years (Clarke 2005). Large and intense wildfires, in all likelihood, wipe out most of the exposed Malleefowl (Benshemesh 2007), and the earliest records of breeding are 17 years after large fires (Tarr 1965). Small patchy burns, on the other hand, may continue to support Malleefowl that use burnt sites to forage and unburnt areas to nest, rest and roost (Benshemesh 1990; Benshemesh 1992a), and nesting in these areas have been observed as early as 6 years after a fire (Benshemesh 2007). It has even been argued that small patchy burns may benefit Malleefowl due to the post-burn recruitment of food plants such as *Acacia* (Brickhill 1987).

Intense grazing by introduced species such as rabbits (*Oryctolagus cuniculus*) and feral goats (*Capra hircus*) or overabundant native species (kangaroos) leads to habitat degradation through loss of understorey and reduced recruitment of vegetation. These species can compete with Malleefowl for food resources and reduce shelter sites for Malleefowl (Benshemesh 2007; Hauser, Southwell *et al.* 2019; Lethbridge 2016). Introduced predators, such as the cat (*Felis catus*) and the red fox (*Vulpes vulpes*), are more abundant (Hradsky, Mildwaters *et al.* 2017; Stobo-Wilson, Stokeld *et al.* 2020) and hunt more successfully (McGregor, Legge *et al.* 2015) in landscapes with reduced understorey cover and have significantly contributed to the extinction of native Australian fauna (Hamer, Gardiner *et al.* 2021; May and Norton 1996). Malleefowl have also suffered high predation by foxes in both disturbed and undisturbed habitats (Priddel, Wheeler *et al.* 2007; Wheeler and Priddel 2009).

Malleefowl now persist in marginal habitats with lower rainfall that support lower densities than in the past (Brickhill 1987; Frith 1962b). Biodiversity loss is closely associated with habitat loss, fragmentation (Fahrig 2003; Fletcher, Didham *et al.* 2018; Haddad, Brudvig *et al.* 2015) and disturbance (Hobbs and Huenneke 1992). This is because habitat loss and fragmentation can split animal populations into smaller and more isolated subpopulations and may result in genetic drift, inbreeding and even local patch extinctions with dire consequences for the overall population in the long term (Ralls, Ballou *et al.* 2018; Stevens, Harrison *et al.* 2018). While the rate of land clearing has decreased since the 1990s, because Malleefowl are long-lived birds and genetic factors contributing to extinction can

take multiple generations to impact, the effects of existing fragmentation can be delayed and have long-term consequences unless appropriate conservation action is taken (Frankham 2005; McAlpine, Syktus *et al.* 2009).

1.4 New emerging threats

Climate change is expected to exacerbate the issues affecting Malleefowl, as weather patterns are projected to change. In the southern parts of Australia, more frequent and prolonged hot and dry periods and more frequent wildfires are predicted for the next decades (BOM and CSIRO 2020; CSIRO and BOM 2015; Williams, Bradstock *et al.* 2009). Higher ambient temperatures and less rainfall affect habitat by decreasing soil moisture which leads to reduced plant growth, flowering, seeding and recruitment (Guerin, Christmas *et al.* 2018; Scheffers, De Meester *et al.* 2016).

Equally importantly, hotter conditions can lead to changes in breeding (Parmesan and Yohe 2003; Visser, Both *et al.* 2004), feeding (Gils, Lisovski *et al.* 2016) and migration (Gienapp, Leimu *et al.* 2007; Travers, Marquardt *et al.* 2015) behaviour in birds, as well as affecting their physiology and morphology (McCoy 2012; McKechnie 2019). Heat dissipation and avoidance behaviours such as panting and increased resting can lead to decreased foraging efficiency, weight loss and worsening overall condition in birds (McKechnie 2019; van de Ven, McKechnie *et al.* 2019). These issues can have damaging effects on the persistence of avian populations (Mac Nally, Bennett *et al.* 2009; McKechnie and Wolf 2010).

Arid-zone birds have shown reduced activity at high temperatures (Cooper, Withers *et al.* 2019; Funghi, McCowan *et al.* 2019; Pattinson, Thompson *et al.* 2020). Similarly, above their heat tolerance of approximately 41 - 42°C (Booth 1984), Malleefowl seek shade and rest more as well as showing heat dissipation behaviours like panting and gular fluttering (Benshemesh 1992). Reduced activity could lead to less time tending mounds or foraging, negatively impacting hatching success, recruitment and survival of Malleefowl.

1.5 Knowledge gaps & aims

Due to the above threats, Malleefowl distribution has declined drastically since European settlement and is very likely to continue to decline further (Benshemesh 2007; Benshemesh, Southwell *et al.* 2020; Frith 1962a). To slow and counteract this decline, it is important to understand how these threats may affect Malleefowl populations in terms of breeding, population trends, genetics, movement patterns and habitat selection.

To commence breeding, Malleefowl are highly reliant on winter rainfall and will not breed until the next season if rainfall is insufficient (Benshemesh 1992a; Booth and Seymour 1983; Frith 1962b). Consequently, with predictions of decreasing winter precipitation and increasing frequency of droughts (CSIRO and BOM 2015), we can expect reduced Malleefowl breeding activity and continued long-term population declines.

Habitat loss and fragmentation have been identified as the main reasons for Malleefowl declines (Benshemesh 2007; Parsons, Short *et al.* 2008; Woinarski 1989). This is likely because fragmentation or destruction of habitat limits movement. Movement is an important part of an animal's life and may be driven by the individuals' need for social interaction (e.g. mating and breeding) or dispersal, migration, resource selection or to escape risks (Nathan 2008). Ultimately, movement is necessary to optimise energy intake, safety and breeding success (Nathan, Getz *et al.* 2008). However, species' movement modes and levels of mobility may also affect gene flow (Teitelbaum and Mueller 2019) and increase extinction risk in fragmented landscapes (Armsworth and Roughgarden 2005; Fahrig 2007). Changing environmental conditions can also influence animal movement (Morales, Moorcroft *et al.* 2010; Shaw 2020). It is therefore important to understand how anthropogenic habitat alterations and climate change may affect movement and impact their fitness. Malleefowl movement has been little studied in the past (Benshemesh 1992a; Booth 1985; Weathers and Seymour 1998) and there are still knowledge gaps about Malleefowl movement ecology. Knowledge gaps include movement in the colder months of the year, when on-ground observation is harder; seasonal and sex or climate-driven differences in movement patterns; occurrence, frequency and timing of long-distance movements and open space use (i.e. cleared land) and habitat preferences. It is important that these gaps are filled by long-term, continuous movement data which were collected in a way that is not influenced by observer bias or changed Malleefowl behaviour through repeated capture and/or stalking. Fine-scale data recorded in this way to document movement with minute behavioural detail (i.e. foraging or resting) would round off existing observational data, however, will not be part of this thesis as our fix frequencies are not suitable for this level of behavioural analysis. This thesis will investigate if Malleefowl remain in their home ranges or show exploratory behaviour outside their home ranges and are able to disperse to new areas. Further, it will examine if movements are influenced by breeding status, sex or type or availability of habitat.

Malleefowl are a long-lived species, therefore their ability to move and resettle in response to danger or changes in their habitats, such as land clearance and wildfires, is especially important. If movement is limited through, for example, habitat fragmentation or a need to conserve energy because of increased ambient temperatures, this may be detrimental to

the animal's breeding success or chances of finding a mate. While effects may not be immediately obvious, in the long-term, the inability to disperse and pass on genetic material may result in the decline of genetic health and ultimately, local or even global extinction. Very little research has been done on Malleefowl genetics to date (Cope, Bertozzi *et al.* 2016; Cope, Mulder *et al.* 2014; Donnellan, Dubach *et al.* 1994). Cope, Mulder *et al.* (2014), using mitochondrial DNA and nuclear microsatellite markers, found that Malleefowl historically dispersed west to east, with two distinct populations west and east of the Eyrean Barrier. Additionally, the authors identified an isolation-by-distance structure in populations and low levels of differentiation but high levels of diversity between mitochondrial haplotypes. To date, no genetic studies have been completed at a finer geographic scale to examine gene flow between fragmented populations, nor to assess levels of genetic diversity in regions with highly fragmented populations.

Consequently, this thesis aims to understand 1) What are the long-term breeding trends of Malleefowl and are they influenced by fragmentation and any environmental factors (likely to be affected by climate change); 2) the effect of fragmentation on genetic diversity and whether movement patterns of Malleefowl on the Eyre Peninsula have changed, i.e., whether dispersal has happened in the past and if so, over what distances and whether there is a sex bias; 3) the current movement patterns of Malleefowl on the Eyre Peninsula and whether they are influenced by temperature, breeding and fragmentation and 4) Habitat use and how this may influence movement patterns.

Results from this thesis may be used to improve conservation planning and outcomes for Malleefowl. Understanding the effects of fragmentation, rainfall, fire and heat on Malleefowl breeding, genetics, movement patterns, population trends and habitat use will enable land managers to make decisions on management options to reduce these effects. Knowing how Malleefowl use and move between remaining patches in agricultural matrices can inform managers about sizes and distances of patches most suitable for the protection and persistence of Malleefowl. Knowing about maximum distances traversed across open agricultural land may also inform about the management of matrix habitat itself, such as improving road-side vegetation strips where Malleefowl have been observed in the past (Benshemesh 2007), which can be valuable habitat (New, Sands *et al.* 2021), or planting paddock trees to encourage dispersal. It can also assist managers in timing management activities, such as weeding, in a way that reduces disturbance or potential exposure to chemicals near mounds.

While this study has its focus on the Eyre Peninsula, the results can be applied to Malleefowl across Australia where Malleefowl can be found in similar semi-arid and highly fragmented habitats. Other terrestrial species that persist in similarly semi-arid landscapes with highly

modified landscapes and pressures from predation and changing environmental conditions may also benefit from this work.

As much habitat has been cleared for farming in Australia many native species now persist in highly fragmented native vegetation patches embedded in agricultural matrices (Tulloch, Barnes *et al.* 2016). Therefore, it is important to know how animals use intact and fragmented habitat and the surrounding matrix, as these habitats may need to be managed differently and movement ecology can be a valuable tool in conservation planning and the identification of suitable management actions (Allen and Singh 2016). Knowing movement patterns and potential seasonal differences or factors influencing them can inform land managers about the appropriate size of connecting habitat corridors and the size of patches needed to support species and ensure long-term persistence. Knowing how species move in the event of a fire may also assist in fire management. The quality of the matrix itself is important too, as it enables species to travel between and use remnant patches (Saura, Bodin *et al.* 2014) and strongly influences the dispersal ability (Fahrig 2007). Lack of movement in fragmented landscapes can lead to decreased gene flow and inbreeding depression, potentially resulting in long-term deterioration and local extinctions of remnant populations (Ribon, Simon *et al.* 2003; Stevens, Harrison *et al.* 2018) Integrating movement ecology and genetic research can improve conservation outcomes and counteract genetic decline by enhancing our ability to make informed management decisions, such as timing and appropriateness of translocations (Frankham, Ballou *et al.* 2011; Jeltsch, Bonte *et al.* 2013; Onley, Austin *et al.* 2021; Weeks, Sgro *et al.* 2011).

1.6 Thesis structure

This thesis consists of a general introduction (chapter 1), followed by four data chapters (chapters 2 - 5) in which I investigate the population trends, past and present dispersal and habitat use of Malleefowl on the Eyre Peninsula and, lastly, a final discussion (chapter 6). Chapters 2 - 4 have been written as stand-alone pieces of work and have been submitted to journals for publication. Consequently, there is some inevitable overlap in the content of the chapters and inconsistencies in style and formatting.

Historic impacts of habitat clearing and degradation for farming have led to an Australia-wide decrease in Malleefowl populations and contraction of their range. In chapter 2, I compare long-term Malleefowl breeding activity on the Eyre Peninsula over 24 years with a comprehensive set of environmental and habitat variables to determine factors that may be related to population persistence and breeding. This chapter has been accepted for publication as:

Stenhouse P, Moseby KE. Trends in breeding activity of the threatened Malleefowl (*Leipoa ocellata*): what can we expect under a changing climate? *Emu - Austral Ornithology*. 2022. <https://www.doi.org/10.1080/01584197.2022.2045870>

Habitat loss and fragmentation can lead to reduced gene flow, increased genetic drift, and inbreeding, all of which may be detrimental to the long-term conservation of species. In chapter 3, I examine the spatial genetic structure and dispersal patterns of Malleefowl on the semi-arid Eyre Peninsula using single nucleotide polymorphisms (SNPs). This chapter has been submitted for publication and is under review as:

Stenhouse P, Onley IR, Mitchell KJ, Moseby KE, Austin JJ. Spatial genetic structure and limited gene flow in fragmented populations of the threatened Malleefowl (*Leipoa ocellata*). *Ecological Genetics and Genomics*. 2022. <https://doi.org/10.1016/j.egg.2022.100127>.

Modern GPS telemetry enables us to collect highly accurate movement data without spatial limitations for long periods without causing undue stress to the animal through repeated capture. In chapter 4, I explore the current movement of Malleefowl on the Eyre Peninsula using solar-powered GPS telemetry. Using up to 50 months' tracking data I investigate the influence of breeding status, patch size and temperature on movement patterns. This chapter has been submitted for publication as:

Stenhouse P, Moseby KE. Patch size and breeding status influence movement patterns in the threatened Malleefowl (*Leipoa ocellata*). *Austral Ecology*. 2022.

Movement can be driven by the need for finding the most suitable habitat for foraging, breeding and escaping predators. For chapter 5, I completed over 240 vegetation surveys at sites (often/rarely/not) visited by non-breeding Malleefowl. I then modelled visitation frequency as a function of a variety of vegetation-related variables using classification tree models to determine whether Malleefowl habitat selection was driven by habitat structure or certain plant genera. This chapter has been written in publication format for future submission to a journal.

1.7 References

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Chapter 2:

Trends in breeding activity of the threatened Malleefowl (*Leipoa ocellata*): what can we expect under a changing climate?



Figure on previous page: All images were taken at Secret Rocks mound 259 between November 2017 and April 2018. At least one of the adults was killed by a feral cat or a fox in late January, after which mound maintenance ceased. This wasn't one of the tracked birds. Credit: Peri Stenhouse

2.1 Statement of Authorship

Title of Paper	Trends in breeding activity of the threatened Malleefowl (<i>Leipoa ocellata</i>): what can we expect under a changing climate?			
Publication status	<input checked="" type="checkbox"/> Published	<input type="checkbox"/> Accepted for Publication	<input type="checkbox"/> Submitted for Publication	<input type="checkbox"/> Publication style
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Author contributions

By signing the Statement of Authorship, each author certifies that the candidate's stated contribution to the publication is accurate, that permission is granted for the candidate to include the publication in the thesis; and that the sum of all co-author contributions is equal to 100% less the candidate's stated contribution.

Name of Principal Author (Candidate)	Peri Stenhouse		
Contribution to the paper	Conceptualization, methodology, data curation, formal analysis, writing – original draft preparation, writing – review and editing. Responses to reviewers.		
Overall percentage	80%		
Certification	This paper reports on original research I conducted during the period of my Higher Degree by Research candidature and is not subject to any obligations or contractual agreements with a third party that would constrain its inclusion in this thesis. I am the primary author of this paper.		
Signature			Date: 04 April 2022

Name of Co-Author	Katherine Moseby		
Contribution to the Paper	Conceptualisation, methodology, writing – review and editing.		
Signature			Date: 04 April 2022

2.2 Abstract

Climate change is expected to significantly impact bird species through changes to breeding and survival. Malleefowl (*Leipoa ocellata*) are threatened mound-building birds that persist in uncleared mallee and shrubland vegetation in semi-arid parts of southern Australia. Our aim was to understand the relationship between environmental factors and mound breeding activity (a proxy for population trends) to identify likely climate change impacts and possible proactive management actions. We compared annual activity at grids that encompassed groups of mounds at six sites in South Australia for up to 24 years with a range of environmental and habitat variables, focussing on variables predicted to change with a warming climate. Mound activity declined significantly over the study period at five of the six sites. Activity was positively associated with increased cumulative rain in the previous 2 years, lower average maximum temperatures in the winter, higher-than-average Southern Oscillation Index (i.e. more rain) 2 years before breeding and greater winter vegetation cover. These results suggest that moisture and vegetation cover are important for higher breeding activity. Climate change is predicted to lead to drier conditions and more frequent fires in our study region, suggesting that Malleefowl populations will continue to decline. We urge conservation practitioners to minimise climate change impacts through implementing proactive management actions that increase habitat quality for Malleefowl: fire management to reduce the scale of fire events and controlling introduced and overabundant native herbivores to preserve vegetation cover, retain soil moisture, increase food resources and protect from temperature extremes.

2.3 Keywords

Population trends; semi-arid zone; conservation ecology; threatened species; mound builders

2.4 Introduction

Climate change is expected to have significant impacts on bird species globally (Conradie, Woodborne *et al.* 2019; Yalcin and Leroux 2018). Increasing heat, changes in precipitation rates and fire frequency are all predicted to lead to increased mortality rates, reduced breeding success and/or changes in frequency or timing of breeding (McKechnie 2019; McKechnie and Wolf 2010). Impacts are expected to be very severe in southern Australia with current climate change scenarios predicting temperatures to increase by up to 5°C by 2090, increased time in drought and generally more erratic weather patterns (Chambers, Hughes *et al.* 2005; Charles and Fu 2015; Wardell-Johnson, Keppel *et al.* 2011). South Australia's April to October rainfall has decreased approximately 12% since the 1990s (BOM and CSIRO 2020) and climate models for the Eyre Peninsula region predict further significant decreases in winter and spring rainfall of up to 40% by 2090 (CSIRO and BOM 2015).

Malleefowl (*Leipoa ocellata*) are large, ground-dwelling birds that live in the arid and semi-arid mallee woodlands and shrublands of southern Australia (Benshemesh 2007; Frith 1962). They build large mounds up to 6 m in diameter in which they incubate their eggs using heat from decomposing leaf litter and the sun (Benshemesh 1992; Booth 1985; Frith 1959). Once found over most of Southern Australia, Malleefowl are now classified globally as Vulnerable on the IUCN list of threatened species (IUCN 2012, criteria VU A1c,e and A2b,c,e). Numerous reasons for the decline have been proposed including predation by foxes and cats (Priddel and Wheeler 1994; Wheeler and Priddel 2009), inappropriate fire regimes (Benshemesh 1992; Parsons and Gosper 2011), high juvenile mortality (Priddel and Wheeler 1994) and road kills (Benshemesh, Barker *et al.* 2007). Habitat loss is the main driver of decreasing population trends, as Malleefowl are habitat dependent and prefer long unburnt and uncleared vegetation (Benshemesh 2007; Parsons, Short *et al.* 2008; Woinarski 1989).

Due to the cryptic nature of the adult birds, monitoring rates of breeding through mound activity is considered the best way to monitor trends in Malleefowl populations (Benshemesh 2004). Analysis of mound monitoring data nationally suggests that birds do not breed every year and that breeding activity appears to be positively influenced by winter rainfall and rainfall in the preceding 4 years (Benshemesh, Barker *et al.* 2007; Benshemesh, Southwell *et al.* 2020; Gillam 2008). Rainfall also influences primary production, providing the laying female with nutrients and energy needed for egg production (Benshemesh 1992; Frith 1959; Weathers and Seymour 1998). Malleefowl densities are highest in areas with higher rainfall (Copley and Williams 1995; Frith 1962) and long unburnt mallee

(Benshemesh 1992; Woinarski 1989). Climate change may exacerbate the pressures on Malleefowl survival by causing an increase in periods of low precipitation and above-average temperatures leading to reduced soil moisture and more frequent and severe wildfires (CSIRO and BOM 2015; Steffen, Hughes *et al.* 2014).

We report on the activity of Malleefowl mounds at six sites across Southern Australia for up to 24 years and investigate possible causes of decline. Wheatley, Beale *et al.* (2017) determined that trends-based methods are more suitable to determine climate change vulnerability of species than traits-based methods. Therefore, we use this approach to compare mound activity trends with a comprehensive set of measured habitat and weather variables including rainfall, temperature, Southern Oscillation Index (SOI), Indian Ocean Dipole (IOD), fox control and remnant patch size. Our aim was to understand the possible drivers behind any correlation between these variables and mound activity and thus population abundance, to determine the likely effects of climate change and identify possible proactive management actions to offset these.

2.5 Methods

2.5.1 Study Sites

The Eyre Peninsula covers about 80,000 km² in the southern coastal region of South Australia. Since European settlement, only about 39% of native vegetation remains - mostly in marginal land unsuitable for agriculture (Brandle 2010; NREP 2017). Malleefowl are patchily distributed on the Eyre Peninsula and are now largely restricted to protected remnant habitat in national parks or private land. The average annual rainfall on the Eyre Peninsula varies from 250 mm in the North East to 500 mm in the South West (BOM 2022). The Eyre Peninsula supports semi-arid and temperate habitats dominated by open mallee scrub with patches of sand dunes. Table 1 lists the predominant canopy species at our sites based on Matthews (2002).

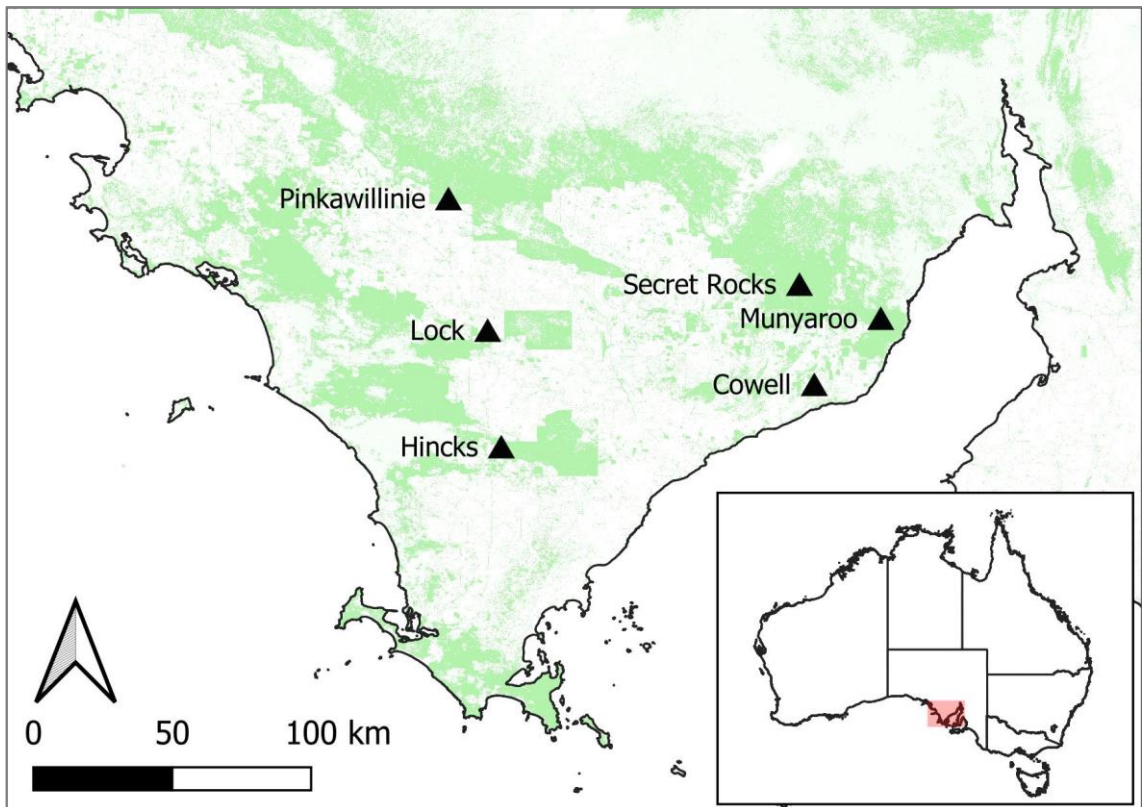


Figure 2-1 Locations of Malleefowl mound activity grids on the Eyre Peninsula. Green areas depict native vegetation cover. Spatial vegetation data (most likely landcover 2010-2015) were obtained from the South Australian Government Data Directory (Government of South Australia 2022). Map was made with QGIS v3.16 (QGIS Development Team 2021).

Malleefowl mound activity is monitored within fixed grids of between 4 and 7 km², located within remnant native vegetation patches that vary in extent between four and several thousand km². The National Malleefowl Recovery Team (NMRT) stores data from a total of 163 grids across Australia. Six of these grids are located on the Eyre Peninsula in Hincks, Munyaroo and Pinkawillinie Conservation Parks; Secret Rocks Nature Reserve and on private Heritage Agreements near Lock and Cowell (Figure 2-1). The grids are between 35 and 168 km apart from each other and are separated by agricultural land, settlements and roads. The Cowell grid has been monitored since 1995 with the latest grid at Secret Rocks established in 2010 (Table 2-1).

Grids are monitored once a year by volunteers and trained personnel according to a standardised methodology (NMRT 2020). Mounds that show clear signs of current use for breeding are recorded as active mounds. A mound can only be active or inactive.

Table 2-1 Attributes of the Malleefowl nest activity monitoring grids on the Eyre Peninsula. Dominant canopy species based on Matthews (2002). Rain = Mean annual rainfall across all years in which monitoring took place [mm]; Species = Predominant canopy species; Size = Total grid size (Patch size) [km²]; Mounds = Number of mounds known in 2018; Fires: Years in which part of the grid was burnt (number of mounds) Note: No mounds were burnt twice.

Grid	Rain	Species	Size	Mounds	Fires	Years monitored
Cowell (2 sections)	257	<i>Eucalyptus socialis</i> , <i>E. gracilis</i>	4.75 (5)	70	na	1995-1997, 1999-2018
Hincks	377	<i>E. diversifolia</i> , <i>E. incrassata</i>	4 (680)	34	1959 (7) 1980 (9)	1998, 2002, 2003, 2005-2007, 2009-2018
Lock	334	<i>E. diversifolia</i> , <i>E.</i> <i>incrassata</i> , <i>Melaleuca</i> <i>uncinata</i>	4 (6.7)	58	na	2003-2007, 2008-2018
Munyaroo	271	<i>E. incrassata</i> , <i>M. uncinata</i> <i>Acacia sowdenii</i> ± <i>Casuarina cristata</i>	4 (846)	44	na	1998, 2003, 2005-2018
Pinkawillinie	290	<i>E. incrassata</i> , <i>M. uncinata</i>	4 (2900)	23	2006 (7)	1998, 2003, 2005, 2006, 2011, 2015-2018
Secret Rocks (3 sections)	317	<i>E. brachycalyx</i> , <i>E. socialis</i> , <i>E. gracilis</i>	7.1 (1600)	66	1972 (17) 1991 (4)	2010-2018

Activity was summarised each year for each grid as the proportion of surveyed mounds that were active (proportional mound activity). The number of monitored mounds per grid varied each year depending on whether new mounds were discovered during monitoring and whether old mounds, which had not been active for more than 5 years, were not visited due to time constraints. Monitoring and activity history for each grid can be found in Supporting Information (SI) 1. Attempts were made to monitor each grid annually, but logistical and funding limitations and fire prevented annual monitoring of some grids. This led to data gaps, especially in the early monitoring phases of some grids and post-fire in the Pinkawillinie grid. Therefore, only data from 2003 to 2018 were used and Pinkawillinie grid data were excluded from modelling.

2.5.2 Environmental and habitat variables

Temperature and rainfall both have a significant impact on ground soil moisture (Huntington 2010; Lvova and Nadporozhskaya 2017) which is likely to influence Malleefowl breeding success through food availability and composting rates.

The SOI is a key indicator for estimating the strength of the El Niño (low precipitation) and La Niña (high precipitation) events (Chiew, Piechota *et al.* 1998). It is a predictor of rainfall in Australia (Feng, Wang *et al.* 2020; Ropelewski and Halpert 1989) and causes large interannual variability in Australian rainfall patterns (Holmgren and Scheffer 2001; McAlpine, Syktus *et al.* 2009; Nicholls 1991) which have profound effects on primary production and terrestrial ecosystems in arid and semi-arid zones (Holmgren, Stapp *et al.* 2006; Letnic, Tamayo *et al.* 2005).

The IOD is one of the main drivers of Australia's weather patterns, particularly in the south of the continent (Ummenhofer, Gupta *et al.* 2011; Ummenhofer, Gupta *et al.* 2009), with the most significant effects observed from June to October (Risbey, Pook *et al.* 2009). Positive IOD values (pIOD) can lead to decreased precipitation from western to south-eastern Australia and have been linked to major bushfires and droughts, while negative values (nIOD) may increase precipitation (Cai, Cowan *et al.* 2009a; McAlpine, Syktus *et al.* 2009; Ummenhofer, Gupta *et al.* 2011).

The SOI and IOD are reliable predictors of drought conditions in Australia (Deo and Şahin 2015; Montazerolghaem, Vervoort *et al.* 2016) and interact with each other, especially from June to October (Risbey, Pook *et al.* 2009; Ummenhofer, Gupta *et al.* 2011). The co-occurrence of a pIOD (nIOD) event with an El Niño (La Niña) event causes the driest (wettest) years, influencing soil moisture and thus vegetation in south-eastern Australia (Ummenhofer, Gupta *et al.* 2011).

Vegetation is important for Malleefowl as a food source, a source of protection from predators and roosting habitat (Benshemesh, Barker *et al.* 2007; Weathers and Seymour 1998).

For further details and a complete list of environmental variables please see Table S 2-3.

2.5.3 Analysis

First, we investigated the probabilities of breeding activity for each grid and each year by applying generalised linear mixed effects models (glmer, Table S 2-4). Where variation among years or among grids was identified, we examined the patterns of difference by estimating marginal means (i.e. least-squares means) and used planned contrasts to investigate where those differences occur (R package emmeans) (Lenth, Singmann *et al.* 2019). Mounds that were inactive in all monitoring years were excluded from the marginal means analysis. Then, we examined the effects of environmental and habitat covariates on variation in breeding activity across space and time by also applying generalised linear mixed effects models. Environmental variables were selected a priori (Table S 2-3). Covariates with a variance inflation factor greater than four were dropped to avoid multicollinearity (Hair, Black *et al.* 2010). Remaining sets of environmental variables were then used to build sets of candidate models and used as fixed effects.

Data from 2010 to 2018 inclusive were used for the five monitoring grids that had been monitored consistently over this time (Cowell, Hincks, Lock, Munyaroo and Secret Rocks). Year and grid were included as random effects. The response variable was the proportional mound activity (activity ratio) per grid for each year. The activity ratio was determined by dividing the number of active mounds recorded on a grid by the total number of mounds

monitored on the same grid in that year. We accounted for differing numbers of monitored mounds each year by adding a weight argument with the number of checked mounds. Environmental covariates were aggregated across all mounds within a grid.

All models were random intercept models with binomial distribution and were optimised using quadratic approximation (*bobyqa*) with a maximum of 200,000 iterations. Model outputs were compared using the corrected Akaike Information Criterion (AICc) (Lenth, Singmann *et al.* 2019). Models within four AICc units of the top model or with a better AICc score than the intercept only model (whichever came first) were considered best models. R² values were calculated using the intercept only model and the model in question. All data analyses were performed with R Studio version 1.2.1335 (R Core Team 2020) and the following packages: data cleaning with *tidyverse* (Wickham, Averick *et al.* 2019); mixed effect modelling with *lme4* (Bates, Mächler *et al.* 2015); R² with *MuMin* (Barton 2019); data visualisation with *flextable* (Gohel 2021) and *sjPlot* (Lüdecke 2021); and graphs with *ggplot2* (Wickham 2016) and *effects* (Fox and Hong 2009).

2.6 Results

2.6.1 Overview

A total of 294 individual mounds were surveyed on the 6 grids over the study period (1995-2018), totalling 3553 mound monitoring events. There was no breeding activity recorded on Cowell and Hincks Grids in 2018; on Lock Grid in 2007, 2017 and 2018; on Munyaroo Grid from 2013 to 2018; and on Secret Rocks Grid from 2013 to 2017 (Table S 2-1).

The average number of mounds per grid across all years was 10.4 per km² and ranged from 4.5 per km² at the Pinkawillinie Grid to 14.8 per km² at the Cowell Grid (Table S 2-2).

2.6.2 Mound activity over time

Over the whole monitoring period from 1995 to 2018, the proportion of active mounds declined in five grids, following a similar trajectory of decline. The Hincks grid was the exception with the number of active mounds on the grid fluctuating but showing a slightly increasing trend (Figure 2-2). The highest proportional activity was observed in Secret Rocks in 2012 (0.300) and 2011 (0.269); in Hincks in 2013 (0.250) and Cowell in 2000 (0.228, 2.74 mounds km⁻²) and 1995 (0.196; 2.11 mounds km⁻²) (Table S 2-1).

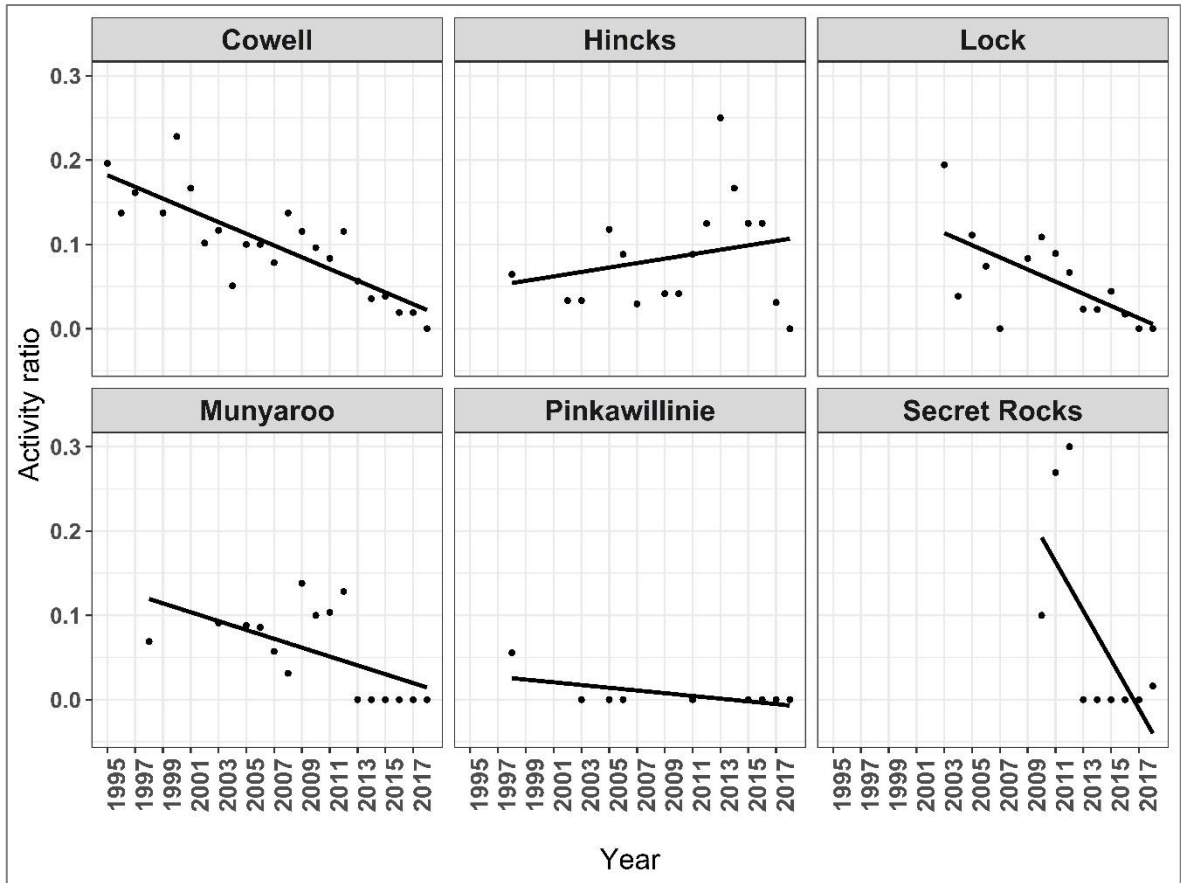


Figure 2-2 Proportional mound activity of Malleefowl in the Eyre Peninsula grids between 1995 and 2018. The proportional mound activity is the number of active mounds divided by the number of monitored mounds.

2.6.3 Probabilities of activity

Estimated means of probabilities differed among years and variation in probability of breeding activity was best explained by a model which only considered year effects (Table S 2-4). For the period between 2003 and 2018, there was an overall decrease in probability of activity in four grids (Cowell, Hincks, Lock and Munyaroo), even though probabilities fluctuated in the first half of observations. This decline over time shows a common pattern over all grids and was characterised by 2017 and 2018 having significantly lower activity than 2003, the initial year in our dataset. The probability of activity was four times lower in 2016 than in 2003 (CI -1.49, -11.4) and 13 times lower in 2017 (CI -2.80, -60.1). In 2018 the estimated probabilities of activity were essentially zero (Table S 2-4, Figure S 2-1).

When the Secret Rocks grid was included in the short-term dataset from 2010 to 2018, the decline in activity over time showed a similar decline trend. The probability of activity was 4 times lower in 2016 than in 2010 (CI -1.46, 10.96), over 13 times lower in 2017 (CI 2.85, 61.56) and over 27 times lower in 2018 (CI 3.47, 224.75) (Figure S 2-2).

2.6.4 Variables influencing activity

Up to 15.4% of the variation in the proportional mound activity from 2010 to 2018 could be explained with the environmental data (Table S 2-5). The model with the best fit included the cumulative rainfall in the 2 years before breeding and the average maximum temperature in the winter before breeding (Figure 2-3). An equivalent model contained the average SOI for the winter months 2 years prior to breeding and the percentage of bare ground in the winter prior to breeding (Figure 2-4). Proportional mound activity in a grid increased with more rain in the 2 years prior to breeding and lower maximum winter temperatures, as well as higher SOI in winter (i.e. more rain) 2 years prior to breeding and less bare ground during winter (i.e. more vegetation cover) prior to breeding.

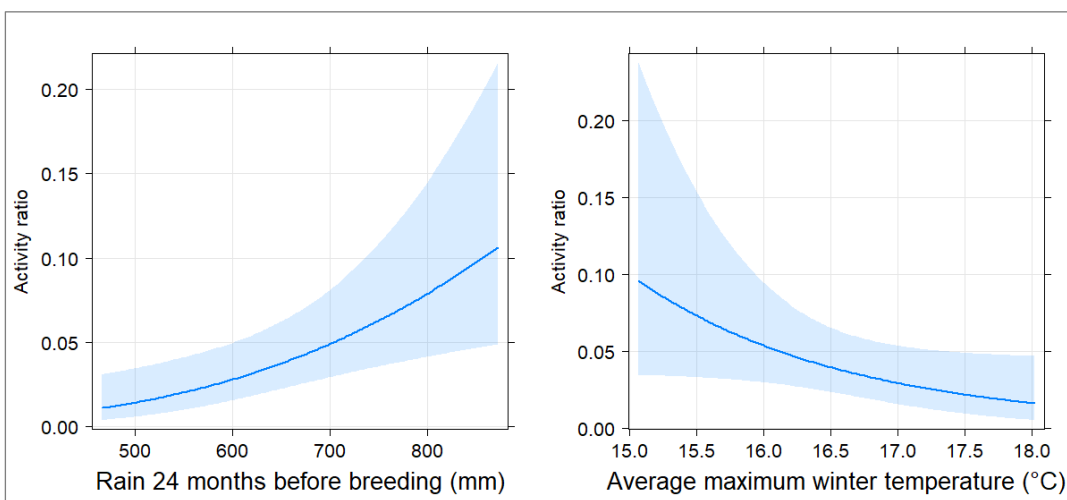


Figure 2-3 Best model: Predicted probabilities of Activity ratio in a grid and, on the left, cumulative rainfall of the 2 years (24 months) prior to breeding, and, on the right, average maximum temperature in the winter months (June, July, August), 1-3 months prior to breeding. The shaded areas are the 95% confidence intervals for the predicted values.

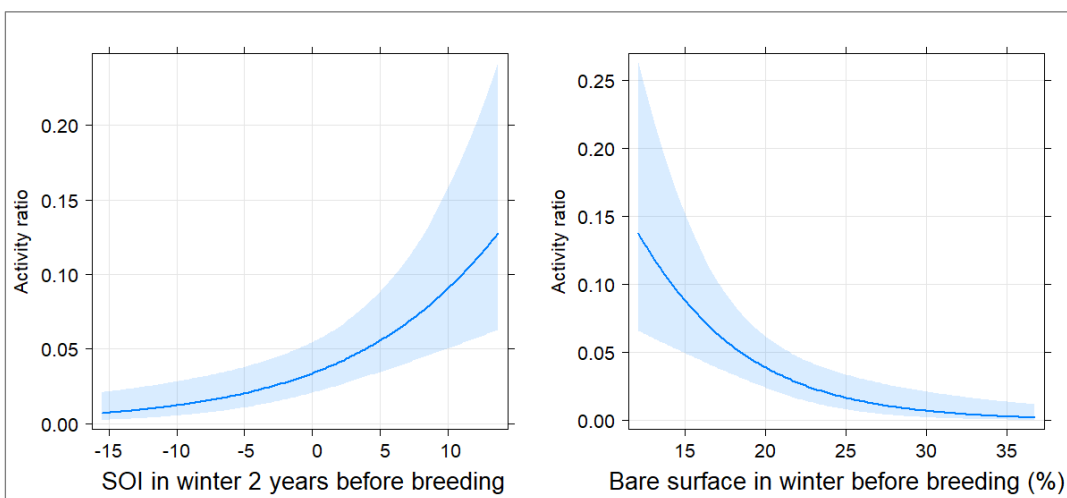


Figure 2-4 Second (equivalent) model: Predicted probabilities of activity ratio in a grid and, on the left, the average SOI in the winter months (June, July, August) 2 years prior to breeding (i.e. 25-30 months before breeding), and, on the right: bare surface (no vegetation) in the winter, 1-3 months before breeding. The shaded areas are the 95% confidence intervals for the predicted values.

Further models within $\Delta AICc \leq 4$ contained the same variables as the best two models but additionally included patch size, fox control and maximum autumn temperature. However, none of these additional variables were statistically significant and thus will not be discussed. Variables such as green cover, minimum temperature or IOD were not present in any of the top models (See Table S 2-3 for a list of all variables).

2.7 Discussion

Our results suggest Malleefowl breeding activity is declining on the Eyre Peninsula, supporting other analyses of national Malleefowl trends which found the highest rate of decline in South Australia (Benshemesh, Barker *et al.* 2007; Benshemesh, Southwell *et al.* 2020). All but one of our grids showed a long-term decline in mound activity over the last two to three decades with a major decline after 2012.

Our results further indicate that Malleefowl breeding activity is significantly influenced by a combination of environmental factors, most of which are related to moisture: rainfall, maximum temperature, SOI and vegetation cover.

Proportional mound activity in the grids was higher if it rained more in the previous 2 years and the winter maximum temperature was lower. This supports previous research in which the positive effect of winter rain on Malleefowl breeding activity is reported (Benshemesh, Southwell *et al.* 2020; Brickhill 1987; Gillam 2008; Priddel and Wheeler 2005) and also time-lagged effects of winter rain of 2-4 years (Benshemesh, Barker *et al.* 2007). Rainfall is crucial for the successful incubation of eggs, as the initial heat for incubation comes from composting leaf litter which needs sufficient moisture to begin microbial decay - in case of a drought, the nests are abandoned for the season (Frith 1959; Lewis 1939). However, even when the moisture level of two mounds was artificially elevated by adding water to them in a drought year, they did not commence breeding (Booth and Seymour 1983; Brickhill 1987), which indicates that other factors related to rainfall, such as food availability or other climatic factors may play a greater role.

Rainfall and temperature have a significant impact on ground soil moisture (CSIRO and BOM 2015; McAlpine, Syktus *et al.* 2009; Steffen, Hughes *et al.* 2014), which in turn affects vegetation. Vegetation cover can include both perennial plants and annual cover stimulated by rainfall, and although both total vegetation cover and green cover (i.e. growing plants) were included in the models, it was the total vegetation cover that was a more important determinant of breeding activity. Herbs, shrubs and their seeds make up a large part of Malleefowl diet and are important for fulfilment of the calorific needs of the egg-laying female (Frith 1962; Harlen and Priddel 1996). Abundant food leads to larger clutch sizes in

Malleefowl (Frith 1959), while food shortages lead to infertility of eggs (Brickhill 1987). Vegetation cover also provides habitat for invertebrate food sources that Malleefowl eat and protects soil moisture levels through shading. Vegetation provides safety from predation in the form of cover for hatchlings and roosting space for both juveniles and adults; however, one study found foxes were killing young Malleefowl at very high rates irrespective of the structural integrity of the mallee scrub (Priddel, Wheeler *et al.* 2007). Plants are likely to produce fewer or no seeds in drought years, thus also reducing chances of recruitment in following years. Decreased plant cover is likely a combination of poor rainfall, but also high herbivore impacts as species such as goats and kangaroos browse the understory and compete for food resources with Malleefowl, especially in drought years (Frith 1962; Hauser, Southwell *et al.* 2019). These impacts can be severe, particularly after a fire (Cohn and Bradstock 2000; Foster, Barton *et al.* 2015). Whilst rainfall is a main driver of vegetation cover, management of fire and herbivore grazing pressure is likely to increase vegetation cover in areas where Malleefowl are present and help boost breeding activity.

A second model, with the same predictive power as the rain and temperature model discussed above, indicates that a higher-than-average winter SOI 2 years before breeding, together with more winter vegetation cover immediately before breeding, may induce an activity response. SOI drives temperature and rainfall (BOM 2022) and high SOI values are indicative of La Niña events with above-average rainfall for several months. On the Eyre Peninsula, where our semi-arid study sites are located, this effect is especially strong in the winter months (Risbey, Pook *et al.* 2009; Ummenhofer, Gupta *et al.* 2011). Significant rainfall events caused by La Niña in winter would therefore encourage plant growth, seeding and recruitment and increase soil moisture for extended periods, consequently improving food availability and lowering ground and air temperatures through shading and transpiration. Therefore, La Niña events may provide disproportionate benefit to the species and might facilitate critical high breeding periods to offset drought conditions. However, the frequency of La Niña events has been decreasing since the beginning of the 20th century and is predicted to decrease further, while El Niño events are predicted to double due to climate change (Dey, Lewis *et al.* 2019; Santoso, McGregor *et al.* 2013). To our knowledge, no other research has investigated the connection between the SOI and Malleefowl activity. The positive effect of (time-lagged) winter rain on Malleefowl breeding activity discussed above underlines our findings, only retrospectively. Distal variables such as SOI may assist with future management planning up to 2 years in advance.

Size of remnant patch, IOD and green cover appeared to have little influence on breeding activity and may reflect the ability of this species to survive and breed in a range of habitats and fragmented patches of vegetation. A comparison with more grids in different habitats

and with different patch sizes might have revealed stronger trends. Nevertheless, Malleefowl are found in a wide range of mallee and shrubland habitats and used to occur over most of semi-arid Australia, suggesting they have broad habitat adaptability.

There was also no evidence of an effect of fox baiting. The reasons for this could be that fox baiting actually has no effect or that baiting regime data were insufficient. Benshemesh, Southwell *et al.* (2020) also recorded no effects using a larger dataset (which included our study grids) with fox abundance and bait intensity as covariates and concluded that fox baiting may not assist Malleefowl conservation. Nonetheless, the absence of a strong response to fox baiting may also be due to poor effectiveness of the fox control methods themselves or the secondary effects of fox baiting, such as an increase in cat, rabbit and kangaroo numbers (Banks, Dickman *et al.* 1998; Banks, Newsome *et al.* 2000). We believe this is the case for our study because five out of nine tagged Malleefowl on the Eyre Peninsula were most likely killed by foxes (chapter 4) and foxes are known to prey heavily on adult and young birds (Priddel and Wheeler 1994; Wheeler and Priddel 2009). A large-scale adaptive management program with before-after-control-impact experiments using large effective baited areas is currently taking place to assess the impacts of predator control on Malleefowl (Hauser, Southwell *et al.* 2019).

Malleefowl mound activity monitoring focuses on breeding adults but disregards non-breeding adults or juveniles. Even during years where no mounds were active, Malleefowl scats and tracks were still recorded at some mounds. Understanding population trends by using monitoring methods that do not rely on breeding rates, such as intensive camera trapping or scats on mounds, may improve our knowledge of factors affecting adult mortality.

In conclusion, our findings suggest that climate change is likely to have a significant impact on Malleefowl and that the documented declines over the last few decades may be related to drying conditions caused by climate change. Driven by large-scale climate drivers, rainfall has decreased by about 12% in South Australia since the 1990s (BOM and CSIRO 2020; Cai, Cowan *et al.* 2009b; Dey, Lewis *et al.* 2019) and while La Niña events would likely benefit Malleefowl breeding, their frequency is decreasing and multiple climate models project substantial decreases in rainfall in the Eyre Peninsula region in the winter months (CSIRO and BOM 2015). Therefore, climate change is likely to exacerbate pressures on Malleefowl survival by causing an increase in periods of low precipitation and above-average temperatures leading to a loss of soil moisture and more frequent and severe wildfires (CSIRO and BOM 2015; McAlpine, Syktus *et al.* 2009; Steffen, Hughes *et al.* 2014).

In arid zones, it is predicted that decreasing rainfall and increasing temperatures will cause range contractions in many bird species (Böhning-Gaese and Lemoine 2004). El Niño-Southern Oscillation events have been shown to affect the breeding phenology of birds (Englert Duursma, Gallagher *et al.* 2018) and these drought events are predicted to become more common under climate change. Extreme heat may reduce fitness (Pattinson, Thompson *et al.* 2020), lead to mortality from dehydration (McKechnie 2019; McKechnie and Wolf 2010) and cause reduced breeding success in arid-zone bird species (van de Ven, McKechnie *et al.* 2019; van de Ven, McKechnie *et al.* 2020). Climate change has already caused global extinctions and is expected to cause more in the future (Conradie, Woodborne *et al.* 2019; Yalcin and Leroux 2018).

Improving the conservation status of the Malleefowl under climate change will require careful fire management and a significant reduction in herbivore grazing pressure to enhance vegetation cover. This will reduce evaporation, improve soil moisture and benefit both adult and juvenile Malleefowl through increased food availability, improved camouflage and a reduction in predation pressure. Although the next prolonged La Niña event will provide an opportunity to test the importance of extended wet conditions on Malleefowl breeding, Malleefowl are still likely to continue to decline on the Eyre Peninsula due to the predicted increase in temperatures and drought conditions. The interacting effects of higher temperatures and drier conditions from climate change, overgrazing by native and introduced herbivores and predation by foxes and cats are key challenges for Malleefowl conservation.

2.8 Miscellaneous

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Data availability statement

Malleefowl activity data are available from DEW upon reasonable request. Fox baiting data are available from the corresponding author upon reasonable request. Other data are freely available from sources as stated in the methods section.

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2.10 Appendix

Table S 2-1 An overview of grid activity of all Eyre Peninsula grids. 'Total' is the total number of mounds; 'Checked' is the number of monitored mounds per grid; 'Not checked' is the number of mounds not monitored that year; 'Active' is the number of active mounds per grid; and 'Ratio' is the proportional mound activity which is calculated as number of active mounds divided by number of monitored mounds.

Year	Cowell			Hincks			Lock			Munjaro			Pinkawilline			Secret Rocks												
	Total	Checked	Not checked	Total	Checked	Not checked	Total	Checked	Not checked	Total	Checked	Not checked	Total	Checked	Not checked	Total	Checked	Not checked										
1995	51	51	0	10	0.2																							
1996	51	51	0	7	0.14																							
1997	51	31	20	5	0.16																							
1998							31	31	0	2	0.06		29	29	0	2	0.07	18	18	0	1	0.06						
1999	51	51	0	7	0.14																							
2000	58	57	1	13	0.23																							
2001	63	54	9	9	0.17																							
2002	65	59	6	6	0.1	31	30	1	1	0.03																		
2003	65	60	5	7	0.12	31	30	1	1	0.03	36	36	0	7	0.19	35	33	2	3	0.09	20	19	1	0	0			
2004	65	59	6	3	0.05						54	52	2	2	0.04													
2005	65	50	15	5	0.1	34	34	0	4	0.12	54	54	0	6	0.11	36	34	2	3	0.09	22	20	2	0	0			
2006	65	50	15	5	0.1	34	34	0	3	0.09	54	54	0	4	0.07	37	35	2	3	0.09	23	21	2	0	0			
2007	65	51	14	4	0.08	34	34	0	1	0.03	55	55	0	0	0	38	35	3	2	0.06								
2008	65	51	14	7	0.14											38	32	6	1	0.03								
2009	66	52	14	6	0.12	34	24	10	1	0.04	57	48	9	4	0.08	38	29	9	4	0.14								
2010	66	52	14	5	0.1	34	24	10	1	0.04	57	46	11	5	0.11	39	30	9	3	0.1								
2011	66	60	6	5	0.08	34	34	0	3	0.09	57	56	1	5	0.09	39	29	10	3	0.1	23	17	6	0	0	0	0	0
2012	66	52	14	6	0.12	34	24	10	3	0.12	57	45	12	3	0.07	40	39	1	5	0.13								
2013	70	53	17	3	0.06	34	24	10	6	0.25	57	43	14	1	0.02	40	30	10	0	0								
2014	70	56	14	2	0.04	34	24	10	4	0.17	58	44	14	1	0.02	41	32	9	0	0								
2015	70	52	18	2	0.04	34	32	2	4	0.12	58	45	13	2	0.04	44	41	3	0	0	23	18	5	0	0	0	0	0
2016	70	52	18	1	0.02	34	32	2	4	0.12	58	58	0	1	0.02	44	41	3	0	0	23	23	0	0	0	0	0	0
2017	70	52	18	1	0.02	34	32	2	1	0.03	58	46	12	0	0	44	43	1	0	0	23	23	0	0	0	0	0	0
2018	70	52	18	0	0	34	32	2	0	0	58	58	0	0	0	44	43	1	0	0	23	23	0	0	0	0	0	0

Table S 2-2 An overview of Malleefowl mound numbers and densities in all Eyre Peninsula grids. 'Total' is the total number of mounds per grid; 'Active' is the number of active mounds per grid. The next two columns show the number of mounds and active mounds per square km.

Year	Cowell		Hincks		Lock		Munyaroo		Pinkawillinie		Secret Rocks													
	Total	Active	Total	Active	Total	Active	Total	Active	Total	Active	Total	Active												
	Mounds per km ²	mounds per km ²	Mounds per km ²	mounds per km ²	Mounds per km ²	mounds per km ²	Mounds per km ²	mounds per km ²	Mounds per km ²	mounds per km ²	Mounds per km ²	mounds per km ²												
1995	51	10	10.74	2.11																				
1996	51	7	10.74	1.47																				
1997	51	5	10.74	1.05																				
1998			31	2	7.75	0.5	29	2	7.25	0.5	18	1	4.5	0.25										
1999	51	7	10.74	1.47																				
2000	58	13	12.21	2.74																				
2001	63	9	13.26	1.89																				
2002	65	6	13.68	1.26	31	1	7.75	0.25																
2003	65	7	13.68	1.47	31	1	7.75	0.25	36	7	9	1.75	8.75	0.75										
2004	65	3	13.68	0.63	54	2	13.5	0.5	54	2	13.5	0.5												
2005	65	5	13.68	1.05	34	4	8.5	1	54	6	13.5	1.5	36	3	9	0.75	22	0	5.5	0				
2006	65	5	13.68	1.05	34	3	8.5	0.75	54	4	13.5	1	37	3	9.25	0.75	23	0	5.75	0				
2007	65	4	13.68	0.84	34	1	8.5	0.25	55	0	13.75	0	38	2	9.5	0.5								
2008	65	7	13.68	1.47					38	1	9.5	0.25	38	1	9.5	0.25								
2009	66	6	13.89	1.26	34	1	8.5	0.25	57	4	14.25	1	38	4	9.5	1								
2010	66	5	13.89	1.05	34	1	8.5	0.25	57	5	14.25	1.25	39	3	9.75	0.75								
2011	66	5	13.89	1.05	34	3	8.5	0.75	57	5	14.25	1.25	39	3	9.75	0.75	23	0	5.75	0	30	3	4.23	0.42
2012	66	6	13.89	1.26	34	3	8.5	0.75	57	3	14.25	0.75	40	5	10	1.25					34	9	4.79	1.27
2013	70	3	14.74	0.63	34	6	8.5	1.5	57	1	14.25	0.25	40	0	10	0					51	0	7.18	0
2014	70	2	14.74	0.42	34	4	8.5	1	58	1	14.5	0.25	41	0	10.25	0					51	0	7.18	0
2015	70	2	14.74	0.42	34	4	8.5	1	58	2	14.5	0.5	44	0	11	0					56	0	7.89	0
2016	70	1	14.74	0.21	34	4	8.5	1	58	1	14.5	0.25	44	0	11	0					59	0	8.31	0
2017	70	1	14.74	0.21	34	1	8.5	0.25	58	0	14.5	0	44	0	11	0					66	0	9.3	0
2018	70	0	14.74	0	34	0	8.5	0	58	0	14.5	0	44	0	11	0					66	1	9.3	0.14

2.10.1 Environmental and habitat variables

General

To calculate lag times we considered the Malleefowl breeding season to begin in September each year, as this is the month when egg laying has usually commenced (Frith 1959).

Rain and Temperature

Long-term monthly rainfall and temperature data were obtained through the Australian Water Availability Project gridded precipitation product (Run 26j). This data product is an interpolation of Bureau of Meteorology data on a 5 km scale (Raupach, Briggs *et al.* 2009; Raupach, Briggs *et al.* 2012).

Monthly rainfall data was used to calculate cumulative seasonal rainfall averages for relevant intervals (, variables 1-7). All rain data were log-transformed and centred to reduce spatial grid effects and avoid scale issues.

Mean monthly minimum and maximum temperatures were used to calculate averages for varying intervals (Table S 2-3, variables 8-15). All temperature data were centred to avoid spatial grid effects.

SOI and IOD

SOI has the strongest effect on south Australian rainfall during the winter months (JJA) therefore we use only these months for analysis (Risbey, Pook *et al.* 2009). SOI data was obtained from the Bureau of Meteorology (BOM 2022) and used to calculate the cumulative averages of relevant intervals (Table S 2-3, variables 16-18).

IOD data was obtained from the Working Group on Surface Pressure (2019) and used to calculate cumulative averages of relevant intervals (Table S 2-3, variables 19-21).

SOI and IOD data were scaled to avoid scale issues but not centred because there was no variation between grids. SOI and IOD were modelled together to look for additive effects, but never together with Rain and Temperature to avoid multicollinearity.

Vegetation

Remotely sensed ground cover data at medium resolution (30 m pixel) was obtained from Vegmachine.net (Beutel, Trevithick *et al.* 2019) and is derived from decades of Landsat satellite imagery. Ground cover data describes vegetation cover at or near ground level and is recorded in January, April, July and October. The three discrete ground cover categories are bare ground, green cover (currently growing plant material) and non-green cover (plant material that is not actively growing). Bare ground and green cover were calculated for relevant periods (Table S 2-3, variables 22-33). Grid vertex coordinates were used to define

polygons to determine cover estimates. Where ground cover data were missing, the average for each location was used.

Fox control

Historical fox control information, to determine if fox baiting occurred within or near the grid, was obtained from Department of Environment and Water (DEW) staff, landowners, or neighbours to the study sites. Fox control was treated as a binary variable, as baiting intensity or frequency could not be determined (Table S 2-3, variables 34-35).

Patch size

The size of the remnant vegetation patch housing each grid was the area around and including the grid that was comprised of immediate, uninterrupted native vegetation. As Malleefowl rarely cross large roads and prefer continuous native vegetation (chapter 4), crops, bitumen roads and structures, e.g. mines, were used as outer boundaries. Patch size was measured with Google Earth and ArcGIS (Table S 2-3, variable 36).

Table S 2-3 All environmental and habitat variables tested in the models and how they were calculated. Description: times are in relation to September of that year when breeding (egg laying) has commenced. Calc: calculation, cu = cumulative, ma = monthly average; Trans: transformation, lc = log-centred, sc = scaled and centred, snc = scaled not centred; na: not applicable; All rainfall was measured in mm, temperatures in °C and distances in km.

	Variable	Description	Cal	Tra
1	RainAut	Rainfall 4 - 6 months prior (March, April, May or MAM)	cu	lc
2	RainWin	Rainfall 1 - 3 months prior (June, July, August or JJA)	cu	lc
3	Rain6Lag1	Rainfall 13 - 18 months prior (JJA)	cu	lc
4	Rain6Lag2	Rainfall 25 - 30 months prior (JJA)	cu	lc
5	Rain12	Rainfall 1 - 12 months prior	cu	lc
6	Rain24	Rainfall 1 - 24 months prior	cu	lc
7	Rain36	Rainfall 1 - 36 months prior	cu	lc
8	TMinAut	Min Temperature 4 - 6 months prior (MAM)	ma	lc
9	TMinWin	Min Temperature 1 - 3 months prior (JJA)	ma	lc
1	TMin12	Min Temperature 1 - 12 months prior	ma	lc
1	TMin24	Min Temperature 1 - 24 months prior	ma	lc
1	TMaxAut	Max Temperature 4 - 6 months prior (MAM)	ma	lc
1	TMaxWin	Max Temperature 1 - 3 months prior (JJA)	ma	lc
1	TMax12	Max Temperature 1 - 12 months prior	ma	lc
1	TMax24	Max Temperature 1 - 24 months prior	ma	lc
1	SOIWin	SOI 1 - 3 months prior	ma	snc
1	SOIWinLag1	SOI 13 - 15 months prior (JJA one-year lag)	ma	snc
1	SOIWinLag2	SOI 25 - 27 months prior (JJA two-year lag)	ma	snc
1	IODWinSpr	IOD JJASO current year	ma	snc
2	IODWinSprL	IOD 11 - 15 months prior (JJASO one-year lag)	ma	snc
2	IODWinSprL	IOD 23 - 27 months prior (JJASO two-year lag)	ma	snc
2	BareSum	Bare ground 7 -9 months prior (December, January, February or	%	sc
2	BareAut	Bare ground 4 - 6 months prior (MAM)	%	sc
2	BareWin	Bare ground 1 - 3 months prior (JJA)	%	sc
2	BareSpr	Bare ground 10 - 12 months prior (September, October,	%	sc
2	Bare12	Bare ground 1 - 12 months prior	%	sc
2	Bare24	Bare ground 1 - 24 months prior	%	sc
2	GreenSum	Green vegetation at ground level 7 - 9 months prior (DJF)	%	sc
2	GreenAut	Green vegetation at ground level 4 - 6 months prior (MAM)	%	sc
3	GreenWin	Green vegetation at ground level 1- 3 months prior (JJA)	%	sc
3	GreenSpr	Green vegetation at ground level 10 - 12 months prior (SON)	%	sc
3	Green12	Green vegetation at ground level 1 - 12 months prior	%	sc
3	Green24	Green vegetation at ground level 1 - 24 months prior	%	sc
3	FoxCtrlGrd	Did fox baiting take place in that grid in the monitoring year?	na	na
3	FoxCtrlNear	Did fox baiting take place within 2km of the grid in the monitoring	na	na
3	PatchSize	Immediate, uninterrupted native vegetation around the grid	na	snc

2.10.2 Probability of activity - models

Table S 2-4 Estimated marginal means, i.e. probabilities of year-to-year and grid-to-grid mound activity differences. The first two rows show the model formulas and remaining rows the modelling results for the 2003-2018 period for 4 grids on the left and the 2010-2018 period for 5 grids on the right. Year-to-year models are on the left and grid-to-grid on the right within each period. Odds: odds ratio, CI: confidence intervals, p: p-value (significant results in bold font).

Formula (Year)	glmer(Active ~ Year + (1 Mound), data= MoundsEMM, family= binomial, control = glmerControl(optimizer = "bobyqa", optCtrl = list(maxfun=2e5)))											
Formula (Grid)	glmer(Active ~ GridName + (1 Mound), data= MoundsEMM, family= binomial, control = glmerControl(optimizer = "bobyqa", optCtrl = list(maxfun=2e5)))											
	2003 - 2018						2010 - 2018					
	Year			Grid			Year			Grid		
Predictors	Active			Active			Active			Active		
	Odds	CI	p	Odds	CI	p	Odds	CI	p	Odds	CI	p
(Intercept)	0.32	0.18-0.55	<0.001	0.15	0.11-0.21	<0.001	0.37	0.21-0.64	<0.001	0.15	0.10-0.23	<0.001
Year [2004]	0.35	0.12-1.02	0.054									
Year [2005]	0.9	0.42-1.93	0.796									
Year [2006]	0.71	0.33-1.56	0.4									
Year [2007]	0.29	0.11-0.75	0.011									
Year [2008]	0.72	0.28-1.86	0.497									
Year [2009]	0.71	0.33-1.56	0.399									
Year [2010]	0.68	0.30-1.50	0.335									
Year [2011]	0.76	0.35-1.65	0.493				1.48	0.70-3.15	0.306			
Year [2012]	0.84	0.39-1.80	0.651				1.85	0.88-3.91	0.106			
Year [2013]	0.44	0.18-1.03	0.058				0.48	0.20-1.16	0.103			
Year [2014]	0.29	0.11-0.75	0.011				0.33	0.12-0.85	0.023			
Year [2015]	0.34	0.14-0.84	0.02				0.37	0.15-0.94	0.037			
Year [2016]	0.24	0.09-0.66	0.005				0.28	0.10-0.78	0.015			
Year [2017]	0.08	0.02-0.35	0.001				0.08	0.02-0.37	0.001			
Year [2018]	0	0.0-2.4E+55	0.815				0.04	0.01-0.31	0.002			
SD (Intercept)	1.47			1.32			1			1		
SD (Observations)	2.72			2.72			2.72			2.72		
GridName [Hincks]				1.54	0.94-2.51	0.084				2.29	1.24-4.23	0.008
GridName [Lock]				0.94	0.60-1.48	0.8				1.45	0.75-2.81	0.272
GridName [Munyaroo]				0.91	0.54-1.52	0.708				1.05	0.49-2.26	0.891
GridName [Secret Rocks]										1.28	0.68-2.42	0.45
Random Effects												
σ ²	3.29			3.29			3.29			3.29		
N	80 Mound			80 Mound			69 Mound			69 Mound		
Observations	1178			1178			588			588		
Marginal R ² / Conditional R ²	0.836 / 0.843			0.009 / 0.032			0.306 / NA			0.025 / NA		

2.10.3 Probability of activity - plots

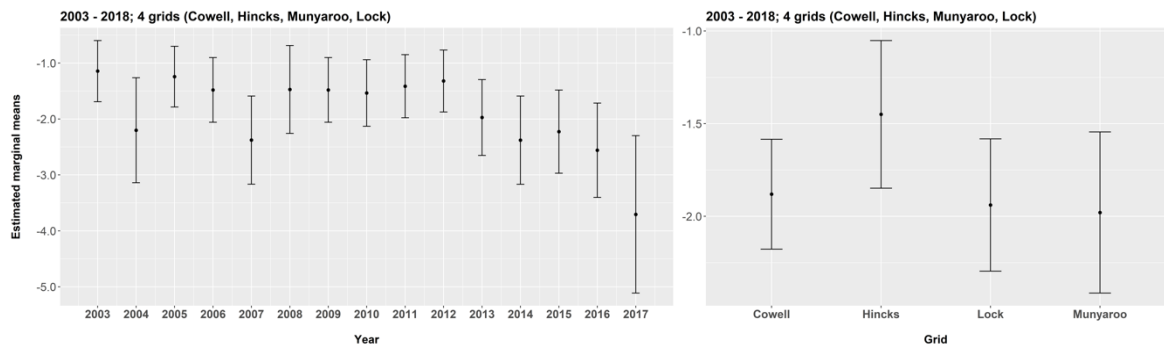


Figure S 2-1 Estimated marginal means, i.e. probabilities of year-to-year mound activity and grid-to-grid differences shown for the period from 2003 – 2018 for Malleefowl monitoring grids in Cowell, Hincks, Munyaroo and Lock. Left: Year to year variation. Right: Grid to grid variation. Error bars show the 95% confidence intervals. 2018 had to be removed for scale reasons.

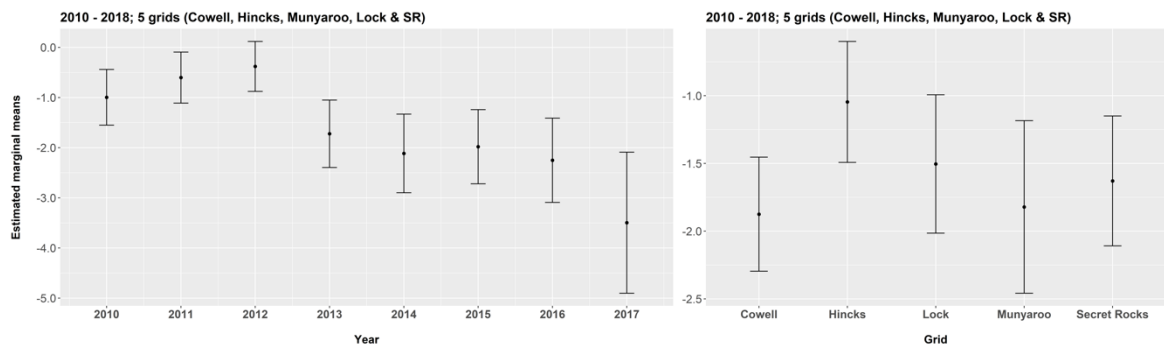


Figure S 2-2 Estimated marginal means, i.e. probabilities of year-to-year and grid-to-grid differences shown for the period from 2010 – 2018 for Malleefowl monitoring grids in Cowell, Hincks, Munyaroo, Lock and Secret Rocks. Left: Year to year variation. Right: Grid to grid variation. Error bars show the 95% confidence intervals. 2018 had to be removed for scale reasons.

2.10.4 Variables influencing activity

Table S 2-5 Model outputs with $\Delta AICc \leq 4$ (5 grids, 2010-2018, $n = 45$). $AICc$ = corrected Akaike Information Criterion; R^2m / R^2c = Marginal / Conditional r-squared (%); SE = Standard Error; LCI/UCI = Lower/Upper confidence intervals; P = P-value.

Model	AICc	R^2m	R^2c	Variables	Estimate	SE	LCI, UCI	p
ActRatioGrid	~ 181.45	14.07	24.18	(Intercept)	-3.21	0.29	(-3.79,-2.64)	0.00
Rain24.lc	+			Rain24.lc	5.09	1.32	(2.5,7.68)	0.00
TMaxWin.lc	+			TMaxWin.lc	-16.67	6.00	(-28.44,-4.91)	0.01
(1 Year) + (1 Grid)				sd__(Intercept) Year	0.49			
				sd__(Intercept) Grid	0.44			
ActRatioGrid	~ 181.73	15.14	25.4	(Intercept)	-3.34	0.31	(-3.94,-2.74)	0.00
SOIWinLag2.snc	+			SOIWinLag2.snc	0.94	0.26	(0.44,1.44)	0.00
BareWin.sc	+			BareWin.sc	-0.64	0.19	(-1.01,-0.26)	0.00
(1 Year) + (1 Grid)				sd__(Intercept) Year	0.49			
				sd__(Intercept) Grid	0.46			
ActRatioGrid	~ 184.19	15.37	25.36	(Intercept)	-3.45	0.38	(-4.2,-2.69)	0.00
SOIWinLag2.snc	+			SOIWinLag2.snc	0.94	0.26	(0.43,1.44)	0.00
BareWin.sc	+			BareWin.sc	-0.63	0.19	(-1.01,-0.26)	0.00
PatchSize	+			PatchSize	0.18	0.38	(-0.57,0.92)	0.64
(1 Year) + (1 Grid)				sd__(Intercept) Year	0.49			
				sd__(Intercept) Grid	0.45			

ActRatioGrid	~	184.33	15.25	25.44	(Intercept)	-3.37	0.33	(-4.01,-2.73)	0.00
SOIWinLag2.snc	+				SOIWinLag2.snc	0.94	0.26	(0.44,1.44)	0.00
BareWin.sc	+				BareWin.sc	-0.64	0.19	(-1.01,-0.26)	0.00
FoxCtrlGrid	+				FoxCtrlGrid1	0.16	0.58	(-0.98,1.29)	0.78
(1 Year) + (1 Grid)					sd__(Intercept) Year	0.49			
					sd__(Intercept) Grid	0.46			
ActRatioGrid	~	184.40	15.14	25.41	(Intercept)	-3.35	0.35	(-4.04,-2.66)	0.00
SOIWinLag2.snc	+				SOIWinLag2.snc	0.94	0.26	(0.44,1.44)	0.00
BareWin.sc	+				BareWin.sc	-0.64	0.19	(-1.01,-0.26)	0.00
FoxCtrlNear	+				FoxCtrlNear1	0.02	0.33	(-0.63,0.66)	0.96
(1 Year) + (1 Grid)					sd__(Intercept) Year	0.49			
					sd__(Intercept) Grid	0.46			
ActRatioGrid	~	184.56	7.66	26.49	(Intercept)	-3.22	0.36	(-3.93,-2.51)	0.00
Rain24.lc	+				Rain24.lc	4.63	1.56	(1.58,7.68)	0.00
(1 Year) + (1 Grid)					sd__(Intercept) Year	0.80			
					sd__(Intercept) Grid	0.45			
ActRatioGrid	~	184.94	15.4	24.83	(Intercept)	-3.32	0.38	(-4.06,-2.59)	0.00
Rain24.lc	+				Rain24.lc	4.33	1.40	(1.58,7.09)	0.00
TMaxWin.lc	+				TMaxWin.lc	-17.73	5.78	(-29.05,-6.4)	0.00
BareWin.sc	+				BareWin.sc	-0.25	0.18	(-0.6,0.11)	0.17
PatchSize	+				PatchSize	0.15	0.38	(-0.6,0.89)	0.70
(1 Year) + (1 Grid)					sd__(Intercept) Year	0.46			
					sd__(Intercept) Grid	0.45			
ActRatioGrid	~	185.07	15.29	24.89	(Intercept)	-3.25	0.32	(-3.87,-2.63)	0.00
Rain24.lc	+				Rain24.lc	4.34	1.40	(1.59,7.08)	0.00
TMaxWin.lc	+				TMaxWin.lc	-17.75	5.77	(-29.06,-6.44)	0.00
BareWin.sc	+				BareWin.sc	-0.25	0.18	(-0.6,0.11)	0.17
FoxCtrlGrid	+				FoxCtrlGrid1	0.09	0.58	(-1.05,1.22)	0.88
(1 Year) + (1 Grid)					sd__(Intercept) Year	0.46			
					sd__(Intercept) Grid	0.46			
ActRatioGrid	~	185.07	15.3	24.88	(Intercept)	-3.26	0.34	(-3.93,-2.59)	0.00
Rain24.lc	+				Rain24.lc	4.33	1.40	(1.59,7.07)	0.00
TMaxWin.lc	+				TMaxWin.lc	-17.86	5.79	(-29.2,-6.51)	0.00
BareWin.sc	+				BareWin.sc	-0.25	0.18	(-0.6,0.1)	0.17
FoxCtrlNear	+				FoxCtrlNear1	0.04	0.33	(-0.6,0.69)	0.89
(1 Year) + (1 Grid)					sd__(Intercept) Year	0.45			
					sd__(Intercept) Grid	0.46			
ActRatioGrid	~	185.35	11.55	27.64	(Intercept)	-3.23	0.35	(-3.91,-2.55)	0.00
Rain24.lc	+				Rain24.lc	4.07	1.56	(1.01,7.14)	0.01
TMaxAut.lc	+				TMaxAut.lc	-8.39	6.25	(-20.65,3.86)	0.18
(1 Year) + (1 Grid)					sd__(Intercept) Year	0.73			
					sd__(Intercept) Grid	0.44			

2.10.5 Additional References

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Chapter 3:

Spatial genetic structure and limited gene flow in fragmented populations of the threatened Malleefowl (*Leipoa ocellata*)



Figure on previous page: Cowell, November 2017, tagged and processed Malleefowl just before its release. This bird died 5 days after capture. It was probably killed by a fox. Credit: Peri Stenhouse

3.1 Statement of Authorship

Title of Paper	Spatial genetic structure and limited gene flow in fragmented populations of the threatened Malleefowl (<i>Leipoa ocellata</i>).			
Publication status	<input checked="" type="checkbox"/> Published	<input type="checkbox"/> Accepted for Publication	<input type="checkbox"/> Submitted for Publication	<input type="checkbox"/> Publication style
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By signing the Statement of Authorship, each author certifies that the candidate's stated contribution to the publication is accurate, that permission is granted for the candidate to include the publication in the thesis; and that the sum of all co-author contributions is equal to 100% less the candidate's stated contribution.

Name of Principal Author (Candidate)	Peri Stenhouse	
Contribution to the paper	Conceptualization, methodology, data curation, formal analysis, laboratory analysis, writing – original draft preparation, writing – review and editing. Responses to reviewers.	
Overall percentage (%)	75%	
Certification	This paper reports on original research I conducted during the period of my Higher Degree by Research candidature and is not subject to any obligations or contractual agreements with a third party that would constrain its inclusion in this thesis. I am the primary author of this paper.	
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Contribution to the Paper	Formal analysis, writing – review and editing.	
Signature		Date: 1/4/22

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Contribution to the Paper	Formal analysis, writing – review and editing.	
Signature		Date: 1/4/22

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Contribution to the Paper	Conceptualisation, Methodology, writing – review and editing.	
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Contribution to the Paper	Conceptualisation, Methodology, laboratory analysis, writing – review and editing.	
Signature		Date: 1/4/22

3.2 Abstract

The Malleefowl (*Leipoa ocellata*) is a threatened megapode bird that persists on the Eyre Peninsula of South Australia—an area that has undergone substantial clearance of native vegetation over the past 200 years. Habitat loss and fragmentation can negatively affect long-term conservation status by creating small and isolated subpopulations that lead to reduced gene flow, increased genetic drift, and inbreeding. In this study, we test whether Malleefowl on the Eyre Peninsula exhibit population structure that may have resulted from recent anthropogenic land cover changes. We used reduced-representation high-throughput DNA sequencing to obtain a dataset of 17,851 single nucleotide polymorphisms (SNPs). Our results suggest that Malleefowl are not genetically homogeneous across the Eyre Peninsula and that habitat fragmentation has likely driven differentiation of at least two distinct populations by decreasing opportunities for gene flow. We also provide preliminary genetic evidence for female-biased dispersal in Malleefowl. Our study highlights the potential problems caused by anthropogenic land cover changes for threatened Malleefowl populations and underscores the need for ongoing monitoring. We emphasise the importance of both large continuous native vegetation patches and small connecting habitat fragments even in highly fragmented landscapes. We encourage land managers to facilitate between-patch movement by improving native vegetation cover and controlling overabundant herbivores to improve habitat quality by allowing natural regrowth of existing native vegetation.

3.3 Keywords

Conservation, habitat fragmentation, semi-arid zone, dispersal, megapodes, DArT-seq

3.4 Introduction

Habitat loss and fragmentation create small, isolated subpopulations that can lead to reduced gene flow, increased genetic drift and inbreeding (Almeida-Rocha, Soares *et al.* 2020; Callens, Galbusera *et al.* 2011; Frankham, Ballou *et al.* 2002; Schlaepfer, Braschler *et al.* 2018; Stevens, Harrisson *et al.* 2018). Inbreeding depression and the loss of genetic diversity contribute to extinction risk and must be considered in conservation management of threatened species (Frankham 2005; Reed, Briscoe *et al.* 2002). Connectivity and thus gene flow between remnant habitat patches, facilitated by habitat corridors or translocations, can be crucial in counteracting these genetic pressures (Biebach and Keller 2012; Blyth, Christmas *et al.* 2020; Christie and Knowles 2015; Frankham, Ballou *et al.* 2011; Weeks, Sgro *et al.* 2011).

To determine the effects of habitat fragmentation and make appropriate management decisions it is important to assess spatial genetic structure among habitat fragments and to identify current and past dispersal and gene flow patterns (Cayuela, Rougemont *et al.* 2018). Dispersal can be studied at different spatial and temporal scales. Radio or GPS tracking can provide accurate information on short-term movement over weeks, months and years, while capture-recapture studies, e.g., bird banding, can provide movement information over generations. However, movement does not equal gene flow and the tracked individuals must first breed in the new region to pass on genetic material (Ronce 2007). Genetic data can provide direct or indirect evidence of dispersal leading to gene flow at a landscape scale, for the recent past or going back thousands of generations.

Our study centres on the effects of fragmentation and loss of native vegetation on Malleefowl (*Leipoa ocellata*), a threatened ground-dwelling bird that is endemic to the semi-arid parts of southern Australia. We focus on the Eyre Peninsula in South Australia, which covers approximately 80,000 km² but encompasses mainly highly fragmented areas of suitable Malleefowl habitat. Over 60% of the native vegetation cover on the Eyre Peninsula has been cleared for agricultural use since European settlement in the 19th Century and the remaining native vegetation is now largely restricted to marginal areas unsuitable for agriculture (Brandle 2010; NREP 2017). Malleefowl persist in remnant native patches on the Eyre Peninsula but their overall population size is declining (Stenhouse and Moseby 2022), and the genetic structure and connectedness of local subpopulations are unclear.

Little research into Malleefowl genetics has been conducted to date (Cope, Bertozzi *et al.* 2016; Donnellan 1997). Previous studies using mitochondrial DNA and nuclear microsatellite markers have suggested that Malleefowl dispersed from western to eastern Australia and—while there are no recognised subspecies of Malleefowl—there is evidence

for genetically distinct western and eastern populations separated by the Flinders Ranges (Cope, Mulder *et al.* 2014). These studies also found high levels of diversity but low levels of differentiation between mitochondrial haplotypes and that populations have an isolation-by-distance structure.

Recent research on Malleefowl movement on the Eyre Peninsula using GPS tracking showed that birds can move up to 10 km a day and range over 23 km while not breeding (chapter 4). Their home ranges vary between 45 and 2,200 ha, limited by available native vegetation. Malleefowl have been recorded traversing closely spaced patches of native vegetation within an agricultural matrix to resettle in new habitat patches (chapter 5). Although not yet proven in Malleefowl, in most birds females are more likely to disperse than males (Greenwood 1980). This has been observed in other gallinaceous birds in the past (Earl, Fuhlendorf *et al.* 2016; Giesen and Braun 1993), albeit with quite different life histories. Male birds are more likely to be philopatric (i.e., show higher site fidelity) with some exceptions in communal living birds and the Anatidae (Clarke, Sæther *et al.* 1997; Williams and Rabenold 2005). Malleefowl males aggressively defend their mounds and female mates against intruders during the breeding season (Cope, Mulder *et al.* 2014; Frith 1959), but when non-breeding, groups of Malleefowl have been seen feeding together. Natal dispersal has been observed in Malleefowl chicks, which were found to disperse up to 6.6 km within a month of release (Benshemesh 1992). However, our knowledge of Malleefowl dispersal within fragmented landscapes is limited and it is not known if there has been gene flow between birds living in isolated habitat patches on the Eyre Peninsula.

Consequently, we aimed to use single nucleotide polymorphisms (SNPs) to test whether anthropogenic land clearing and habitat fragmentation have affected the population structure of Malleefowl on the Eyre Peninsula. We expected lower genetic diversity, evidence of genetic drift including the formation of distinct populations, and higher degrees of inbreeding in individuals living in smaller and more isolated habitat fragments. Another aim was to examine whether there is evidence for sex bias in past Malleefowl dispersal. Based on previous reports of predominantly female-biased dispersal in birds, we also expected to see female dispersal in Malleefowl. Furthermore, we assessed the effects of habitat intactness on the genetic structure of Eyre Peninsula Malleefowl by studying the remaining native vegetation cover at and around sample collection sites. Combining these different approaches and temporal perspectives on Malleefowl movement will help fill knowledge gaps and result in better-informed management decisions that will help conserve the species.

3.5 Methods

3.5.1 Study area

Specimens were collected on or near the Eyre Peninsula in South Australia where Malleefowl persist in patches of native vegetation. The maximum distance between sample locations was approximately 750 km with the densest concentration of samples in the northeast, in an area of approximately 4200 km² (inset Figure 3-1).

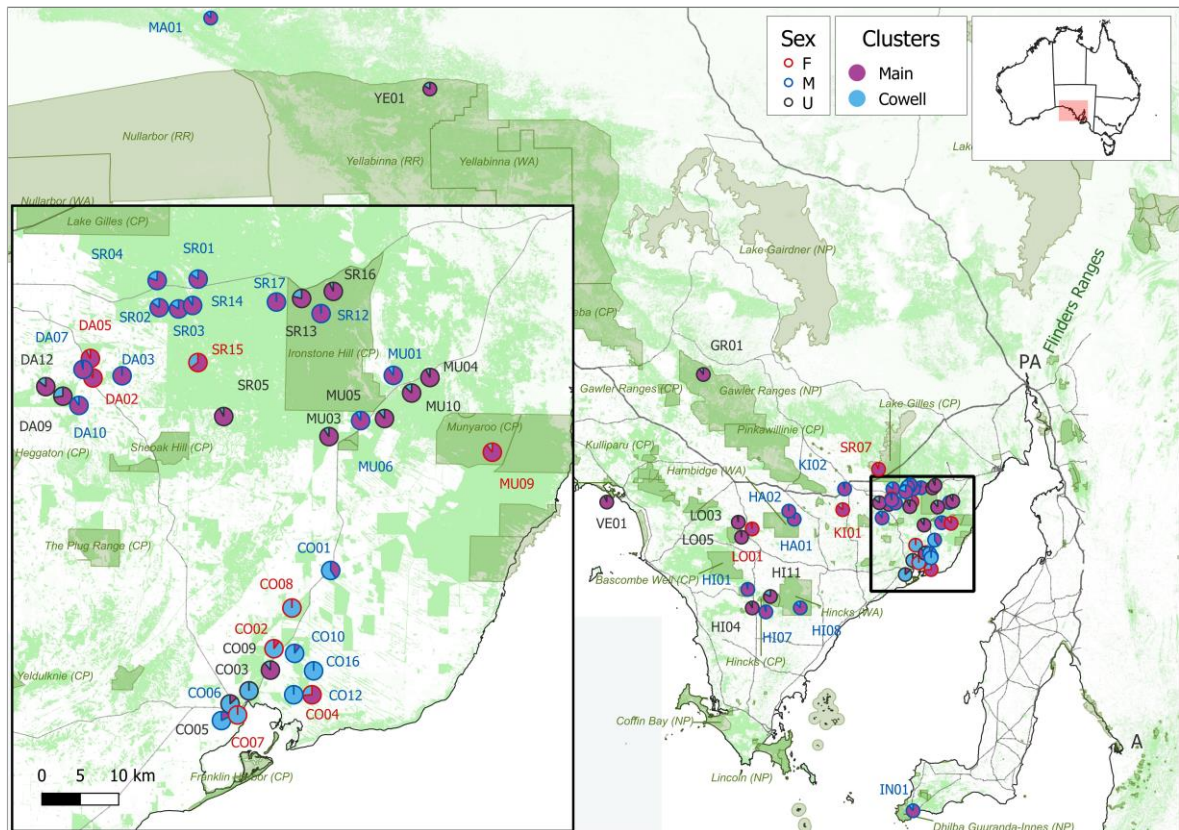


Figure 3-1 Malleefowl sample collection sites on and near the Eyre Peninsula, South Australia. The predominant colour of the pie-chart matches that of the clusters identified by the PCA. The proportions within each pie chart reflect the LEA plot ancestry composition of samples ($K = 2$). Pie-chart line and label colours indicate the sex of the individual. The light green areas are native vegetation, the darker green highlights are nature protection and wilderness areas under South Australian Government management.

3.5.2 Sample collection

We assembled a total of 285 samples from Malleefowl collected between 1911 and 2019. These included:

- Blood and feathers from nine Malleefowl that were live captured between 2016 and 2019 and subsequently included in a GPS tracking study (chapter 4). Blood was collected using glass capillaries (Booth 1985; Booth 1987; King, Kirkpatrick *et al.* 1996) from the metatarsal vein above the inter-tarsal joint and stored in Queen's lysis buffer solution (Seutin, White *et al.* 1991).

- Feathers, carcasses and egg membranes collected opportunistically during field surveys in the region.
- Skin and toepad samples from preserved museum skins held in the South Australian Museum (SAM), the Australian Museum (AM), the Natural Resource Management Board (NRM) and the South Australian Department for Environment and Water (DEW).
- Frozen and ethanol preserved feathers and tissue samples from the SAM.
- Feathers and tissue (from road-killed Malleefowl) collected by local residents, volunteers, NRM staff, and DEW staff.

We selected 117 of the 285 samples for DNA extraction based on age, condition and the best spatiotemporal coverage across the Eyre Peninsula.

3.5.3 DNA extraction

We extracted DNA using a Qiagen DNeasy kit (QIAGEN Inc., Valencia, CA, USA). As starting material, we used approximately 5-10 mm of the base of a feather calamus, 20-30 mm² of egg membrane, 2-3 mm³ of tissue (liver, muscle or toe pad), 20 µl of yolk sac, or 20 µl of blood in buffer solution. We added 20 µl of 1M DTT to feather samples but otherwise followed the manufacturer's instructions. DNA concentration was measured with a Quantus™ fluorometer, following the manufacturer's instructions (Promega).

Some samples returned very low DNA concentrations, therefore only 94 samples (Table S 3-1) with the highest DNA concentration were sent to Diversity Arrays Pty Ltd (DART), a private company, for single nucleotide polymorphism (SNP) genotyping. Diversity Arrays produced SNP data using DART-seq, a reduced representation sequencing method (Akbari, Wenzl *et al.* 2006; Cruz, Kilian *et al.* 2013; Kilian, Wenzl *et al.* 2012). The output was made available as a coded matrix of SNP loci by sample and raw FASTQ files for each sample. From the 94 samples, nine failed to generate any data due to lack of Malleefowl DNA and/or microbial contamination, leaving 85 samples for further analyses.

3.5.4 SNP Filtering

Preparation of the SNP dataset was performed in the R package *dartR* (Gruber, Unmack *et al.* 2018). Preliminary visualisation of the dataset using a smear plot showed several individuals with a high degree of missingness, and so SNP filtering on call rate was performed iteratively in three steps as described in O'Leary, Puritz *et al.* (2018): 1. Locus call rate <0.50, individual call rate <0.10; 2. Locus call rate <0.60, individual call rate <0.30; and 3. Locus call rate <0.70, individual call rate <0.50. SNP and sample metrics were recalculated after each filtering step. The dataset was then filtered once more based on

Massault, Jones *et al.* (2021) on call rate with a threshold of 0.80 and a reproducibility threshold of 0.90, monomorphs and secondaries were removed, and SNPs with a minor allele frequency (MAF) <0.05 were filtered out.

3.5.5 Kinship analysis to identify duplicate samples

To identify duplicate samples from the same bird, a kinship analysis was performed using an identity-by-descent approach (KING method of moment) in the R package *SNPRelate* (Zheng and Zheng 2013), producing kinship coefficients for pairs of samples within the dataset. Although a kinship coefficient of ~0.5 typically indicates identical genotypes (and ~0.25 immediate siblings or parent/offspring) (Lopes, Silva *et al.* 2013), due to the high degree of missingness in the dataset we determined pairings with a kinship coefficient >0.3 to be duplicates. In the case of pairings producing a coefficient above this threshold, the sample with the worst DNA concentration after extraction was removed from the dataset before further analysis.

3.5.6 Sex assignment

We used two methods to identify the sex of birds included in the genetic analyses. Initially, we performed DNA-based sexing on nine samples that were live captured (see section 2.2) using polymerase chain reaction (PCR) and primers 2550F/2718R following Çakmak, Pekşen *et al.* (2017). Second, to confirm the PCR results and determine the gender of the remaining samples, we used the DArT-seq FASTQ data for all samples in a genetic sex assignment approach following Onley, Austin *et al.* (2021a). Briefly, we used the PALEOMIX v1.2.9 (Schubert, Ermini *et al.* 2014) pipeline to process the FASTQ data: AdapterRemoval v2.3.1 (Schubert, Lindgreen *et al.* 2016) was used to trim residual adapter sequences (using default parameter values) and we aligned the FASTQ data to the genome of the domestic chicken (*Gallus gallus domesticus*; galGal5) using the *mem* algorithm (with default parameter values) in BWA v0.7.15 (Li and Durbin 2009). Next, we extracted per-scaffold read counts using the *idxstats* command in SAMtools v1.9 (Li, Handsaker *et al.* 2009) and constructed two binomial models (one for each sex) comparing the Z chromosome with the read count mapping to the autosomes using the Python script *sexassign* from Gower, Fenderson *et al.* (2019). We assigned a sex only when the likelihood ratio test between the intercept only model and the binomial model for a sex was highly significant ($p < 0.001$). Our sex assignment protocol differs from Onley, Austin *et al.* (2021a) in that we considered samples with lower read-dosage for the Z to be females because in birds females are the heterogametic sex (as opposed to males in mammals). Like Onley, Austin *et al.* (2021a), we observed a depletion of sex chromosome linked markers in our DArT-seq data versus the number expected if markers are randomly distributed throughout

the genome (though read-dosage of the autosomes was positively correlated with chromosome length, as expected). Following their protocol, we calculated and applied a correction of 1.655 to the Z chromosome read counts prior to analysis with *sexassign*.

3.5.7 Genetic diversity and spatial genetic structure

Principal component analysis (PCA) was used to identify and visualise population structure. PCA was performed using the R package *smartsnp* (Herrando-Pérez, Tobler *et al.* 2021), a package designed for fast analysis of large genetic datasets. To determine genetic clustering in the samples, the PCA was performed without *a priori* assignment of sample locations.

Using the R package *LEA* (version 3.4.0) (Frichot and François 2015), we then estimated the number of ancestral populations (K) via sparse non-negative matrix factorisation (sNMF) (Frichot, Mathieu *et al.* 2014). Ten repetitions were performed for each K value from 1 to 10. Cross-entropy was then applied to the most appropriate K value(s), with ancestry proportions for the run with the lowest cross-entropy visualised using R package *ggplot2* (Wickham 2016). We also used R package *hierfstat* (Goudet 2005) to estimate the genetic differentiation (F_{ST}) between the identified clusters and to estimate the inbreeding coefficient (F_{IS}) and the observed (H_O) and expected (H_E) heterozygosity for each cluster.

We quantified correlations between geographic (Euclidean) and genetic distance (calculated using the percentage pairwise method), thereby testing isolation-by-distance (IBD), by applying a Mantel Test with 999 permutations (Mantel 1967), using R package *ade4* (Dray and Dufour 2007). We used the natural logarithm of the geographic distance according to Rousset (2000). IBD analyses were run on all samples combined, on samples from each genetic cluster identified with $K=2$, and on samples from each genetic cluster identified with $K=3$.

3.5.8 Sex-biased dispersal

Spatial genetic correlation tests were then performed using GenAlEx6 (Peakall and Smouse 2006) to identify any evidence of sex-biased dispersal following Onley, Austin *et al.* (2021b). To meet memory requirements of GenAlEx, we randomly subset the data to 5,000 SNPs before analysing all samples combined ($n=35$), and then males ($n=24$) and females ($n=11$) separately (see section 3.2 for justification of sample size). Pairwise geographic distance was calculated from the GPS coordinates of each sample and correlated with a matrix of genetic distance to produce a correlation coefficient (r). GenAlEx performs 999 permutations of this procedure and then produces a spatial correlogram, where if r exceeds

the upper and lower confidence intervals of spatial homogeneity, the correlation is considered significant and spatial heterogeneity declared.

3.5.9 Effects of habitat intactness on spatial genetic structure

We used the percentage of native vegetation cover around each sample location as a measure of habitat intactness. Using QGIS (v3.16; QGIS Development Team 2021), we calculated the percentage of native vegetation in a 12 km radius around each sample location, based on chapter 4 where the furthest distance travelled by a tracked Malleefowl was over 23.3 km (i.e. roughly the diameter of a circle with a radius of 12 km). The Shapiro-Wilk test for normality and Levene's test showed that the native vegetation cover data did not meet normality and equal variance assumptions, so we applied the non-parametric Mann-Whitney U Test to determine if habitat intactness played a role in the clusters identified by the PCA. Spatial vegetation data (most likely landcover 2010-2015) were obtained from the South Australian Government Data Directory (Government of South Australia 2022). See Figure S 3-5 for relationship between ancestry proportions and native vegetation distribution.

3.5.10 Effects of storage and sample type

We used logistic regression (glm) with a binomial error distribution to investigate the effects of sample type, storage method and age on the suitability of DNA for use in SNP genotyping using the 94 samples that were sent to Diversity Arrays. The response variable was DNA usability (1 if successfully used after all filtering stages, 0 if not). We removed one sample that was the only sample of type "yolk". Explanatory variables were *sample type* with three categories (tissue, containing blood, liver or muscle samples; *others*, containing egg membrane and toe samples; and *feathers*); storage type with three categories (*chemical*, containing samples stored in buffer solution or ethanol; *room*, containing samples stored at room temperature or taxidermied samples; and *frozen*) and age (years since collection date). We used the Hosmer-Lemeshow Goodness of Fit Test to assess the fit of our glm.

3.5.11 Data analysis

All data analyses were performed with R Studio v1.4.1106 (R Core Team 2020) with *tidyverse* (Wickham, Averick *et al.* 2019) for data cleaning, *car* (Fox and Weisberg 2019) for equal variance testing, *ggstatsplot* (Patil 2021) for comparative statistics, *ResourceSelection* (Lele, Keim *et al.* 2019) for the goodness of fit test (Hosmer Jr, Lemeshow *et al.* 2013) and *effects* (Fox and Hong 2009) for the glm plot. Maps were made with QGIS v3.16 (QGIS Development Team 2021).

3.6 Results

3.6.1 SNP Filtering

Of the 94 samples sent to Diversity Arrays, only 85 returned sequence data. The dataset for the 85 individuals had 43,441 SNPs and 30.98% missing data before filtering. After filtering and kinship analysis, 18 individuals were excluded due to excessive missing data, 11 were excluded due to being duplicates and one (from Lincoln National Park) was excluded because it was possibly a translocated bird with an unknown origin (Table S 3-1), leaving 55 unique individuals remaining with 17,851 SNPs and 7.9% missing data. Ten of the 11 duplicate samples were collected within 260 m of each other and were therefore almost certainly from the same bird. One pairing was 45 km apart and is most likely due to a labelling error.

3.6.2 Sex assignment

PCR sex assignment of the nine trapped individuals identified eight males and one female. For the *sexassign* pipeline assignment, 41 of the 85 samples (in the final SNP dataset) had sufficient mapped sequencing reads (>40,000) to generate a consistent pattern of read dosage per chromosome and to assign a sex: 12 females and 29 males. Five males and one female had to be removed because they were duplicates, leaving 35 unique individuals, with 11 females and 24 males. The *sexassign* results for eight of the nine trapped birds were in 100% concordance with the PCR sex assignment results. The ninth trapped bird was identified as a male via PCR but could not be sexed using the *sexassign* pipeline. In summary, we were able to sex 36 unique individuals, with 11 females and 25 males.

3.6.3 Genetic diversity and spatial genetic structure

A Mantel test showed significant correlation between the genetic distance and the natural logarithm of the geographic distance for all individuals in our dataset ($p = 0.001$, Figure S 3-2). Further, the PCA indicated two genetic clusters separated along PC1, which accounted for 48.23% of the variance. One cluster comprised ten birds collected near the town of Cowell (on the east coast of the Eyre Peninsula; Inset Figure 3-1) between 1985 and 2017 (*Cowell*; blue in Figure 3-1 and Figure 3-2), with the second comprising all remaining samples (*Main*; collected between 1994 and 2019; purple in Figure 3-1 and Figure 3-2). A Mantel test applied to the *Main* individuals revealed significant isolation-by-distance ($p = 0.001$, Figure S 3-2), but no significant correlation was observed between genetic and geographic distance for the individuals from the *Cowell* cluster (Figure S 3-2). There was also some evidence for residual structure within the *Main* cluster, with eight birds (*Dart* cluster) separating from the rest along PC2, which comprised 29.6% of the variance (Figure 3-2)—these birds were sampled near Yalanda, about 30 km south of Kimba on the

Cowell Kimba Road, plus one from Secret Rocks Nature Reserve (also see Figure S 3-1, Figure S 3-4). For this three cluster scenario, a Mantel test applied to the individuals from the *Dart* and *Main* clusters separately revealed significant isolation-by-distance ($p = 0.001$ for *Dart* and $p = 0.002$ for *Main*, Figure S 3-2). We hereafter present results for two (*Main* and *Cowell*) and three (*Main*, *Dart* and *Cowell*) genetic clusters.

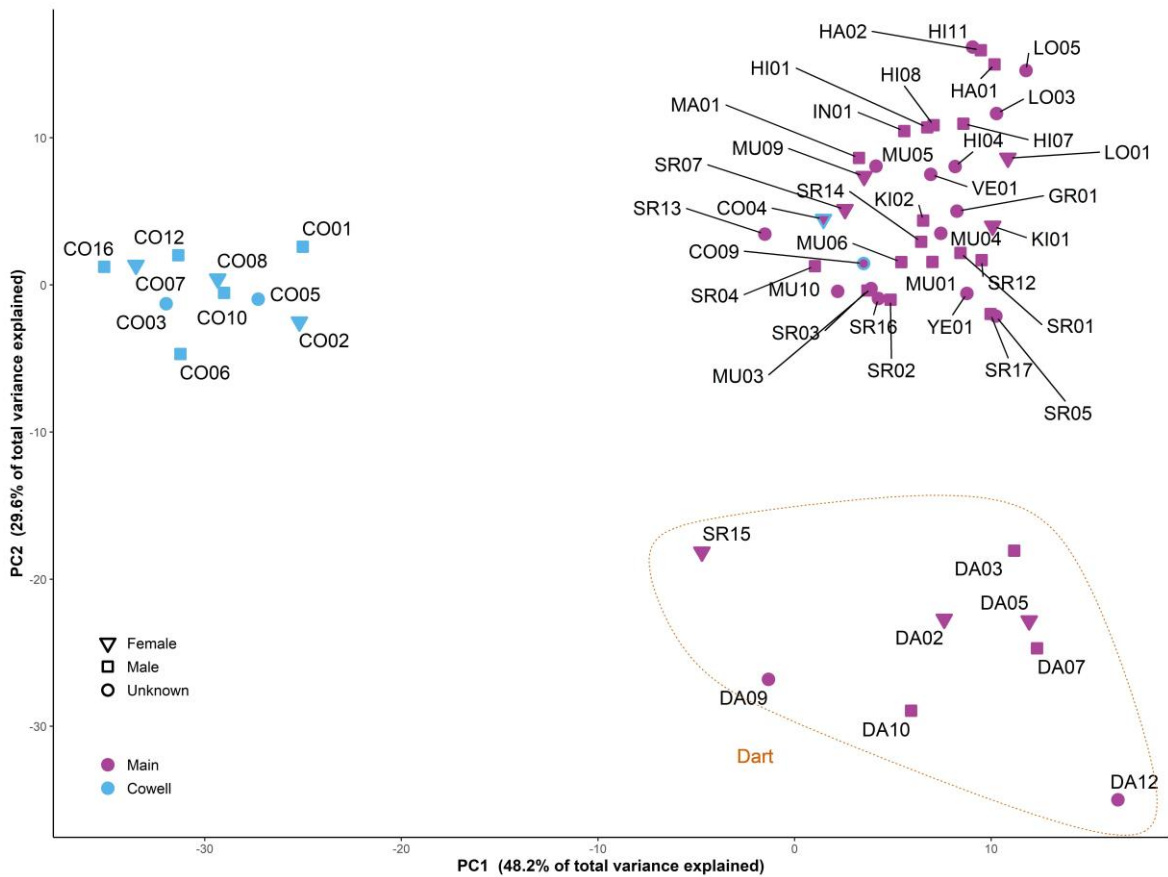


Figure 3-2 Principal Component Analysis of population genetic structure of 55 Malleefowl samples from the Eyre Peninsula, South Australia. The fill colours depict the genetic clusters identified in the PCA. Different outer colours show two samples (CO04 and CO09) where the collection location did not match the PCA cluster assignments. Shapes show the gender of the individual. Also, see PC3 and PC4 in Figure S 3-1.

The observed heterozygosity for all three genetic clusters was significantly lower than the expected heterozygosity (Table 3-1), and all three clusters showed positive inbreeding coefficients (F_{IS}). Pairwise genetic differentiation (F_{ST}) between the Main (including Dart) and Cowell clusters was 0.042. When the Dart individuals were considered separately, the F_{ST} between Main and Cowell increased to 0.043, while their F_{ST} with Dart was 0.019 and 0.061, respectively.

Table 3-1 Cluster name, size after filtering (n), observed (H_o) and expected (H_e) heterozygosity (both values range from 0 to 1, with 0 meaning no diversity) and the inbreeding coefficient (F_{is} , range 0 to 1 with 0 meaning no inbreeding). Results are presented with one standard deviation in brackets.

	Cluster	n	H_o	H_e	F_{is}
K = 2	Main	45	0.228 (\pm 0.121)	0.315 (\pm 0.135)	0.266 (\pm 0.242)
	Cowell	10	0.216 (\pm 0.202)	0.259 (\pm 0.204)	0.140 (\pm 0.356)
K = 3	Main	37	0.230 (\pm 0.125)	0.314 (\pm 0.138)	0.255 (\pm 0.257)
	Dart	8	0.218 (\pm 0.200)	0.289 (\pm 0.202)	0.199 (\pm 0.409)
	Cowell	10	0.216 (\pm 0.202)	0.259 (\pm 0.204)	0.140 (\pm 0.356)

Cross-validation error values for the LEA cross-entropy analysis showed the greatest support for two ancestral populations (i.e. $K = 2$; Figure S 3-3). At $K = 2$, most individuals were inferred to have derived the majority of their ancestry from only one of the two hypothetical ancestral populations (Figure 3-3). These inferred ancestry proportions corresponded closely with the two groups identified on PC1 in the PCA. However, several individuals (e.g. CO01) were inferred to derive moderate ancestry from both hypothetical populations. Notably, two individuals that were collected close to Cowell (CO04 and CO09) predominantly shared ancestry with individuals from the *Main* population as opposed to the *Cowell* population. CO04 and CO09 also clustered with *Main* in the PCA.

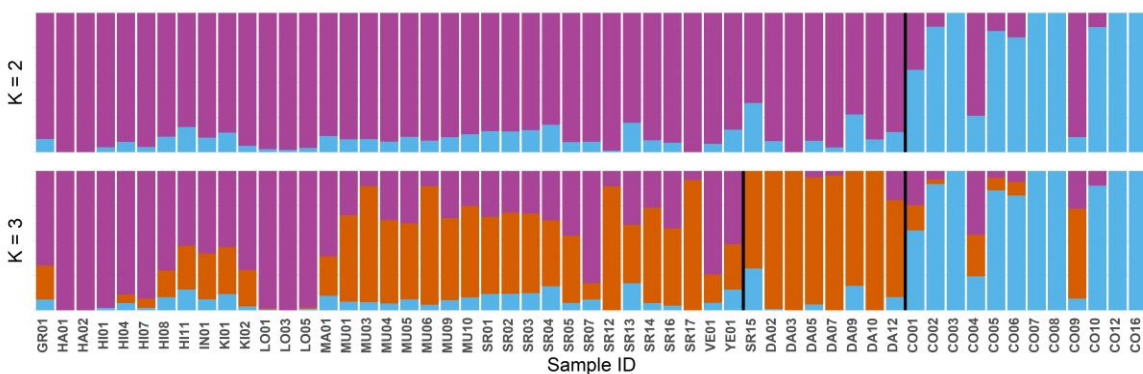


Figure 3-3 Bar plots show individual genetic cluster assignment from LEA results from $K = 2$ and $K = 3$. Samples are grouped horizontally according to the clusters observed in the PCA (separated by black vertical lines).

Even though the LEA cross-entropy analysis showed the greatest support for $K = 2$, we also plotted the results for $K = 3$ to investigate the possibility that the separation of some *Main* samples on PC2 represented individuals from a third genetic cluster. However, the addition of a third hypothetical ancestral population did not result in inferred ancestry proportions that matched exactly with the clustering of samples observed in the PCA—while the eight *Main* samples that separated from the rest on PC2 were inferred to derive most of their ancestry from the hypothetical third population, so were several other samples (e.g. SR12

and SR17) that clustered with the remaining *Main* samples in the PCA (Figure 3-2, and also see Figure S 3-1).

3.6.4 Sex-biased dispersal

Spatial genetic correlation tests for all samples, and males and females analysed separately produced different results. The spatial correlogram for all samples combined and males separately produced a significant result (p -value = 0.001), with males within ~15 km from one another demonstrating r values above the confidence intervals of no spatial genetic structure. Conversely, females showed a low value of r even in shared locations and the correlogram was not considered significant (p -value = 0.131). For females, r always fell within the confidence intervals of spatial homogeneity, indicating a low level of genetic structure across the landscape (Figure 3-4).

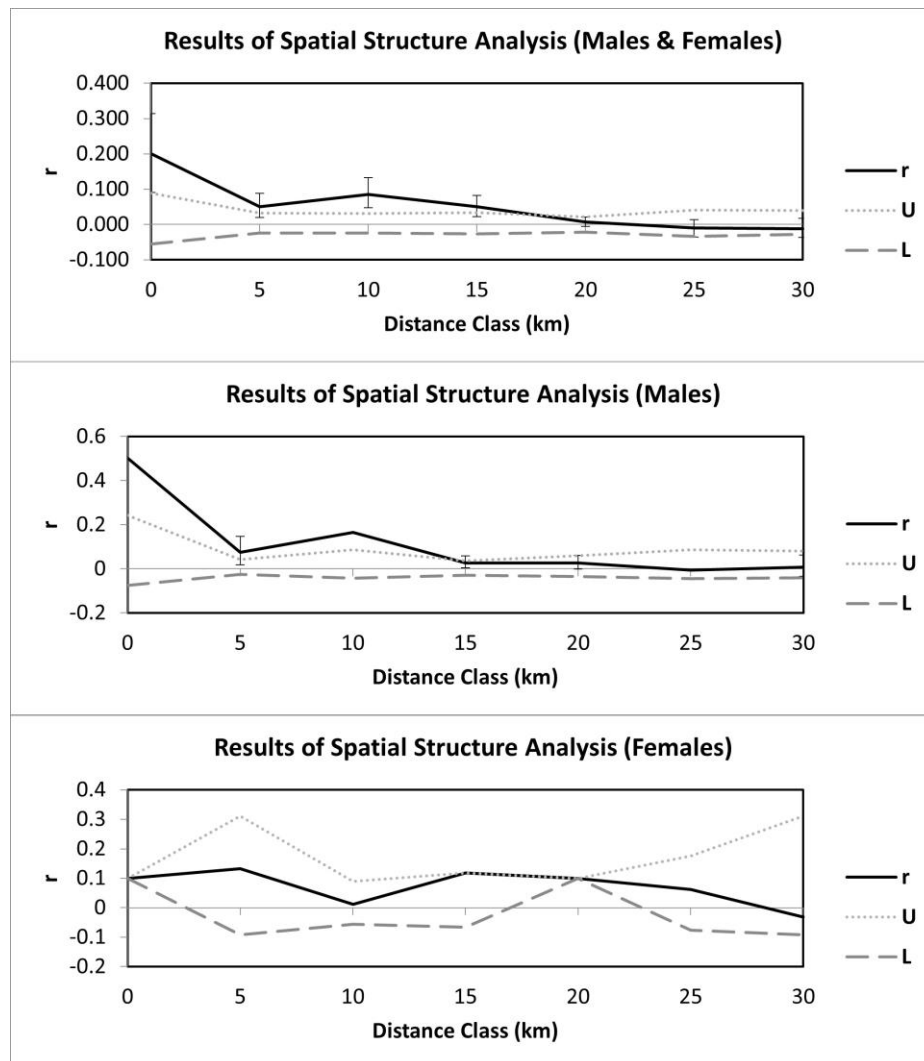


Figure 3-4 Correlograms showing spatial genetic structure in male and female Malleefowl on the Eyre Peninsula. Genetic correlation coefficient (r) is displayed with 95% confidence intervals (U = upper, L = lower) and error bars determined by bootstrapping

3.6.5 Effects of habitat intactness on spatial genetic structure

The proportional native vegetation cover around all sample locations ranged between 21 and 100% ($67.2 \pm 22.5\%$ s.d., median 62.1%). Native vegetation cover was the highest but also the most variable around samples from the *Main* cluster with a range between 21 and 100% ($69\% \pm 24.7\%$, median 78.3%). For the *Cowell* cluster, proportional native landcover averaged 60% ($\pm 3.7\%$). We excluded samples CO04 and CO09 from this analysis because they clustered with *Main* individuals in the PCA but were distributed in the same area as the *Cowell* individuals (discussed further below). A Mann-Whitney Test showed no significant difference between the proportional vegetation cover of the *Main* ($n = 43$) and *Cowell* ($n = 10$) samples.

The Kruskal-Wallis test (for comparing all three clusters) revealed that there was a statistically significant difference in proportional native vegetation cover between the 52 samples that genetically matched their geographic locations (excluding CO04, CO09 and SR15; $p = 0.002$, $\varepsilon = 0.25$, $CI_{95\%} [0.14, 0.42]$, $n = 52$). Pairwise comparison showed that the *Main* cluster proportional vegetation was significantly higher than for the *Dart* cluster ($p = 0.001$). But although the native vegetation cover in the *Main* cluster was 24% higher than in the *Cowell* cluster, and the *Cowell* cover 45% higher than in the *Dart* cluster, these differences were not statistically significant, presumably due to the small sample size in the *Cowell* and *Dart* clusters. However, when we compared the *Main* cluster to the combined dataset from *Cowell* and *Dart* the difference was highly significant with a large effect size (Mann-Whitney, $p = 0.003$, $r_{\text{rank biserial}} = 0.52$, $CI_{95\%} [0.28, 0.69]$, $n = 52$).

3.6.6 Effects of storage and sample type on genotyping

Sixty-seven samples had sufficient quality DNA for SNP genotyping. See Table S 3-2 for further details on sample types, ages and storage methods. Linear regression of grouped sample type, grouped storage type and age showed a statistically significant relationship between SNP typing success and type of sample, but not with storage method or age (Table S 3-3 and Figure S 3-6). Tissue samples (i.e. blood, muscle or liver tissue) were 38 times more likely to be successfully genotyped (probability of 0.97) than feathers or other sample types.

3.7 Discussion

3.7.1 Spatial genetic structure on the Eyre Peninsula

We found evidence for spatial clustering of genetic diversity among 55 Malleefowl samples from across the Eyre Peninsula. First, a Mantel test for isolation-by-distance (IBD) revealed a positive correlation between genetic distance and geographical distance across our larger

data subset (*Main*) and the whole dataset—in general, more geographically proximate individuals were more closely related, consistent with previous findings based on microsatellite data (Cope, Mulder *et al.* 2014). Second, our PCA and LEA results both supported the existence of two distinct genetic clusters (K=2): one geographically restricted cluster comprising ten individuals sampled near the town of Cowell on the east coast of the Eyre Peninsula (*Cowell*), and a second more widespread cluster comprising the remaining 45 individuals (*Main*).

There was also some evidence for residual population structure within the *Main* cluster—eight individuals separated from *Main* along PC2 of our PCA (*Dart*). Despite our LEA results suggesting that K=2 was the best fit for our data, we also considered the results for K=3 to explore whether the *Dart* cluster might represent a third incipient population. However, unlike the *Main* vs. *Cowell* split for K=2, the ancestry proportions inferred by LEA at K=3 do not correspond closely to the clusters observed in the PCA. For example, some samples from the *Main* cluster—e.g. MU03, SR12, SR17—actually have a higher inferred contribution from the third ancestry component than some of the *Dart* samples (e.g. SR15, SR09). Additionally, the genetic diversity measures for *Main* do not change substantially when the *Dart* individuals are analysed separately (Table 3-1). Consequently, while our results do not exclude the possibility of additional cryptic population structure among Malleefowl on the Eyre Peninsula, we only find strong evidence for two populations—*Main* and *Cowell*.

The *Cowell* cluster is distributed over a relatively small area with approximately 60% native vegetation cover, which is separated by nearly 20 kilometres of cleared agricultural land from the closest individuals in the much more broadly distributed *Main* cluster (comprising on average 69% native vegetation cover). Even so, two individuals sampled from near Cowell (CO04 sampled in 1996 & CO09 sampled in 2001) cluster with the *Main* individuals in the PCA and are inferred to share a substantial proportion of ancestry with the *Main* individuals in our LEA analyses. These two samples—CO04 and CO09—may represent first-generation migrants, as their ancestry is markedly different from that of *Cowell* individuals sampled at around the same time (i.e., CO02 in 1995; CO03, CO05, CO06, and CO07 in 1996; CO08 in 2000). However, CO04 and CO09 do not appear to have contributed substantial ancestry to individuals sampled near Cowell more recently (i.e., CO10 sampled in 2007; CO12 & CO16 sampled in 2017).

The oldest of our *Cowell* individuals—and oldest individual overall—was sampled in 1985 (CO01). Interestingly, this sample appears to share the most ancestry with *Main* individuals after CO04 and CO09. CO01 could thus be interpreted as the result of admixture between *Cowell* and *Main* individuals, or possibly may pre-date much of the accumulation of allele

frequency differences between more recently sampled individuals from *Cowell* and *Main*. The latter scenario would also violate some of the assumptions of programs like ADMIXTURE and LEA, namely that sampled individuals post-date the establishment of distinct populations, and so care should be taken not to over-interpret the inference of multiple ancestry components as evidence of recent admixture (Lawson, van Dorp *et al.* 2018).

In any case, our data support the existence of two largely separate populations of Malleefowl on the Eyre Peninsula. Despite some evidence for migration (at least historically in 1996 & 2001), our data do not reveal compelling evidence for recent gene flow between these two populations. Indeed, tracking has shown that Malleefowl prefer staying in native vegetation where they can range over 23 km in continuous native habitat, which is consistent with the significantly elevated relatedness between male birds (and all individuals) at spatial scales ≤ 20 km revealed by our correlograms (Figure 3-4). However, Malleefowl have only been recorded crossing relatively short (e.g. 250 m) tracts of agricultural land (chapter 4). Thus, we suggest that 19th-20th Century vegetation clearance and changes in land use on the Eyre Peninsula (i.e., following European settlement) have resulted in a reduction of genetic connectivity—and establishment of spatial structure—in local Malleefowl. This is consistent with data that suggest Malleefowl population numbers on the Eyre Peninsula have declined significantly over the last 20 years (Stenhouse and Moseby 2022).

Recent population decline is also consistent with our observation that heterozygosity is lower than expected for both *Cowell* and *Main* genetic clusters, and that both have positive inbreeding coefficients. Our results further suggest that inbreeding is higher in the *Main* cluster than in the *Cowell* cluster, which is counterintuitive because the range of the *Cowell* cluster is smaller (and the population presumably smaller). This result does not appear to be driven by residual population structure within the *Main* cluster (i.e. Wahlund effect), because removing the *Dart* individuals only slightly reduces the inbreeding coefficient for the *Main* individuals. However, if population decline is still ongoing then this result may alternatively reflect that our *Cowell* samples are older overall than our *Main* samples (e.g. 70% of our *Cowell* samples were collected during or before 2000, compared to only 4.4% of our *Main* samples).

3.7.2 Sex-biased dispersal

We found preliminary evidence suggesting female-biased dispersal, although the small sample size for females limits confidence in this conclusion. Spatial genetic correlograms for our female samples showed limited genetic structure across the landscape, indicative of

dispersal—notably one of the putative migrants from *Main* to *Cowell* discussed above (CO04) was female (the sex of the other, CO09, was undetermined). In contrast, spatial correlograms showed spatial heterogeneity for males, with individuals within 15 km of one another demonstrating a significant degree of relatedness. Previous studies on other ground-dwelling birds living in fragmented habitats have also shown long-range dispersal in females (Earl, Fuhlendorf *et al.* 2016; Vogel 2015). Female natal and breeding dispersal and male philopatry is the most common dispersal pattern in birds (Greenwood 1980). Greenwood (1980) reasoned that this was because males are “resource defenders” (in the case of Malleefowl, the mound and its familiar surroundings), while females travel to increase access to mates and avoid inbreeding, thus improving reproductive success and genetic health. However, recent research argues that drivers of sex-biased dispersal in birds are more complex and that male-biased dispersal is more common than previously thought (Clarke, Sæther *et al.* 1997).

Studies have revealed that Malleefowl are not strictly monogamous as previously believed (Cope, Mulder *et al.* 2011) and males can be polygynous (Weathers, Weathers *et al.* 1990). Female breeding dispersal could be related to breeding success, predation of eggs and young, vegetation (Greenwood and Harvey 1982) and other factors such as fire and disturbance. Benshemesh (1992) reported a female Malleefowl 10-11 km from the study area eight months after a fire, as well as a pair and a male approximately 9 km from the study site two months after the fire. In chapter 4 we found that a female Malleefowl abandoned her mound after being trapped and resettled in a new area over 7 km away, crossing open farmland. And while tracked males in the same study ranged further overall, they moved less directionally than the female. Captive-bred Malleefowl chicks of unknown gender were found to disperse over 6 km when released back at their mounds of origin (Benshemesh 1992). It should be noted that Malleefowl chicks are precocial and there is no parental care after emergence from the mound (Benshemesh 1992; Frith 1959).

3.7.3 Limitations

Our study was hampered by the number and quality of available samples. For instance, when many feathers were collected from a mound and its vicinity at different times, it was impossible to know the exact source individual as the feathers could be from the parent(s) or chick(s). DNA extraction of these samples then led to duplicates. Further, visual inspection did not always show how long a sample had been exposed to the environment before collection and thus many samples were old and degraded. Some were also stored at room temperature for many years which may have contributed to the degradation and/or bacterial contamination. There was a male bias in samples (69% of samples with assigned sexes were male), which could be explained for samples collected from mounds, as males

tend to spend more time at the mound than females, but not from other samples found randomly on the landscape. This bias implies that our spatial autocorrelation relied on only 11 females with a limited number of pairwise genotypes for each distance category, which means our results can only be considered preliminary. Additionally, the samples from the *Main* cluster had a much broader geographic range (hundreds of km) than the *Cowell* samples. Any existing sub-structures within the *Main* cluster that were not registered due to large distances between sample locations (and/or sparse local sampling) could possibly lead to increased divergence between expected and observed heterozygosity and biased apparent inbreeding coefficients. For example, eight individuals from near Yalanda ($46.6 \pm 15.5\%$ native vegetation) in the north-eastern Eyre Peninsula were separated from the other *Main* samples along PC2 in our PCA and may reflect finer-scale population structure within the *Main* samples. However, our LEA results suggested $K=2$ was a better fit for our data than $K=3$. Denser sampling may provide greater resolution of such finer-scale spatial structure in future studies. Finally, we took a simplified approach to the habitat intactness analysis by using the observed maximum range of one Malleefowl to calculate native vegetation around samples. This may not reflect the true dispersal distance for Malleefowl and may only be regarded as preliminary.

3.7.4 Management implications

We recommend that Malleefowl conservation includes management for habitat fragmentation and lack of gene flow. Gene flow could be encouraged by building corridors between habitat fragments. While the conservation of larger and connected patches has been favoured in the past (Margules and Pressey 2000; Moilanen, Franco *et al.* 2005), the importance and high conservation value of smaller and more fragmented vegetation patches has recently been highlighted (Fahrig, Arroyo-Rodríguez *et al.* 2019; Wintle, Kujala *et al.* 2018). Small native vegetation patches can be important habitat for Malleefowl, one tracked Malleefowl bred successfully in and rarely left a 107 ha patch in over 4 years (chapter 4). In chapter 4 we also found that patch size restricts non-breeding home range and may negatively influence Malleefowl dispersal. Research on other ground-dwelling birds suggests occupancy is higher if a patch is bigger or closer to other patches (Bollmann, Graf *et al.* 2011). Therefore, we recommend revegetation of corridors and habitat patches to connect highly fragmented areas with low proportional native vegetation cover of below 60%, with gaps between patches ideally of no more than 250 m. Translocations are another option for improving gene flow. However, at this stage, given the relatively weak genetic spatial structuring on the Eyre Peninsula and the evidence for female-biased dispersal, translocations are probably not necessary. We recommend continued genetic monitoring

as part of the overall species management plan and to guide potential future translocations to assist gene flow between isolated habitats (Weeks, Sgro *et al.* 2011).

3.7.5 Conclusions

Our study is the first to use genome-wide SNP markers to study past Malleefowl movement. Results suggest that anthropogenic habitat fragmentation is affecting Malleefowl population structure on the Eyre Peninsula. Habitat fragmentation and subsequent isolation have led to a lack of gene flow, so that genetic drift has contributed largely to the differentiation of at least one new population on the Eyre Peninsula. However, the differentiation between populations is still relatively low, suggesting this is a relatively recent development. This may have consequences for the long-term viability of these isolated populations. We also provide preliminary genetic evidence of female-biased dispersal in a megapode in a highly fragmented landscape. Ultimately, we suggest that management actions include genetic targets, and we encourage land managers to facilitate between-patch movement by improving native vegetation cover. Translocations may be necessary to improve genetic health in the future if continued monitoring shows no improvement in gene flow between fragmented populations.

3.8 Miscellaneous

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Ethics

This project was undertaken with approval from the University of Adelaide Animal Ethics Committee (S-2016-105) and the SA Department of Environment and Water and the Environment (permits A26564-1 to 5).

Data accessibility

Raw sequences and final dataset are available via Figshare (DOI: 10.25909/19322261 and 10.25909/19407491)

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3.10 Appendix

3.10.1 Samples

Table S 3-1 Sequenced samples Eyre Peninsula Malleefowl. We present sample ID, collection date, location (Hamb = Hambidge CP, Mara = Maralinga, Muny = Munyaroo CP, SR/Iron = Secret Rocks/Ironstone NP, Yala = Yalanda, Yella = Yellabinna), sex (S = sex, F = female, M = male, U = unknown), sample type (Toe = toepad, Mus = muscle, Liv = liver, Yolk = yolk sac, Fea = feather, Egg = egg membrane, Blo = blood), storage method (Sto = Storage, ta = taxidermied, fr = frozen (-80°C if from museum, -20°C for others), ro = room temperature, bu = lysis buffer, et = 100% EtOH), origin of sample (Sou = Source, SAM = South Australia Museum, PS = Author, Gov = DEW or NRM, Misc = Miscellaneous, AM = Australian Museum), collection event (Field = found during field work or monitoring, Trap = from captured bird at active mound, Chick = chick hatched while author at mound and feather sample collected, Active = from active mound), whether sample was from immediate proximity of the mound (M = Mound), and reason why sample wasn't used for analysis (Rem = Removed, Kin = kinship >0.3, Trans = four Malleefowl from Lock were translocated to Lincoln NP in 2001, making the heritage of this bird unclear), DNA = DNA absent or contaminated, Data = >50% missing data). Dup = Duplicate sample(s).

ID	Date	Loc	S	Type	Sto	Sou	Event	M	Rem	Dup
CO01	Dec-1985	Cowell	M	Toe	ta	SAM	Road	N	-	-
CO02	Feb-1995	Cowell	F	Mus	fr	SAM	Road	N	-	-
CO03	Jan-1996	Cowell	U	Liv	fr	SAM	Road	N	-	-
CO04	Jan-1996	Cowell	F	Yolk	fr	SAM	Field	Y	-	-
CO05	Jan-1996	Cowell	U	Liv	fr	SAM	Road	N	-	-
CO06	Jan-1996	Cowell	M	Liv	fr	SAM	Road	N	-	-
CO07	Jan-1996	Cowell	F	Liv	fr	SAM	Road	N	-	-
CO08	May-2000	Cowell	F	Mus	fr	SAM	Road	N	-	-
CO09	Apr-2001	Cowell	U	Fea	fr	SAM	Road	N	-	-
CO10	Mar-2007	Cowell	M	Egg	ro	SAM	Field	Y	-	-
CO12	Feb-2017	Cowell	M	Blo	bu	PS	Trap	Y	-	-
CO16	Nov-2017	Cowell	M	Blo	bu	PS	Trap	Y	-	-
DA02	Nov-2016	Yala	F	Liv	fr	PS	Road	N	-	-
DA03	Jan-2017	Yala	M	Blo	bu	PS	Trap	Y	-	-
DA05	Feb-2017	Yala	F	Fea	fr	PS	Field	Y	-	-
DA07	May-2017	Yala	M	Fea	fr	PS	Field	Y	-	-
DA09	Oct-2018	Yala	U	Fea	fr	PS	Field	Y	-	-
DA10	Oct-2018	Yala	M	Fea	fr	PS	Field	Y	-	-
DA12	Nov-2018	Yala	U	Fea	fr	PS	Field	Y	-	-
GR01	Oct-2019	GRNP	U	Fea	fr	GOV	Field	Y	-	-
HA01	Nov-2016	Hamb	M	Fea	fr	PS	Chick	Y	-	-
HA02	Dec-2016	Hamb	M	Blo	bu	PS	Trap	Y	-	-
HI01	Dec-1994	Hincks	M	Liv	fr	SAM	Road	N	-	-
HI04	Nov-2016	Hincks	U	Fea	fr	PS	Chick	Y	-	-
HI07	Dec-2016	Hincks	M	Blo	bu	PS	Trap	Y	-	-
HI08	May-2017	Hincks	M	Fea	fr	PS	Field	Y	-	-
HI11	Jul-2017	Hincks	U	Egg	fr	PS	Active	Y	-	-
IN01	Apr-2007	Innes	M	Fea	fr	SAM	Field	N	-	-
KI01	Jan-2003	Kimba	F	Toe	ta	GOV	Road	N	-	-
KI02	Aug-2019	Kimba	M	Mus	fr	Misc	Road	N	-	-
LO01	Feb-2017	Lock	F	Fea	fr	PS	Trap	Y	-	-

LO03	Jun-2017	Lock	U	Fea	fr	PS	Road	N	-	-
LO05	Apr-2018	Lock	U	Toe	fr	PS	Road	N	-	-
MA01	Feb-2010	Mara	M	Fea	ro	SAM	Field	Y	-	-
MU01	Apr-2001	Muny	M	Mus	fr	SAM	Road	N	-	-
MU03	Nov-2011	Muny	U	Fea	ro	SAM	Road	N	-	-
MU04	Jan-2012	Muny	U	Fea	ro	Misc	Road	N	-	-
MU05	Aug-2012	Muny	U	Fea	ro	Misc	Road	N	-	-
MU06	Dec-2012	Muny	M	Mus	fr	SAM	Road	N	-	-
MU09	Jan-2014	Muny	F	Fea	ro	Misc	Field	Y	-	-
MU10	Dec-2018	Muny	U	Mus	fr	Misc	Road	N	-	-
SR01	Jan-2011	SR/Iron	M	Toe	ro	PS	Road	N	-	-
SR02	Jan-2011	SR/Iron	M	Fea	ro	SAM	Field	Y	-	-
SR03	Oct-2011	SR/Iron	M	Fea	ro	SAM	Field	Y	-	-
SR04	Oct-2011	SR/Iron	M	Fea	ro	SAM	Field	Y	-	-
SR05	Oct-2011	SR/Iron	U	Fea	ro	SAM	Field	Y	-	-
SR07	Sep-2012	SR/Iron	F	Mus	fr	SAM	Road	N	-	-
SR12	Dec-2017	SR/Iron	M	Blo	bu	PS	Trap	Y	-	-
SR13	Feb-2018	SR/Iron	U	Fea	fr	PS	Field	Y	-	-
SR14	Nov-2018	SR/Iron	M	Blo	bu	PS	Trap	Y	-	-
SR15	Oct-2019	SR/Iron	F	Fea	fr	Misc	Field	Y	-	-
SR16	Oct-2019	SR/Iron	U	Fea	fr	Misc	Field	Y	-	-
SR17	Dec-2019	SR/Iron	M	Fea	fr	PS	Trap	Y	-	-
VE01	Oct-2019	Venus	U	Fea	fr	GOV	Field	Y	-	-
YE01	Aug-2008	Yella	U	Fea	et	SAM	Road	N	-	-
CO11	Jan-2017	Cowell	M	Fea	fr	PS	Field	Y	Kin	DA04/CO11 (45)
CO13	Jul-2017	Cowell	M	Fea	fr	PS	Field	N	Kin	CO15/16/17 (0-0.26)
CO15	Oct-2017	Cowell	U	Fea	fr	PS	Field	Y	Kin	CO13/16/17 (0.26)
CO17	Nov-2017	Cowell	M	Fea	fr	PS	Trap	Y	Kin	CO16 (0)
DA01	Oct-2016	Yala	M	Fea	fr	PS	Field	Y	Kin	DA07 (0)
DA04	Jan-2017	Yala	M	Fea	fr	PS	Trap	Y	Kin	DA03/CO11 (45)
LO02	Feb-2017	Lock	F	Blo	bu	PS	Trap	Y	Kin	LO01 (0)
LO04	Apr-2018	Lock	U	Fea	ro	PS	Road	N	Kin	LO05/06 (0)
LO06	Apr-2018	Lock	M	Fea	ro	PS	Road	N	Kin	LO04/05 (0)
MA02	Feb-2010	Mara	U	Fea	ro	SAM	Field	Y	Kin	MA01 (0)
SR11	Dec-2017	SR/Iron	M	Fea	fr	PS	Trap	Y	Kin	SR12 (0)
LI01	Oct-2016	Lincoln	U	Mus	fr	GOV	Road	N	Trans	-
CL01	Jul-1911	Cleve	U	Toe	ta	SAM	Historic	N	DNA	-
CO18	Nov-2016	Cowell	U	Fea	fr	PS	Road	N	DNA	-
GR02	Apr-2006	GRNP	U	Fea	fr	SAM	Field	N	DNA	-
GR03	Nov-2018	GRNP	U	Egg	et	PS	Field	Y	DNA	-
HA04	Mar-2017	Hamb	U	Fea	fr	PS	Field	Y	DNA	-
HI12	Mar-2007	Hincks	U	Egg	ro	SAM	Field	Y	DNA	-
HI13	Feb-2017	Hincks	U	Fea	fr	PS	Field (kill)	N	DNA	-
HI14	Nov-2017	Hincks	U	Fea	fr	PS	Field	Y	DNA	-

WH01	Jan-1952	Whyalla	U	Toe	ta	AM	Historic	N	DNA	-
CO14	Oct-2017	Cowell	U	Fea	fr	PS	Field	Y	Data	-
DA06	Mar-2017	Yala	U	Fea	fr	PS	Active	Y	Data	-
DA08	Oct-2017	Yala	U	Fea	fr	PS	Field	Y	Data	-
DA11	Nov-2018	Yala	U	Fea	fr	PS	Field	Y	Data	-
HA03	Jul-2017	Hamb	U	Fea	fr	PS	Active	Y	Data	-
HI02	Nov-2016	Hincks	U	Fea	fr	PS	Field	Y	Data	-
HI03	Nov-2016	Hincks	U	Fea	fr	PS	Field (kill)	N	Data	-
HI05	Nov-2016	Hincks	U	Fea	fr	PS	Field	Y	Data	-
HI06	Nov-2016	Hincks	U	Fea	fr	PS	Field (kill)	N	Data	-
HI09	Jul-2017	Hincks	U	Fea	fr	PS	Active	Y	Data	-
HI10	Jul-2017	Hincks	U	Egg	fr	PS	Active	Y	Data	-
MU02	Apr-2001	Muny	U	Liv	et	SAM	Road	N	Data	-
MU07	Feb-2013	Muny	U	Fea	ro	Misc	Road	N	Data	-
MU08	Mar-2013	Muny	U	Fea	ro	Misc	Road	N	Data	-
SR06	Nov-2011	SR/Iron	U	Fea	ro	SAM	Field	Y	Data	-
SR08	Oct-2016	SR/Iron	U	Fea	fr	PS	Field	Y	Data	-
SR09	Jul-2017	SR/Iron	U	Fea	fr	PS	Field	N	Data	-
SR10	Oct-2017	SR/Iron	U	Fea	fr	PS	Field	Y	Data	-

3.10.2 PCA Results

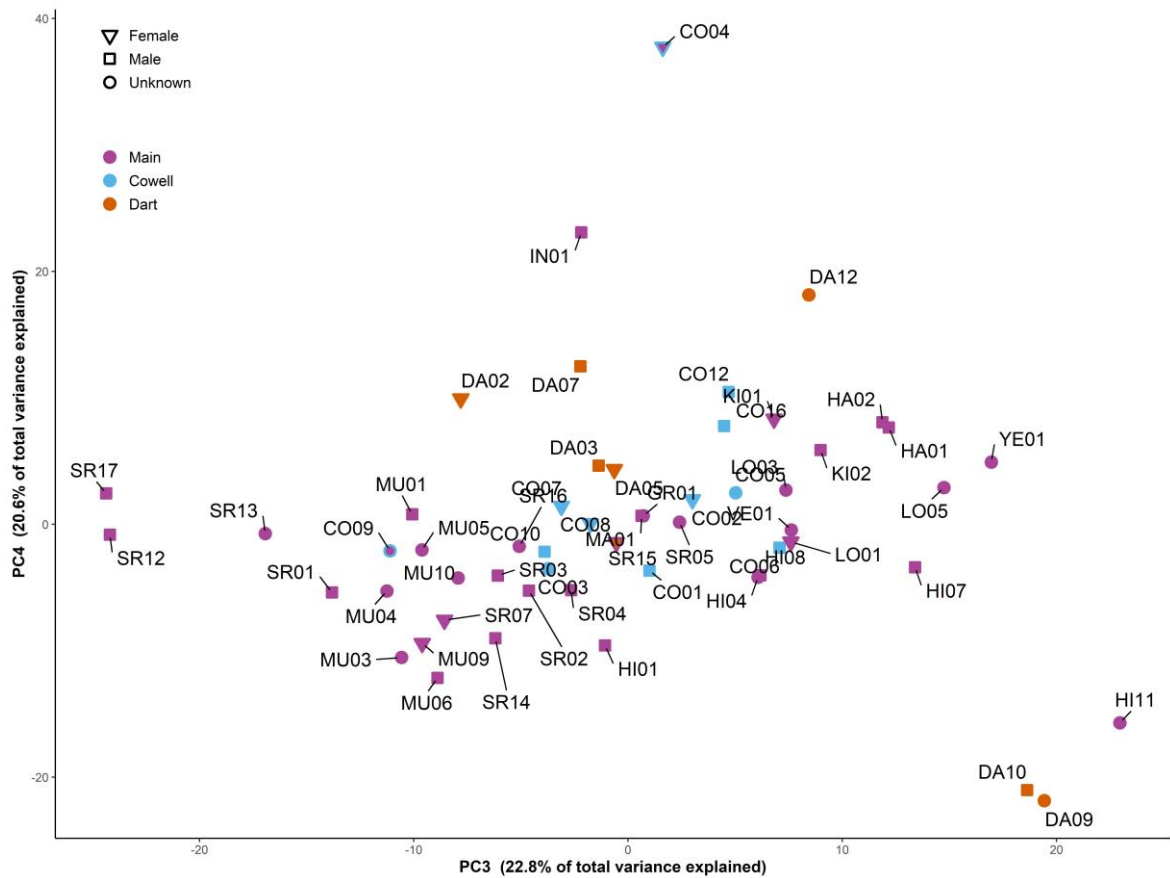


Figure S 3-1 Principal Component Analysis (Axes PC3 and PC4) of population genetic structure of 55 Malleefowl samples from the Eyre Peninsula, South Australia. The fill colours depict the genetic clusters identified in the PCA. Different outer colours show two samples (CO04 and CO09) where collection location did not match the PCA cluster assignments. Shapes show the gender of the individual.

3.10.3 Isolation-by-distance plots

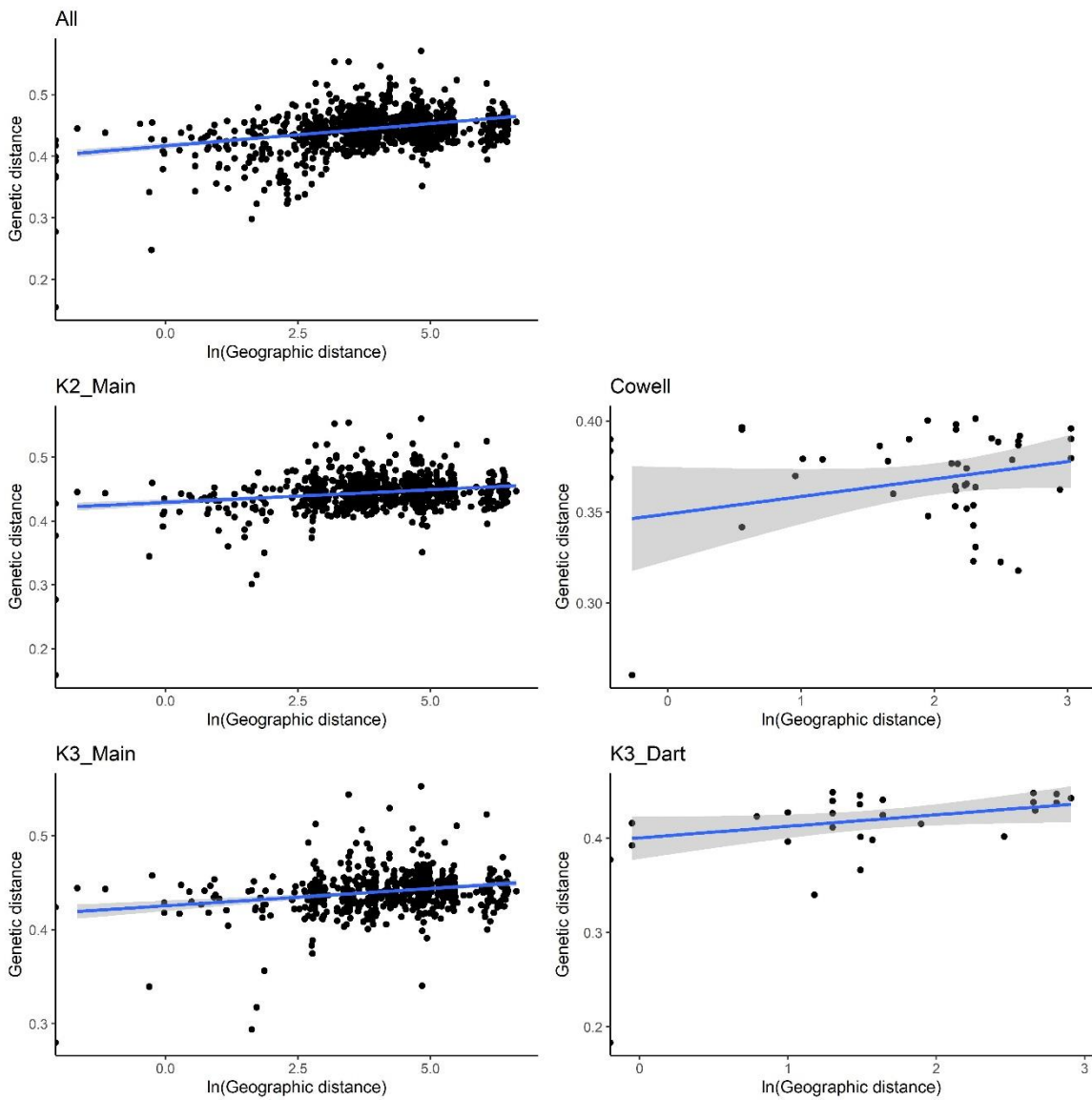


Figure S 3-2 Isolation-by-distance scatterplots for all birds, K = 2 and K = 3. The x-axis shows the natural logarithm of the geographic distance after Rousset (2000). The y-axis shows the pairwise genetic distance calculated as a percentage.

3.10.4 ADMIXTURE error results

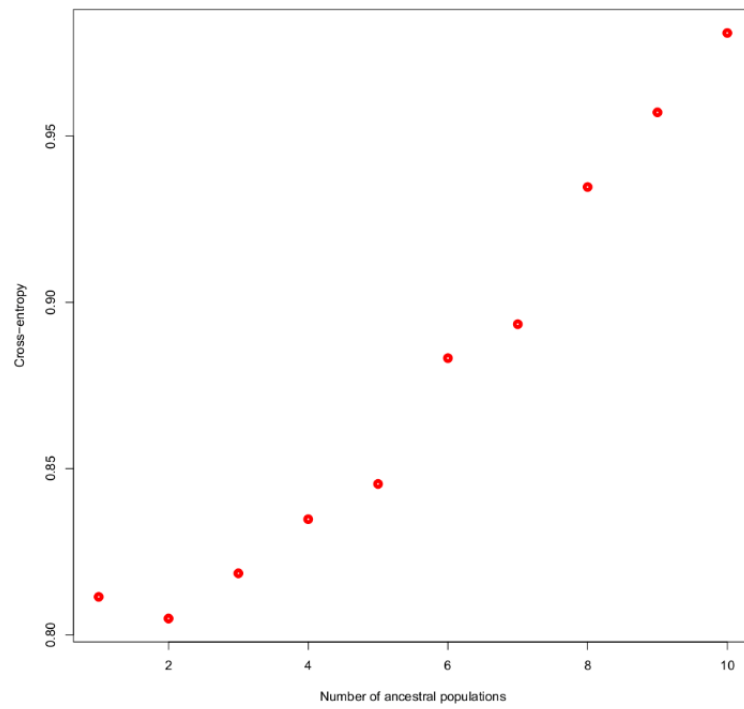


Figure S 3-3 LEA cross-entropy results for Malleefowl on the Eyre Peninsula, South Australia. The cross-entropy validation errors are plotted for K values of 1 to 10. The lowest error indicates the greatest support at K = 2.

3.10.5 Study sites with $K = 3$

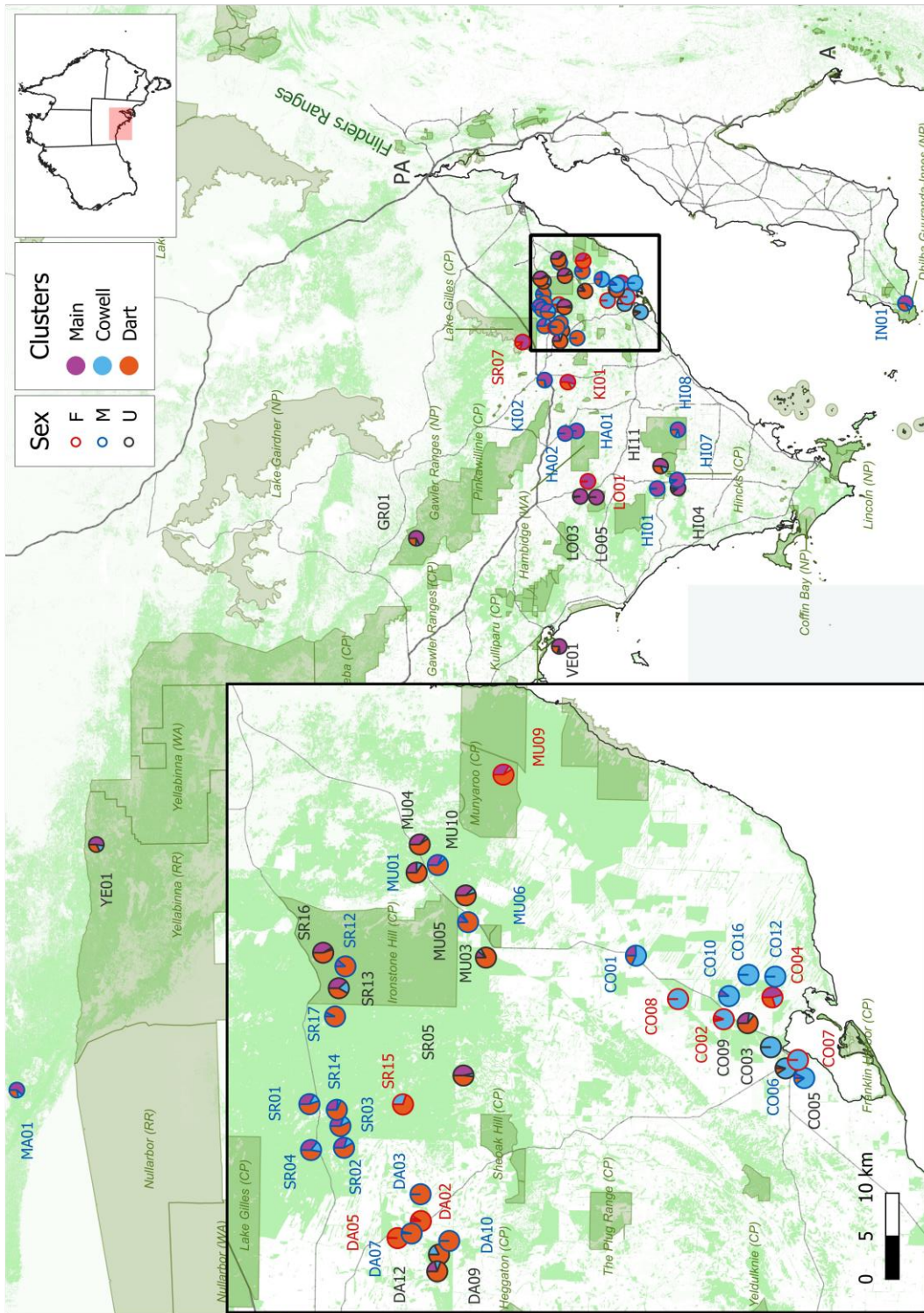


Figure S 3-4 Malleefowl sample collection sites on and near the Eyre Peninsula, South Australia. The predominant colour of the pie-chart matches that of the clusters identified by the PCA. The proportions within each pie chart reflect the LEA plot ancestry composition of samples ($K = 3$). Pie-chart line and label colours indicate the sex of the individual. The light green areas are native vegetation, the darker green highlights are nature protection and wilderness areas under South Australian Government management.

3.10.6 Relationship between ancestry proportions and native vegetation cover

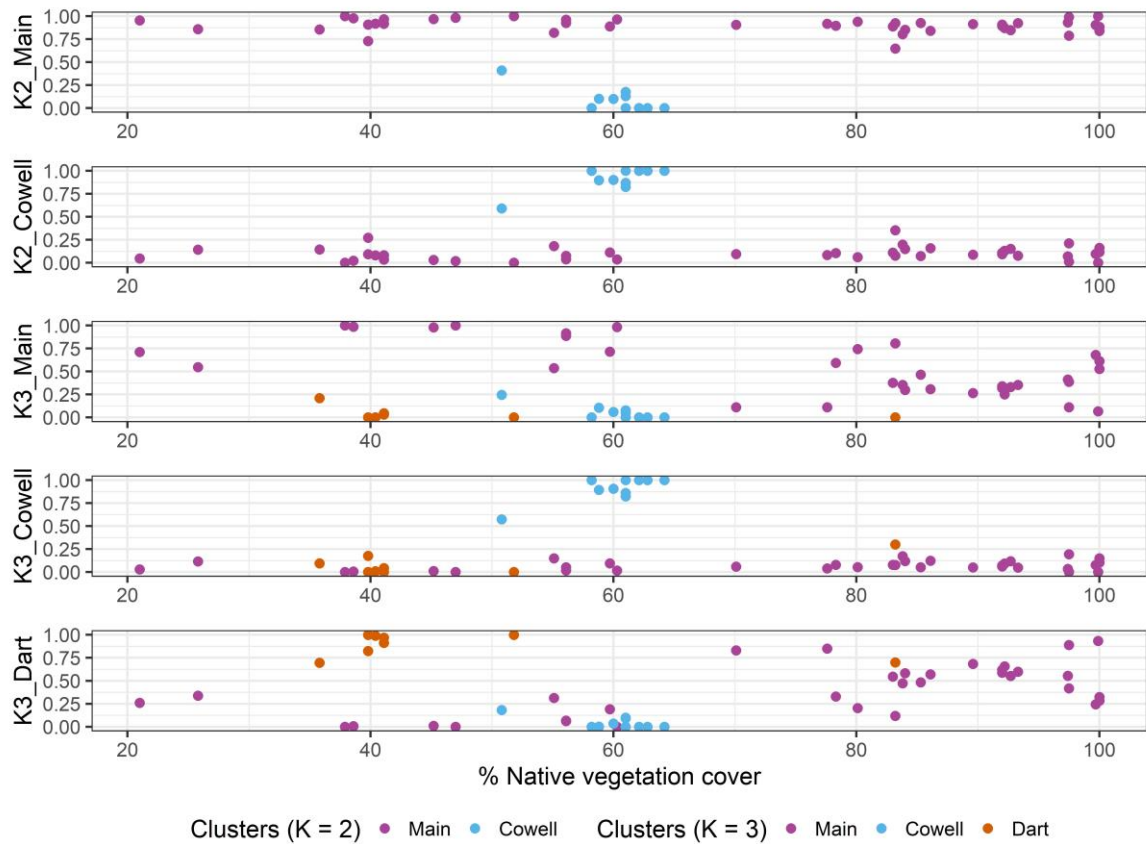


Figure S 3-5 Visualisation of the relationship between ancestry proportions and native vegetation cover of Malleefowl on the Eyre Peninsula, South Australia. Each plot shows the proportion of the named ancestry proportion on the y-axis and the % native vegetation cover on the x-axis. The colours indicate the cluster assignments as calculated by the PCA. Three samples (SR15, CO04 and CO09) where collection location did not match the PCA cluster assignments were excluded.

3.10.7 Effects of storage and sample type

Table S 3-2 Overview of sample type and storage methods for Eyre Peninsula Malleefowl samples. We present DNA quality (good (bad)= sufficient (insufficient) for SNP genotyping), sample types (Toe = toepad, Yolk = yolk sac, Egg m.= egg membrane), storage method (Storage, bu = lysis buffer, fr = frozen (-80°C if from museum, -20°C for others), ro = room temperature, ta = taxidermy, et = 100% EtOH), age classes (1 = under 5, 2 = 5 - 10; 3 = 11 -20; 4 = over 20 years) and number of samples of that type (Count). The table is sorted by most successful sample type.

DNA quality	Type	Storage	Age class	Count	% of type by storage and age	% of type by storage	% of type
Good	Blood	bu	1	8	100	100	100
	Yolk	fr	4	1	100	100	100
	Liver	fr	4	5	100	83.33	71.43
	Feather	fr	1	23	57.5	53.49	38.98
	Muscle	fr	1	3	100	37.5	37.5
	Muscle	fr	2	2	100	25	25
	Muscle	fr	3	2	100	25	25
	Egg m.	fr	1	1	50	50	20
	Egg m.	ro	3	1	50	50	20
	Feather	ro	2	10	76.92	66.67	16.95
	Toe	fr	1	1	100	100	16.67
	Toe	ro	2	1	100	100	16.67
	Toe	ta	3	1	100	25	16.67
	Toe	ta	4	1	33.33	25	16.67
	Liver	fr	1	1	100	16.67	14.29
	Muscle	fr	4	1	100	12.5	12.5
	Feather	fr	3	2	66.67	4.65	3.39
	Feather	ro	1	2	100	13.33	3.39
	Feather	et	3	1	100	100	1.69
Bad	Toe	ta	4	2	66.67	50	33.33
	Feather	fr	1	17	42.5	39.53	28.81
	Egg m.	et	1	1	100	100	20
	Egg m.	fr	1	1	50	50	20
	Egg m.	ro	3	1	50	50	20
	Liver	et	3	1	100	100	14.29
	Feather	ro	2	3	23.08	20	5.08
	Feather	fr	3	1	33.33	2.33	1.69

Table S 3-3 Parameter estimates of the generalised linear model investigating the relationship between sample type, storage and age on DNA usability. SE = Standard Error; z = z-value; LCI/UCI = Lower/Upper confidence intervals; p = P-value. We also present the Hosmer and Lemeshow goodness of fit test results (p <0.05 means data is not a good fit).

Model	Variables	Estimate	SE	z	LCI, UCI	p
glm(DNA_Quality ~ Type + Storage + Age, family = "binomial", data = dat)	(Intercept)	-0.66	1.30	-0.50	(-3.83,1.82)	0.61
	TypeOther	0.01	0.85	0.01	(-1.64,1.83)	0.99
	TypeTissue	3.65	1.32	2.77	(1.54,7.08)	0.01
	StorageFrozen	1.22	1.31	0.93	(-1.27,4.41)	0.35
	StorageRoom	2.48	1.45	1.71	(-0.23,5.87)	0.09
	Age	-0.05	0.03	-1.54	(-0.13,0)	0.12
Null deviance: 112.735 on 93 degrees of freedom						
Residual deviance: 95.371 on 88 degrees of freedom						
Number of Fisher Scoring iterations: 5						
Hosmer and Lemeshow goodness of fit test: data: dat\$DNA_Quality, fitted(m2) X-squared = 8.0969, df = 8, p-value = 0.4241						

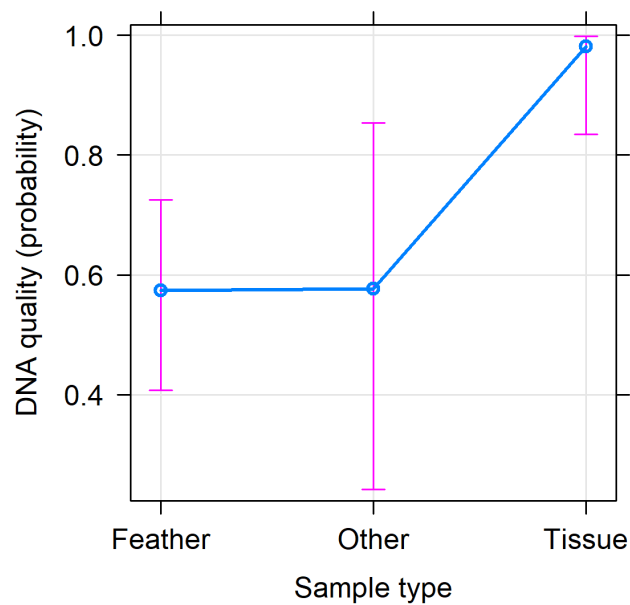


Figure S 3-6 Probabilities of three sample type groups being successfully genotyped for the Eyre Peninsula Malleefowl. Pink bars are the confidence intervals.

3.10.8 Additional References

Rousset (2000) Genetic differentiation between individuals. *Journal of Evolutionary Biology* **13**(1), 58-62. DOI: <https://doi.org/10.1046/j.1420-9101.2000.00137.x>.

Chapter 4:

Patch size and breeding status influence movement patterns in the threatened Malleefowl (*Leipoa ocellata*)



Figure on previous page: Hambidge, December 2016. This male (HAM1) successfully completed the breeding season but was later killed by a cat in May 2017. Credit: Peri Stenhouse

4.1 Statement of Authorship

Title of Paper	Stenhouse P, Moseby KE. Patch size and breeding status influence movement patterns in the threatened Malleefowl (<i>Leipoa ocellata</i>).			
Publication status	<input type="checkbox"/> Published	<input type="checkbox"/> Accepted for Publication	<input checked="" type="checkbox"/> Submitted for Publication	<input type="checkbox"/> Publication style
Publication Details	Submitted to the journal Austral Ecology on 24 December 2021.			

Author contributions

By signing the Statement of Authorship, each author certifies that the candidate's stated contribution to the publication is accurate, that permission is granted for the candidate to include the publication in the thesis; and that the sum of all co-author contributions is equal to 100% less the candidate's stated contribution.

Name of Principal Author (Candidate)	Peri Stenhouse	
Contribution to the paper	Conceptualization, methodology, data curation, formal analysis, writing – original draft preparation, writing – review and editing.	
Overall percentage	80%	
Certification	This paper reports on original research I conducted during the period of my Higher Degree by Research candidature and is not subject to any obligations or contractual agreements with a third party that would constrain its inclusion in this thesis. I am the primary author of this paper.	
Signature	Date: 04 April 2022	

Name of Co-Author	Katherine Moseby	
Contribution to the Paper	Conceptualisation, methodology, writing – review and editing.	
Signature	Date: 04 April 2022	

4.2 Abstract

Information on the movement ecology of threatened species can assist with identifying potential barriers to dispersal and appropriate management units and actions. We focus on the movement of the threatened Malleefowl (*Leipoa ocellata*) in fragmented landscapes and the possible effects of climate change on movement. We used solar-powered GPS telemetry to collect movement data and determine the influence of breeding status, remnant vegetation patches and environmental variables. Six males and one female were tracked between one and fifty months, resulting in 20,932 fixes. While breeding, Malleefowl had significantly smaller home ranges (92 ± 43 ha breeding; 609 ± 708 ha non-breeding), moved shorter daily distances (1283 ± 605 breeding; 1567 ± 841 non-breeding) and stayed closer to the mound (349 ± 324 m breeding; 3293 ± 2715 m non-breeding). Most Malleefowl effectively disassociated from the incubation mound once breeding stopped, with two birds dispersing up to 10.2 km. Movement patterns were significantly influenced by the size of the remnant native vegetation patch, with smaller home ranges being utilised in small patches than in large patches. One male almost exclusively remained within a 107-ha patch for over four years, but a female crossed between closely spaced uncleared patches. Long-range movements of nearly 10 km daily displacement were recorded in large remnants almost exclusively when not breeding. Temperature and rain had a significant but small effect on movement: modelling suggests daily distance travelled declines from 1.3 km at 25°C to 0.9 km at 45°C. The influence of patch size on movement patterns suggests that Malleefowl movement may be governed by the size of remnant habitat patches and that habitat continuity may be important for maintaining genetic diversity or facilitating recolonization after drought or other catastrophic events. Climate change may reduce Malleefowl movement during hot, dry periods with possible effects on breeding success.

4.3 Keywords

Habitat fragmentation, conservation ecology, semi-arid zone, dispersal, utilisation distribution, GPS telemetry, dynamic Brownian bridge movement models

4.4 Introduction

Malleefowl (*Leipoa ocellata*) are large, ground-dwelling birds that build mounds from soil and leaf litter to incubate their eggs. Endemic to Australia, historically abundant and widely distributed, they are now a threatened species listed as Vulnerable in Australia and globally (EPBC Act 1999; IUCN 2012). Malleefowl populations have been decreasing at least since European settlement and continue to decline in many parts of Australia (Benshemesh, Southwell *et al.* 2020). Declines are due to a combination of factors such as habitat loss and fragmentation, predation by the introduced fox and cat, competition with overabundant herbivores and the effects of dry conditions on soil moisture and vegetation (Benshemesh 2007; Ford, Barrett *et al.* 2001; Harlen and Priddel 1996; Priddel, Wheeler *et al.* 2007; Wheeler and Priddel 2009). Climate change is expected to exacerbate these pressures by reducing precipitation, increasing the frequency and intensity of periods with hot temperatures, and consequently leading to more droughts and wildfires (CSIRO and BOM 2015; Garnett and Baker 2021; Guerin, Christmas *et al.* 2018; Head, Adams *et al.* 2014; Hughes 2011; McKechnie and Wolf 2010).

Malleefowl are diurnal and prefer to walk, flying only to roost in trees or escape predation (Benshemesh 1992; Frith 1962b; Priddel and Wheeler 1997). They can spend up to eleven months a year (Frith 1959) and 44% of daylight hours (Weathers and Seymour 1998) at their breeding mound, with Frith (1959) observing maximum distances of 230 and 90 m from the mound for females and males, respectively. Non-breeding Malleefowl move further, and adult Malleefowl have been observed up to 11 km from their mounds, some after a wildfire destroyed their habitat (Benshemesh 1992; Booth 1985). Home range estimates vary widely from 3 to 370 ha for breeding and from 170 to 240 ha for non-breeding birds (Benshemesh 1992; Booth 1987a). Little is known of Malleefowl movement when they are not breeding and are thought to be less closely associated with their mounds (Benshemesh 1992; Booth 1985). We will investigate daily patterns in Malleefowl movement but also study their long-range movements. These could be classified as excursions which are temporary exploratory movements outside of the home range, but an endpoint within the home range (Dingle and Drake 2007; Earl, Fuhlendorf *et al.* 2016) or dispersal which is more permanent, with an endpoint outside of the home range, resulting in resettlement and often followed by reproduction (Clobert, Baguette *et al.* 2012). Knowing Malleefowl movement patterns, preferred habitats and use of croplands, as well as seasonal and climate or sex-driven differences will assist managers in identifying, protecting and restoring the most valuable habitat for Malleefowl conservation.

How Malleefowl use and move between habitat patches in agricultural matrices can inform managers about sizes and distances of patches most suitable for the protection and persistence of Malleefowl. How much time Malleefowl spend in open agricultural land and what distances of open land they are prepared to cross may inform about the size of patches and functional habitat corridors connecting these or about the management of matrix habitat itself to improve dispersal (e.g. planting paddock trees as stepping stones or improving road-side vegetation strips). It can also assist managers in timing management activities to decrease disturbance or potential exposure to chemicals via food plants (e.g. spraying weeds near the mound that could be eaten by Malleefowl during the breeding season).

Size and proximity of habitat patches and environmental variables may influence movement patterns. Many Malleefowl live in areas of fragmented native vegetation scattered within a matrix of agricultural land. How Malleefowl move between habitat patches and use cleared agricultural land is largely unknown although they have been observed feeding at the edge of wheat paddocks (Short and Parsons 2008; van der Waag 2004) and in roadside vegetation strips, suggesting roadside vegetation may facilitate movement between habitat patches (Benshemesh 2007). The ability of Malleefowl to move and disperse within and between habitat patches is critical to their survival, especially after drought, heatwaves or wildfire, all of which are predicted to increase in frequency. Understanding how such environmental factors influence movement patterns is thus imperative for implementing actions to protect or improve refuge habitat to enhance Malleefowl survival. Malleefowl have a heat tolerance of approximately 41 - 42°C (Booth 1984) and above these temperatures they show heat dissipation and avoidance behaviours like panting, gular fluttering, seeking shade and increasing rest periods (Benshemesh 1992). These behaviours can lead to reduced foraging efficiency, loss of weight and overall condition in birds (McKechnie 2019; van de Ven, McKechnie *et al.* 2019). Reduced movement is a known technique used to reduce heat stress in other animals (Shepard, Wilson *et al.* 2013) and arid-zone birds have shown reduced activity at high temperatures (Cooper, Withers *et al.* 2019; Funghi, McCowan *et al.* 2019; Pattinson, Thompson *et al.* 2020). In Malleefowl, reduced activity could lead to less time tending mounds or foraging, negatively impacting hatching success, recruitment and survival.

An understanding of how extrinsic and intrinsic factors influence movement patterns is essential to inform the appropriate scale of regional management. Most knowledge of Malleefowl ecology and behaviour comes from mound-based surveillance using cameras or direct observation (Benshemesh 1992; Frith 1962a; Neilly, Wells *et al.* 2021; Priddel and Wheeler 2003; Weathers and Seymour 1998). Malleefowl are well camouflaged and have acute hearing (Bellchambers 1916), making them difficult to observe away from the mound.

An initial understanding of Malleefowl movement ecology and time budgets was gained by using VHF radio tags (Benshemesh 1992; Booth 1985; Booth 1987a; Priddel and Wheeler 1996). In these studies, although transmitters were attached for up to two years, data were recorded intermittently and only for up to 15 consecutive days, and some transmitters could not be located after short periods. Benshemesh (1992) and Booth (1987a) lost 30% and 50% of VHF-tagged adults, respectively. These losses are indicative of the limitations of VHF tracking, as it is difficult to keep frequent track of birds on the ground for long periods, especially if they move out of their existing range. Malleefowl are difficult to catch and handle as they are prone to stress and become wary after capture, changing their movement patterns and making it difficult to collect unaffected movement data (Benshemesh 1992; Booth 1987a). This is problematic as patchy data can cause inaccurate home range estimates (Silva, Crane *et al.* 2018).

Solar-powered GPS trackers can be fitted to birds for long periods with remote data download and without the need for recapture, thus enabling the collection of continuous fine-scale data. We fitted GPS transmitters to Malleefowl in a semi-arid environment to compare movement patterns during the breeding and non-breeding period and investigate the influence of patch size and environmental variables. We predicted that patch size would limit movement and that Malleefowl would rarely use cleared land, as, to our knowledge, they have not been observed far away from native vegetation. We also predicted that Malleefowl would move less with increasing temperatures to avoid heat stress. Our research outcomes may inform managers about management actions including the size of habitat patches required to protect viable populations of the species.

4.5 Methods

4.5.1 Study Sites

Study sites were on the Eyre Peninsula in South Australia (Figure 4-1), where Malleefowl persist in fragmented native vegetation patches. About 39% of native vegetation cover on the Eyre Peninsula remains, mostly in marginal areas unsuitable for agriculture (Brandle 2010; NREP 2017).

We trapped Malleefowl at six mallee sites: Hincks and Hambidge Conservation Parks; one Heritage Agreement property near Lock (HA 370) and one near Cowell (HA 172), all with low to medium rainfall (320-400 mm); and at Secret Rocks Nature Reserve and a nearby private property (310 mm). Vegetation and rainfall details are in Table S 4-1.

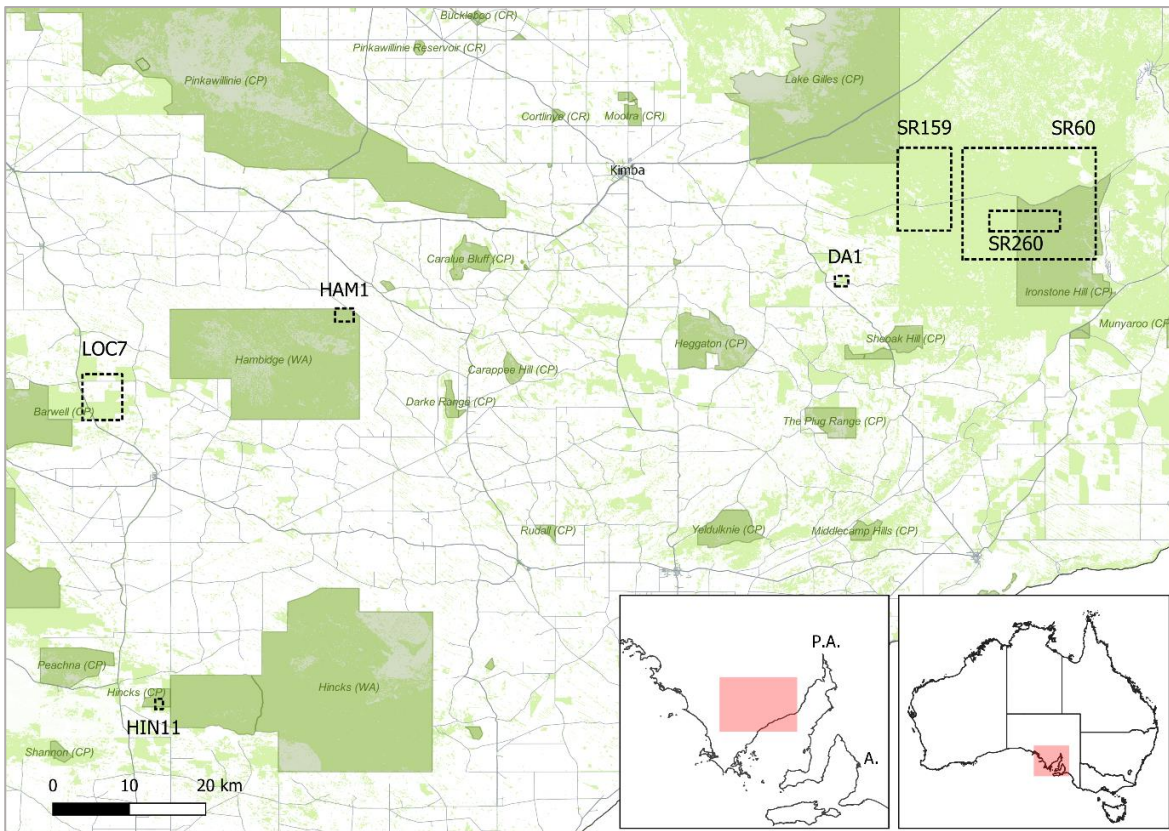


Figure 4-1 Overview map of the Eyre Peninsula study sites. Darker green shows government conservation areas and light green native vegetation cover. Dashed lines indicate outer boundaries of Malleefowl movement ranges and correspond to sections in Figure 4-5. P.A. = Port Augusta, A. = Adelaide.

4.5.2 Data collection

Malleefowl were trapped using a cage trap set on an active mound (i.e. while breeding) following Priddel and Wheeler (2003) but with a soft polyester net top (see 4.10.1). Captured birds were fitted with solar-powered GPS transmitters (Solar Argos/GPS 30 g PTT, accuracy ± 18 m; Microwave Telemetry Inc. (2018)). High-resolution locations (fixes) were calculated six to twelve times a day (4.10.1) and transmitted via satellite (Argos CLS 2021).

After capture, we determined whether birds returned to their mounds through GPS fixes and camera traps (HC600 HyperFire, Reconyx). In subsequent seasons, we determined breeding status through ground visits to areas with high-frequency fixes and cameras at mounds. Malleefowl were considered *breeding* when visiting the mound at least every two days and *non-breeding* when they remained more than 100 m away from the mound for five or more successive days. As capture causes stress and birds may subsequently move erratically, we disregarded all data until they returned to their mounds, or the first three days after capture if they did not. Two birds that died within five days of capture were excluded from all analyses.

Originally, home range was defined as the area an animal uses during its normal activities like foraging (Burt 1943). Modern tracking technology added a temporal dimension, enabling us to determine the intensity of use of areas within the home range, known as the utilisation distribution (UD; Worton 1989). Here, we use the term *home range* for the 95% UD (areas used 95% of the time), estimated with dynamic Brownian bridge movement models (dBBMM; Kranstauber, Smolla *et al.* 2020). dBBMMs perform better in estimating home ranges than other methods by strongly reducing type-2 errors (inclusion of unused areas), allowing for irregular sampling, behavioural heterogeneity and high data volumes (Kranstauber, Kays *et al.* 2012; Walter, Onorato *et al.* 2015). For comparison with previous studies, we calculated the 95% minimum convex polygon (MCP) with `adehabitatHR` (Calenge 2006). *Total range length* was the linear distance between the two most distant fixes per patch, breeding stage or the whole tracking period.

4.5.3 Movement patterns

To investigate Malleefowl movement patterns, we calculated a range of summary statistics using days where at least 75% of scheduled fixes were successful (4.10.1) and looked at variation between breeding states, sexes, seasons and times of day (4.10.1, Comparative statistics). A “bird day” was 24 hours beginning just prior to the first dawn record. We recorded the *daily distance* (sum of all distances moved between fixes (non-linear) for a bird day), *daily nett displacement* (the distance between the first and last fix of that bird day, the greater the nett displacement, the greater the directionality of movement; henceforth *daily displacement*); *distance to mound* (distance from a fix to the current mound when breeding and the previous season’s mound if not breeding) and *hourly nett movement* (henceforth *hourly movement*, straight-line distance between two consecutive fixes (m) divided by the time (h) between the same two fixes). Further details can be found in 4.10.1.

We also investigated the occurrence and frequency of Malleefowl long-range movements. Movement was considered g-range when the daily displacement of a bird exceeded four times the mean daily displacement of all birds (502 m). Long-range movements could be excursions or dispersal. We defined excursions as temporary exploratory movements outside of the home range, with an endpoint within the home range (Dingle and Drake 2007; Earl, Fuhlendorf *et al.* 2016) and dispersal as more permanent movements, with an endpoint outside of the home range, resulting in settlement in a new habitat and typically (but not necessarily) followed by reproduction (Clobert, Baguette *et al.* 2012).

4.5.4 Effect of environmental factors on movement patterns

To examine the effects of temperature and rainfall on movement we used linear mixed-effects models. As one individual (DA1) was tracked over multiple seasons, we used a

continuous correlation structure that tests for autocorrelation between subsequent observations within individuals. To allow for individual behavioural differences in different periods, we used an interaction term of bird-ID and breeding season as the random effect. We used a Box-Cox transformation model to determine the best transformation parameter to ensure model linearity. The response variables were daily distance or hourly movement. The fixed effects were daily minimum and maximum temperature (°C) and rainfall (yes/no). For hourly movement, we focused on daytime (4.10.1). For night-time hourly movement, we included 'moon illumination' (new(0) - full(1) moon) as an explanatory variable.

4.5.5 Effect of breeding status and patch size on home range and total range length

We investigated the effect of breeding and patch size on home range and total range length. We modelled the response variables separately as a function of patch size using mixed-effects models (lmer). We used bird id as the random effect and breeding state and patch size and their interaction term as the main effects. Response variables and patch size were log transformed to fulfil model linearity assumptions and improve model fit. To reduce the influence of short tracking periods, we only used patches where birds remained longer than 12 days. Patch size was the size of the area of uninterrupted remnant native vegetation available to each Malleefowl.

Data analyses were performed with R Studio v1.4.1106 (R Core Team 2020) and the following packages: tidyverse (data cleaning and graphs; Wickham, Averick *et al.* 2019), nlme (mixed effect modelling; Pinheiro, Bates *et al.* 2006), MuMin (R²; Barton 2019), car (Box-Cox and variance testing; Fox and Weisberg 2019), ggstatsplot (comparative statistics; Patil 2021), suncalc (times of day; Thieurmel and Elmarhraoui 2019) and effects (model plots; Fox and Hong 2009). Maps were made with QGIS 3.16 (QGIS Development Team 2021).

4.6 Results

We trapped eight male and one female Malleefowl between December 2016 and December 2019. Of the nine tagged birds, eight died during the study. Two died within five days of release, one likely from stress and the other from fox predation but stress may have been a contributing factor. The remaining six birds died between 36 and 452 days after release. Three birds died from cat predation (one confirmed with DNA), one from fox or raptor predation, and one probably of heat stress during a drought. One transmitter stopped and the bird could not be found. Only one bird remained at the end of the study and had been tracked for over 1530 days. We collected a total of 20,932 GPS data points with a fix success rate of 88.8%, resulting in 18,645 successful fixes. One bird (DA1) had particularly

low fix success rates in winters, with, at worst, no fixes recorded from May to July 2019 (Table 4-1). After data cleaning (4.10.1) 17,356 records remained. The cause of death was determined by the condition and location of any remains found. Chewed feather ends and protected kill sites indicate cat predation but cut feather ends, teeth marks on tags and scattered feathers are more likely indicative of fox predation, while plucked feathers are a sign of raptor predation (Priddel and Wheeler 1994). DNA swabs were taken from four sets of remains and sent to an external laboratory (www.helixsolutions.com.au) to determine the presence of predator DNA (Berry and Sarre 2007). Two swabs returned cat DNA (Table 4-1).

Table 4-1 Overview of tracked Malleefowl on the Eyre Peninsula. We present the trapping location, bird-ID, tracking period and duration, cause of death (fate) and number (percentage) of successful day and night-time fixes for each bird. F = Female. ¹Cat DNA present. ²DNA Failed to amplify. ³Chewed feather ends. ⁴Cut feather ends. ⁵Tooth mark on transmitter. ⁶Feathers plucked. ⁷Kill cached. ⁸Feathers scattered

Location	ID	Tracking period	Days tracked	Fate	Day fixes	Night fixes
Cowell	COW17	15/02/2017–16/02/2017	1	Suspected fox ^{4,8}	-	-
Cowell	COW41	27/11/2017–03/12/2017	5	Suspected fox ^{2,4,8}	-	-
Yalanda	DA1	15/01/2017–25/03/2021	1530	Alive	5451 (77.5%)	1401 (66.7%)
Hambidge	HAM1	01/12/2016–21/05/2017	171	Cat ^{1,3}	912	250
Hincks	HIN11	10/12/2016–15/01/2017	36	Suspected cat ³	196	32
Lock (F)	LOC7	13/02/2017–22/10/2017	251	Suspected fox/raptor ^{2,5,6,8}	1643 (99.8%)	1239
Secret Rocks	SR159	18/11/2018–17/11/2019	364	Suspected heat stress in drought	1795	568
Secret Rocks	SR260	02/12/2017–27/02/2019	452	Cat ^{1,7}	2307	656
Secret Rocks	SR60	13/12/2019–17/11/2020	340	Unknown	1649 (99.9%)	546 (31.2%)
Total			3150		13953	4692

Malleefowl spent at least 97.5% of their time in native vegetation. Three Malleefowl were caught at mounds that were within 300 m of agricultural land. DA1 was recorded moving up to 77 m onto agricultural land, but fixes on agricultural land accounted for only 1.3% of 5201 fixes (only October to March, as most complete monthly data). Fixes were most frequently found on agricultural land in November (2.5% of monthly fixes), followed by October (<2%) and December (1.5%), all just before and during crop harvesting time in December (confirmed by landowner, pers. comm). In contrast, January to March only had 0.6 to 0.9% of fixes on agricultural land. Only 1.5% of HA1's fixes were on agricultural land and all fixes

were less than 20 m from the edge of native vegetation, suggesting they could be an artefact of GPS inaccuracy (± 18 m). LOC7, the female, only used agricultural land to cross to other patches of native vegetation.

4.6.1 Movement after capture

Capturing Malleefowl at their mounds affected their movement behaviour for various periods after trapping and only four males of the nine tagged Malleefowl returned to their mounds to resume breeding activities. Three of these birds returned after two (DA1), four (HAM1) and eleven (HIN11) days, and recommenced mound maintenance with their mate. Two of these three pairs completed their breeding season, and the third pair (HIN11) continued until the tagged male died 36 days after release (the female continued tending the mound for eleven days afterwards). The fourth male (SR260) returned to the mound after 24 days by which time his mate, who had been tending the mound by herself for eight days after his capture, had left the mound. He resumed mound tending activities by himself for a further 22 days (Table S 4-2).

The five birds that did not return to the mound included the female that was tracked for 251 days, two males that were tracked for around a year each and two males that died within five days of capture.

During the three days after capture that were removed from analyses, the two males that did not return to their mounds moved up to 1500 m and the female up to 1280 m from the mound. Of the four males that did eventually return to their mounds, SR260 moved over 3.2 km away from their mound in the first three days, while the other three stayed within 1000 m.

In the week before trapping, of the males that returned to their mound, SR260 was recorded at the mound daily by camera-trap (for four days only, as the SD card was full before that for about four weeks, before which the mound was tended daily), DA1 daily except on one day, HIN11 on two days and HAM1 was not recorded at all as the camera was only set up two days before capture. Of the birds that did not return after capture, LOC7, SR159 and COW41 were at the mound every day, COW17 was there on all but two days and SR60 was only seen once the day before trapping. Camera trap photos indicate that all non-tagged breeding mates continued visiting the mound when their mates were absent (Table S 4-2).

4.6.2 Home range and total range length

Malleefowl home range and total range length were influenced by breeding status and patch size. Non-breeding home ranges averaged 609 ha (± 708 ha s.d.) and ranged from 41 to 2,168 ha. Breeding home ranges were much smaller, averaging 92 ha (± 43 ha) and ranging from 44 to 176 ha (Table 4-2). The female had the largest breeding home range (176 ha) compared with the males (44 - 100 ha) but low sample size precluded statistical significance. The female used three patches but two were excluded to reduce the influence of short tracking periods (Table S 4-3). All males remained within one patch.

Table 4-2 Summary of movement metrics of Malleefowl on the Eyre Peninsula. We present sex (F = Female, M = Male); breeding stage (B = Breeding, NB = Non-breeding (number of stages used for analyses); home range (95% UD, ha); total range length (km); and mean \pm 1 s.d. and maximum of daily distance, daily displacement, distance to mound and hourly movement (m). Maxima for each sex are in bold. Details in Table S 4-3.

		Home range	Total range length	Daily distance	Daily displacement		Distance to mound		Hourly movement		
		mean \pm sd	mean \pm sd	mean \pm sd	max	mean \pm sd	max	mean \pm sd	max	mean \pm sd	max
F	B (1)	176	2.2	1830 \pm 517	2956	566 \pm 324	1396	470 \pm 267	1507	117 \pm 82	484
	NB (1)	234	4.7	1565 \pm 557	3720	501 \pm 378	1812	5599 \pm 1178	7096	102 \pm 96	697
M	B (7)	79 \pm 23	1.7 \pm 0.4	1256 \pm 596	3896	296 \pm 247	1098	338 \pm 326	2062	66 \pm 78	537
	NB (7)	591 \pm 721	5.7 \pm 5.6	1571 \pm 880	10394	647 \pm 686	9738	2639 \pm 2670	12969	87 \pm 98	959
	B	92 \pm 43	1.8 \pm 0.5	1283 \pm 605	3896	310 \pm 258	1396	349 \pm 324	2062	69 \pm 79	537
	NB	609 \pm 708	6.1 \pm 5.3	1567 \pm 841	10394	624 \pm 653	9738	3293 \pm 2715	12969	89 \pm 97	959

Total range length was calculated as the distance between the two most distant fixes for each bird in the breeding and non-breeding period and both combined. Total range lengths varied from 0.9 to 14.4 km when not breeding and were larger than when breeding (1.1 to 2.4 km). The greatest total range length for the complete tracking period was 23.1 km for a male (SR60). The female's total range length over the complete tracking period was 7.8 km (Table S 4-3).

Over 77% of the variation in home range size ($R^2 = 0.774$) and total range length ($R^2 = 0.786$) could be explained by the factors in the mixed-effects models. Models showed that breeding status and patch size were significant predictors of both home range size and total range length. Home range size was positively related to patch size for non-breeders ($r = 0.25$, 95% CI [0.11, 0.39], $p < 0.001$), but not for breeding birds ($p = 0.3$, Figure 4-2). Home ranges ($r = -1.45$ [-2.44, -0.47], $p = 0.01$) and total range lengths ($r = -1.33$ [-2.25, -0.42], p

= 0.01) were significantly smaller at small patch sizes during non-breeding periods but not during the breeding season (Table S 4-4, Figure S 4-1). For example, DA1 (tracked 1530 days) was resident in a 107 ha patch and his home range never exceeded 98 ha. In contrast, SR60 (tracked 337 days) lived in a patch of over 200,000 ha and his non-breeding home range was more than 20 times larger at 2,168 ha.

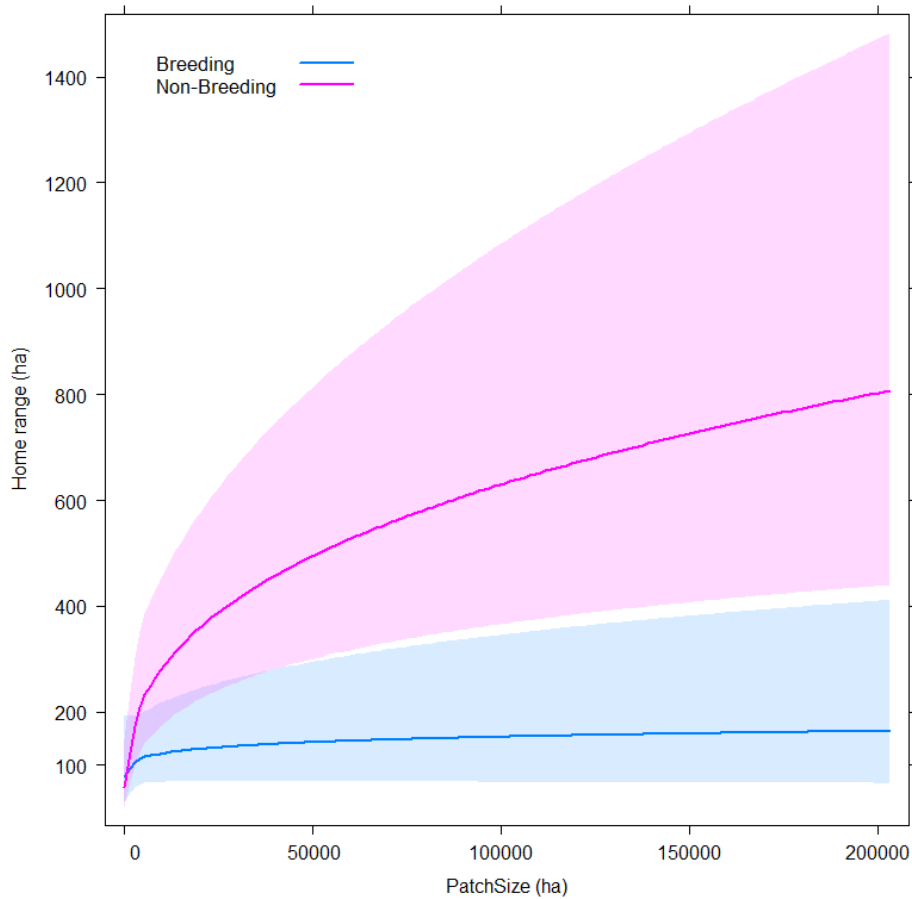


Figure 4-2 Predicted home range (ha) values by patch size (ha) for Malleefowl on the Eyre Peninsula (n = 7) while breeding and non-breeding. The lines are the fitted multiple linear regression and shaded areas are the 95% confidence intervals.

4.6.3 Movement metrics

The results of the comparative statistics presented in the below four sub-sections can be found in Table S 4-5.

4.6.3.1 Breeding

Non-breeding Malleefowl moved significantly more, further from the mound and more directionally than breeding birds (Figure 4-3). Non-breeding birds were located on average nearly an order of magnitude further from the mound than breeding birds (NB: 3292 ± 2715 m; B: 349 ± 324 m; $r_{tb} = -0.95$). Non-breeders (624 ± 653 m) showed significantly higher

daily displacement ($r_{rb} = -0.49$) than breeding birds (310 ± 258 m, Figure S 4-2). Non-breeding birds travelled 1568 ± 841 m each day, which was significantly further ($r = -0.28$) than breeding birds 1283 ± 605 m. A small effect ($r_{rb} = -0.1$) on hourly movement was observed with breeding birds travelling less per hour than non-breeders (NB: 89 ± 97 m; B: 69 ± 79 m).

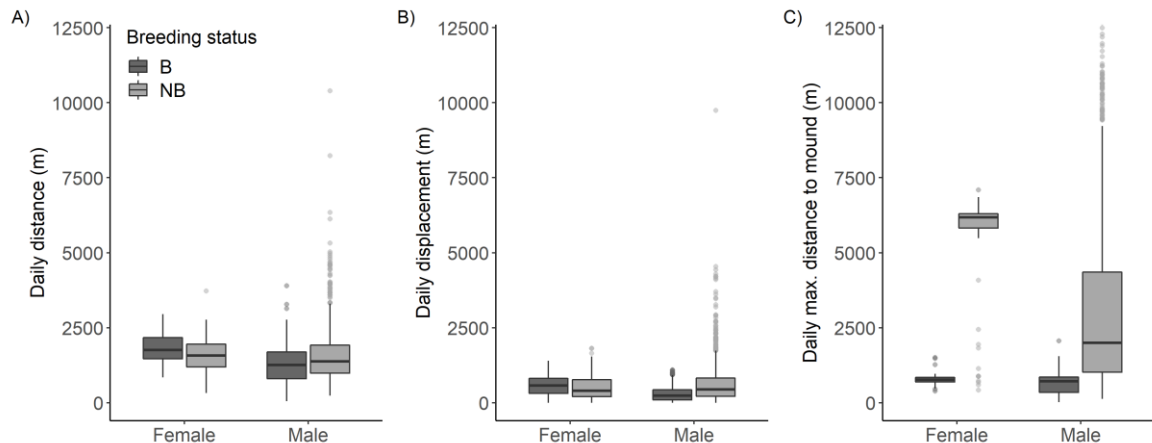


Figure 4-3 Daily movement statistics of Malleefowl on the Eyre Peninsula by sex and breeding status. A) daily distance (sum of distances between all consecutive fixes during a day), B) daily displacement (distance between first and last fix of a day) and C) maximum distance away from the mound recorded per day.

4.6.3.2 Sex

The breeding female moved significantly further in all measured distance categories than the breeding males ($n = 3$). The female travelled 1829 ± 517 m each day which was significantly further ($r_{rb} = 0.52$) than 1256 ± 596 m for males. The female's movements were also more directional with 566 ± 324 m per day versus 296 ± 247 m for males ($r_{rb} = 0.49$). The female moved 117 ± 82 m per hour and males 66 ± 78 m ($r_{rb} = 0.43$), with maxima of 697 and 959 m, respectively. The female was on average 470 ± 267 m away from the mound and the males 338 ± 326 m ($r_{rb} = 0.28$).

The non-breeding female moved significantly further than the non-breeding males in three movement metrics but there was no influence of sex on daily displacement. The largest effect ($r_{rb} = 0.64$) was observed for distance to mound, with the non-breeding female moving up to 5599 ± 1178 m away from the mound and the males an average 2639 ± 2670 m. Effect sizes for daily distance and hourly movement were weak ($r_{rb} = 0.1$).

4.6.3.3 Seasons

Non-breeding males ($n = 5$) showed significant seasonal differences in daily and hourly movement ($W_K = 0.24$) as well as daily displacement ($W_K = 0.29$). Average daily distances

travelled were significantly longer during spring ($1,770 \pm 1,136$ m, $p = 0.031$) and summer ($1,624 \pm 899$ m, $p = 0.031$) than during winter ($1,475 \pm 804$ m) but not significantly different than autumn ($1,463 \pm 678$ m). Daily displacement was significantly higher in spring (850 ± 1025 m) than in autumn (533 ± 479 m, $p = 0.028$) and winter (612 ± 618 m, $p = 0.028$). Daily displacement in summer (647 ± 570 m) was not significantly different from the other seasons.

4.6.3.4 Time of day

Hourly movement patterns (Figure 4-4-A) changed significantly throughout the day in all individuals and both breeding stages ($W_K = 0.24$): Average hourly movement was shortest at night when roosting (9 ± 18 m, likely attributable to GPS precision variability) and increased throughout the day from 23 ± 34 m at dawn to 83 ± 91 m during the day and 133 ± 102 m at dusk (pairwise comparisons were all significant with $p < 0.001$). While the mean hourly movement of all Malleefowl was 63 ± 86 m, they walked as far as 959 m per hour.

Distance to mound also differed significantly throughout the day ($W_K = 0.24$; Figure 4-4-B). Breeding Malleefowl were closest to the mound during the day (240 ± 288 m), significantly further away at dawn (406 ± 315 m; DC: $p < 0.001$), dusk (492 ± 321 m; DC: $p < 0.001$) and night (506 ± 315 m; DC: $p < 0.001$). When we further split the breeding birds by sex, we found a major difference in the daytime unlike the other times of day, when the breeding female was furthest from the mound (550 ± 238 m), while the males were the closest (217 ± 278 m).

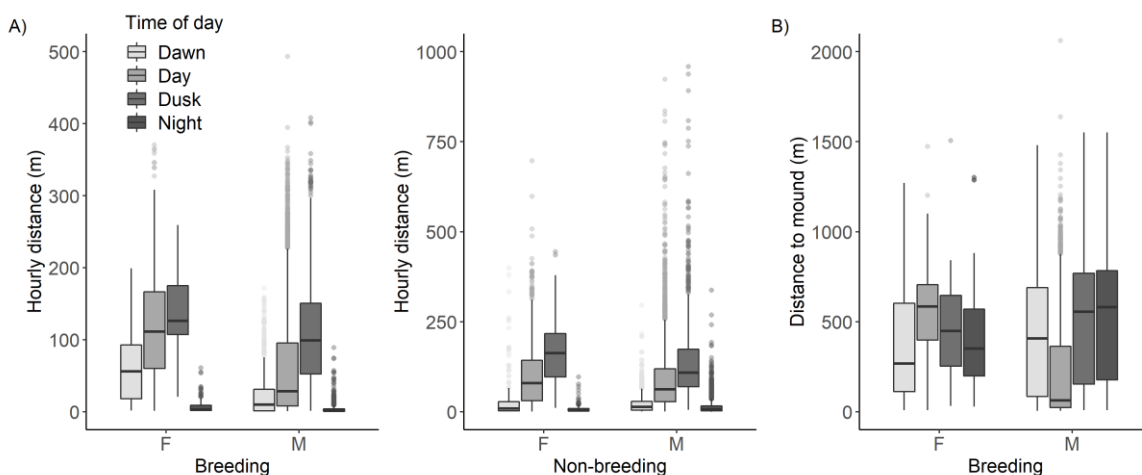


Figure 4-4 Hourly movement statistics of Malleefowl on the Eyre Peninsula by sex. A) hourly distance travelled by breeding status, B) distance away from the mound only while breeding. Note the different y-axis scales.

4.6.4 Movement patterns

We observed three modes of movement in the Eyre Peninsula Malleefowl. Some birds were sedentary and didn't move outside their home range, others showed short-term exploratory movements either within or outside of their home range, and others dispersed to new areas. Dispersal and excursions occurred almost exclusively when birds were not breeding, and the extent of these movements seemed to be influenced by patch size.

4.6.4.1 Dispersal

Dispersal was observed in two non-breeding Malleefowl. The female (LOC7) never went back to her mound after trapping and after seven days left her initial patch. From there, she first moved to a small patch of native vegetation 1.7 km away, and after three days, moved to a large patch 4.2 km away, where she settled and maintained a mound in the following breeding season. As fixes were two hours apart in each case, we cannot know the exact path, but the patches of uncleared habitat present suggested that the longest distance traversed in the open was about 250 m (Figure 4-5). LOC7 died 52 days into her new breeding season. SR60 also did not return to his mound after capture and moved to an area north of his trapping site for over eight months where a home range was established of over 2,000 ha. Ultimately, he resettled approximately 10 km away to the east of the capture site in September 2020 and established a new home range (Figure 4-5 and Figure S 4-3).

4.6.4.2 Excursions

Exploratory movement with large daily displacement occurred much more frequently than dispersal. Three Malleefowl that lived in large habitat patches made short-term excursions outside and within their home ranges, almost exclusively while not breeding (exception: SR260 while single). For example, SR159, in 362 days of tracking, had two excursions outside of his home range with a daily displacement of up to 9.8 km (Figure 4-5; Table S 4-6). The second excursion was particularly noteworthy because SR159 travelled a linear distance of 10 km away from his home range in three days before returning to it the next day. In the next two days, SR519 moved daily total distances of 0.9 and 2 km on the outside of the enclosure, then crossed into the enclosure and moved 1.3 and 1.6 km over the next two days and died on the morning of the fifth day of suspected heat stress (Table S 4-6 and Figure 4-5, top-right). SR159 also left and returned to the pest-proof enclosure (4.10.1) it was captured in on many occasions (Figure 4-5). In contrast, SR260 showed high displacement movement predominantly inside his home range. He initially frequently commuted between the western and central parts of his home range approximately 3-4 km apart. Later, from winter onwards, he ranged further and moved between the eastern and western parts of his range which were approximately 7 km apart (Figure 4-5; Figure S 4-4;

Table S 4-6). SR260 made a total of 21 long-range movements of one to three days with a daily displacement of up to 4.5 km in 426 days of tracking (Table S 4-6). SR60 had 17 long-range movements of one to two days with a maximum daily displacement of 4.3 km in 337 days of tracking. Two excursions were outside the home range, of which one was part of a dispersal (Figure 4-5; Figure S 4-4).

4.6.4.3 Sedentarism

The male in the smallest patch of native vegetation (DA1) was the most sedentary and showed no noteworthy exploratory behaviour. His average home range was the smallest at 64 ± 22 ha, and he very rarely left his 107 ha patch of native vegetation in four years (Figure 4-5). Two birds that lived in large patches, HAM1 and HIN11 also did not show exploratory behaviour.

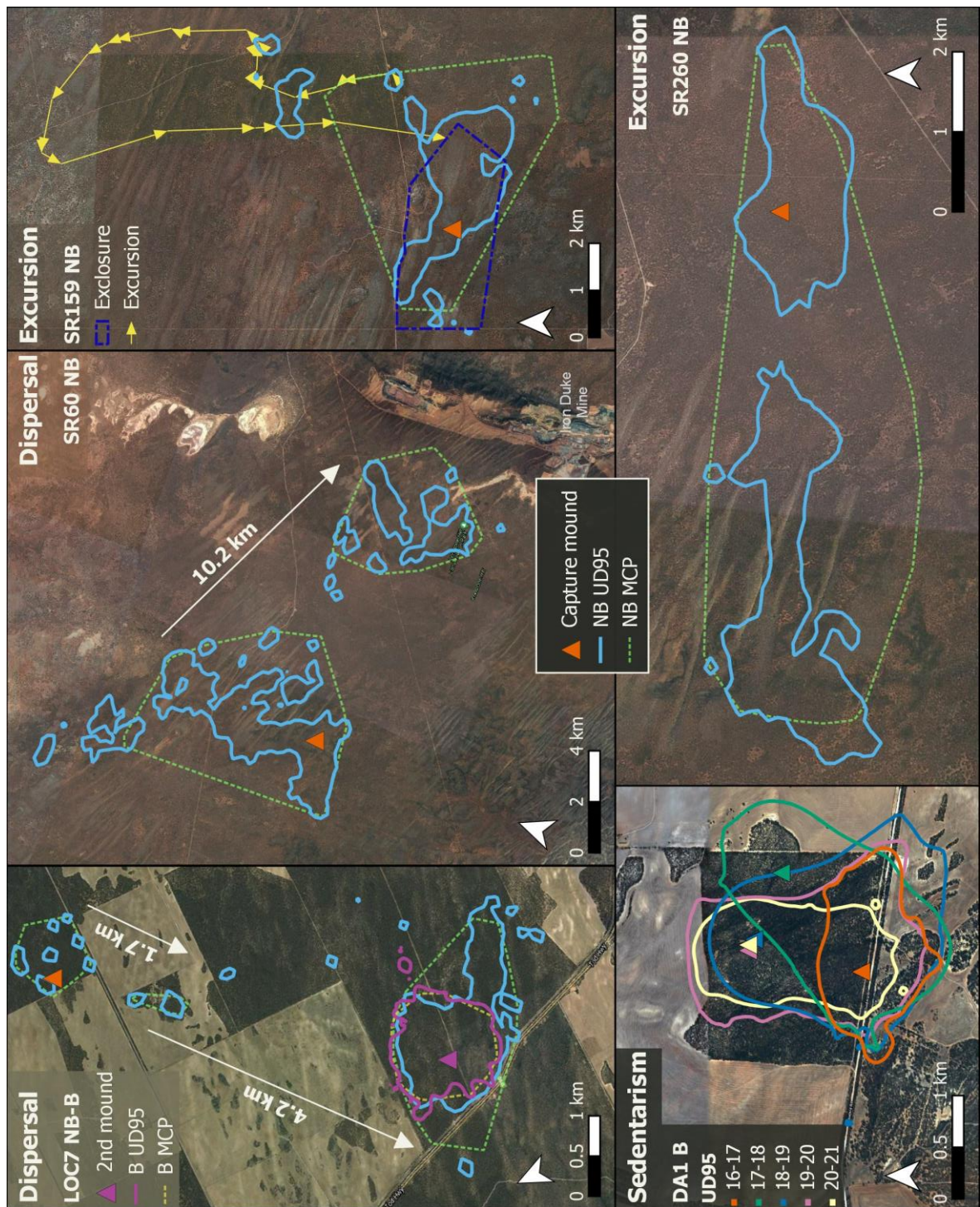


Figure 4-5 Examples of movement patterns recorded in the Eyre Peninsula Malleefowl (clockwise from top left): **Dispersal** of LOC7 in fragmented habitat (initially NB, then B); **Dispersal** of SR60 in continuous habitat (NB); **Excursion** outside of the home range in SR159 in continuous habitat (NB); home range in SR260 (NB); **Sedentarism** in DA1 over five years in fragmented habitat (B). *Symbology*: Light blue contours: NB 95% UD; green dashed contours: NB 95% MCP; orange triangles: mound where bird was caught; white arrows: dispersal distance and direction. LOC7 - Magenta contours & triangle: 95% UD and mound of the breeding season after dispersal; yellow dashed contours: B 95% MCP. SR159 - Dark blue dashed line: pest-proof enclosure. Edges of each map correspond to the dashed lines in Figure 1.

4.6.5 Environmental effects

Only 5.7% of the variation in daily distances moved could be explained by the environmental data while breeding, where daily distance decreased with higher maximum temperature (Figure 4-6, Table S 4-7). However, only 1.8% of the variation was explained when birds were not breeding, with daily distance decreasing with higher maximum temperature but increasing with rising minimum temperature or during rain (Figure 4-6). While breeding, with a minimum temperature of 13°C (mean for breeding) and no rain, our models predict a daily movement of 1314 m [1090,1550] at 25°C, 1106 m [894,1332] at 35°C, and 910 m [692,1145] at 45°C. For a 1°C increase in maximum temperature from 24 to 25°C daily movement decreased by 1.6%, from 34 to 35°C by 1.8%, and from 44 to 45°C by 2.1%.

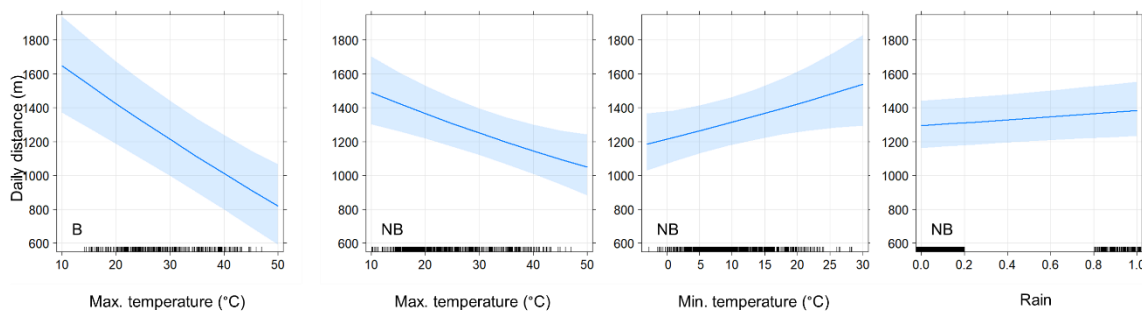


Figure 4-6 Linear mixed-effects model predictions of daily distance (m) travelled by Malleefowl on the Eyre Peninsula. The left panel shows the modelled estimates for the breeding period vs maximum temperature (°C). The right three panels show the estimates while non-breeding for maximum and minimum temperature and the presence or absence of rain. Data were analysed on a transformed scale (SI table 5), but effects are displayed on the scale of the response (m) for easier interpretation. The shaded areas are the 95% confidence intervals for the predicted values. The rug plot at the bottom shows the location of the explanatory values.

Up to 7.7% of the difference in hourly movement during the daytime could be explained by environmental factors (Table S 4-7). Increasing maximum temperature led to a decrease in hourly movement during the day (Figure 4-6). While breeding, with a minimum temperature of 13°C (mean for breeding) and no rain, our model predicts an hourly movement of 69 m [53,87] at 25°C and 34 m [22,48] at 45°C. For a 1°C increase in maximum temperature from 24 to 25°C models predict that daily movement will decrease by 3%, from 34 to 35°C by 3.4%, and from 44 to 45°C by 4.1%. While not breeding, the influence of maximum temperature was less pronounced with a 2% decrease in hourly movement for a 1°C increase between all maximum temperatures over 23°C. Our model predicts an hourly movement of 72 m [62,83] at 25°C and 48 m [40,58] at 45°C. When not breeding, the presence of rain (at mean temperatures for the non-breeding period) increased hourly movement by 8.7% from 73 m [63,85] to 79 m [68,93] (Figure 4-6). For dawn, dusk or night only a negligible portion of the variation (<1.5%) could be explained. Moon illumination did

not affect night-time movement. For all models, most of the variation was explained by differences in individual behaviour in each breeding period (R^2c , Table S 4-7).

4.7 Discussion

We found that breeding status and patch size had the most significant influence on Malleefowl movement, together accounting for 83% of the recorded variation in Malleefowl movement. Breeding birds were more sedentary, always stayed within their home ranges and did not undertake any long-range movements. As a result, breeding birds had significantly smaller home ranges, shorter total range lengths, daily and hourly movement, and remained closer to the mound than non-breeding birds. Average home range estimates of 92 ha (95% UD) and 83 ha (95% MCP) for breeding birds are smaller than those recorded by Booth (1985) (M: 260 ha, n=1; F: 370 ha, n=1) but larger than those of Frith (1962b) (M: 3-20 ha, F: 17 ha; n unknown) and Benshemesh (1992) (F: 49-75 ha, n=2), possibly due to differences in rainfall, habitat or methodology.

When not breeding, the home range size of our Malleefowl increased sixfold, daily displacement more than doubled to over 600 m, and the average distance to the mound increased tenfold. These results support previous studies that found Malleefowl are tightly bonded (philopatric) to their mound during the breeding season and roam further from the mound during the non-breeding season (Benshemesh 1992; Booth 1985). Interestingly, mean daily movement increased to a much lesser extent during the non-breeding season, from 1.3 km per day when breeding to 1.6 km, but the highest daily movement and displacement were recorded in spring. This suggests that the distance travelled by Malleefowl each day is fairly consistent year-round but that non-breeding birds move either for a longer period each day or in a more directional manner and roam over a larger area, especially in spring, which Malleefowl usually spend in preparation for breeding. This seasonal increase in movement and disassociation from the mound may enable Malleefowl to search for better nesting grounds, locate potential mates, escape disturbance or reduce pressure on food resources close to mounds. It may also reflect a decline in populations (see long-range discussion below). Such disassociation was suspected by Benshemesh (1992) when following non-breeding Malleefowl became very difficult, and our study shows Malleefowl can move up to 13 km from their breeding mound after breeding has finished.

Sex differences were also apparent during the breeding season when the female moved significantly more, further from the mound and more directionally than males. Males remained closest to the mound during the day whereas the female was furthest from the mound at this time. This confirms the males' tighter bond to the mound than the females' (Neilly, Wells *et al.* 2021; Weathers and Seymour 1998). Breeding females presumably

move further to forage and cover the energy demand of egg production (Weathers and Seymour 1998; Weathers, Seymour *et al.* 1993). However, as only one female was tracked, it is hard to know if movement patterns of this individual were different because it was a female or for other reasons. However, we think presenting and contrasting the female and male data still has merit, as this female *may be* representative of a larger number of females. Additionally, only two birds showed dispersal behaviour and while the female's dispersal was likely triggered by capture, the male dispersed many months after capture and the reason for dispersal is unclear. This makes interpretation and generalisation of these findings difficult, and more research needs to be done in this area to enable more definitive conclusions. Similarly, all three birds that displayed long-range movement and never resumed breeding were all in the largest continuous native vegetation patch, which may have confounded the results. Conversely, one male that was also in a large patch did not move long distances.

The consistent sedentary nature of Malleefowl recorded during the breeding season was unrelated to patch size. In contrast, a wider range of movement patterns was recorded when Malleefowl were not breeding including sedentarism (Berbert and Fagan 2012), excursions (Bell 1990) and dispersal (Clobert, Baguette *et al.* 2012) which were related to patch size. One Malleefowl in the smallest patch of uncleared vegetation exhibited sedentarism when not breeding, but in contrast, long-range excursions were observed in three non-breeding Malleefowl that lived within the largest areas of continuous habitat. Mound activity has declined across the Eyre Peninsula over the last few decades (Stenhouse and Moseby 2022) and thus long-range movements may be an effort to locate mates where they occur in low densities. Other excursions are likely related to searching for food, water or familiarisation with the surrounding topography (Bell 1990). Malleefowl usually fulfil their water needs through food and drink little even when it is available, however, high ambient temperature may increase the need for water (Booth 1987b). Two Malleefowl went to areas where satellite surface water records (CSIRO Data61 2019) showed that water was present intermittently up until 2018 (but not at the time of excursion) in ephemeral lakes, dams and other surfaces. One Malleefowl died after an excursion to such an area in November 2019, the end of the driest year on record in South Australia. Drought-related Malleefowl deaths have been reported in the past (Priddel and Wheeler 2003). Such excursions suggest these long-lived birds may have spatial knowledge over an area much larger than their home range, acquired over many years, as also suggested by Berbert and Fagan (2012).

Two Malleefowl dispersed during the study. The female resettled seven days after being caught and commenced breeding in a new patch the following season. The female's late capture in the breeding season may have triggered dispersal, as late captures have caused

cessation of egg-laying and abandonment of breeding activities previously (Benshemesh 1992; Booth 1987a). However, a male dispersed only nine months after capture and other captured birds did not disperse at all, suggesting dispersal may also be triggered by other events. Malleefowl are known to move to other mounds within continuous native vegetation due to habitat clearance, fire, trapping and available food sources, but some mound changes also appear to occur randomly as mounds abandoned by one pair after one season were used by another pair successfully in the next (Benshemesh 1992; Frith 1959). Only Benshemesh (1992) reported the 'emigration' (i.e. dispersal to new patches) of four Malleefowl after a fire. In contrast, Frith (1962a) reported that most Malleefowl struggle to colonise new areas when their habitat is heavily cleared and perish. Seasonal movements and (predominantly female) long-range dispersal in other medium-sized ground-dwelling birds that live in similarly fragmented habitats highlight the importance of dispersal in population connectivity and resulting gene flow (Earl, Fuhlendorf *et al.* 2016; Vogel 2015).

Although Malleefowl in our study were regularly recorded crossing unsealed roads up to 20 m wide, they very rarely used open agricultural land and only crossed cleared areas that were less than 250 m wide. Benshemesh (1992) also found Malleefowl used corridors of vegetation instead of travelling over burnt country. Avoiding open spaces is likely a self-defence mechanism against raptors, which have been reported to attack Malleefowl (Korn 1986; Priddel and Wheeler 1994; Priddel and Wheeler 1996) and caused Malleefowl to hide in dense scrub (Frith 1962a). Although Malleefowl have been recorded at the edge of paddocks and roads feeding on grain (Benshemesh, Southwell *et al.* 2020; Short and Parsons 2008; van der Waag 2004), our results show they rarely ventured into cleared areas and when they did, they remained within 80 m of uncleared vegetation.

While conservation preference is given to larger and connected vegetation patches (Margules and Pressey 2000; Moilanen, Franco *et al.* 2005) research shows that smaller, fragmented patches are also important and have high conservation value (Fahrig, Arroyo-Rodríguez *et al.* 2019; Volenec and Dobson 2020; Wintle, Kujala *et al.* 2018). One Malleefowl in our study remained in an isolated 107 ha patch of vegetation for four years and maintained a mound each year suggesting that relatively small habitat patches can represent important habitat for the species. Benshemesh, Southwell *et al.* (2020) found Malleefowl had higher breeding activity in smaller rather than larger patches. However, although Malleefowl were able to persist and breed in small patches, these isolated patches restricted movement, particularly in the non-breeding season. The effect of this restriction is unknown but is likely to include difficulty in recolonising patches after fire or drought, inbreeding depression and increased risk of extinction due to stochastic events (Lacy 2000). Research on other ground-dwelling birds has shown that occupancy is related to patch size

and proximity to other patches (Bollmann, Graf *et al.* 2011). We, therefore, encourage conservation managers to include and even emphasize the reconnection and restoration of small habitat patches which will not only benefit Malleefowl but biodiversity more broadly. Our results suggest habitat patches should be as close as 250 m to facilitate adult dispersal. Although movement patterns were strongly driven by breeding and patch size, temperature and rainfall had a small but significant influence on Malleefowl movement. The predicted reduction in movement is substantial. Malleefowl have a heat tolerance of around 41 - 42°C and start showing heat dissipation and avoidance behaviours above this temperature but can also show these behaviours at lower temperatures if they have to tend the mound unexpectedly at a time when they would normally be resting (Booth 1985). High temperatures are detrimental to the survival and health of arid-zone birds (Conradie, Woodborne *et al.* 2019; van de Ven, McKechnie *et al.* 2020) and studies show how thermal refuges under taller vegetation are preferred by other gallinaceous birds to limit exposure to high temperatures (Carroll, Davis *et al.* 2017; Carroll, Davis *et al.* 2015). With the number of days over 40°C expected to double in South Australia by the end of the century (CSIRO and BOM 2015), climate change will almost certainly have an impact on Malleefowl movement and flow-on effects on foraging and breeding success.

Our data showed a clear pattern of movement throughout the day. Birds were largely sedentary at their roost sites, including on moonlit nights, moved slightly more in the mornings, more again during the day and most at dusk. Many authors report peaks in activity in the late afternoon (Bellchambers 1916; Benshemesh 1992; Frith 1957), which possibly reflects movement towards favoured roosting sites. Benshemesh (1992) and Frith (1959) also reported increased activity in the early morning, which we did not record. This is very likely because we can only measure the linear distance between two fixes and any divergent movement is missed, meaning the linear distance between fixes is unlikely to reflect the actual movement and thus isn't suitable for inferring activity as such. For the same reason, our study has recorded far shorter daytime distances moved than Weathers and Seymour (1998), who, through continuous observation, calculated an average walking speed of 1.2 km/hr (maximum 8.3 km/hr) and a daily travelling distance of 2.1 km. However, walking *per se* only made up 12% of their birds' daily activities. We recorded the greatest linear movement during the day, while other studies report Malleefowl resting more in the middle of the day, typically in the shade, due to hot ambient temperature or after prolonged periods of digging (Frith 1956; Frith 1959; Weathers and Seymour 1998; Weathers, Seymour *et al.* 1993). This is likely because our daytime hourly distance was based on multiple fixes throughout the day and not solely on midday.

We observed high mortality in birds tagged for this study, with 67% dying within a year of tagging and most mortality caused by fox and cat predation. This supports past findings of high mortality in adults (Booth 1987a; Priddel and Wheeler 2003). For a long-lived bird that is thought to live for up to 25 years in the wild (Benshemesh 2007), such a high mortality rate is concerning. Movement in unknown habitats (for dispersal) increases predation risk (Yoder, Marschall *et al.* 2004) and habitat fragmentation increases nest predation (Kurki, Nikula *et al.* 2000) in other ground-dwelling birds and may play a role. Only two individuals from this study survived for over twelve months: one in the smallest and one in the largest patch of habitat.

Our study was limited by a small sample size (seven birds) only one of which was female. Birds were wary and difficult to trap at the mound and changes in immediate post-trapping behaviour and mortality in some birds suggest that this method is not ideal for trapping Malleefowl. Individual Malleefowl reacted differently to capture; whilst some Malleefowl returned to their mounds after a few days, others only returned after three weeks and 55% never returned. The untagged breeding mates continued to tend the mound for up to three weeks when birds were missing. Loosening the mound substrate regularly is crucial, as otherwise, the soil gets compacted and chicks will suffocate trying to get out of the mound (Benshemesh 1992; Frith 1959). However, Benshemesh (1992) observed chicks hatching up to 6-8 weeks after the parents deserted the mound after a fire. The effect of trapping on behaviour and mortality suggests that other methods like soft netting around all sides of the trap or drop-frame trapping should be considered. A recent review of the effects of tracking devices on birds recommends a tag mass of no more than 3% of the bird mass (Geen, Robinson *et al.* 2019). Our tags were less than 2.5% of the Malleefowl's body weight, and once they were attached, they were well tolerated with one individual carrying a tag for over four years without issues. The female abandoned her breeding mound after capture but started breeding again in the next breeding season. A male returned to his mound after four days and continued his usual mound tending activities. We interpret the return to breeding activities as an indication for the bird being healthy and not impeded by the backpack harness. Camera traps at mounds also confirm this, with no visible changes to plumage or posture. Two birds carried the tag for 12 and 14 months, in 2019 and 2018, respectively, and while these birds never bred again, this may have been due to the hot and dry conditions in these years. A third bird carried the tag for 11 months through most of 2020 and - although breeding attempts may have been expected as 2020 was wetter than the previous two - also did not breed. Many factors may have stopped it from breeding, such as a lack of suitable mounds or females, but the possible effect of the tag cannot be completely dismissed. Increased divorce rates for harness-wearing waterfowl have been

reported (Lameris, Müskens *et al.* 2018) and may similarly play a role in Malleefowl. We interpreted statistical results cautiously, as there were few samples in the intermediate patch size range, and the regression fit was largely driven by the largest patches. However, visual inspection confirms that most Malleefowl in larger patches travelled further and had larger home ranges.

In conclusion, Malleefowl movement in our study was influenced mainly by breeding status and the size of available native vegetation. We recorded high mortality due to predation from introduced predators. Malleefowl were closely tied to patches of native vegetation, only moving up to 250 m across cleared land and making very little use of cleared agricultural land. Movement patterns suggest that Malleefowl in low-rainfall areas such as the Eyre Peninsula may be seasonal nomads (Lenz, Böhning-Gaese *et al.* 2015) that are tightly associated with their mounds while breeding but move significantly further afield when not breeding to explore and disperse to new areas if patches of native vegetation were contiguous or closely spaced. Results suggest that even small remnant patches of native vegetation can support breeding Malleefowl pairs, although perhaps not indefinitely so as genetic processes may lead to inbreeding and extinction in bottlenecked Malleefowl populations (Priddel and Wheeler 1999; Priddel and Wheeler 2005; Stenhouse, Onley *et al.* 2022, in prep). As Malleefowl are long-lived birds, their ability to move and disperse within and between habitat patches is critical to their survival and recolonization of patches after drought, heatwaves or wildfire, all of which are predicted to increase in frequency with climate change. More GPS tracking, particularly of female birds, would assist in determining the factors influencing survival but initial results suggest that controlling introduced cats and foxes, improving existing native vegetation through herbivore control, and building corridors connecting remnant patches of vegetation to enable dispersal could improve Malleefowl conservation.

4.8 Miscellaneous

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Ethics

This project was undertaken with approval from the University of Adelaide Animal Ethics Committee (S-2016-105) and the SA Department of Environment and Water and the Environment (permits A26564-1 to 5).

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4.10 Appendix

4.10.1 Methods

4.10.1.1 Study sites

4.10.1.1.1 Vegetation

Table S 4-1 Attributes of the study sites on the Eyre Peninsula. Predominant vegetation types (Berkinshaw and McGregor 2010; Matthews 2002; Wilderness Advisory Committee 2014). Rain = Mean annual rainfall from the nearest BOM weather station for 2016 to 2020 (mm); Columns 3 and 4 show the predominant plant species in the upper and lower vegetation levels, respectively.

Site	Rain	Upper storey species	Mid-/lower storey species
Hambidge	403, 344,	<i>Melaleuca lanceolata</i> , <i>Allocasuarina</i>	<i>M. lanceolata</i> , <i>Melaleuca acuminata</i> ,
	311, 221,	<i>verticillata</i> , <i>Eucalyptus porosa</i> , <i>E.</i>	<i>Acacia</i> spp., <i>Dodonea</i> spp., <i>Triodia</i> spp.,
	318	<i>diversifolia</i> , <i>E. socialis</i> , <i>E. dumosa</i>	<i>Hibbertia</i> spp., <i>Boronia</i> spp.
Hincks	515, 401,	<i>E. incrassata</i> ± <i>Callitris gracilis</i>	<i>M. lanceolata</i> , <i>M. uncinata</i> , <i>Senna</i>
	361, 326,	<i>E. diversifolia</i> , <i>E. calcareana</i> , <i>E. gracilis</i> ,	<i>artemisioides</i> , <i>Acacia</i> spp.
	392	<i>Myoporum platycarpum</i> , <i>M. lanceolata</i>	
Lock	403, 344,	<i>M. lanceolata</i> , <i>E. diversifolia</i> , <i>E.</i>	<i>Acacia</i> spp., <i>M. lanceolata</i> , <i>M.</i>
	311, 221,	<i>incrassata</i> , <i>E. calcareana</i> , <i>E. gracilis</i> ,	<i>acuminata</i> , <i>Triodia</i> spp.
	318	<i>Melaleuca uncinata</i>	
Dart	545, 325,	<i>M. uncinata</i> , <i>E. dumosa</i> , <i>E. socialis</i> ,	<i>M. acuminata</i> , <i>M. uncinata</i> ,
	220, 148,	<i>E. incrassata</i> , <i>E. calycogona</i>	<i>M. pauperifolia</i> , <i>Acacia</i> spp., <i>Triodia</i> spp.,
	351		<i>Maireana</i> spp., <i>Enchylaena tomentosa</i>
Secret	492, 358,	<i>E. brachycalyx</i> , <i>E. socialis</i> , <i>E. gracilis</i> ,	<i>M. platycarpum</i> , <i>S. artemisioides</i> , <i>Alyxia</i>
Rocks	235, 150,	<i>E. incrassata</i>	<i>buxifolia</i> , <i>Acacia</i> spp., <i>Maireana</i> spp.,
(3 birds)	333		<i>E. tomentosa</i> , <i>Atriplex</i> spp., <i>Roepera</i> spp., <i>Triodia</i> spp.

4.10.1.1.2 Secret Rocks enclosure

Secret Rocks has a pest-proof enclosure in the northwest of the property, to the south of and bordering Middleback Road. The enclosure is made of a 180 cm high wire netting fence with a curved 'floppy' overhang and a foot apron (Moseby and Read 2006). It is approximately 14 km long and effectively keeps out predators (e.g. cats and foxes) and herbivores (e.g. goats, rabbits and kangaroos).

4.10.1.2 Data collection

Cage trapping and processing

For this study, one of the original traps by Priddel & Wheeler (2003) was used but the steel "ceiling" of the trap was replaced with a 20 mm mesh polyester fishing net to reduce potential injury to birds, as well as reducing the overall weight of the trap. We measured weight, head-bill length and tarsus and collected feather and blood samples from each bird.

Birds were sexed via polymerase chain reaction, using primer pairs 2550F/ 2718R (Insee, Kamolnorranath *et al.* 2014).

GPS telemetry

A 6 mm neoprene layer was glued under the tag to better raise it above the feathers to improve solar charging. The tags were attached around the wing base, like a backpack, using a soft polyester shoelace as a harness as described by Benshemesh (1992) and Priddel and Wheeler (1996); Priddel and Wheeler (1997). The weight of the whole backpack, including harness, was approximately 37 grams which was under 2.5% of the body mass of an adult Malleefowl (1520 - 2500 grams (Jones and Goth 2008)).

High-resolution locations (fixes) were calculated six to twelve times a day (6 times from 22 May to 21 August (00:00, 07:00, 10:00, 13:00, 16:00, 19:00), six times from 22 August to 21 October (00:00, 06:00, 09:00, 12:00, 16:00, 19:00), seven times from 22 October to 14 March (00:00, 05:00, 08:00, 11:00, 14:00, 17:00, 20:00) and seven times from 15 March to 21 May (00:00, 06:00, 09:00, 12:00, 15:00, 18:00, 20:00), LOC7 12 fixes every 2 hours).

Data cleaning

As capture causes stress and birds may move erratically, we disregarded all data after capture until they returned to their mounds, or the first three days after capture if they did not. Sometimes fixes were not successful due to low battery or poor reception of the tag, most likely caused by a lack of insolation due to dense vegetation or plumage. These 'zero fixes' recorded the time of the fix but "0.00" instead of the proper coordinates. We removed all zero fix records, outliers due to satellite errors and all distances from fixes that were more than seven hours apart (maximum possible time between scheduled fixes) to reduce bias towards longer distances. For one bird (ID = DA) with numerous data gaps, we also removed all weeks where more than 15% of days were missing, which resulted in splitting its data up into individual seasons with complete data.

Movement patterns

Any zero hourly distances were replaced with a random value between 1.8 and 6 (10th to a 3rd of tag accuracy $\pm 18\text{m}$) to avoid transformation of zeros in models. Using examples and recommendations from previous studies (Byrne, Clint McCoy *et al.* 2014; Kranstauber, Kays *et al.* 2012) and taking into account the different fix rates of the LOC7 tag, we used the following parameters to fit the dBBMM to the Malleefowl tracks: Window size: 9, margin: 3; ext: 1; location error: 18; dimsize: 200.

Times of day

Location and time-dependent sunlight phases, such as nautical dawn, dawn, sunrise and the lunar phase were calculated for each day and fix. We specified day times as the interval

between nautical dawn (c. 1 hour before sunset) and astronomical dusk (c. 1.5 hours after sunset, considered the onset of complete darkness), as our camera trap images show Malleefowl on mounds in these periods. Night was the period between astronomical dusk and nautical dawn. For a more detailed analysis, we split each day further into dawn, day and dusk. Dawn was the period between nautical dawn and golden hour end, day was between golden hour end and sunset, and dusk between sunset and astronomical dusk. Depending on the time of year, fix times did not always fall into sunlight phases we used for our classifications, resulting in some days not having dawn or dusk fixes. If a dawn fix was missing, the first day fix was reclassified as a dawn fix. If a dusk fix was missing, the last day fix was reclassified as a dusk fix. Dusk and night distances were adjusted according to proportion of time before and after the first night fix. E.g., if the time difference between a dusk and night fix was 180 min, and the distance travelled in this time was 100 m, but the dusk proportion of the time difference was 160 min (89%), the night distance was adjusted to 11 m (100 - 89-m) and 89 m was added to the dusk distance. Dawn distances were not adjusted like this because considerable distances would have been added to the night distances, which does not conform with observational data (i.e. that Malleefowl roost and not move during the night).

Comparative statistics

As all our movement metrics were not normally distributed, we used non-parametric Mann Whitney U test (MWU) to compare two independent groups (i.e. sexes) and the Wilcoxon signed-rank test (Wilcoxon) for two dependent groups (e.g. breeding and non-breeding distances across birds). For more than two dependent groups (e.g. times of day), we applied Friedman rank sum tests (Friedman) and followed up with Durbin-Conover (DC) pairwise comparison test to determine which of the times of day differed from others.

4.10.2 Effect of breeding status and patch size on home range and total range length

Patch size was the size of the area of uninterrupted remnant native vegetation available to each Malleefowl. We used Google Earth and ArcGIS to measure patch size and used agricultural land, bitumen roads, and structures such as mines as outer boundaries. Temperature and rainfall data were obtained from the Australian Bureau of Meteorology (BOM 2022) using the nearest weather station to the capture mound. Spatial vegetation and reserve data was obtained from the South Australian Government Data Directory (Government of South Australia 2022).

4.10.3 Movement after capture

Table S 4-2 Movement after capture of the Eyre Peninsula Malleefowl in the initial season of capture. We present the trapping date; the date on which the tagged bird returned to the mound (d: number of days between trapping and return); date until which the mate continued to maintain the mound in the absence of the tagged bird (d: days); date on which breeding activities ceased; whether the breeding season was completed. ¹Tagged bird killed. ²Tagged bird tending mound without mate.

Bird	Trapping date	Return to mound (d)	Mate at mound alone until (d)	End breeding tagged bird	Completed breeding?
DA1	15/01/2017	17/01/2017 (2)	Return of tagged bird, then both	6/4/2017	Yes
HAM1	01/12/2016	05/12/2016 (4)	Return of tagged bird, then both	26/3/2017	Yes
HIN11	10/12/2016	21/12/2016 (11)	Return of tagged bird, then both	15/1/2017 ¹	No
LOC7	13/02/2017	-	18/2/2017 (5)	trapping	No
SR159	18/11/2018	-	22/11/2018 (4)	trapping	No
SR260	02/12/2017	26/12/2017 (24)	10/12/2017 (8)	11/1/2017 ²	No
SR60	13/12/2019	-	20/12/2019 (7)	trapping	No

4.10.4 Movement overview

Table S 4-3 Basic movement statistics of Malleefowl on the Eyre Peninsula. We present the years for the bird observed over multiple years and the different patches used by the birds that resettled (Period), the breeding status (B), the days tracked (Days), the patch size (Patch; ha), the home range (MCP = 95% MCP, HR = 95% UD; ha), the total range length (TRL; km), and the mean \pm 1 s.d. and the maximum of the daily total distance, the daily displacement, the distance to mound and the hourly movement in meters. B = Breeding, NB = Non-Breeding; F = Female; S = Single Malleefowl tending mound without its mate; ° = were not used for analyses due to short tracking time; * = MCP bootstraps did not converge; DA has 5 periods because of missing fixes in winter, Lock has 3 patches with a total range length of 7.8 km over the whole

ID	Period	B	Days	Patch	Daily distance			TRL	HR	Daily displacement			Distance to mound			Hourly movement		
					Mean \pm sd	Max	Max			Mean \pm sd	Max	Max	Mean \pm sd	Max	Max	Mean \pm sd	Max	Max
DA1	1617	B	78	107	39	44	1.1	931 \pm 326	1,840	224 \pm 140	584	183 \pm 128	667	49 \pm 43	199			
	1617	NB	24	49*	62	62	1	1,182 \pm 342	1,749	184 \pm 148	461	365 \pm 190	756	70 \pm 64	229			
	1718	B	152	76	88	88	1.7	1,583 \pm 419	2,545	326 \pm 248	1,078	434 \pm 342	1,355	84 \pm 89	361			
	1718	NB	32	46*	62	62	1.1	1,323 \pm 539	2,149	224 \pm 137	420	684 \pm 195	1,024	69 \pm 71	292			
	1819	B	215	74	98	98	1.8	1,509 \pm 525	3,273	311 \pm 221	1,033	474 \pm 351	1,551	81 \pm 86	400			
	1819	NB	12	30*	41	41	0.9	912 \pm 296	1,491	144 \pm 65	250	656 \pm 145	997	47 \pm 38	183			
	1920	B	166	76	84	84	1.7	1,277 \pm 520	2,771	354 \pm 281	1,098	392 \pm 315	1,293	66 \pm 73	408			
	1920	NB	3°	16**	76°	76°	1.1	1,395 \pm 874	2,192	32 \pm 28	64	652 \pm 267	1,052	68 \pm 69	202			
	2021	B	163	72	52	52	1.3	827 \pm 574	2,170	217 \pm 271	892	161 \pm 250	961	41 \pm 60	298			
HAM1	B	111	37,789	83	100	100	2.4	1,188 \pm 699	3,896	290 \pm 207	922	258 \pm 296	2,062	63 \pm 81	537			
	NB	57	210*	214	214	214	2.3	1,546 \pm 561	3,114	461 \pm 272	1,102	620 \pm 379	1,901	93 \pm 92	499			
HIN11	B	25	68,009	70	90	90	1.8	1,085 \pm 580	3,134	364 \pm 299	841	276 \pm 248	1,380	59 \pm 73	402			
LOC07 ^F	1	NB	8°	1,662	74**	98°	1.3	1,874 \pm 981	3,720	660 \pm 410	1,235	498 \pm 223	888	106 \pm 127	508			
	2	NB	3°	33	12**	17	0.8	1,162 \pm 237	1,311	459 \pm 320	777	1,584 \pm 260	1,944	71 \pm 94	378			
	3	B	52	1,942	164	176	2.2	1,829 \pm 517	2,956	566 \pm 324	1,396	470 \pm 267	1,507	117 \pm 82	484			
	3	NB	186	430	234	234	4.7	1,562 \pm 540	2,768	495 \pm 378	1,812	5844 \pm 463	7,096	102 \pm 95	697			
SR159	NB	362	202,821	2017	699	699	13.1	1,215 \pm 776	10,394	581 \pm 702	9,738	1,797 \pm 1,091	10,492	68 \pm 81	937			
SR260	NB	404	202,821	1931	837	837	9.4	1,749 \pm 963	8,237	584 \pm 726	4,538	2,542 \pm 2,489	6,965	96 \pm 108	959			
	S	22	-	-	502	502	4.6	2,210 \pm 1,305	4,931	900 \pm 821	2,479	908 \pm 887	3,093	118 \pm 171	915			
SR60	1	NB	270	202,821	5101	2,168	14.4	1,744 \pm 763	5,033	875 \pm 614	4,288	3,275 \pm 2,580	12,969	95 \pm 96	644			
	2	NB	67	-	2,390*	1,164	8.4	2,045 \pm 1,040	4,903	1,043 \pm 722	3,265	9473 \pm 1,194	12,664	108 \pm 126	717			
F	B	52	-	164	176	176	2.2	1,829 \pm 517	2,956	566 \pm 324	1,396	470 \pm 267	1,507	117 \pm 82	484			
	NB	197	-	430	234	234	4.7	1,565 \pm 557	3,720	501 \pm 378	1,812	5,599 \pm 1,178	7,096	102 \pm 96	697			
M	B	910	-	70 \pm 14	79 \pm 23	79 \pm 23	1.7 \pm 0.4	1,256 \pm 596	3,896	296 \pm 247	1,098	338 \pm 326	2,062	66 \pm 78	537			
	NB	1236	-	1310 \pm 1,737	591 \pm 721	5.7 \pm 5.6	1567 \pm 877	10,394	644 \pm 684	9,738	2639 \pm 2,670	12,969	86 \pm 97	959				
B	962	-	83 \pm 38	92 \pm 43	92 \pm 43	1.8 \pm 0.4	1,283 \pm 605	3,896	310 \pm 258	1,396	349 \pm 324	2,062	69 \pm 79	537				
NB	1428	-	1356 \pm 1,704	609 \pm 708	4.9 \pm 5.1	1567 \pm 841	10,394	624 \pm 653	9,738	3,293 \pm 2,715	12,969	89 \pm 97	959					

4.10.5 Home / total range model results

Table S 4-4 Model summary showing the effect of breeding status and patch size on home range (UD95) and total range (km) for Malleefowl on the Eyre Peninsula. We present model formulas, marginal R² values (in%), terms used, model estimates, lower and upper confidence levels, standard errors (SE) and p-values. Significance is indicated in bold.

Formula	R ²	Variables	Est	LCI	UCI	SE	p
lmer(log(UD95) ~ Breeding*log(PatchSize.ha) + (1 id), data = dat)	77.4	(Intercept)	3.88	2.31	5.45	0.80	0.01
		BreedingNB	-1.45	-2.44	-0.47	0.50	0.01
		log(PatchSize.ha)	0.10	-0.08	0.28	0.09	0.31
		BreedingNB:log(PatchSize.ha)	0.25	0.11	0.39	0.07	< 0.001
lmer(log(TotalRange) ~ Breeding*log(PatchSize.ha) + (1 id), data = dat)	78.6	(Intercept)	0.10	-1.00	1.20	0.56	0.87
		BreedingNB	-1.33	-2.25	-0.42	0.46	0.01
		log(PatchSize.ha)	0.07	-0.06	0.21	0.07	0.32
		BreedingNB:log(PatchSize.ha)	0.21	0.09	0.33	0.06	0.01

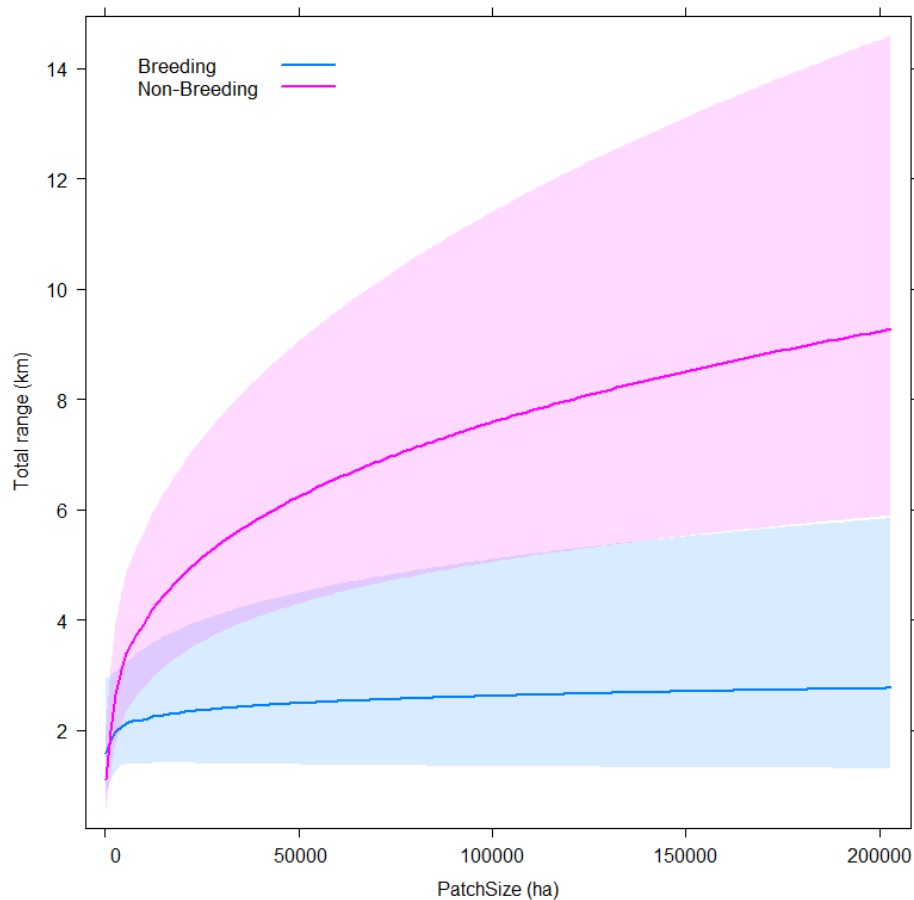


Figure S 4-1 Predicted total range (km) values by patch size (ha) for Malleefowl on the Eyre Peninsula (n = 7) while breeding (blue) and non-breeding (red). The shaded areas are the 95% confidence intervals for the fitted values.

4.10.6 Movement metrics

4.10.6.1 Daily displacement and distances

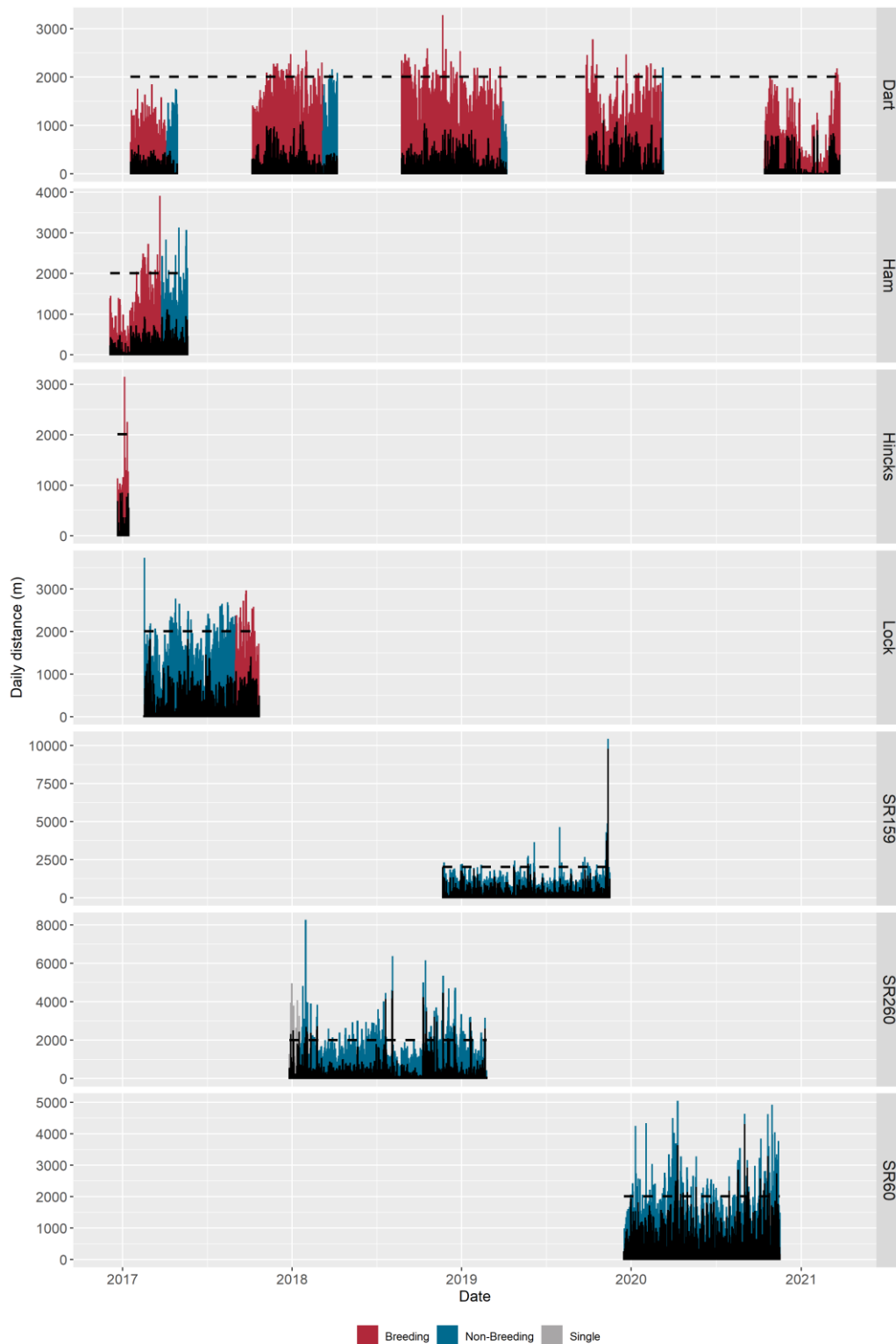


Figure S 4-2 Timeline of GPS tracked Malleefowl on the Eyre Peninsula. The coloured vertical bars show the daily total distance moved (i.e. sum of all distances between fixes per day) and the black bars in front show the daily displacement (i.e. distance between the first and last fix of a bird day). The horizontal line shows the long-distance movement threshold of $4 \times \text{mean daily displacement of all birds}$ (4×502 m).

4.10.6.2 Distance metrics - comparative statistics

Table S 4-5 Comparative statistics of distance metrics. Comparisons are between breeding (B) and non-breeding (NB) periods; between the female and the males while breeding (Sex - B) and non-breeding (Sex - NB); between seasons (spring, summer, autumn and winter) and between the times of day (dawn, day, dusk and night). We present the effect size (Sullivan and Feinn 2012) with lower and upper confidence interval limits (r_{rb} = rank biserial correlation, W_K = Kendall's coefficient of concordance). Significant p-values in bold. n = number of records. Please note there was only one female tracked!

	Metric	Test	Effect size [LCI, UCI]	p	n
B v NB	Daily distance	Wilcoxon	$r_{rb} = -0.28 [-0.34, -0.22]$	p < 0.001	1,880
	Daily displacement	Wilcoxon	$r_{rb} = -0.49 [-0.55, -0.43]$	p < 0.001	1,912
	Hourly movement	Wilcoxon	$r_{rb} = -0.1 [-0.13, -0.08]$	p < 0.001	13,546
	Distance to mound	Wilcoxon	$r_{rb} = -0.95 [-0.96, -0.94]$	p < 0.001	13,606
Sex – B	Daily distance	MWU	$r_{rb} = 0.52 [0.39, 0.62]$	p < 0.001	940
	Daily displacement	MWU	$r_{rb} = 0.49 [0.37, 0.64]$	p < 0.001	956
	Hourly movement	MWU	$r_{rb} = 0.43 [0.40, 0.49]$	p < 0.001	5,412
	Distance to mound	MWU	$r_{rb} = 0.28 [0.25, 0.32]$	p < 0.001	6,803
Sex – NB	Daily distance	MWU	$r_{rb} = 0.10 [0.03, 0.17]$	p = 0.026	1,407
	Daily displacement	MWU	$r_{rb} = -0.08 [-0.17, 0.00]$	p = 0.089	1,430
	Hourly movement	MWU	$r_{rb} = 0.10 [0.07, 0.14]$	p < 0.001	7,437
	Distance to mound	MWU	$r_{rb} = 0.64 [0.62, 0.66]$	p < 0.001	10,400
Seasons	Daily distance	Friedman	$W_K = 0.24 [0.24, 0.83]$	p = 0.012	1,004
	Hourly movement	Friedman	$W_K = 0.24 [0.24, 0.83]$	p = 0.001	6,464
	Daily displacement	Friedman	$W_K = 0.29 [0.29, 1]$	P = 0.025	1,016
Time of day	Hourly movement	Friedman	$W_K = 0.24 [0.24, 1]$	p < 0.001	2,385
	Distance to mound	Friedman	$W_K = 0.24 [0.24, 0.63]$	p < 0.001	3,804

4.10.7 Long range movement

Table S 4-6 Long-range movement with daily displacement greater than four times the mean displacement of all birds (4*502 m). We present the breeding status (B = Breeding, NB = Non-breeding, S = Single), the date, the type of movement (Type, O = outside of home range, I = inside of home range, R = resettlement), the excursion number (Also see Fig. 3 and 4 for SR260 and SR60) and their length in days. Bold font indicates the maxima for each bird.

ID	Breeding status	Date	Type	Daily displacement	Daily distance	Excursion #	Excursion length
SR159	NB	24/05/2019	O	2200	2718	1	1
SR159	NB	09/11/2019	O	3698	3944	2	4
SR159	NB	10/11/2019	O	2177	3060	2	4
SR159	NB	11/11/2019	O	4207	4841	2	4
SR159	NB	12/11/2019	O	9738	10393	2	4
SR260	S	29/12/2017	I	2292	3927	1	1
SR260	S	03/01/2018	I	2479	3291	2	1
SR260	S	16/01/2018	I	2411	3233	3	1
SR260	NB	30/01/2018	I	2114	8237	4	2
SR260	NB	31/01/2018	I	2657	3969	4	2
SR260	NB	02/02/2018	I	2410	2986	5	2
SR260	NB	03/02/2018	I	2285	3937	5	2
SR260	NB	10/02/2018	I	2349	3872	6	1
SR260	NB	23/02/2018	I	2505	3177	7	2
SR260	NB	24/02/2018	I	2701	3811	7	2
SR260	NB	21/07/2018	I	4105	4429	8	1
SR260	NB	04/08/2018	I	4147	4580	9	2
SR260	NB	05/08/2018	I	4538	6346	9	2
SR260	NB	10/10/2018	I	4212	4972	10	2
SR260	NB	11/10/2018	I	3494	3809	10	2
SR260	NB	15/10/2018	I	2306	6121	11	1
SR260	NB	17/10/2018	I	3465	3675	12	1
SR260	NB	27/10/2018	I	2145	2715	13	1
SR260	NB	03/11/2018	I	3189	3495	14	1
SR260	NB	08/11/2018	I	2930	3342	15	1
SR260	NB	22/11/2018	I	4431	5320	16	1
SR260	NB	02/12/2018	I	2070	2973	17	1
SR260	NB	05/12/2018	I	2047	2412	18	1
SR260	NB	16/12/2018	I	2710	3078	19	1
SR260	NB	20/01/2019	O	2906	3187	20	3
SR260	NB	21/01/2019	O	2159	2275	20	3
SR260	NB	22/01/2019	O	2085	2455	20	3
SR260	NB	20/02/2019	I	2575	3133	21	1
SR60	NB	05/04/2020	I	2325	2948	1	2
SR60	NB	06/04/2020	I	2479	3674	1	2
SR60	NB	10/04/2020	I	3616	5033	2	1
SR60	NB	16/04/2020	I	2069	2794	3	1
SR60	NB	22/04/2020	I	2108	2425	4	1
SR60	NB	01/05/2020	I	2128	2529	5	1
SR60	NB	20/05/2020	I	2287	3250	6	1
SR60	NB	30/07/2020	I	2170	2629	7	1
SR60	NB	17/08/2020	I	2427	3071	8	2
SR60	NB	18/08/2020	I	2838	3142	8	2
SR60	NB	01/09/2020	I	4288	4615	9	1
SR60	NB	05/09/2020	I	2010	2640	10	1
SR60	NB	07/09/2020	I	2906	3149	11	1
SR60	NB	10/09/2020	R	2136	2298	12	1
SR60	NB	05/10/2020	I	2560	2673	13	1
SR60	NB	14/10/2020	O	2404	2787	14	1
SR60	NB	21/10/2020	I	3265	4602	15	1
SR60	NB	25/10/2020	I	2094	2755	16	1
SR60	NB	09/11/2020	I	2721	3326	17	1



Figure S 4-3 Long-range movements of SR60 between April and November 2020 (NB). The white arrow depicts the length and direction of resettlement. The arrows are colour coded by season, with autumn brown to purple, winter in blues and spring in greens. The orange triangle shows the mound at which the bird was captured, the blue contours the 95% UD, the grey contours the 50% UD and the green dashed line the 95% MCP. Compare excursion numbers to Table S 4-6.

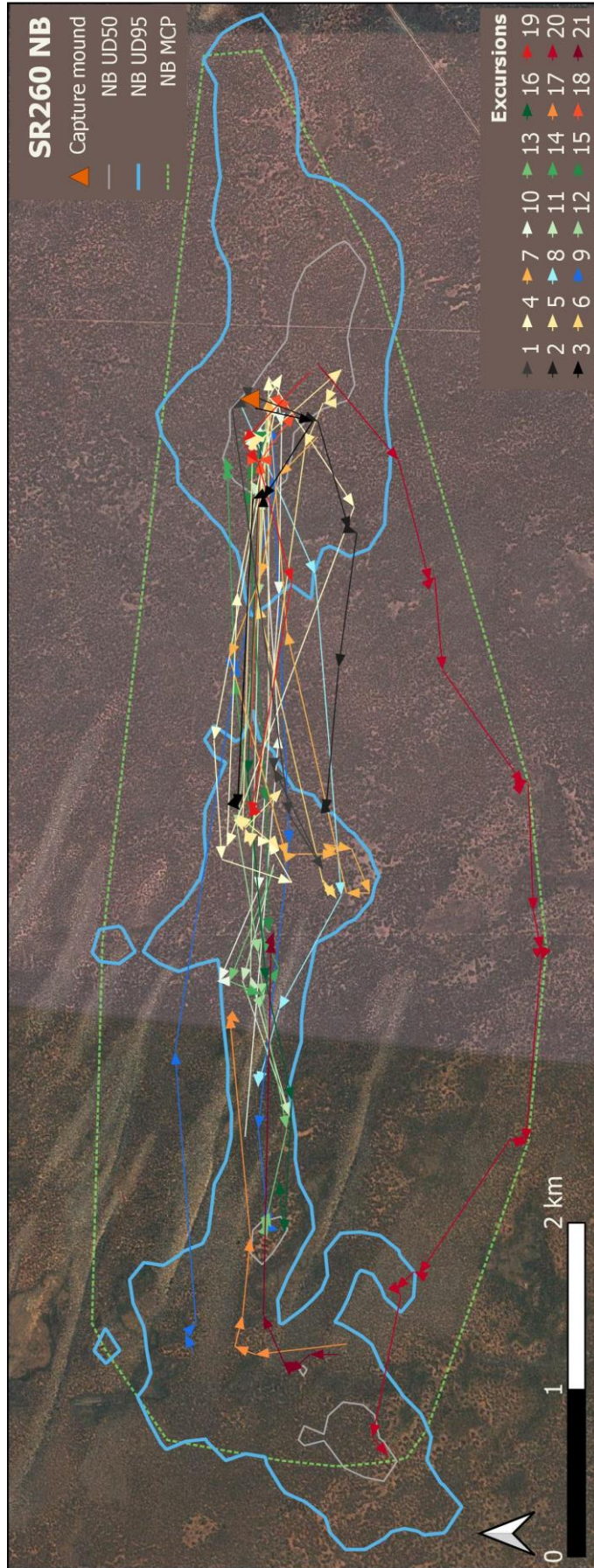


Figure S 4-4 Long-range movements of SR260 between December 2017 and February 2019 (NB). The first three excursions occurred when this male was tending the mound by himself. The excursion arrows are colour coded by season, with summer in yellow to red, winter in blues and spring in greens. The orange triangle shows the mound at which the bird was captured, the blue contours the 50% UD, the grey contours the 95% UD and the green dashed line the 95% MCP. Compare excursion numbers to Table S 4-6

4.10.8 Environmental effects

Table S 4-7 Model summaries showing the effect of environmental features on daily and hourly movement. R2m = Marginal r-squared (in %); R2c = Conditional r-squared (in %); Est= Estimate, LCI/UCI = Upper/Lower Confidence Interval; SE = Standard Error; DF = degrees of freedom, All models (lme) have random = ~1|idSeason, correlation = corCAR1(form = ~ timeSeq | idSeason). Significant values in bold.

Season		Formula	R2m	R2c	Term	Est	LCI	UCI	SE	DF	t-value	p-value		
Daily distance	B	(DayDist^0.71) ~	5.7	30.2	(Intercept)	214.4258	188.0888	240.7628	13.42	929	15.9781	< 0.001		
		TMax + TMin + Rain01	TMax	-1.8798	-2.5221	-1.2374	0.3273	929	-5.7431	< 0.001				
			TMin	-0.2811	-1.1222	0.56	0.4286	929	-0.6559	0.512				
	NB	TMax + TMin + Rain01 +	Rain01	2.1831	-5.9164	10.2826	4.1271	929	0.529	0.597				
			(DayDist^0.02) ~	1.8	11.7	(Intercept)	1.1572	1.1535	1.1609	0.0019	1388	609.2469	< 0.001	
			TMax	-0.0002	-0.0003	-0.0001	0.0001	1388	-3.2124	0.0013				
		TMin	0.0002	0	0.0003	0.0001	1388	2.214	0.027					
		Rain01	0.0016	0.0002	0.0029	0.0007	1388	2.324	0.0203					
Hourly movement	Dawn	B	(DistPerHour^0.06) ~	TMax + TMin + Rain01	(Intercept)	0.7	21.2	1.1722	1.1174	1.2269	0.0279	936	42.0304	< 0.001
					TMax	-0.0006	-0.002	0.0008	0.0007	936	-0.8019	0.4228		
					TMin	-0.0014	-0.0033	0.0004	0.0009	936	-1.5661	0.1177		
			Rain01	-0.0021	-0.019	0.0148	0.0086	936	-0.2489	0.8035				
	NB	(DistPerHour^0.1) ~	TMax + TMin + Rain01	(Intercept)	0.5	25.8	1.2782	1.2026	1.3538	0.0385	1401	33.1758	< 0.001	
				TMax	-0.0024	-0.0043	-0.0004	0.001	1401	-2.3979	0.0166			
				TMin	0.0035	0.001	0.006	0.0013	1401	2.7339	0.0063			
			Rain01	-0.0287	-0.0491	-0.0082	0.0104	1401	-2.7492	0.0061				
	Day	B	(DistPerHour^0.46) ~	TMax + TMin + Rain01	(Intercept)	7.7	31.9	9.2021	8.1756	10.2285	0.523	943	17.5936	< 0.001
					TMax	-0.0977	-0.1231	-0.0722	0.013	943	-7.5263	< 0.001		
					TMin	0.0193	-0.014	0.0526	0.017	943	1.1365	0.2561		
				Rain01	0.2327	-0.092	0.5573	0.1654	943	1.4065	0.1599			
		NB	(DistPerHour^0.06) ~	TMax + TMin + Rain01	(Intercept)	5.5	18.2	1.3241	1.3083	1.34	0.0081	1405	164.2358	< 0.001
					TMax	-0.0015	-0.0021	-0.001	0.0003	1405	-5.7517	< 0.001		
	TMin				0.0005	-0.0002	0.0012	0.0004	1405	1.3631	0.1731			
			Rain01	0.0065	0.0004	0.0126	0.0031	1405	2.0793	0.0378				
	Dusk	B	(DistPerHour^0.4) ~	TMax + TMin + Rain01	(Intercept)	1	8.6	6.9091	6.1833	7.635	0.3699	941	18.6804	< 0.001
					TMax	-0.0043	-0.0294	0.0208	0.0128	941	-0.3376	0.7358		
TMin					-0.0352	-0.0677	-0.0027	0.0166	941	-2.1243	0.0339			
			Rain01	-0.0891	-0.4278	0.2495	0.1726	941	-0.5165	0.6057				
NB		(DistPerHour^0.14) ~	TMax + TMin + Rain01	(Intercept)	0.5	4.5	1.8993	1.8428	1.9558	0.0288	1399	65.9814	< 0.001	
				TMax	0.0012	-0.0011	0.0034	0.0012	1399	1.0077	0.3138			
	TMin			-0.0002	-0.0033	0.0029	0.0016	1399	-0.1177	0.9063				
		Rain01	0.0343	0.0077	0.0609	0.0136	1399	2.5294	0.0115					
Night	B	(DistPerHour^-0.2) ~	TMax + TMin + Rain01 +	(Intercept)	0.7	14.9	0.8032	0.7266	0.8798	0.039	945	20.5795	< 0.001	
				TMax	0.0015	-0.0007	0.0038	0.0011	945	1.3262	0.1851			
				TMin	0.0014	-0.0016	0.0043	0.0015	945	0.9084	0.3639			
			Rain01	0.0124	-0.017	0.0419	0.015	945	0.8284	0.4076				
			MoonFrac	0.0025	-0.0329	0.038	0.0181	945	0.1411	0.8878				
	NB	(DistPerHour^0.06) ~	TMax + TMin + Rain01 +	(Intercept)	1.4	19.4	1.1405	1.1116	1.1694	0.0147	1405	77.4614	< 0.001	
TMax				-0.0015	-0.0023	-0.0007	0.0004	1405	-3.691	0.0002				
TMin				0.0008	-0.0003	0.0018	0.0005	1405	1.4502	0.1472				
		Rain01	-0.001	-0.0096	0.0076	0.0044	1405	-0.2229	0.8237					
		MoonFrac	-0.0003	-0.0155	0.0149	0.0077	1405	-0.0342	0.9727					

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Chapter 5:

Malleefowl movement and fine-scale habitat selection driven by canopy



Figure on previous page: A Secret Rocks panorama. Credit: Peri Stenhouse

5.1 Statement of Authorship

Title of paper	Malleefowl movement and fine-scale habitat selection driven by canopy			
Publication status	<input type="checkbox"/> Published	<input type="checkbox"/> Accepted for Publication	<input type="checkbox"/> Submitted for Publication	<input checked="" type="checkbox"/> Publication style
Publication details	-			

Author contributions

By signing the Statement of Authorship, each author certifies that the candidate's stated contribution to the publication is accurate, that permission is granted for the candidate to include the publication in the thesis; and that the sum of all co-author contributions is equal to 100% less the candidate's stated contribution.

Name of Principal Author (Candidate)	Peri Stenhouse		
Contribution to the paper	Conceptualization, methodology, data curation, formal analysis, writing – original draft preparation, writing – review and editing.		
Overall percentage (%)	80%		
Certification	This paper reports on original research I conducted during the period of my Higher Degree by Research candidature and is not subject to any obligations or contractual agreements with a third party that would constrain its inclusion in this thesis. I am the primary author of this paper.		
Signature			Date: 04 April 2022

Name of Co-Author	Katherine Moseby		
Contribution to the Paper	Conceptualisation, methodology, writing – review and editing.		
Signature			Date: 04 April 2022

5.2 Introduction

Animals do not use their environment randomly, and movement is driven by a range of factors including the individuals' need for social interaction (e.g. for breeding), dispersal, migration, resource selection or to escape risks (Dingle 2014; Stephens and Krebs 2019; Swingland and Greenwood 1983). Ultimately, movement is necessary to optimise space and resource use and the fitness and persistence of a species. An animal's habitat is an important determinant of its movement and extensive research has shown that animals select certain habitats or vegetation characteristics for shelter (Carroll, Davis *et al.* 2015), food (Crawford and Long 2017; Davies, Gramotnev *et al.* 2013) or breeding (Bell 1990; Dean 1997).

Understanding how vegetation characteristics influence space use by threatened species can assist in conserving critical habitat or restoring suboptimal areas (Bowen, McAlpine *et al.* 2007; Stobo-Wilson, Stokeld *et al.* 2020). The Malleefowl is a long-lived threatened Australian bird that builds large incubation mounds and uses external sources of heat to incubate its eggs. Malleefowl persist in long-unburnt remnant mallee woodlands and shrublands of semi-arid to arid southern Australia but can also be found in other habitats such as mulga shrublands (Benshemesh, Dennings *et al.* 2007). While Malleefowl prefer higher rainfall areas with good quality soils, these areas have been heavily cleared and degraded since colonisation for agriculture and pastoralism (Parsons, Short *et al.* 2009). Today, Malleefowl largely persist in small remnant pockets of native vegetation in semi-arid to arid parts of the country and their numbers and range have noticeably declined (Benshemesh, Southwell *et al.* 2020; Stenhouse and Moseby 2022).

Although found over a range of habitats, little is known about the specific habitat requirements of Malleefowl other than their need for long unburnt vegetation (Benshemesh 1992; Clarke 2005; Woinarski 1989). While long term habitat choices for breeding are predominantly driven by the need for leaf litter for nesting, short term movement patterns may be related to habitat preferences driven by food availability, shelter from predators or thermal regulation. The midstorey is likely used as protection from heat as Malleefowl have been observed resting in the shade of shrubs to cool down on hot days or after tending their mounds intensively (Benshemesh 1992; Weathers and Seymour 1998). Other ground-dwelling birds also avoid bare ground in the hottest parts of the day (Kline 2015). As leaf litter slows evaporation and reduces maximum soil temperatures (Evans and Young 1970; MacKinney 1929), Malleefowl may prefer areas with more leaf litter for assistance with thermoregulation. Habitat selection may also be driven by predation pressure, as predators can be more numerous and more successful in habitats with

reduced understoreys (Hradsky, Mildwaters *et al.* 2017; McGregor, Legge *et al.* 2015; Stobo-Wilson, Stokeld *et al.* 2020) and canopy cover can be used by Malleefowl to hide from raptors (Frith 1962a). Both Frith (1962b) and Benshemesh (1992) found that Malleefowl preferred an uninterrupted and dense canopy and Frith (1962b) and Harlen and Priddel (1996) emphasized the importance of a dense and diverse shrub layer as protection from predators as well as a food source for Malleefowl.

In many birds, habitat requirements are strongly linked to dietary preferences (Buelow and Sheaves 2015; Korpimäki 1986; Robinson and Holmes 1982). Malleefowl are ground-dwelling birds and have a variable, flexible and opportunistic diet consisting of plants (flowers, foliage, buds, seeds, tubers, pods, berries and other fruit), fungi and invertebrates. Previous studies have found 58 plant genera in 34 families are palatable to Malleefowl (Table S 5-1, Benshemesh 1992; Booth 1986; Brickhill 1987; Frith 1956; Frith 1959; Frith 1962b; Harlen and Priddel 1996; Kentish and Westbrooke 1994; Reichelt and Jones 2008; van der Waag 2004). The most commonly reported plant genera by Malleefowl in these studies were *Acacia*, *Dodonaea* and *Thysanotus*, followed by *Astroloma*, *Beyeria*, *Brassica*, *Cassytha*, *Daucus*, *Daviesia*, *Eremophila*, *Goodenia*, *Helichrysum*, *Hypochaeris*, *Lomandra*, *Pterostylis* and *Sclerolaena*. Frith (1959) identified *Acacia* and *Senna* as the most important food plants in his study area and observed the influence of rainfall on the productivity of these shrubs. Chandler (1913) reported a strong liking for *Beyeria* seeds, Booth (1986) for *Cassytha* seeds, Bellchambers (1916) for *Acacia* seeds, Reichelt and Jones (2008) for all parts of *Acacias* except the leaves and flowers of various species and van der Waag (2004) for seeds in general.

While these studies showed that Malleefowl feed on a wide variety of plant species and parts, they also showed that some plants are favoured and that seeds were preferred. The distribution of favoured food plants is thus likely to influence movement patterns and site selection by Malleefowl in addition to other vegetation characteristics such as protection from predators or heat extremes. As Malleefowl remain close to their mounds during the breeding season (chapter 4), we studied their movement patterns and site selection when they were not breeding to determine whether Malleefowl movement was driven by their preference for certain plants or habitat characteristics such as plant cover, richness or diversity. Such knowledge may assist in the conservation of the species by helping select optimum habitat for conservation or may guide revegetation and restoration programs in Malleefowl habitat.

5.3 Methods

5.3.1 Study areas

We GPS tracked Malleefowl on the Eyre Peninsula in South Australia (Figure 5-1), where 39% of native vegetation cover remains, mostly in marginal areas unsuitable for agriculture (Brandle, 2010, NREP, 2017). We trapped two birds at Secret Rocks Nature Reserve and one at a Heritage Agreement property near Lock (HA 370). The study areas are approximately 120 km apart and include a variety of mallee eucalypt shrubland communities (SI-Table 2). Both areas are semi-arid with a rainfall gradient of 290-400mm (BOM 2022, also see SI-Table 2). Fires were recorded in 1972 and 1990 in the eastern part of Secret Rocks where one Malleefowl was tracked (SR260), but not in areas used by the other two Malleefowl.

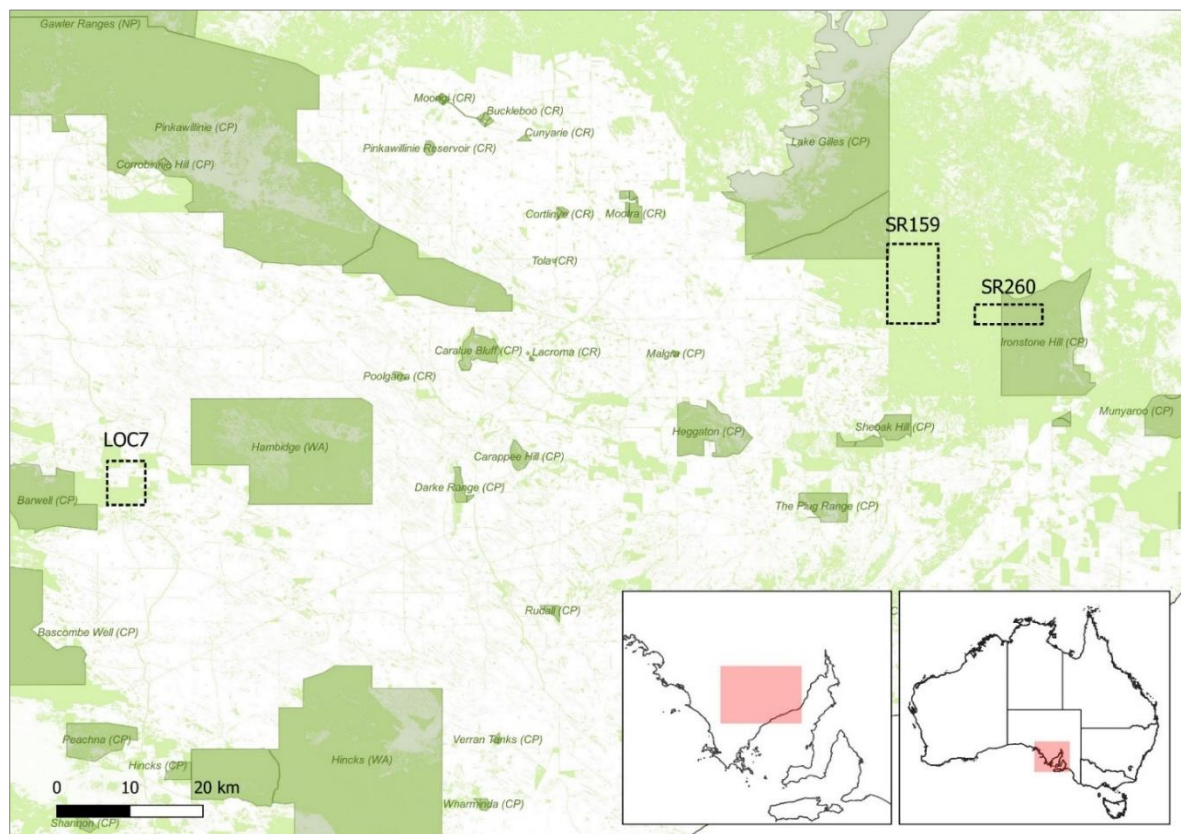


Figure 5-1 Overview map of the Eyre Peninsula Malleefowl study sites. Darker green shows government conservation areas and light green native vegetation cover. Dashed lines indicate the outer boundaries of Malleefowl movement ranges.

5.3.2 Trapping and tracking

Breeding Malleefowl were trapped using a cage trap set on the mound following Priddel and Wheeler (2003) but with a soft top. We tracked birds with solar-powered GPS transmitters (Solar Argos/GPS 30 g PTT, horizontal accuracy ± 18 m; Microwave

Telemetry Inc. (2018) (MTI). Locations (fixes) were calculated six to twelve times a day and transmitted via satellite (Argos CLS 2021). For this chapter, we only used fixes made during daylight hours, because Malleefowl most likely select their habitat based on visual cues and need light to do so. For more details on trapping and movement, data see chapter 4. We discarded the first three days of movement data after trapping to avoid recording aberrant behaviour caused by the stress of being caught. We considered Malleefowl to be breeding if they visited a mound at least every second day.

5.3.3 Movement periods considered

Non-breeding birds largely disassociate from their mounds and move up to 23 km if native habitat is available, while breeding birds usually stay within 500 m of their mound (chapter 4). As we were interested in habitat-driven movement, we focused on three non-breeding Malleefowl in large native habitat patches: a female from Lock (LOC7) and two males from Secret Rocks (SR260 and SR159). Sex was determined through DNA analysis of blood (see chapter 3).

LOC7 was trapped on 13 February 2017 and did not return to her mound. A week after capture, she moved away from the initial patch and explored other remnant patches up to 7 km away from the capture site. On 27 February 2017 she arrived in a final patch and from 9 April 2017 onwards, she focussed her attention on a new mound within this patch, visiting it intermittently on 51 days before starting breeding (i.e. visits to the mound at least every second days) at this new mound on 31 August 2017. Before 9 April, her median distance to the new mound was 1.5 km (718 m interquartile range) with a maximum of 2.3 km, but afterwards, the median was 353 m (454 m IQR) with a maximum of 1.5 km. Therefore, to avoid bias towards areas near the mound, we disregarded sites and fixes within 150 m of the mound after 9 April until breeding began but used all data before that date. This resulted in 52 days of movement data from 16 February to 9 April and 144 days from 9 April to 31 August 2017.

SR159 was trapped on 18 November 2018 and did not return to his mound. We used tracking data from 21 November 2018 to 17 November 2019 (361 days) when he died of natural causes (possibly because of drought conditions, as 2019 was the driest year on record in South Australia).

SR260 was trapped on 2 December 2017 and abandoned the mound on 16 January 2018. We used tracking data from 17 January 2018 to 24 February 2019 (403 days) when he was killed by a feral cat.

5.3.4 Vegetation surveys

We conducted vegetation surveys at locations (sites) used by Malleefowl during daylight hours (day sites) chosen based on the number of fixes at those sites. For each bird, we aimed to survey the 40 most and least frequently visited sites. To identify low and high use sites, we counted the number of days each site was visited. Multiple visits to a site on the same day were counted as one visit. To account for differing tracking periods, proportional days visited were calculated as the number of days visited divided by the number of days the bird was tracked. Next, the visit frequency of a day site was determined depending on the proportional days visited: out of all sites that were visited at least once, the bottom 25% were classified as low visit day sites, and the top 25% as high visit day sites, but only if they were visited on more than four days. Additionally, for each bird, we surveyed up to 20 control sites that were not visited but were randomly selected from within the boundary of a minimum convex polygon around each bird's fixes. This totalled 87 (73 Day + 14 Controls), 96 (76 + 20) and 97 (80 + 17) sites for LOC7, SR159 and SR260, respectively. However, to take GPS inaccuracy into account, site visits had to be recalculated during data analysis which reduced the number of control sites. Field surveys were conducted blind, i.e. site type and frequency of use were unknown. Surveyed sites were at least 18 m apart to allow for GPS inaccuracy. Randomly selected control sites were checked against GPS records to ensure they were never visited by birds during the monitoring period.

At each site, we recorded vegetation and habitat characteristics in a 10m x 10m quadrat with the GPS location forming the centre of the quadrat. The horizontal accuracy of the GPS trackers was determined by the manufacturer as ± 18 m for ~96% of known locations ($n = 2,236,404$, pers. comm with MTI). Although covering this area would suggest a square quadrat with a side length of 25 m, we reduced this to 10m for practical reasons and ease of cover estimation, but also because over 83% of fixes in the manufacturer's test were within 10 m of the known location. In each 100 m² quadrat, we measured the height of the lowest and highest points of the canopy, the leaf litter cover, the total plant cover in 3 strata based on plant height (<1 m (ground cover), 1-2 m (midstorey), >2 m (canopy) and cover of individual plants down to species level. Ground cover and midstorey were mutually exclusive (plants were measured in one or the other strata, never in both). If canopy Eucalypts had regrowth from lignotubers under 1 m, these were considered part of the ground cover and counted separately (as well as being counted in the canopy). Cover estimates and leaf density were estimated visually using the same observer at all sites. Leaf density was assessed at the densest part of the canopy based on a crown density scale from Schomaker (2007). All cover was measured in per cent which is equivalent to m², as the quadrats were 100 m².

5.3.5 Data analysis

5.3.5.1 Summary stats

To investigate differences in habitat used by each Malleefowl during the day, we first determined the most common plant taxa (top 30%) of the frequency of site occurrence (A) and proportional cover (B) in each of the three strata of the high use sites. We then determined their site occurrence (A) and proportional cover (B) at the low and control sites.

5.3.5.2 Classification tree models

Over 130 plant species were recorded so data were aggregated to genus level. To investigate whether the three visit frequency categories (high, low and control) were related to the proportional cover of certain genera or other habitat characteristics we applied classification tree models from R package *rpart* (Therneau and Atkinson 2019) with 10-fold default cross-validation. For a robust prediction that avoids overfitting, we pruned all trees according to the 1-SE rule, which says that the best tree is the simplest tree (i.e., with the least splits) that is within one standard deviation of the tree with the smallest cross-validation error. To improve predictive power, we fitted each model 100 times and used the modal tree as the final tree. There were no missing data in any of the data subsets used for classification trees.

We modelled ground and midstorey cover together but did not combine the covers of identical genera in the two strata, as both combined are essential for foraging and most likely predator avoidance too, but the midstorey is likely to be more important as a heat refuge than the ground cover and may be indicative of fully grown plants that may provide more food. In the plots, we called this combined stratum *understorey*. Additionally, we modelled the canopy by itself and all strata together to investigate possible common trends. We fitted all three strata datasets to each Malleefowl separately as the vegetation composition at each bird's site differed. However, we also fitted it to all three birds combined, because even though diet may vary depending on the available vegetation, there may be underlying structural or compositional (e.g. plant diversity) preferences. Visit frequency (High/Low/Control) was used as the response variable. Explanatory variables always included the cover of each plant genus (in %) per site, the number of genera (richness) per site, the total plant cover (abundance, in %) per site and the diversity of genera per site (Shannon index calculated with the *vegan* package (Oksanen, Blanchet *et al.* 2020)). Raised litter (dead branches or whole plants raised off the ground) was considered a genus with one species. Additional explanatory variables used in the models were leaf litter (plant material that has fallen to the ground in %) for the ground cover and

lowest height and maximum height of the canopy in m. We included leaf density in all strata to determine if the GPS signal was poor in areas of dense foliage.

5.4 Results

5.4.1 Locations

Visit frequency classification by proportional days visited resulted in 49 sites for LOC7 (19 high, 19 low and 11 control), and 54 (21, 21 and 12) for both SR159 and SR260. This totalled 61 high use sites, 61 low use sites and 35 controls for all three birds.

5.4.2 Plant taxa counts

In the day and control sites, we recorded 133 unique plant species in 60 genera overall. Most species were recorded for SR159 with 105 unique species from 54 genera. Please see Table 5-1 for the other sites and Table S 5-3 and section 5.7.5 for a complete list of species.

In the dataset classified by visit frequency, we recorded the most species and genera in the low use sites, followed by high use and control sites. There were 97 unique species from 50 genera in the low use sites, 88 species from 47 genera in the high use sites and 76 species from 41 genera in the control sites (Table 5-1).

Table 5-1 Overview of species and genera found in each visit frequency category

ID	Overall		High use		Low use		Control	
	Species	Genera	Species	Genera	Species	Genera	Species	Genera
Lock	46	25	28	17	35	21	25	15
SR159	105	54	60	40	71	42	51	31
SR260	67	33	42	25	43	24	32	21
Total	133	60	88	47	97	50	76	41

5.4.3 Percentage occurrence and cover of plant taxa

The most common plant species at a site varied strongly between birds, except for raised litter, which was present in most sites (>76%) as ground cover. The midstorey of all sites was primarily made up of Eucalypts but Melaleuca had an equally strong presence at the LOC7 sites, as did *Eremophila* at the SR260 sites. The canopy strata of all sites consisted predominantly of Eucalypts. See Table S 5-4 for details.

The percentage cover data across all the sites were slightly positively skewed. The median ground cover was low (8%, IQR = 7%), the midstorey cover slightly lower (6%, IQR = 7%)

and the canopy provided the most cover with a median of 15% (IQR = 19%) (section 5.7.5).

Plant genera with the most ground and midstorey cover varied strongly between birds. Eucalypts dominated the canopy for all birds with the average cover changing between 14 and 25% in the three strata (Table 5-2, also section 5.7.5 for further details).

Table 5-2 Top 30% genera and species by cover (average cover per bird, height and visit frequency in %) found in the high use day sites and how much the average cover of the genera/species was in the low day and control sites. H = Height, G = Ground cover, M = Midstorey, C = Canopy. Genera that were included in the top models are highlighted in bold font.

	H	Genus	High	Low	Control	Species	High	Low	Control	
Lock Nr. of sites: H = 19 L = 19 C = 11	G	<i>Melaleuca</i>	2.4	7	4.9	<i>Eremophila weldii</i>	2.5		0.1	
		Triodia	2.3	6.5	5.8	<i>Melaleuca lanceolata</i>	2.4	5.9	4.7	
		Raised litter	2.2	2.4	2.2	<i>Triodia</i> sp.	2.3	6.5	5.8	
		<i>Eremophila</i>	1.8	1.1	0.3	Raised litter	2.2	2.4	2.2	
						<i>Microcybe pauciflora</i>	1.7	1.7	1.3	
						<i>Santalum acuminatum</i>	1.6	0	0	
	M	<i>Melaleuca</i>	6.9	8.4	5.8	<i>Melaleuca acuminata</i>	6.4	6.7	2	
		<i>Eucalyptus</i>	3.6	4.3	4.6	<i>Melaleuca lanceolata</i>	6.1	6.9	5.6	
						<i>Eucalyptus calcareana</i>	2.8	4.7	3.9	
						<i>Eucalyptus</i> sp.	2.7	1.1	3	
	C	<i>Eucalyptus</i>	17.3	17.1	14.2	<i>Acacia anceps</i>	2.3	0	0	
						<i>Eucalyptus brachycalyx</i>	25	10.5	6.5	
						<i>Eucalyptus oleosa</i>	16.2	3.6	1.6	
	SR159 Nr. of sites: H = 21 L = 21 C = 12	G	<i>Triodia</i>	6.2	10.8	17.2	<i>Triodia</i> sp.	6.2	10.8	17.2
			<i>Beyeria</i>	2.5	3.2	3	<i>Dodonea bursariifolia</i>	3.2	0.7	0.5
Raised litter			2	1.7	2.2	<i>Senna artemisioides ssp flifolia</i>	2.6	0.9	0	
<i>Dodonea</i>			1.5	1	3.4	<i>Beyeria lechenaultii</i>	2.5	3.2	3	
<i>Acacia</i>			1.4	1.1	1.4	Raised litter	2	1.7	2.2	
<i>Olearia</i>			1.3	1.6	1.1	<i>Dodonea hexandra</i>	1.8	0.7	1.1	
<i>Alyxia</i>			1.2	0.8	0.8	<i>Eucalyptus socialis</i>	1.5	4	1.7	
Atriplex			1.1	1.1	1.2	<i>Eucalyptus brachycalyx</i>	1.5	3.9	1.8	
<i>Eucalyptus</i>			1.1	4.3	1.8	<i>Acacia sclerophylla</i>	1.4	1.4	0	
<i>Senna</i>			1	1	0.8	<i>Olearia muelleri</i>	1.4	1.3	1.1	
<i>Myoporum</i>			0.9	0	0	<i>Dodonea stenozyga</i>	1.2	1.3	0.8	
						<i>Alyxia buxifolia</i>	1.2	0.8	0.8	
						<i>Atriplex vesicaria</i>	1.1	1.6	1.3	
						<i>Rhagodia crassifolia</i>	1	0.6	0.6	
					<i>Eremophila glabra</i>	1	0.1	3		
M		<i>Eucalyptus</i>	6.9	5.8	11.1	<i>Eucalyptus incrassata</i>	14.7	2.2	5	
		<i>Alyxia</i>	4.4	1.9	0.6	<i>Alyxia buxifolia</i>	4.4	1.9	0.6	
		<i>Beyeria</i>	4.2	1.8	1.8	<i>Beyeria lechenaultii</i>	4.2	1.8	1.8	
						<i>Eucalyptus brachycalyx</i>	3.9	4.3	10.3	
					<i>Eucalyptus</i> sp.	3.6	0	3		
C		<i>Eucalyptus</i>	20.7	19	25.1	<i>Eucalyptus oleosa</i>	24.1	16.5	28.4	
						<i>Myoporum platycarpum</i>	16	4	0	
						<i>Eucalyptus brachycalyx</i>	13.1	14.6	12	
SR260		G	<i>Acacia</i>	4.1	12.8	3.9	<i>Acacia sclerophylla</i>	4.1	18.2	3.9
			<i>Cratystylis</i>	3.6	4.2	0.9	<i>Cratystylis conocephala</i>	3.6	4.2	0.9

Nr. of sites: H = 21 L = 21 C = 12		<i>Microcybe</i>	3.5	0	2.2	<i>Microcybe multiflora</i>	3.5	0	2.2
		<i>Melaleuca</i>	2.7	1.2	15.9	<i>Rhagodia ulicina</i>	2.8	1	1.2
		<i>Eremophila</i>	2.5	1.9	2	<i>Melaleuca pauperiflora</i>	2.7	1.2	22.7
		<i>Westringia</i>	2.1	0.9	3.4	<i>Eucalyptus socialis</i>	2.6	1.2	0
						<i>Eremophila scoparia</i>	2.4	1.9	2
						<i>Westringia rigida</i>	2.1	0.9	3.4
						Raised litter	2	1.1	1.1
						<i>Scaevola spinescens</i>	2	0.2	0
	M	<i>Eremophila</i>	4.1	0.9	10.5	<i>Eremophila scoparia</i>	4.1	1	10.5
		<i>Templetonia</i>	3.4	1.2	1	<i>Santalum spicatum</i>	3.8	0.8	1.6
		<i>Cratystylis</i>	2.9	3	0	<i>Templetonia retusa</i>	3.4	1.2	1
						<i>Melaleuca lanceolata</i>	3.2	1.5	3.2
	C	<i>Eucalyptus</i>	18.9	20.8	16.6	<i>Eucalyptus</i> sp.	14.2	1.8	8.6

5.4.4 Classification tree models

5.4.4.1 General

On the next page, we present the results of the twelve modal classification trees (all birds/strata combined and separately; Figure 5-2). We focus on covariates that best predicted the likelihood of observations being from high use sites if certain conditions were met. If they were not met, observations were more likely to be from low use sites (unless stated differently). Control sites could not be discriminated with any covariates of any of the twelve modal trees, suggesting that control sites were not defined by a particular set of characteristics and were random. A summary of the classification tree findings and their statistics can be found in Table 5-3.

Table 5-3 Variables that predicted high site use among Malleefowl on the Eyre Peninsula, using rpart classification tree models. G = Ground cover, M = Midstorey, C = Canopy. Understorey is the combined data of ground cover and midstorey. We also present variance explained, prediction accuracy and misclassification rate in %. CI = 95% Confidence intervals for hundred repetitions.

		All birds	LOC7	SR159	SR260	
All strata	Canopy starting height	>2.1 m				
	<i>Senna</i> (G)	<0.2				
	Canopy maximum height	>6.3 m				
	<i>Atriplex</i> (G)	<0.2				
	Ground cover	<16				
	<i>Geijera</i>	<0.3				
	Variance explained / Prediction accuracy	28.1 / 56.1	40 / 63.3	36.4 / 61.1	24.2 / 53.7	
	Misclassification rate (CI)	60.4 (59.6-61.3)	40.8 (40.1-41.4)	66.8 (65.5-68.1)	59.8 (58.5-61.2)	
	Understorey	<i>Eremophila</i> (M)	>1.3			
		Ground cover	<16			
Raised litter (G)		>1.6				
<i>Triodia</i> (G)		<3.3				
<i>Olearia</i> (G)		<0.1				
<i>Atriplex</i> (G)		<0.2				
Total cover (U)		<21				
<i>Geijera</i> (G)		<0.3				
Variance explained / Prediction accuracy		27.1 / 55.4	43.3 / 65.3	36.4 / 61.1	24.2 / 53.7	
Misclassification rate (CI)		63.3 (62.5-64)	60.4 (59-61.9)	66.7 (65.3-68.1)	59.1 (57.8-60.5)	
Canopy	Canopy starting height	>2.1 m				
	Canopy maximum height	>6.3 m				
	Leaf density	>36 & <29				
	Variance explained / Prediction accuracy	18.9 / 50.3	40 / 63.3	30.3 / 57.4	21.9 / 51.9	
	Misclassification rate (CI)	50.9 (50.5-51.2)	40.4 (39.7-41.1)	64.4 (63.1-65.8)	59.4 (58.3-60.6)	

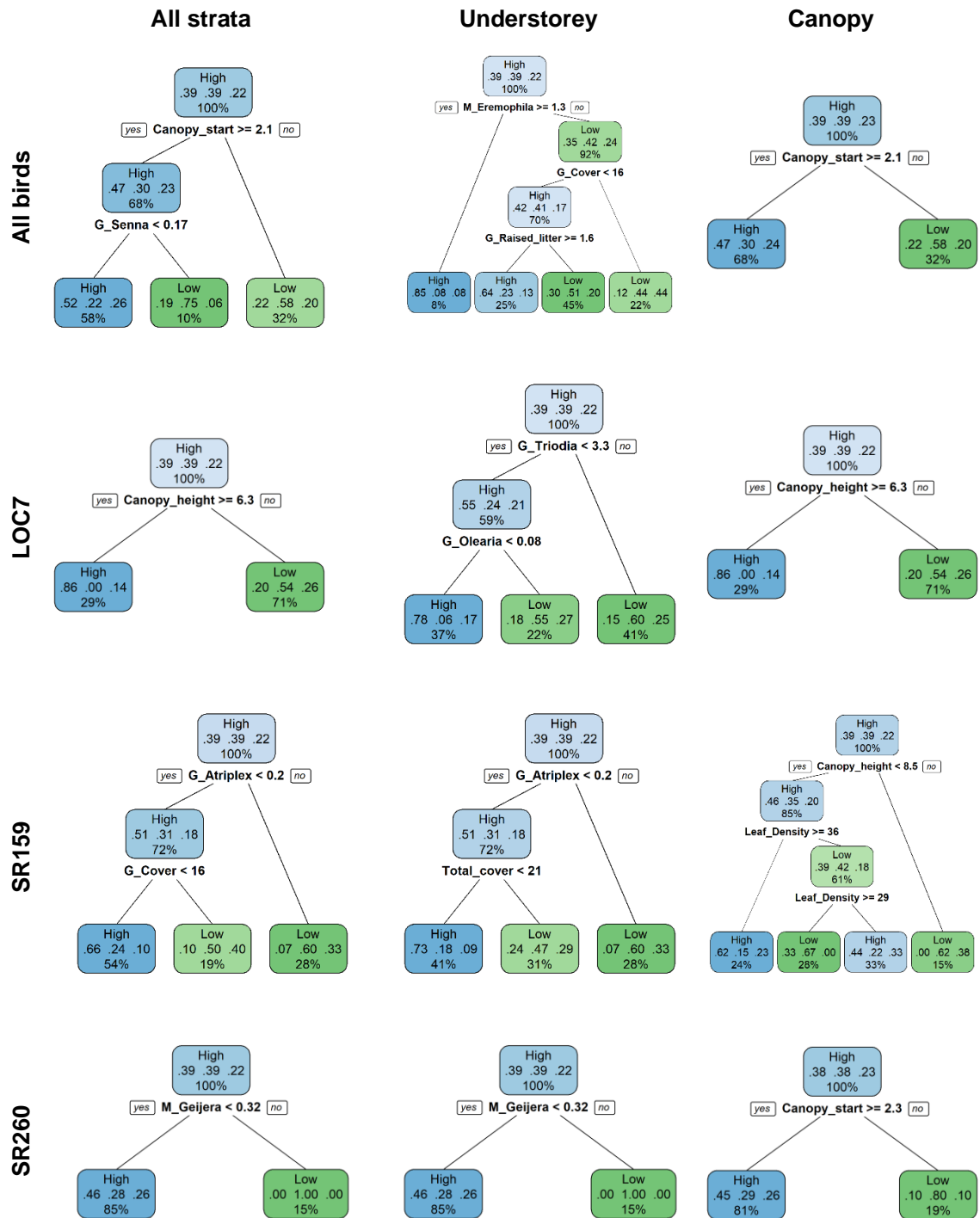


Figure 5-2 Optimal classification trees for the habitat selection of three non-breeding Malleefowl on the Eyre Peninsula. The left plots show the best trees for all three strata combined, the middle plots for the combination of ground cover and midstorey, and the right plots for the canopy. The first row shows the data for all three birds combined, the remaining rows for each bird, as labelled. Each node shows the predicted (most likely) class (High/Low/Control) in the top row; the predicted probability of each of these three classes in the middle row, and the percentage of observations in that node in the bottom row. Blue colours indicate high use sites and green colours low use sites. Controls were never predicted. Stronger colours indicate higher probabilities of prediction. G = Ground cover, M = Midstorey, C = Canopy. Understorey is the combined data of ground cover and midstorey.

5.4.4.2 All birds

The classification tree for all strata and all birds combined indicated that high use sites were more likely to be those with a canopy starting height of over 2.1 m and a ground cover of *Senna* less than 0.2%. The average starting height for tree canopies across all birds was 2.9 ± 1.4 m and *Senna* was present as ground cover in small amounts (0 - 2.6%) at the two Secret Rocks sites (section 5.7.5).

Increased cover of *Eremophila* was a predictor of high use sites for the combined understorey (ground cover + midstorey). If observations had a midstorey *Eremophila* cover of over 1.3%, they were likely to be from a high use site. The average *Eremophila* midstorey across all birds and sites was $0.4 \pm 1.4\%$ but could be as high as 15% (section 5.7.5). If, on the other hand, *Eremophila* was less than 1.3%, the total ground cover could be used to classify observations into high and low. Observations were more likely to be from high use sites if the total ground cover was under 16% and raised litter above 1.6%. Canopy high use sites were best predicted by a canopy starting height of over 2.1 m.

5.4.4.3 Individual birds - LOC7

The classification tree for all strata combined (also the canopy only dataset) for LOC7 indicated that if observations have a canopy maximum height of over 6.3 m, they were more likely to be from a high use site. Average canopy heights for this bird were 5.3 ± 1.8 m with a maximum of 12 m (section 5.7.5).

In the combined understorey, *Triodia* and *Olearia* ground cover of less than 3.3% and 0.1%, respectively, predicted a high use site. The average *Triodia* ground cover for LOC7 was $1.8 \pm 2\%$ in high and $6.2 \pm 5.7\%$ in low use sites. The average *Olearia* ground cover was very low with $0.1 \pm 0.3\%$ for high and $0.6 \pm 1\%$ for low use sites (section 5.7.5).

5.4.4.4 Individual birds - SR159

For all strata combined, *Atriplex* ground cover of below 0.2% (practically zero) and a total ground cover of less than 16% predicted high use site best. The average *Atriplex* ground cover for SR159 was very low, with $0.2 \pm 0.7\%$ in high and $0.6 \pm 1.1\%$ in low use sites (section 5.7.5).

Atriplex ground cover (<0.2%) and total understorey cover (ground + midstorey cover) below 21% were the best predictors for the combined understorey for this bird. The average understorey cover for SR159 at high use sites was $17.8 \pm 12.3\%$ with a range of 2.5 to 75%. It was slightly higher at low use sites with $22 \pm 15\%$ and a range of 3 to 65% (section 5.7.5).

For the canopy, high use sites were best predicted by a canopy maximum height of below 8.5 m and a leaf density of over 36% or below 29%. If the leaf density was between 29 and 36%, they were more likely to be in low use sites. The average leaf density for this bird was 30.7 ± 13.2 % with a maximum of 65%. Average canopy heights at high use sites were 4.7 ± 2.6 m and 5.8 ± 2.8 m at low use sites with a maximum of 10 m overall (section 5.7.5).

5.4.4.5 Individual birds - SR260

For all strata combined and the combined understorey, a low midstorey cover of *Geijera* (< 0.3%) predicted high use sites best. None of the SR260 high use sites had *Geijera* recorded. The low use sites had an average *Geijera* midstorey cover of 1.3 ± 2.7 %. A canopy starting height of over 2.3 m was the best predictor for the canopy stratum. The average canopy starting height for SR260 was 3 ± 1 m) (section 5.7.5).

5.4.4.6 Summary

The data of all birds combined showed that Malleefowl were more likely to visit sites if the starting canopy height was above 2.1 m and there was only very little *Senna* cover. In the understorey, they preferred sites if midstorey *Eremophila* cover was higher, or ground cover was low with some raised litter on the ground.

LOC7 preferred sites where the canopy was tall and there was less *Triodia* and very little *Olearia* cover.

SR159 preferred sites where *Atriplex* ground cover was very low, total ground cover was low, the canopy height was below 8.5 m, and the leaf density was below 29 or above 36%.

SR260 was more likely to visit sites if their midstorey cover of *Geijera* was low and the canopy started above 2.3 m.

In summary, all birds preferred less understorey cover and tall canopies. Each bird avoided different ground cover plants, depending on availability, but there was a common preference for *Eremophila* and an aversion for *Senna*. Species richness or diversity, leaf litter or canopy cover were not predictors of high or low use sites in any of the optimal trees. Starting canopy height was highly correlated with maximum canopy height (0.7) suggesting that the preference for higher starting heights was also a preference for taller total canopy.

5.5 Discussion

5.5.1 All birds

Studying the fine-scale habitat use of Malleefowl in our study suggested that the species does not have strong associations with particular plant genera. We found that the canopy structure was the overall central driver of fine-scale Malleefowl habitat selection, with a preference for higher canopies starting over 2.1 m. Two-thirds of all observations were in this branch of the classification tree. A high canopy of approximately 5 - 7 m is an indicator of mature mallee eucalypts over 30 years post-fire with individual trees having a well-developed and close-spaced canopy (Gosper, Yates *et al.* 2012; Haslem, Kelly *et al.* 2011; Kenny, Bennett *et al.* 2018). Mallee eucalypt growth slows after approximately 50 years (Kenny, Bennett *et al.* 2018) and the canopy between 2 and 4 m is densest about 60+ years post-fire (Doherty, van Etten *et al.* 2016). However, in our study, all habitat sampled was at least 28 years post-fire with two of the three birds inhabiting mallee at least 60 years old, hence the vegetation at all sites could be regarded as mature. Thus the preference for taller canopies was at a finer scale within a mature *Eucalyptus* matrix. However, fires can be very patchy, and some trees are likely older than others within unburnt habitat of the same age. Additionally, trees grow at different rates depending on their access to water, and drainage, elevation and groundwater are likely unevenly distributed across the landscape. The Malleefowl's preference for canopies over two meters may be an indication that they actively seek out taller, possibly older mallee trees, because the denser canopy attributed to this height profile may act as a heat refuge and/or offer protection from avian predators (Benshemesh 1992; Frith 1962a). Other ground-dwelling birds also prefer taller canopies as thermal refuges (Carroll, Davis *et al.* 2015; Rakowski, Elmore *et al.* 2019) or prefer cooler, denser vegetation to bare ground (Londe, Elmore *et al.* 2021). This is supported by Frith (1962b) and Benshemesh (1992) who found that Malleefowl breeding densities were highest in areas with an uninterrupted and dense canopy. However, a denser canopy, which we measured as leaf density, was not a predictor in the models for this combined dataset and perhaps warrants further research. Leaf density was part of the modal decision tree for only one bird and is discussed below. An alternative explanation for the preference for trees with taller canopy may be related to night-time roosting sites. GPS fixes taken in the early morning and late evening may capture bird locations just before or just after they are roosting thus reflecting roosting preferences rather than foraging or temperature and predator avoidance preferences. Apart from limiting fixes to day-time fixes, we did not further discriminate fix times and included fixes between approximately 1 hour before sunset and 1.5 hours after sunset.

Additionally, sites were more likely to be used less often if *Senna* was present, indicating a dislike for this genus. *Senna* is a leguminous woody shrub with unpalatable leaves, but its seeds have been mentioned as a food source for Malleefowl (Frith 1959; Frith 1962a). However, *Senna* was only available at the SR sites where the birds were tracked in 2018 and into 2019, which were both very dry and hot years (BOM 2019; BOM 2020). It is possible that the *Senna* plants did not set seed in these years thus providing no incentive to visit often.

In the combined understorey, Malleefowl strongly preferred sites with an abundance of *Eremophila*. There was an 85% possibility of a site being a high use site if it contained *Eremophila* between 1-2 m high. This could be explained by the popularity of *Eremophila* as a food plant, which provides a variety of edible parts nearly year-round (Harlen and Priddel 1996; van der Waag 2004). If there was less *Eremophila*, Malleefowl preferred areas with less ground cover and some raised litter on ground level. Raised litter possibly plays a role as a habitat for decomposers like fungi and termites but also a structural component that provides a hiding place for invertebrates, such as grasshoppers or cockroaches. All these organisms are food sources for Malleefowl (Benshemesh 1992; Booth 1986; Brickhill 1987; Reichelt and Jones 2008) and could thus explain their preference for these sites with raised litter. Gosper, Prober *et al.* (2013) identified ground fuel (their definition includes leaf litter and raised litter) as highest and the understorey as suppressed (open) in the 35 - 125 year post-fire age class. While our classification trees did not include leaf litter as a predictor, high use sites were more likely to have raised litter and reduced ground cover, possibly a result of a dense, healthy canopy which are indicators of the 35-125 age bracket (Gosper, Prober *et al.* 2013), and this possibly further supports the Malleefowl's preference for patches of mallee with a mature canopy.

5.5.2 Individual birds

When we investigated visit frequencies to the lower vegetation strata of individual birds, we found that each bird showed a dislike for different understorey plants. For LOC7 and SR159 this was *Triodia* and *Atriplex* at ground level respectively, while for SR260 it was *Geijera* in the midstorey. Interestingly, none of these plants were listed as food plants in previous studies (Table S 5-1), which could explain their lower cover at high use sites. *Triodia* could be found in all three study areas, while *Atriplex* and *Geijera* were only present in the Secret Rocks sites (Table S 5-2). *Triodia* cover gradually decreases from approximately 30 years post-fire (Kenny, Bennett *et al.* 2018), therefore this dislike might be an indication that Malleefowl favour areas that are long unburnt.

When we studied the habitat preferences in the canopy level, both starting height and maximum height of the canopy were the most important variables. However, all three birds had varying preferences, with LOC7 preferring high canopies over 6 m, SR159 canopy heights below 8.5 m and SR260 canopies that start over 2.3 m. The common factor was reflected in the top results from the analysis of all three birds combined, where canopy starting height over 2.1 m was the most important variable and was discussed above. Leaf density was an additional factor for only one of the birds: SR159 preferred sites that had a canopy height less than 8.5 m *and* a leaf density of below 29 or over 36%. This leaf density range was surprising and may be related to unrecorded correlated variables. The birds' preference for denser canopy also indicates that GPS triangulation and battery charging were not hindered by dense tree canopies.

5.5.3 Shortcomings of study

While our number of observations was not particularly large, especially when testing the individual birds, we tried to alleviate this issue by repeating the classification models 100 times which produced small confidence intervals of the misclassification rates. We also only have a small sample size of three non-breeding birds, and future studies should attempt to obtain data from more individuals to increase the size of the dataset. This might increase the chances of uncovering more common attributes that are preferred by Malleefowl and temporal aspects such as time of year or associated environmental aspects such as temperature and rainfall. The size of our vegetation survey sites was 10m x 10m, which is slightly smaller than the average accuracy of our GPS locations, thus our results should be used with some caution. However, while resource and technology limitations currently constrain studies such as these, future studies using more accurate tracking technology with higher temporal frequency fixes may enable a more accurate selection of highly-used habitats, while remote sensing technologies may be useful to provide broader-scale habitat analyses. For smaller-scale habitat analyses, using species-level data as opposed to genus level data may be beneficial in discovering finer habitat use preferences, for example, we combined all Eucalypt species in the canopy due to low sample size but some species may be favoured over others.

5.5.4 Conclusion

In this chapter, we show that Malleefowl did not have strong associations with particular plant genera supporting other studies that found Malleefowl have a broad diet and feed opportunistically on a range of plant species. Although some genera such as *Atriplex*, *Triodia*, *Senna* and *Geijera* were in lower cover at high use sites this may reflect a seasonal absence of food items rather than a broader scale avoidance. Equally, the higher cover of

Eremophila could be related to a more regular and dependable supply of food items. Although the plant composition in each study area varied and pointed to individual preferences, there were also commonalities that influenced Malleefowl movement and habitat selection. The most significant commonalities indicate that Malleefowl preferred micropatches of habitat with taller canopies that start above 2.1 m, less ground cover and more raised litter perhaps reflecting older trees within the landscape. These sites may be used for heat or predator avoidance, or simply reflect proximity to favoured roosting sites. Climate change is expected to lead to more frequent hotter and drier periods and increased high-intensity wildfires, which may remove taller, older trees. Conservation managers can help to maintain these patches within Malleefowl habitat by ensuring fires are patchy and herbivores are controlled, which will also encourage the re-establishment of a large variety of native shrubs that are palatable to Malleefowl and provide year-round food.

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5.7 Appendix

5.7.1 Previous studies

Table S 5-1 Plants consumed by Malleefowl (modified after Reichelt and Jones 2008). Header row: O = observation, C & G = crop and gizzard content, F = faecal matter, P = palatability study. Plant parts consumed: F = flowers, f = foliage, b = buds, p = pods, berries, other fruit, s = seeds, t = tubers.

Family	Genus	Frith (1962) Griffith, NSW (O)	Booth (1986) Renmark, SA (C & G)	Brickhill (1987) Yalgogrin, NSW (F)	Benshemesh (1992) Wyperfeld, Vic. (O)	Kentish and Westbrooke (1994) Ouyen, Vic. (C & G)	Harlen and Priddel (1996) Yathong, NSW (P)	van der Waag (2004) Narembeen, WA (Chick, C & G)	Reichelt and Jones (2008) Little Desert, Vic. (O)
Amaranthaceae	<i>Einadia</i>			f, s					
Amaranthaceae	<i>Enchylaena</i>		s						
Amaranthaceae	<i>Maireana</i>						e		
Amaranthaceae	<i>Sclerolaena</i>		f				e		
Apiaceae	<i>Daucus</i>					f, p	e		
Asparagaceae	<i>Arthropodium</i>		F, f, p						
Asparagaceae	<i>Lomandra</i>				F				F
Asparagaceae	<i>Thysanotus</i>				f		e		t
Asteraceae	<i>Actinoble</i>						e		
Asteraceae	<i>Brachyscome</i>				f				
Asteraceae	<i>Calotis</i>						f		
Asteraceae	<i>Carthamus</i>			s					
Asteraceae	<i>Helichrysum</i>				f		f		
Asteraceae	<i>Helipterum</i>						e		
Asteraceae	<i>Hypochaeris</i>					F, f	f		
Asteraceae	<i>Olearia</i>						b, F		
Asteraceae	<i>Sonchus</i>						f		
Bertyaceae	<i>Bertya</i>					p			
Brassicaceae	<i>Brassica</i>				s	p			
Brassicaceae	<i>Stenopetalum</i>				f				
Campanulaceae	<i>Wahlenbergia</i>					p			
Caryophyllaceae	<i>Silene</i>					F, f, p			
Celastraceae	<i>Stackhousia</i>						e		
Crassulaceae	<i>Crassula</i>						e		
Cupressaceae	<i>Callitris</i>								f
Dilleniaceae	<i>Hibbertia</i>							s	
Ericaceae	<i>Astroloma</i>							s	F
Ericaceae	<i>Brachyloma</i>								F
Euphorbiaceae	<i>Beyeria</i>	p				p			
Fabaceae	<i>Acacia</i>	s		s			p, s		F, b, p
Fabaceae	<i>Bossiaea</i>						b, F, p		
Fabaceae	<i>Daviesia</i>						F	s	
Fabaceae	<i>Eutaxia</i>						b, F		
Fabaceae	<i>Medicago</i>						e		
Fabaceae	<i>Senna</i>	s							
Geraniaceae	<i>Erodium</i>						e		
Goodeniaceae	<i>Goodenia</i>						e		F
Haloragaceae	<i>Glischrocaryon</i>								F
Juncaginaceae	<i>Triglochin</i>					p			

Family	Genus	Frith (1962) Griffith, NSW (O)	Booth (1986) Renmark, SA (C & G)	Brickhill (1987) Yalgogrin, NSW (F)	Benshemesh (1992) Wyperfeld, Vic. (O)	Kentish and Westbrooke (1994) Ouyen, Vic. (C & G)	Harlen and Priddey (1996) Yathong, NSW (P)	van der Waag (2004) Narembeen, WA (Chick, C & G)	Reichelt and Jones (2008) Little Desert, Vic. (O)
Lamiaceae	<i>Prostanthera</i>								F, f
Lauraceae	<i>Cassytha</i>		s	f, s					
Malvaceae	<i>Sida</i>			s					
Myrtaceae	<i>Eucalyptus</i>								F, f
Myrtaceae	<i>Melaleuca</i>								s
Orchidaceae	<i>Pterostylis</i>						e		t
Pittosporaceae	<i>Pittosporum</i>	f							
Plantaginaceae	<i>Plantago</i>						f		
Poaceae	<i>Poa</i>						f		
Poaceae	<i>Stipa</i>						f		
Poaceae	<i>Triticum</i>			s					
Proteaceae	<i>Grevillea</i>						b, F		
Ranunculaceae	<i>Ranunculus</i>			s					
Rutaceae	<i>Philothea</i>	f							
Sapindaceae	<i>Dodonaea</i>		s				f, s		f
Scrophulariaceae	<i>Eremophila</i>						b, F, f	s	
Scrophulariaceae	<i>Myoporum</i>						b, F, f		
Solanaceae	<i>Solanum</i>			s					
Zygophyllaceae	<i>Zygophyllum</i>		f, s, p						

5.7.2 Study sites plant composition

Table S 5-2 Predominant plant communities in the home ranges three tracked Malleefowl on the Eyre Peninsula. Table adapted from Brandl, McDonald *et al.* (2009). Rainfall is the cumulative rainfall for the years the birds were tracked (BOM 2022). We used QGIS (QGIS Development Team 2021) to plot sites and report the intersecting Native Vegetation Floristic Areas sourced from the Government of South Australia (2022).

Bird	Rain	SA VEG ID	Broad Vegetation Community Description	Dominant Species Description
LOC7	2017: 344	[EP1901] presumably MM1901	Eucalyptus mallee forest and mallee woodland	<i>Eucalyptus incrassata</i> mid mallee woodland over <i>Leptospermum coriaceum</i> (mixed) shrubs and <i>Hibbertia australis</i> (mixed) shrubs
		MN2505	Eucalyptus mallee forest and mallee woodland	<i>Eucalyptus gracilis</i> mid open mallee forest over <i>Geijera linearifolia</i> (mixed) shrubs and <i>Carrichtera annua</i> (mixed) shrubs
SR159	2018: 235 2019: 150	MN2505	As above	As above
		EP2503	Eucalyptus mallee forest and mallee woodland	<i>Eucalyptus gracilis</i> , <i>E. oleosa</i> , &/or <i>E. dumosa</i> open Mallee over <i>Maireana sedifolia</i> or <i>Cratystylis conocephala</i> low shrubs
		EP2404	Eucalyptus mallee forest and mallee woodland	<i>Eucalyptus incrassata</i> mid mallee woodland over <i>Melaleuca uncinata</i> tall shrubland and <i>Thryptomene micrantha</i> (mixed) low open shrubland and <i>Triodia lanata</i> low open hummock grassland
		EP1001	Callitris forest and woodland	<i>Callitris verrucosa</i> low woodland over <i>Leptospermum coriaceum</i> tall shrubs
		EP3501	Melaleuca shrubland >1m	<i>Melaleuca uncinata</i> tall open shrubland over <i>Hysterobaeckea behrii</i> (mixed) low shrubs and <i>Triodia irritans</i> low shrubs
		MN6201	shrubland >1m	<i>Geijera linearifolia</i> (mixed) mid open shrubland over <i>Nitraria billardierei</i> shrubs and <i>Maireana</i> sp. (mixed) shrubs
		MM0801	Callitris forest and woodland	<i>Callitris gracilis</i> low open woodland over <i>Austrostipa</i> sp. (mixed) tussock grasses
EP1603	Eucalyptus mallee forest and mallee woodland	<i>Eucalyptus brachycalyx</i> mid mallee woodland over <i>Melaleuca pauperiflora</i> ssp. <i>mutica</i> (mixed) tall shrubs and <i>Olearia muelleri</i> (mixed) low shrubs		
SR260	2017: 358 2018: 235 2019: 150	EP1602	Eucalyptus mallee forest and mallee woodland	<i>Eucalyptus brachycalyx</i> mid mallee woodland over <i>Eremophila scoparia</i> (mixed) tall shrubs and <i>Olearia muelleri</i> (mixed) low shrubs
		EP2404	As above	As above
		EP1001	As above	As above
		EP3501	As above	As above

5.7.3 Species counts

Table S 5-3 List of all plant species recorded at all sites independently of visit frequency. Species are listed by height class. Count is the number of locations at which a species was found, and n is the total number of day sites surveyed (including control sites). S = Stratum.

S	LOC7 (n = 87)	Count	SR159 (n = 96)	Count	SR260 (n = 97)	Count
Ground cover	Raised litter	83	Raised litter	86	Raised litter	91
	<i>Melaleuca lanceolata</i>	76	<i>Alyxia buxifolia</i>	35	<i>Rhagodia crassifolia</i>	60
	<i>Triodia</i> sp.	74	<i>Olearia muelleri</i>	35	<i>Eremophila scoparia</i>	48
	<i>Rhagodia crassifolia</i>	49	<i>Triodia</i> sp.	34	<i>Grevillea huegelii</i>	36
	<i>Microcybe pauciflora</i>	35	<i>Senna artemisioides</i> ssp <i>coriacea</i>	25	<i>Acacia sclerophylla</i>	28
	<i>Acrotriche patula</i>	22	<i>Rhagodia crassifolia</i>	22	<i>Eucalyptus</i> sp.	26
	<i>Eremophila crassifolia</i>	15	<i>Lomandra effusa</i>	20	<i>Melaleuca pauperiflora</i>	26
	<i>Beyeria lechenaultii</i>	14	<i>Atriplex vesicaria</i>	19	<i>Westringia rigida</i>	23
	<i>Olearia muelleri</i>	14	<i>Roepera aurantiaca</i>	19	<i>Olearia</i> sp.	20
	<i>Acacia anceps</i>	13	<i>Geijera linearifolia</i>	18	<i>Atriplex stipitata</i>	19
	<i>Dodonea humilis</i>	13	<i>Maireana pentatropis</i>	18	<i>Rhagodia ulicina</i>	19
	<i>Eucalyptus</i> sp.	13	<i>Roepera apiculata</i>	17	<i>Cratystylis conocephala</i>	18
	<i>Lomandra effusa</i>	10	<i>Beyeria lechenaultii</i>	16	<i>Templetonia retusa</i>	14
	<i>Eremophila weldii</i>	9	<i>Cratystylis conocephala</i>	16	<i>Atriplex</i> sp.	11
	<i>Melaleuca acuminata</i>	9	<i>Enchylaena tomentosa</i>	15	<i>Beyeria lechenaultii</i>	11
	<i>Olearia minor</i>	9	<i>Podolepis capillaris</i>	15	<i>Olearia muelleri</i>	11
	<i>Dianella revoluta</i>	7	<i>Senna artemisioides</i> ssp <i>petiolaris</i>	14	<i>Senna artemisioides</i> ssp <i>coriacea</i>	9
	<i>Eucalyptus calcareana</i>	6	<i>Dodonea stenozyga</i>	13	<i>Alyxia buxifolia</i>	8
	<i>Eucalyptus yalatensis</i>	5	<i>Eucalyptus brachycalyx</i>	13	<i>Atriplex vesicaria</i>	8
	<i>Eucalyptus phenax</i>	4	Grass sp.	12	<i>Maireana pentatropis</i>	8
	Grass sp.	4	<i>Rhagodia preissii</i>	12	<i>Geijera linearifolia</i>	6
	<i>Eucalyptus brachycalyx</i>	3	<i>Maireana erioclada</i>	11	<i>Lomandra effusa</i>	5
	<i>Eucalyptus oleosa</i>	3	<i>Grevillea huegelii</i>	9	<i>Maireana georgei</i>	5
	<i>Helichrysum leucopsidium</i>	3	<i>Acacia sclerophylla</i>	8	<i>Triodia</i> sp.	5
	<i>Pimelea glauca</i>	2	<i>Atriplex stipitata</i>	8	<i>Dianella revoluta</i>	4
	<i>Santalum acuminatum</i>	2	<i>Dodonea hexandra</i>	8	<i>Eremophila glabra</i>	4
	<i>Westringia rigida</i>	2	<i>Eremophila glabra</i>	8	<i>Eucalyptus gracilis</i>	4
	<i>Acacia spinescens</i>	1	<i>Westringia rigida</i>	8	<i>Eucalyptus oleosa</i>	4
	<i>Billardiera cymosa</i>	1	<i>Dianella revoluta</i>	7	<i>Eucalyptus socialis</i>	4
	<i>Callitris canescens</i>	1	<i>Dodonea bursariifolia</i>	7	<i>Maireana radiata</i>	4
	<i>Eremophila deserti</i>	1	<i>Rhagodia ulicina</i>	7	<i>Carpobrotus glaucescens</i>	3
	<i>Eremophila glabra</i>	1	<i>Sclerolaena uniflora</i>	7	<i>Enchylaena tomentosa</i>	3
	<i>Eremophila</i> sp.	1	<i>Dissocarpus paradoxa</i>	6	<i>Microcybe multiflora</i>	3
	<i>Eutaxia microphylla</i>	1	<i>Eucalyptus socialis</i>	6	<i>Olearia calcarea</i>	3
	<i>Lasiopetalum discolor</i>	1	<i>Dodonea viscosa</i> ssp <i>angustissima</i>	5	<i>Scaevola spinescens</i>	3
	<i>Lepidosperma congestum</i>	1	<i>Eucalyptus incrassata</i>	5	<i>Sclerolaena diacantha</i>	3
	<i>Olearia ciliata</i>	1	<i>Eucalyptus</i> sp.	5	<i>Senna artemisioides</i> ssp <i>petiolaris</i>	3
	<i>Ozothamnus decurrens</i>	1	<i>Melaleuca lanceolata</i>	5	<i>Lasiopetalum behrii</i>	2
	<i>Pimelea flava</i>	1	<i>Olearia minor</i>	5	<i>Melaleuca lanceolata</i>	2
	<i>Roepera aurantiaca</i>	1	<i>Calytrix tetragona</i>	4	<i>Myoporum platycarpum</i>	2
			<i>Exocarpos aphyllus</i>	4	<i>Roepera aurantiaca</i>	2
			<i>Goodenia willisiana</i>	4	<i>Acacia halliana</i>	1
		<i>Maireana georgei</i>	4	<i>Acacia notabilis</i>	1	

S	LOC7 (n = 87)	Count	SR159 (n = 96)	Count	SR260 (n = 97)	Count
			<i>Maireana turbinata</i>	4	<i>Acacia nyssophylla</i>	1
			<i>Olearia decurrens</i>	4	<i>Acacia</i> sp.	1
			<i>Eremophila scoparia</i>	3	<i>Calytrix tetragona</i>	1
			<i>Olearia</i> sp.	3	<i>Dodonea stenozyga</i>	1
			<i>Rhagodia parabolica</i>	3	<i>Eremophila crassifolia</i>	1
			<i>Rhagodia spinescens</i>	3	<i>Eremophila deserti</i>	1
			<i>Senna artemisioides</i> ssp <i>filifolia</i>	3	<i>Eucalyptus incrassata</i>	1
			<i>Trachymene cyanopetala</i>	3	<i>Eucalyptus leptophylla</i>	1
			<i>Actinoble uliginosum</i>	2	<i>Melaleuca uncinata</i>	1
			<i>Atriplex</i> sp.	2	<i>Minuria leptophylla</i>	1
			<i>Daisy</i> sp.	2	<i>Olearia decurrens</i>	1
			<i>Eremophila crassifolia</i>	2	<i>Rhagodia preissii</i>	1
			<i>Eriochiton sclerolaenoides</i>	2	<i>Roepera angustifolia</i>	1
			<i>Goodenia havilandii</i>	2	<i>Roepera apiculata</i>	1
			<i>Lasiopetalum behrii</i>	2	<i>Santalum acuminatum</i>	1
			<i>Lepidosperma congestum</i>	2	<i>Santalum spicatum</i>	1
			<i>Microcybe pauciflora</i>	2	<i>Sclerolaena uniflora</i>	1
			<i>Myoporum platycarpum</i>	2	<i>Thryptomene micrantha</i>	1
			<i>Olearia calcarea</i>	2		
			<i>Pimelea microcephala</i>	2		
			<i>Pittosporum angustifolium</i>	2		
			<i>Solanum hystrix</i>	2		
			<i>Acacia euthycarpa</i>	1		
			<i>Acacia ligulata</i>	1		
			<i>Acacia notabilis</i>	1		
			<i>Acacia spinescens</i>	1		
			<i>Blennospora drummondii</i>	1		
			<i>Callitris verrucosa</i>	1		
			<i>Carpobrotus glaucescens</i>	1		
			<i>Cassytha</i> sp.	1		
			<i>Dodonea</i> sp.	1		
			<i>Eucalyptus oleosa</i>	1		
			<i>Eucalyptus phenax</i>	1		
			<i>Eutaxia microphylla</i>	1		
			<i>Halgania</i> sp.	1		
			<i>Helichrysum leucopsidium</i>	1		
			<i>Hyalosperma glutinosum</i>	1		
			<i>Leptospermum coriaceum</i>	1		
			<i>Maireana radiata</i>	1		
			<i>Maireana</i> sp.	1		
			<i>Maireana trichoptera</i>	1		
			<i>Maireana triptera</i>	1		
			<i>Melaleuca uncinata</i>	1		
			<i>Millotia muelleri</i>	1		
			<i>Minuria leptophylla</i>	1		
			<i>Roepera ammophila</i>	1		
			<i>Roepera angustifolia</i>	1		
			<i>Roepera glauca</i>	1		
			<i>Senecio glossanthus</i>	1		
			<i>Thysanotus patersonii</i>	1		
			<i>Vittadinia dissecta</i>	1		

S	LOC7 (n = 87)	Count	SR159 (n = 96)	Count	SR260 (n = 97)	Count
			<i>Waitzia acuminata</i>	1		
			<i>Xerochrysum bracteatum</i>	1		
Midstorey	<i>Melaleuca lanceolata</i>	78	<i>Eucalyptus brachycalyx</i>	33	<i>Eremophila scoparia</i>	36
	<i>Eucalyptus calcareana</i>	26	<i>Geijera linearifolia</i>	30	<i>Eucalyptus</i> sp.	36
	<i>Eucalyptus phenax</i>	23	Raised litter	30	<i>Melaleuca pauperiflora</i>	34
	Raised litter	22	<i>Alyxia buxifolia</i>	20	Raised litter	27
	<i>Eucalyptus</i> sp.	19	<i>Eucalyptus incrassata</i>	16	<i>Alyxia buxifolia</i>	14
	<i>Melaleuca acuminata</i>	14	<i>Senna artemisioides</i> ssp <i>coriacea</i>	16	<i>Eucalyptus socialis</i>	10
	<i>Eucalyptus oleosa</i>	13	<i>Eucalyptus socialis</i>	13	<i>Geijera linearifolia</i>	10
	<i>Eucalyptus gracilis</i>	10	<i>Cassytha</i> sp.	10	<i>Templetonia retusa</i>	8
	<i>Eucalyptus yalataensis</i>	8	<i>Exocarpos aphyllus</i>	10	<i>Myoporum platycarpum</i>	7
	<i>Beyeria lechenaultii</i>	6	<i>Senna artemisioides</i> ssp <i>petiolaris</i>	10	<i>Acacia sclerophylla</i>	6
	<i>Acacia anceps</i>	4	<i>Eucalyptus oleosa</i>	9	<i>Eucalyptus oleosa</i>	6
	<i>Eucalyptus brachycalyx</i>	4	<i>Beyeria lechenaultii</i>	8	<i>Eucalyptus phenax</i>	5
	<i>Eucalyptus diversifolia</i>	4	<i>Dodonea stenozyga</i>	7	<i>Santalum spicatum</i>	5
	<i>Eremophila weldii</i>	3	<i>Eucalyptus</i> sp.	5	<i>Eucalyptus gracilis</i>	4
	<i>Pittosporum angustifolium</i>	3	<i>Melaleuca lanceolata</i>	5	<i>Melaleuca lanceolata</i>	4
	<i>Callitris canescens</i>	2	<i>Melaleuca uncinata</i>	4	<i>Santalum acuminatum</i>	4
	<i>Cassytha</i> sp.	2	<i>Myoporum platycarpum</i>	4	<i>Beyeria lechenaultii</i>	3
	<i>Eucalyptus leptophylla</i>	2	<i>Acacia lanceolata</i>	3	<i>Cratystylis conocephala</i>	3
	<i>Acacia spinescens</i>	1	<i>Callitris verrucosa</i>	3	<i>Melaleuca uncinata</i>	2
	<i>Eucalyptus socialis</i>	1	<i>Eremophila scoparia</i>	3	<i>Senna artemisioides</i> ssp <i>petiolaris</i>	2
	<i>Microcybe pauciflora</i>	1	<i>Eucalyptus leptophylla</i>	3	<i>Acacia</i> sp.	1
	<i>Olearia minor</i>	1	<i>Acacia spinescens</i>	2	<i>Acacia spinescens</i>	1
	<i>Santalum acuminatum</i>	1	<i>Cratystylis conocephala</i>	2	<i>Acacia tetragonophylla</i>	1
			<i>Dodonea viscosa</i> ssp <i>angustissima</i>	2	<i>Acacia wilhelmiana</i>	1
			<i>Eucalyptus gracilis</i>	2	<i>Dodonea stenozyga</i>	1
			<i>Eucalyptus phenax</i>	2	<i>Eremophila deserti</i>	1
			<i>Acacia hakeoides</i>	1	<i>Eucalyptus incrassata</i>	1
			<i>Acacia nyssophylla</i>	1	<i>Eucalyptus leptophylla</i>	1
			<i>Acacia rigens</i>	1	<i>Grevillea huegelii</i>	1
			<i>Atriplex vesicaria</i>	1	<i>Microcybe multiflora</i>	1
			<i>Dodonea bursariifolia</i>	1	<i>Pittosporum angustifolium</i>	1
		<i>Grevillea huegelii</i>	1	<i>Rhagodia crassifolia</i>	1	
		<i>Lasiopetalum behrii</i>	1	<i>Thryptomene micrantha</i>	1	
		<i>Olearia decurrens</i>	1			
		<i>Pittosporum angustifolium</i>	1			
		<i>Senna artemisioides</i> ssp <i>filifolia</i>	1			
Canopy	<i>Eucalyptus calcareana</i>	37	<i>Eucalyptus brachycalyx</i>	41	<i>Eucalyptus socialis</i>	49
	<i>Eucalyptus phenax</i>	31	<i>Eucalyptus oleosa</i>	32	<i>Eucalyptus gracilis</i>	29
	<i>Eucalyptus gracilis</i>	18	<i>Eucalyptus socialis</i>	19	<i>Eucalyptus oleosa</i>	29
	<i>Eucalyptus brachycalyx</i>	11	<i>Eucalyptus incrassata</i>	14	<i>Eucalyptus</i> sp.	12
	<i>Eucalyptus oleosa</i>	6	<i>Eucalyptus gracilis</i>	7	<i>Eucalyptus phenax</i>	11
	<i>Eucalyptus yalataensis</i>	6	<i>Eucalyptus phenax</i>	6	<i>Eucalyptus incrassata</i>	3
	<i>Eucalyptus socialis</i>	5	<i>Cassytha</i> sp.	5	<i>Eucalyptus brachycalyx</i>	2
	<i>Eucalyptus diversifolia</i>	3	<i>Myoporum platycarpum</i>	5	<i>Eucalyptus leptophylla</i>	1
	<i>Eucalyptus leptophylla</i>	1	<i>Eucalyptus leptophylla</i>	4	<i>Melaleuca uncinata</i>	1
	<i>Eucalyptus</i> sp.	1	<i>Melaleuca uncinata</i>	3	<i>Santalum spicatum</i>	1

S	LOC7 (n = 87)	Count	SR159 (n = 96)	Count	SR260 (n = 97)	Count
			<i>Acacia lanceolata</i>	1		
			<i>Acacia oswaldii</i>	1		
			<i>Callitris verrucosa</i>	1		
			<i>Eucalyptus calycogona</i>	1		
			<i>Eucalyptus</i> sp.	1		
			<i>Melaleuca lanceolata</i>	1		
			Raised litter	1		
			<i>Santalum acuminatum</i>	1		

5.7.4 Presence/absence of plant taxa

Table S 5-4 Top 30% most common genera and species recorded as present at high use day sites (i.e. number of sites found at) and their percent occurrence at high, low and control sites. S = Stratum, G = Ground cover, M = Midstorey, C = Canopy

	S	Genus	High	Low	Control	Species	High	Low	Control
LOC7	G	Raised	19	16 (84%)	11	Raised litter	19	16 (84%)	11
		<i>Triodia</i>	16 (84%)	18 (95%)	11	<i>Triodia</i> sp.	16 (84%)	18 (95%)	11
		<i>Melaleuca</i>	16 (84%)	17 (89%)	10 (91%)	<i>Melaleuca lanceolata</i>	16 (84%)	17 (89%)	10 (91%)
		<i>Rhagodia</i>	12 (63%)	7 (37%)	6 (55%)	<i>Rhagodia crassifolia</i>	12 (63%)	7 (37%)	6 (55%)
						<i>Microcybe pauciflora</i>	11 (58%)	4 (21%)	3 (27%)
	M					<i>Acrotiche patula</i>	5 (26%)	7 (37%)	3 (27%)
						<i>Melaleuca lanceolata</i>	16 (84%)	18 (95%)	10 (91%)
						<i>Eucalyptus phenax</i>	7 (37%)	3 (16%)	4 (36%)
						<i>Eucalyptus calcareana</i>	5 (26%)	8 (42%)	2 (18%)
						<i>Eucalyptus</i> sp.	5 (26%)	2 (11%)	3 (27%)
C					<i>Eucalyptus gracilis</i>	5 (26%)	2 (11%)	0	
					<i>Eucalyptus phenax</i>	10 (53%)	2 (11%)	3 (27%)	
SR 159	G	Raised	16 (76%)	19 (90%)	11 (92%)	Raised litter	16 (76%)	19 (90%)	11 (92%)
		<i>Alyxia</i>	10 (48%)	8 (38%)	4 (33%)	<i>Alyxia buxifolia</i>	10 (48%)	8 (38%)	4 (33%)
		<i>Roepera</i>	9 (43%)	8 (38%)	2 (17%)	<i>Triodia</i> sp.	8 (38%)	6 (29%)	3 (25%)
		<i>Maireana</i>	9 (43%)	7 (33%)	3 (25%)	<i>Olearia muelleri</i>	7 (33%)	8 (38%)	5 (42%)
		<i>Dodonea</i>	9 (43%)	5 (24%)	6 (50%)	<i>Roepera apiculata</i>	7 (33%)	3 (14%)	1 (8%)
		<i>Olearia</i>	8 (38%)	11 (52%)	5 (42%)	<i>Roepera aurantiaca</i>	6 (29%)	7 (33%)	0
		<i>Triodia</i>	8 (38%)	6 (29%)	3 (25%)	<i>Beyeria lechenaultii</i>	6 (29%)	6 (29%)	1 (8%)
		<i>Eucalyptus</i>	8 (38%)	3 (14%)	5 (42%)	<i>Maireana pentatropis</i>	6 (29%)	3 (14%)	1 (8%)
		<i>Rhagodia</i>	6 (29%)	11 (52%)	6 (50%)	<i>Senna artemisioides ssp</i>	4 (19%)	10 (48%)	3 (25%)
		<i>Beyeria</i>	6 (29%)	6 (29%)	1 (8%)	<i>Lomandra effusa</i>	4 (19%)	4 (19%)	1 (8%)
		<i>Senna</i>	5 (24%)	10 (48%)	4 (33%)	<i>Dodonea stenozyga</i>	4 (19%)	3 (14%)	1 (8%)
						<i>Maireana erioclada</i>	4 (19%)	2 (10%)	1 (8%)
						<i>Eucalyptus brachycalyx</i>	4 (19%)	1 (5%)	1 (8%)
						<i>Atriplex vesicaria</i>	3 (14%)	6 (29%)	4 (33%)
						<i>Dodonea hexandra</i>	3 (14%)	1 (5%)	1 (8%)
SR 260	M	<i>Eucalyptus</i>	18 (86%)	12 (57%)	5 (42%)	<i>Geijera linearifolia</i>	6 (29%)	10 (48%)	3 (25%)
		<i>Geijera</i>	6 (29%)	10 (48%)	3 (25%)	<i>Eucalyptus brachycalyx</i>	6 (29%)	5 (24%)	3 (25%)
		Raised	5 (24%)	8 (38%)	5 (42%)	Raised litter	5 (24%)	8 (38%)	5 (42%)
	C					<i>Eucalyptus incrassata</i>	5 (24%)	3 (14%)	1 (8%)
						<i>Eucalyptus oleosa</i>	4 (19%)	2 (10%)	
						<i>Eucalyptus brachycalyx</i>	10 (48%)	5 (24%)	5 (42%)
						<i>Eucalyptus oleosa</i>	8 (38%)	7 (33%)	2 (17%)
				<i>Cassyltha</i> sp.	3 (14%)	2 (10%)	0		
SR 260	G	Raised	19 (90%)	21	10 (83%)	Raised litter	19 (90%)	21	10 (83%)
		<i>Rhagodia</i>	17 (81%)	17 (81%)	7 (58%)	<i>Rhagodia crassifolia</i>	14 (67%)	12 (57%)	6 (50%)
		<i>Eremophila</i>	13 (62%)	12 (57%)	4 (33%)	<i>Eremophila scoparia</i>	13 (62%)	11 (52%)	4 (33%)
		<i>Grevillea</i>	10 (48%)	10 (48%)	4 (33%)	<i>Grevillea huegelii</i>	10 (48%)	10 (48%)	4 (33%)
		<i>Olearia</i>	10 (48%)	7 (33%)	0	<i>Westringia rigida</i>	8 (38%)	6 (29%)	2 (17%)
		<i>Acacia</i>	8 (38%)	3 (14%)	3 (25%)	<i>Acacia sclerophylla</i>	8 (38%)	2 (10%)	3 (25%)
						<i>Eucalyptus</i> sp.	7 (33%)	6 (29%)	3 (25%)
				<i>Atriplex stipitata</i>	6 (29%)	3 (14%)	0		

	S	Genus	High	Low	Control	Species	High	Low	Control
SR 260						<i>Melaleuca pauperiflora</i>	5 (24%)	6 (29%)	2 (17%)
						<i>Beyeria lechenaultii</i>	5 (24%)	1 (5%)	0
	M	<i>Eucalyptus</i>	13 (62%)	13 (62%)	7 (58%)	<i>Eremophila scoparia</i>	13 (62%)	7 (33%)	1 (8%)
		<i>Eremophila</i>	13 (62%)	8 (38%)	1 (8%)	<i>Eucalyptus</i> sp.	9 (43%)	8 (38%)	2 (17%)
		<i>Raised</i>	9 (43%)	7 (33%)	0	Raised litter	9 (43%)	7 (33%)	0
						<i>Melaleuca pauperiflora</i>	6 (29%)	6 (29%)	5 (42%)
	C	<i>Eucalyptus</i>	20 (95%)	21	11 (92%)	<i>Eucalyptus socialis</i>	12 (57%)	10 (48%)	3 (25%)

5.7.5 Explanatory variables

Table S 5-5 Summary statistics of explanatory variables (Structure, diversity and visits). We present the percentage cover mean with 1 standard deviation (mean), the median (med), the interquartile range (IQR) and the percentage cover range (range). Rows with *All* visits show statistics of the complete dataset with all visits taken into account. Rows with *High/Low/Ctrl* visits show the statistics of the reduced subset of the data with high (top 25% visited) and low use (bottom 25% visited) sites as well as controls (not visited) which were used for the decision tree models. V = Visits, A = All, H/L = High/Low use, C = Control, G = Ground cover, M = Midstorey, C = Canopy

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
Canopy height	A	5.4 ± 2.1	5.5	2.5	0 - 12	5.3 ± 1.8	5.5	1.5	0 - 12	5.7 ± 2.7	5.7	4	0 - 10	5.3 ± 1.9	5	2	2.5 - 12
	H	5.5 ± 2	5.5	2	0 - 12	6.7 ± 1.7	6.5	1.8	4.5 - 12	4.7 ± 2.6	5	3.5	0 - 8	5.2 ± 1.1	5	1.5	3.5 - 7
	L	5.3 ± 2.4	5.5	2.7	0 - 11	4.3 ± 1.9	5	2.2	0 - 6	5.8 ± 2.8	5.5	4	0 - 10	5.7 ± 2.2	5	2.5	3.5 - 11
	C	4.9 ± 2.3	5	2.2	0 - 10	5.3 ± 1	5.5	1.2	4 - 7	5 ± 3.8	5	4.4	0 - 10	4.4 ± 1.1	4.5	1.5	3 - 6.5
Canopy start	A	2.9 ± 1.4	2.5	1.5	0 - 10	3 ± 1.4	2.8	1.5	0 - 10	2.8 ± 1.6	2.5	2.4	0 - 7	3 ± 1	3	1	1.5 - 7
	H	3.1 ± 1.5	3	1.5	0 - 10	4 ± 1.7	4	1.5	2 - 10	2.3 ± 1.5	2.5	1.5	0 - 5	3.1 ± 0.8	3	1	2 - 5
	L	2.7 ± 1.5	2.5	1.9	0 - 7	2.3 ± 1.3	2	1.5	0 - 4.5	2.6 ± 1.7	2.5	2	0 - 6	3.1 ± 1.4	3	2	1.5 - 7
	C	2.6 ± 1.2	2.5	1	0 - 5	2.9 ± 0.9	2.7	1	2 - 5	2 ± 1.5	2.2	1.9	0 - 4.5	3 ± 1	2.5	0.6	2 - 5
Leaf Density	A	29.3 ± 10.7	30	10	0 - 65	25.8 ± 9.9	25	15	0 - 50	30.7 ± 13.2	32	13	0 - 65	31.1 ± 7.7	30	10	15 - 55
	H	28.4 ± 11.3	30	10	0 - 55	25.3 ± 8.4	25	12.5	10 - 40	28.7 ± 15.7	35	18	0 - 55	31 ± 7.6	30	10	15 - 50
	L	28.6 ± 11.3	30	10	0 - 55	23 ± 12.4	25	15	0 - 50	29.6 ± 11	30	5	0 - 45	32.5 ± 8.7	32	7	15 - 55
	C	26.2 ± 11.1	25	12.5	0 - 45	26.4 ± 7.1	25	7.5	15 - 40	25.3 ± 17	26	23.5	0 - 45	26.9 ± 6.7	28	6.8	15 - 37
Leaf litter	A	41.8 ± 22.1	41.5	30.8	1 - 98	40.4 ± 20.4	40	27	3 - 85	44.2 ± 23.5	43.5	33.5	4 - 95	40.6 ± 22.3	42	28	1 - 98
	H	45.6 ± 25.8	45	42	5 - 98	47.3 ± 22.3	45	34	15 - 85	51.5 ± 26.1	57	40	5 - 95	38.2 ± 27.6	32	40	6 - 98
	L	37.7 ± 23.1	40	35	3 - 85	37.2 ± 23.7	40	37.5	3 - 75	34 ± 23.4	28	36	4 - 85	41.9 ± 22.8	45	20	6 - 80
	C	38.1 ± 23.5	35	32	1 - 90	40 ± 18.6	35	22	10 - 75	42.2 ± 30.6	42.5	52.5	4 - 90	32.2 ± 19.9	30.5	21.2	1 - 73
Ground Cover	A	11.1 ± 8.8	9	8	1 - 45	13.6 ± 10.2	10	8	2.9 - 45	10.2 ± 7.3	8	8	1 - 40	9.7 ± 8.2	8	6	1 - 45
	H	9.5 ± 7	8	7	1 - 45	9.6 ± 4.5	9	6	2.9 - 20	8.3 ± 6.3	7.8	7	1 - 30	10.5 ± 9.3	9	6	2 - 45
	L	13.5 ± 11.3	9	10	1.5 - 45	19.3 ± 14.4	11	25	3 - 45	13 ± 8.5	11.5	8	3 - 40	8.8 ± 8.3	7	4.5	1.5 - 40
	C	15.2 ± 11.8	12	15.8	1 - 45	15.7 ± 11.9	9	12	4 - 40	15 ± 10.3	15.5	18.1	3.5 - 30	15 ± 14	11	16.8	1 - 45
Midstorey Cover	A	8.2 ± 7.7	6	7	0 - 60	10.4 ± 6.4	9	5	1 - 33	8.5 ± 10.2	5	6	0 - 60	5.8 ± 4.8	5	6	0 - 22
	H	8.3 ± 8.6	7	7	0 - 60	9.9 ± 5.5	9	4.5	1.5 - 25	8.8 ± 12.8	5	8	1 - 60	6.5 ± 5.2	5	6	0 - 22
	L	8.5 ± 8	5	8	0 - 40	12.2 ± 7.2	11	8.5	1 - 25	8 ± 9.7	5	4.5	0 - 40	5.8 ± 5.5	5	4	0 - 22
	C	8.5 ± 7.7	7	8	0 - 40	10.1 ± 4.4	8	4.5	5 - 20	8.8 ± 11.2	4.5	5.2	0 - 40	6.8 ± 6	5.5	7.2	0 - 20
Canopy Cover	A	17.1 ± 13.5	15	19	0 - 60	14.1 ± 9.1	12	13	0 - 40	18.2 ± 15.9	15	23.2	0 - 60	18.9 ± 14	17	18	0 - 55
	H	17.4 ± 14	16	19	0 - 60	17.3 ± 7.5	17	13.5	5 - 30	17.7 ± 18.9	10	25.5	0 - 60	17.1 ± 13.3	16	19	0 - 50
	L	17.1 ± 14.4	13	20	0 - 50	15.3 ± 12.2	13	17	0 - 40	16 ± 16.1	10	21	0 - 50	19.8 ± 14.8	18	19	0 - 50
	C	16.8 ± 16	10	19.5	0 - 55	14.2 ± 8.6	10	12	5 - 30	20.7 ± 21.4	14	36	0 - 55	15.4 ± 15.4	10.5	11.2	0 - 50
Total cover (G+M)	A	19.3 ± 12.3	16	12	2.5 - 74.8	24 ± 13	20	15	7 - 65	18.7 ± 12.8	14	13.8	2.5 - 74.8	15.5 ± 9.5	14	9	3 - 60
	H	17.8 ± 12.3	16	10	2.5 - 74.8	19.5 ± 7.3	18	10	7 - 37	17.1 ± 16	13	13	2.5 - 74.8	17 ± 12	16	9	3.5 - 60
	L	22 ± 14.9	18	14	3 - 65	31.4 ± 17.5	26	27.1	11 - 65	20.9 ± 12.3	16.1	12.5	5.5 - 50	14.6 ± 9.6	12	9	3 - 46
	C	23.8 ± 13.6	21	18.2	4 - 51	25.8 ± 13.9	20	19	10 - 50	23.8 ± 13.7	20.5	20	5.5 - 45	21.8 ± 14.1	21.5	17.2	4 - 51
Total cover (G+M+C)	A	36.4 ± 16.7	34	20.9	4.5 - 100	38.2 ± 16.3	34	18	12 - 94	36.9 ± 18.5	35	25.4	4.5 - 100	34.3 ± 15	31.8	18	6 - 85
	H	35.2 ± 16.1	32	19.5	4.5 - 85	36.7 ± 9.9	34.9	11.2	24 - 62	34.9 ± 19.9	35	27	4.5 - 76.8	34.1 ± 17.2	31	16	6 - 85
	L	39.1 ± 20.5	33	33.5	10 - 94	46.7 ± 24.7	36	39	16 - 94	37 ± 18.8	32	27	14 - 74	34.4 ± 16.5	34.5	25	10 - 66
	C	40.6 ± 20.8	37	25.5	11.5 - 100	40 ± 14.1	38	11.5	22 - 75	44.5 ± 28.9	35.8	37.9	11.5 - 100	37.2 ± 16.9	36.5	28.8	12 - 60
Ground Richness	A	6.1 ± 2	6	2	2 - 13	5.7 ± 1.4	6	2	3 - 10	6.9 ± 2.4	7	3	3 - 13	5.8 ± 1.8	6	2	2 - 12
	H	6.2 ± 2.2	6	3	2 - 13	5.7 ± 1.6	5	1	4 - 10	6.9 ± 2.9	7	5	3 - 13	6 ± 1.7	6	2	2 - 9
	L	6.6 ± 2.2	6	3	3 - 12	5.6 ± 1.2	6	1	3 - 8	8 ± 2.3	8	3	4 - 12	6.1 ± 2.1	6	1	3 - 12
	C	5.9 ± 1.6	6	2	2 - 10	6 ± 0.9	6	2	5 - 7	6.9 ± 1.6	7	2	4 - 10	4.8 ± 1.4	5.5	2	2 - 6
Midstorey Richness	A	2.4 ± 1.1	2	1	0 - 6	2.3 ± 0.8	2	1	1 - 4	2.5 ± 1.3	2	1	0 - 6	2.4 ± 1.1	2	1	0 - 5
	H	2.4 ± 1	2	1	0 - 5	2.4 ± 0.9	2	1	1 - 4	2.2 ± 1.1	2	2	1 - 4	2.6 ± 1.1	3	1	0 - 5
	L	2.5 ± 1.1	2	1	0 - 5	2.1 ± 0.5	2	0	1 - 3	2.7 ± 1.3	3	1	0 - 5	2.8 ± 1.2	3	2	0 - 5
	C	2.3 ± 1.2	2	1	0 - 6	2.5 ± 0.9	2	1	1 - 4	2.6 ± 1.6	2	1.2	0 - 6	2 ± 1.1	2	0.2	0 - 4

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
Canopy Richness	A	1±0.3	1	0	0-3	1±0.2	1	0	0-1	1.1±0.5	1	0	0-3	1±0.2	1	0	0-2
	H	1±0.4	1	0	0-2	1±0	1	0	1-1	1±0.6	1	0	0-2	0.9±0.3	1	0	0-1
	L	1±0.4	1	0	0-3	0.9±0.3	1	0	0-1	1.1±0.7	1	0	0-3	1±0.2	1	0	0-1
	C	0.9±0.4	1	0	0-2	1±0	1	0	1-1	0.8±0.6	1	0.2	0-2	1±0.4	1	0	0-2
Total richness (G+M)	A	8.5±2.4	8	3	4-19	8±1.5	8	2	5-12	9.4±3	9	4	4-19	8.2±2.2	8	2	4-15
	H	8.6±2.5	8	3	4-15	8.1±1.9	7	2	5-12	9±3.2	9	5	4-15	8.5±2.2	9	3	4-12
	L	9.1±2.7	8	3	5-16	7.6±1.2	8	1	6-10	10.7±3.1	10	5	6-16	8.9±2.6	9	3	5-15
	C	8.3±2.2	8	2.5	4-14	8.5±1.5	8	2	6-11	9.5±2.5	9	2.2	6-14	6.8±1.7	7	2.2	4-10
Total richness (G+M+C)	A	9.5±2.4	9	3	4-20	9±1.6	9	2	6-13	10.4±3	10	3.2	5-20	9.2±2.2	9	2	4-16
	H	9.5±2.6	9	3	4-16	9.1±1.9	8	2	6-13	10.1±3.3	10	5	5-16	9.4±2.3	9	3	4-13
	L	10.1±2.7	10	3	6-17	8.5±1.3	9	1.5	6-11	11.8±2.7	11	4	8-17	9.9±2.7	10	3	6-16
	C	9.2±2.2	9	3	5-15	9.5±1.5	9	2	7-12	10.3±2.5	10	2.2	7-15	7.8±1.7	8	3	5-11
Ground Diversity	A	1.4±0.4	1.4	0.4	0.2-2.2	1.4±0.3	1.4	0.4	0.9-2	1.3±0.4	1.4	0.5	0.2-2.2	1.3±0.4	1.4	0.5	0.3-2
	H	1.4±0.4	1.4	0.4	0.2-2.2	1.4±0.3	1.4	0.3	0.9-2	1.3±0.5	1.4	0.7	0.2-2.2	1.4±0.4	1.4	0.5	0.3-2
	L	1.4±0.3	1.4	0.5	0.7-2.1	1.3±0.3	1.3	0.5	0.9-1.8	1.4±0.4	1.6	0.6	0.7-2.1	1.4±0.3	1.4	0.4	0.8-2
	C	1.3±0.4	1.3	0.4	0.5-2	1.4±0.3	1.5	0.5	1.1-1.8	1.3±0.4	1.3	0.4	0.7-2	1.1±0.4	1.3	0.8	0.5-1.6
Midstorey Diversity	A	0.6±0.4	0.6	0.6	0-1.6	0.6±0.3	0.6	0.5	0-1.4	0.6±0.5	0.6	0.8	0-1.6	0.7±0.4	0.7	0.7	0-1.6
	H	0.6±0.4	0.6	0.5	0-1.6	0.6±0.3	0.6	0.4	0-1.3	0.5±0.5	0.6	0.8	0-1.3	0.7±0.4	0.8	0.5	0-1.6
	L	0.6±0.4	0.7	0.6	0-1.4	0.5±0.3	0.7	0.4	0-1	0.6±0.5	0.6	0.8	0-1.4	0.8±0.4	0.9	0.4	0-1.4
	C	0.6±0.4	0.6	0.5	0-1.5	0.6±0.4	0.6	0.6	0-1.4	0.7±0.5	0.7	0.5	0-1.5	0.5±0.5	0.3	0.5	0-1.4
Canopy Diversity	A	0±0.1	0	0	0-0.9	0±0	0	0	0-0	0.1±0.2	0	0	0-0.9	0±0	0	0	0-0.2
	H	0±0.1	0	0	0-0.7	0±0	0	0	0-0	0.1±0.2	0	0	0-0.7	0±0	0	0	0-0
	L	0±0.2	0	0	0-0.9	0±0	0	0	0-0	0.1±0.3	0	0	0-0.9	0±0	0	0	0-0
	C	0±0.1	0	0	0-0.7	0±0	0	0	0-0	0.1±0.2	0	0	0-0.7	0±0.1	0	0	0-0.2
Total diversity (G+M)	A	1.6±0.4	1.6	0.4	0.5-2.4	1.7±0.3	1.7	0.3	0.9-2.2	1.6±0.4	1.6	0.6	0.6-2.4	1.6±0.4	1.7	0.5	0.5-2.3
	H	1.7±0.4	1.7	0.4	0.6-2.3	1.6±0.3	1.7	0.3	0.9-2.2	1.6±0.4	1.6	0.6	0.6-2.3	1.8±0.4	1.7	0.5	1-2.3
	L	1.7±0.4	1.6	0.5	0.7-2.4	1.6±0.2	1.5	0.3	1.1-2.1	1.7±0.5	1.7	0.6	0.7-2.4	1.7±0.3	1.7	0.4	1.1-2.3
	C	1.6±0.4	1.6	0.5	0.5-2.3	1.7±0.3	1.7	0.4	1.4-2.1	1.5±0.4	1.5	0.6	0.8-2.3	1.4±0.5	1.5	0.7	0.5-2
Total diversity (G+M+C)	A	1.4±0.4	1.5	0.6	0.4-2.4	1.6±0.3	1.7	0.3	0.7-2.1	1.4±0.4	1.3	0.6	0.4-2.4	1.3±0.4	1.3	0.6	0.4-2.2
	H	1.4±0.4	1.4	0.6	0.4-2.4	1.5±0.3	1.5	0.4	1-2.1	1.3±0.5	1.3	0.7	0.4-2.4	1.5±0.5	1.6	0.6	0.4-2.2
	L	1.5±0.4	1.6	0.6	0.5-2.4	1.6±0.2	1.7	0.2	1.1-2	1.5±0.5	1.6	0.8	0.8-2.4	1.3±0.4	1.4	0.6	0.5-2
	C	1.4±0.4	1.6	0.6	0.7-2.1	1.7±0.3	1.7	0.2	0.9-2.1	1.4±0.4	1.4	0.5	0.8-2	1.3±0.4	1.2	0.5	0.7-1.8
Days Visited	A	4.2±4	3	5	0-22	3.8±4.5	2	4	0-22	4±3.3	3	5	0-16	4.9±4.1	4	5	0-19
	H	10.3±3.7	9	3	6-22	10.8±4.9	10	6	6-22	9±2.3	8	3	7-16	11±3.2	10	3	8-19
	L	1.2±0.5	1	0	1-3	1±0	1	0	1-1	1.2±0.4	1	0	1-2	1.5±0.6	1	1	1-3
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
PropDays Visited	A	1.4±1.6	1	1.5	0-11.3	1.9±2.3	1	2.1	0-11.3	1.1±0.9	0.8	1.4	0-4.4	1.2±1	1	1.2	0-4.7
	H	3.5±2	2.8	1.4	1.9-11.3	5.6±2.5	5.1	3.1	3.1-11.3	2.5±0.6	2.2	0.8	1.9-4.4	2.7±0.8	2.5	0.7	2-4.7
	L	0.4±0.1	0.5	0.2	0.2-0.7	0.5±0	0.5	0	0.5-0.5	0.3±0.1	0.3	0	0.3-0.6	0.4±0.1	0.2	0.2	0.2-0.7
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Visits	A	4.7±4.4	4	6	0-23	4.1±4.7	3	4	0-23	4.6±4	4	6	0-16	5.2±4.5	5	5	0-21
	H	11.2±3.9	10	5	6-23	11.5±5	10	7	6-23	10.4±2.7	10	5	7-16	11.8±3.9	10	4	8-21
	L	1.4±0.7	1	1	1-4	1.2±0.6	1	0	1-3	1.3±0.8	1	0	1-4	1.6±0.7	2	1	1-3
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
PropVisits	A	0.3±0.3	0.2	0.3	0-2.2	0.4±0.5	0.3	0.4	0-2.2	0.3±0.2	0.2	0.3	0-0.9	0.3±0.2	0.2	0.2	0-1
	H	0.7±0.4	0.6	0.4	0.4-2.2	1.1±0.5	1	0.7	0.6-2.2	0.6±0.2	0.6	0.3	0.4-0.9	0.6±0.2	0.5	0.2	0.4-1
	L	0.1±0	0.1	0	0-0.3	0.1±0.1	0.1	0	0.1-0.3	0.1±0	0.1	0	0.1-0.2	0.1±0	0.1	0	0-0.1
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0

Table S 5-6 Summary statistics of explanatory variables (Ground cover). We present the percentage cover mean with 1 standard deviation (mean), the median (med), the interquartile range (IQR) and the percentage cover range (range). Rows with *All* visits show statistics of the complete dataset with all visits taken into account. Rows with *High/Low/Ctrl* visits show the statistics of the reduced subset of the data with high (top 25% visited) and low use (bottom 25% visited) sites as well as controls (not visited) which were used for the decision tree models. V = Visits, A = All, H/L = High/Low use, C = Control, G = Ground cover, M = Midstorey, C = Canopy

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
<i>Acacia</i>	A	0.5 ± 2.5	0	0	0 - 30.8	0.1 ± 0.3	0	0	0 - 1.8	0.1 ± 0.5	0	0	0 - 3.1	1.2 ± 4.2	0	0.4	0 - 30.8
	H	0.5 ± 2.9	0	0	0 - 22.5	0.1 ± 0.2	0	0	0 - 0.8	0.1 ± 0.4	0	0	0 - 1.5	1.4 ± 4.9	0	0.6	0 - 22.5
	L	0.7 ± 4	0	0	0 - 30.8	0.1 ± 0.2	0	0	0 - 0.9	0.2 ± 0.4	0	0	0 - 1.8	1.8 ± 6.8	0	0	0 - 30.8
	C	0.4 ± 1.5	0	0	0 - 8.5	0.1 ± 0.4	0	0	0 - 1.2	0.1 ± 0.4	0	0	0 - 1.4	1 ± 2.5	0	0.2	0 - 8.5
<i>Acrotriche</i>	A	0.1 ± 0.7	0	0	0 - 5.6	0.4 ± 1.1	0	0.2	0 - 5.6	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	H	0 ± 0.1	0	0	0 - 1	0.1 ± 0.2	0	0	0 - 1	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0.3 ± 0.8	0	0	0 - 4	0.8 ± 1.4	0	0.9	0 - 4	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0.2 ± 0.8	0	0	0 - 4.4	0.6 ± 1.4	0	0.2	0 - 4.4	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Actinoble</i>	A	0 ± 0	0	0	0 - 0.2	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.2	0 ± 0	0	0	0 - 0
	H	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0.2	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.2	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Alyxia</i>	A	0.2 ± 0.5	0	0	0 - 4.5	0 ± 0	0	0	0 - 0	0.4 ± 0.8	0	0.4	0 - 4.5	0 ± 0.2	0	0	0 - 1.8
	H	0.2 ± 0.7	0	0	0 - 4.5	0 ± 0	0	0	0 - 0	0.6 ± 1.1	0	0.6	0 - 4.5	0 ± 0.2	0	0	0 - 0.7
	L	0.1 ± 0.4	0	0	0 - 1.8	0 ± 0	0	0	0 - 0	0.3 ± 0.5	0	0.4	0 - 1.8	0.1 ± 0.4	0	0	0 - 1.8
	C	0.1 ± 0.3	0	0	0 - 1.2	0 ± 0	0	0	0 - 0	0.3 ± 0.5	0	0.4	0 - 1.2	0 ± 0.1	0	0	0 - 0.2
<i>Atriplex</i>	A	0.2 ± 0.7	0	0	0 - 4.8	0 ± 0	0	0	0 - 0	0.3 ± 0.8	0	0.1	0 - 4.8	0.4 ± 0.8	0	0.5	0 - 3.6
	H	0.2 ± 0.6	0	0	0 - 3.2	0 ± 0	0	0	0 - 0	0.2 ± 0.7	0	0	0 - 3.2	0.3 ± 0.6	0	0.5	0 - 2.2
	L	0.3 ± 0.8	0	0.1	0 - 4.8	0 ± 0	0	0	0 - 0	0.6 ± 1.1	0	0.6	0 - 4.8	0.3 ± 0.8	0	0.3	0 - 3.6
	C	0.4 ± 0.9	0	0.3	0 - 3.5	0 ± 0	0	0	0 - 0	0.5 ± 0.8	0	0.7	0 - 2.5	0.8 ± 1.3	0.2	0.6	0 - 3.5
<i>Beyeria</i>	A	0.4 ± 1.4	0	0	0 - 11.2	0.5 ± 1.6	0	0	0 - 11.2	0.5 ± 1.5	0	0	0 - 9	0.2 ± 1	0	0	0 - 7.8
	H	0.3 ± 0.9	0	0	0 - 4.5	0 ± 0	0	0	0 - 0	0.7 ± 1.4	0	0.3	0 - 4.5	0.2 ± 0.6	0	0	0 - 2.2
	L	0.7 ± 2.1	0	0	0 - 11.2	0.9 ± 2.6	0	0	0 - 11.2	0.9 ± 2	0	1	0 - 8.5	0.4 ± 1.7	0	0	0 - 7.8
	C	0.2 ± 0.8	0	0	0 - 3.1	0.4 ± 1	0	0	0 - 3.1	0.2 ± 0.9	0	0	0 - 3	0 ± 0	0	0	0 - 0
<i>Billardiera</i>	A	0 ± 0.3	0	0	0 - 4.5	0 ± 0.5	0	0	0 - 4.5	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0.1 ± 0.6	0	0	0 - 4.5	0.2 ± 1	0	0	0 - 4.5	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Blennospora</i>	A	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0
	H	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Callitris</i>	A	0 ± 0.2	0	0	0 - 3.8	0 ± 0.2	0	0	0 - 1.7	0 ± 0.4	0	0	0 - 3.8	0 ± 0	0	0	0 - 0
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0.1 ± 0.5	0	0	0 - 3.8	0 ± 0	0	0	0 - 0	0.2 ± 0.8	0	0	0 - 3.8	0 ± 0	0	0	0 - 0
	C	0 ± 0.3	0	0	0 - 1.7	0.2 ± 0.5	0	0	0 - 1.7	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Calytrix</i>	A	0 ± 0.1	0	0	0 - 1.8	0 ± 0	0	0	0 - 0	0 ± 0.2	0	0	0 - 1.8	0 ± 0	0	0	0 - 0.3
	H	0 ± 0.1	0	0	0 - 0.6	0 ± 0	0	0	0 - 0	0 ± 0.2	0	0	0 - 0.6	0 ± 0	0	0	0 - 0
	L	0 ± 0.2	0	0	0 - 1.8	0 ± 0	0	0	0 - 0	0.1 ± 0.4	0	0	0 - 1.8	0 ± 0.1	0	0	0 - 0.3
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Carpobrotus</i>	A	0 ± 0	0	0	0 - 0.6	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.1	0 ± 0.1	0	0	0 - 0.6
	H	0 ± 0.1	0	0	0 - 0.6	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.1	0 ± 0.2	0	0	0 - 0.6
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
<i>Cassytha</i>	A	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	H	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
<i>Cratystylis</i>	A	0.4±1.7	0	0	0-15	0±0	0	0	0-0	0.6±2.2	0	0	0-15	0.6±1.8	0	0	0-11.1
	H	0.2±1	0	0	0-7.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0.5±1.6	0	0	0-7.2
	L	0.8±2.4	0	0	0-15	0±0	0	0	0-0	1.4±3.4	0	0.8	0-15	1±2.1	0	0	0-6.8
	C	0.7±2.4	0	0	0-13.6	0±0	0	0	0-0	1.9±4	0	1.8	0-13.6	0.1±0.4	0	0	0-1
<i>Daisy</i>	A	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
<i>Dianella</i>	A	0±0.2	0	0	0-2	0±0.1	0	0	0-0.8	0.1±0.3	0	0	0-2	0±0.2	0	0	0-1.6
	H	0±0	0	0	0-0.2	0±0	0	0	0-0.1	0±0	0	0	0-0.2	0±0	0	0	0-0
	L	0±0.1	0	0	0-0.6	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0.1	0	0	0-0.6
	C	0±0.2	0	0	0-0.9	0±0	0	0	0-0	0.1±0.3	0	0	0-0.9	0±0.2	0	0	0-0.6
<i>Dissocarpus</i>	A	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0.1	0	0	0-0.2	0±0	0	0	0-0
	C	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
<i>Dodonea</i>	A	0.3±1.1	0	0	0-14.9	0.2±0.6	0	0	0-3.8	0.6±1.7	0	0.5	0-14.9	0±0	0	0	0-0.1
	H	0.3±0.8	0	0	0-4	0.1±0.5	0	0	0-2	0.7±1.2	0	0.6	0-4	0±0	0	0	0-0.1
	L	0.2±0.5	0	0	0-3	0.3±0.7	0	0.1	0-2	0.2±0.7	0	0	0-3	0±0	0	0	0-0
	C	0.6±2.6	0	0	0-14.9	0±0	0	0	0-0	1.7±4.3	0.1	0.9	0-14.9	0±0	0	0	0-0
<i>Enchylaena</i>	A	0±0.1	0	0	0-1.5	0±0	0	0	0-0	0±0.2	0	0	0-1.5	0±0	0	0	0-0.3
	H	0±0.1	0	0	0-0.5	0±0	0	0	0-0	0±0.1	0	0	0-0.5	0±0.1	0	0	0-0.3
	L	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0.1	0	0	0-0.4	0±0	0	0	0-0
	C	0.1±0.3	0	0	0-1.5	0±0	0	0	0-0	0.2±0.5	0	0	0-1.5	0±0	0	0	0-0
<i>Eremophila</i>	A	0.4±1.1	0	0.2	0-6.4	0.3±0.7	0	0.1	0-4.8	0.1±0.5	0	0	0-3	0.9±1.6	0.1	1.2	0-6.4
	H	0.8±1.4	0	1.2	0-6	0.9±1.3	0	1.6	0-4.8	0.1±0.2	0	0	0-1	1.4±1.8	0.9	2.2	0-6
	L	0.4±1.2	0	0.2	0-6.3	0.2±0.6	0	0	0-2.2	0±0.1	0	0	0-0.4	1.1±1.8	0.2	1.4	0-6.3
	C	0.4±1.1	0	0	0-5	0.1±0.1	0	0	0-0.4	0.5±1.2	0	0	0-3	0.7±1.5	0	0.7	0-5
<i>Eriochiton</i>	A	0±0	0	0	0-0.3	0±0	0	0	0-0	0±0	0	0	0-0.3	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0.3	0±0	0	0	0-0	0±0.1	0	0	0-0.3	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
<i>Eucalyptus</i>	A	0.5±1.1	0	0.4	0-8.4	0.4±0.9	0	0.5	0-5.2	0.5±1.2	0	0.4	0-7.8	0.4±1.3	0	0.2	0-8.4
	H	0.4±1.1	0	0.3	0-7.2	0.3±0.8	0	0.2	0-3.6	0.4±0.7	0	0.4	0-2.2	0.6±1.6	0	0.2	0-7.2
	L	0.5±1.4	0	0.2	0-7.8	0.4±0.8	0	0	0-3	0.6±1.9	0	0	0-7.8	0.5±1.3	0	0.3	0-6
	C	0.5±1.1	0	0.6	0-5.2	0.7±1.5	0.1	0.6	0-5.2	0.8±1	0	1.6	0-2.5	0.1±0.3	0	0.1	0-1.2
<i>Eutaxia</i>	A	0±0.1	0	0	0-2	0±0	0	0	0-0.4	0±0.2	0	0	0-2	0±0	0	0	0-0
	H	0±0.1	0	0	0-0.4	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
<i>Exocarpos</i>	A	0±0.5	0	0	0-7.2	0±0	0	0	0-0	0.1±0.8	0	0	0-7.2	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0.2±1	0	0	0-7.2	0±0	0	0	0-0	0.4±1.6	0	0	0-7.2	0±0	0	0	0-0
	C	0±0.2	0	0	0-1	0±0	0	0	0-0	0.1±0.3	0	0	0-1	0±0	0	0	0-0
<i>Geijera</i>	A	0±0.2	0	0	0-2.1	0±0	0	0	0-0	0.1±0.3	0	0	0-2.1	0±0.1	0	0	0-0.3
	H	0±0.1	0	0	0-0.7	0±0	0	0	0-0	0.1±0.2	0	0	0-0.7	0±0	0	0	0-0
	L	0.1±0.3	0	0	0-1.4	0±0	0	0	0-0	0.2±0.4	0	0.2	0-1.4	0±0.1	0	0	0-0.3
	C	0.1±0.4	0	0	0-2.1	0±0	0	0	0-0	0.2±0.6	0	0	0-2.1	0±0	0	0	0-0

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
Goodenia	A	0±0	0	0	0-0.4	0±0	0	0	0-0	0±0.1	0	0	0-0.4	0±0	0	0	0-0
	H	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	L	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0.1	0	0	0-0.4	0±0	0	0	0-0
	C	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0.1	0	0	0-0.4	0±0	0	0	0-0
Grass	A	0±0.3	0	0	0-3.8	0.1±0.4	0	0	0-3.8	0±0.1	0	0	0-0.3	0±0	0	0	0-0
	H	0±0.1	0	0	0-0.6	0±0.1	0	0	0-0.6	0±0	0	0	0-0.1	0±0	0	0	0-0
	L	0±0.3	0	0	0-2	0.1±0.5	0	0	0-2	0±0.1	0	0	0-0.3	0±0	0	0	0-0
	C	0±0.1	0	0	0-0.3	0±0	0	0	0-0	0±0.1	0	0	0-0.3	0±0	0	0	0-0
Grevillea	A	0.1±0.4	0	0	0-4	0±0	0	0	0-0	0.1±0.2	0	0	0-1.2	0.3±0.6	0	0.4	0-4
	H	0.2±0.6	0	0	0-4	0±0	0	0	0-0	0±0	0	0	0-0	0.5±0.9	0	0.8	0-4
	L	0.1±0.3	0	0	0-2.4	0±0	0	0	0-0	0±0	0	0	0-0	0.3±0.6	0	0.3	0-2.4
	C	0.1±0.3	0	0	0-1.2	0±0	0	0	0-0	0.2±0.3	0	0.1	0-0.8	0.2±0.4	0	0.3	0-1.2
Halgania	A	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Helichrysum	A	0±0	0	0	0-0.3	0±0	0	0	0-0.3	0±0	0	0	0-0.1	0±0	0	0	0-0
	H	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	L	0±0	0	0	0-0.3	0±0.1	0	0	0-0.3	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0.1	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0
Hyalosperma	A	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	H	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0.1	0	0	0-0.2	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Lasiopetalum	A	0.1±0.9	0	0	0-12	0±0	0	0	0-0.3	0.1±0.9	0	0	0-9.2	0.1±1.2	0	0	0-12
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0.2±1.2	0	0	0-9.2	0±0	0	0	0-0	0.4±2	0	0	0-9.2	0±0	0	0	0-0
	C	0.4±2	0	0	0-12	0±0	0	0	0-0	0±0	0	0	0-0	1.1±3.5	0	0	0-12
Lepidosperma	A	0±0	0	0	0-0.5	0±0.1	0	0	0-0.5	0±0.1	0	0	0-0.5	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0.1	0	0	0-0.5	0±0.1	0	0	0-0.5	0±0.1	0	0	0-0.3	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Leptospermum	A	0±0	0	0	0-0.7	0±0	0	0	0-0	0±0.1	0	0	0-0.7	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0.1	0	0	0-0.7	0±0	0	0	0-0	0±0.1	0	0	0-0.7	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Lomandra	A	0.1±0.3	0	0	0-3	0.1±0.3	0	0	0-2.2	0.1±0.4	0	0	0-3	0±0.2	0	0	0-1.4
	H	0.1±0.3	0	0	0-2.1	0±0.1	0	0	0-0.6	0.1±0.5	0	0	0-2.1	0±0	0	0	0-0
	L	0.1±0.4	0	0	0-2.2	0.1±0.5	0	0	0-2.2	0.1±0.2	0	0	0-0.8	0.1±0.3	0	0	0-1.4
	C	0.1±0.6	0	0	0-3	0.2±0.5	0	0	0-1.8	0.2±0.9	0	0	0-3	0±0	0	0	0-0
Maireana	A	0±0.2	0	0	0-3	0±0	0	0	0-0	0.1±0.3	0	0.1	0-3	0±0.1	0	0	0-0.3
	H	0.1±0.4	0	0	0-3	0±0	0	0	0-0	0.2±0.7	0	0.1	0-3	0±0.1	0	0	0-0.2
	L	0±0.1	0	0	0-1	0±0	0	0	0-0	0.1±0.2	0	0.1	0-1	0±0.1	0	0	0-0.2
	C	0±0.1	0	0	0-0.3	0±0	0	0	0-0	0±0.1	0	0	0-0.3	0±0	0	0	0-0
Melaleuca	A	1.6±3.9	0	1.3	0-30.4	3.6±5.1	1.9	3.7	0-29.2	0.1±0.3	0	0	0-2.7	1.1±3.8	0	0.4	0-30.4
	H	0.9±1.7	0	0.9	0-7.4	2±2.2	1.2	1.5	0-7.4	0±0.2	0	0	0-0.8	0.8±1.6	0	0.1	0-5.1
	L	2.1±5.5	0	1	0-29.2	6.3±8.5	2.1	7.6	0-29.2	0±0	0	0	0-0	0.3±0.8	0	0.3	0-3
	C	2.8±6.6	0	1.5	0-30.4	4.5±6.2	1.7	3.4	0-20	0.2±0.4	0	0	0-1	4±9.4	0	0.6	0-30.4
Microcybe	A	0.3±1	0	0	0-9.5	0.8±1.5	0	1.2	0-9.5	0±0.4	0	0	0-3.2	0.1±0.5	0	0	0-3.5
	H	0.4±1	0	0	0-5.2	1±1.4	0.8	1.4	0-5.2	0±0	0	0	0-0	0.2±0.8	0	0	0-3.5
	L	0.2±0.6	0	0	0-3.2	0.4±0.8	0	0	0-3	0.2±0.7	0	0	0-3.2	0±0	0	0	0-0
	C	0.2±0.5	0	0	0-2.2	0.3±0.6	0	0.4	0-1.8	0±0	0	0	0-0	0.2±0.6	0	0	0-2.2

		All				Lock				SR159				SR260			
	V	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
Millotia	A	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	H	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Minuria	A	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0.1
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Myoporium	A	0±0.2	0	0	0-2.5	0±0	0	0	0-0	0±0.1	0	0	0-0.9	0±0.3	0	0	0-2.5
	H	0±0.1	0	0	0-0.9	0±0	0	0	0-0	0±0.2	0	0	0-0.9	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0.1±0.4	0	0	0-2.5	0±0	0	0	0-0	0±0	0	0	0-0	0.2±0.7	0	0	0-2.5
Olearia	A	0.4±0.8	0	0.4	0-6	0.2±0.6	0	0.2	0-3.8	0.6±1	0	1	0-6	0.3±0.6	0	0.3	0-3.3
	H	0.3±0.7	0	0.2	0-3.2	0.1±0.3	0	0	0-1	0.5±0.9	0	0.4	0-3.2	0.3±0.6	0	0.3	0-2.2
	L	0.6±1.1	0	0.4	0-6	0.6±1	0	0.5	0-3.1	0.8±1.5	0.2	1	0-6	0.3±0.6	0	0.3	0-2.1
	C	0.2±0.5	0	0.1	0-2.1	0.2±0.4	0	0.2	0-1.2	0.4±0.7	0	0.8	0-2.1	0±0	0	0	0-0
Ozothamnus	A	0±0	0	0	0-0.2	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Pimelea	A	0±0.2	0	0	0-3.9	0±0.4	0	0	0-3.9	0±0	0	0	0-0.4	0±0	0	0	0-0
	H	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0.1	0	0	0-0.4	0±0	0	0	0-0
	L	0.1±0.5	0	0	0-3.9	0.2±0.9	0	0	0-3.9	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Pittosporum	A	0±0	0	0	0-0.4	0±0	0	0	0-0	0±0	0	0	0-0.4	0±0	0	0	0-0
	H	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0.1	0	0	0-0.4	0±0	0	0	0-0
	L	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Podolepis	A	0±0	0	0	0-0.5	0±0	0	0	0-0	0±0.1	0	0	0-0.5	0±0	0	0	0-0
	H	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	L	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0.1	0	0	0-0.4	0±0	0	0	0-0
	C	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0.1	0	0	0-0.2	0±0	0	0	0-0
Raised	A	1.7±1.9	1.2	1.6	0-16.2	2.1±1.7	1.8	1.7	0-10.5	1.6±2.4	0.8	1.5	0-16.2	1.4±1.2	1.1	1.5	0-6.8
	H	1.8±1.8	1.5	2	0-9.6	2.2±1.3	1.8	1	0.7-6	1.5±2.1	1.4	2	0-9.6	1.7±1.8	1.2	1.9	0-6.8
	L	1.5±1.6	1.1	1.8	0-7.6	2±2	1.8	1.5	0-7.6	1.5±1.6	1.2	2	0-6	1.1±0.9	0.9	1	0-3.6
	C	1.7±2.7	1.2	1.8	0-16.2	2.2±0.9	2.4	1	0.8-3.4	2.1±4.5	0.8	1	0-16.2	0.9±0.9	0.7	1.6	0-2.4
Rhagodia	A	0.7±1	0.2	1	0-7.2	0.5±0.8	0.2	0.8	0-3.4	0.4±0.7	0	0.4	0-3.8	1.1±1.3	0.6	1.3	0-7.2
	H	0.8±1.3	0.2	1	0-7.2	0.8±1	0.3	1.1	0-3.4	0.2±0.4	0	0	0-1.6	1.3±1.8	0.7	1.8	0-7.2
	L	0.5±0.7	0.1	1	0-2.6	0.3±0.6	0	0.4	0-2	0.6±0.8	0.1	1	0-2.6	0.7±0.6	0.6	1.1	0-2
	C	0.6±0.8	0.4	0.8	0-3.8	0.4±0.4	0.4	0.7	0-1.2	0.6±1.1	0	0.9	0-3.8	0.6±0.7	0.6	0.8	0-2
Roepora	A	0±0.1	0	0	0-1.4	0±0	0	0	0-0	0.1±0.2	0	0	0-0.9	0±0.1	0	0	0-1.4
	H	0±0.1	0	0	0-0.9	0±0	0	0	0-0	0.1±0.2	0	0.1	0-0.9	0±0	0	0	0-0.2
	L	0±0.1	0	0	0-0.8	0±0	0	0	0-0	0.1±0.2	0	0.1	0-0.8	0±0	0	0	0-0
	C	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0.1	0	0	0-0.4	0±0	0	0	0-0
Santalum	A	0±0.1	0	0	0-1.6	0±0.2	0	0	0-1.6	0±0	0	0	0-0	0±0	0	0	0-0.4
	H	0±0.2	0	0	0-1.6	0.1±0.4	0	0	0-1.6	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
Scaevola	A	0±0.1	0	0	0-2	0±0	0	0	0-0	0±0	0	0	0-0	0±0.2	0	0	0-2
	H	0±0.3	0	0	0-2	0±0	0	0	0-0	0±0	0	0	0-0	0.1±0.4	0	0	0-2
	L	0±0	0	0	0-0.3	0±0	0	0	0-0	0±0	0	0	0-0	0±0.1	0	0	0-0.3
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
<i>Sclerolaena</i>	A	0±0	0	0	0-0.5	0±0	0	0	0-0	0±0.1	0	0	0-0.5	0±0.1	0	0	0-0.4
	H	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0.1	0	0	0-0.4
	L	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	C	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
<i>Senecio</i>	A	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0.1	0	0	0-0.2	0±0	0	0	0-0
<i>Senna</i>	A	0.1±0.4	0	0	0-2.6	0±0	0	0	0-0	0.3±0.6	0	0.3	0-2.6	0±0.2	0	0	0-1
	H	0.1±0.4	0	0	0-2.6	0±0	0	0	0-0	0.2±0.7	0	0	0-2.6	0.1±0.2	0	0	0-0.6
	L	0.2±0.4	0	0.2	0-2.1	0±0	0	0	0-0	0.5±0.6	0	0.8	0-2.1	0.1±0.3	0	0.2	0-1
	C	0.1±0.3	0	0	0-1.5	0±0	0	0	0-0	0.3±0.5	0	0.2	0-1.5	0±0	0	0	0-0
<i>Solanum</i>	A	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
<i>Templetonia</i>	A	0.1±0.4	0	0	0-6	0±0	0	0	0-0	0±0	0	0	0-0	0.2±0.7	0	0	0-6
	H	0.1±0.3	0	0	0-2.2	0±0	0	0	0-0	0±0	0	0	0-0	0.2±0.5	0	0	0-2.2
	L	0.1±0.8	0	0	0-6	0±0	0	0	0-0	0±0	0	0	0-0	0.4±1.3	0	0	0-6
	C	0.1±0.4	0	0	0-2	0±0	0	0	0-0	0±0	0	0	0-0	0.3±0.6	0	0.2	0-2
<i>Thryptomene</i>	A	0±0.1	0	0	0-1.2	0±0	0	0	0-0	0±0	0	0	0-0	0±0.1	0	0	0-1.2
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0.2	0	0	0-1.2	0±0	0	0	0-0	0±0	0	0	0-0	0.1±0.3	0	0	0-1.2
<i>Thysanotus</i>	A	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
<i>Trachymene</i>	A	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	H	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	L	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
<i>Triodia</i>	A	2.4±5	0	2.4	0-38.2	3.9±4.9	2.2	3.9	0-19.6	2.8±5.3	0	4.6	0-33.2	0.5±4	0	0	0-38.2
	H	1.4±2.9	0	1.5	0-11.9	1.8±2	1.5	2.3	0-7	2.4±4.2	0	1.6	0-11.9	0±0	0	0	0-0
	L	3±5.9	0	4	0-33.2	6.2±5.7	4.5	7.9	0-16.5	3.1±7.6	0	3	0-33.2	0±0.1	0	0	0-0.3
	C	4.7±8.4	0	6	0-38.2	5.8±6.1	2.4	7.5	0.6-19.6	4.3±7.8	0	4	0-18	4±11.2	0	0	0-38.2
<i>Vittadinia</i>	A	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0.1	0	0	0-0.2	0±0	0	0	0-0
<i>Waitzia</i>	A	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0.1	0	0	0-0.2	0±0	0	0	0-0
<i>Westringia</i>	A	0.2±0.7	0	0	0-6.4	0±0.3	0	0	0-3.1	0.1±0.3	0	0	0-2.4	0.4±1.1	0	0	0-6.4
	H	0.3±0.8	0	0	0-3.4	0±0	0	0	0-0	0.1±0.2	0	0	0-0.8	0.7±1.2	0	0.9	0-3.4
	L	0.2±0.6	0	0	0-3.1	0.2±0.7	0	0	0-3.1	0.1±0.4	0	0	0-1.5	0.2±0.6	0	0.2	0-2.7
	C	0.2±0.9	0	0	0-4.8	0±0.1	0	0	0-0.3	0±0	0	0	0-0	0.6±1.5	0	0	0-4.8
<i>Xerochrysum</i>	A	0±0	0	0	0-0.1	0±0	0	0	0-0	0±0	0	0	0-0.1	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0

Table S 5-7 Summary statistics of explanatory variables (Midstorey). We present the percentage cover mean with 1 standard deviation (mean), the median (med), the interquartile range (IQR) and the percentage cover range (range). Rows with *All* visits show statistics of the complete dataset with all visits taken into account. Rows with *High/Low/Ctrl* visits show the statistics of the reduced subset of the data with high (top 25% visited) and low use (bottom 25% visited) sites as well as controls (not visited) which were used for the decision tree models. V = Visits, A = All, H/L = High/Low use, C = Control, G = Ground cover, M = Midstorey, C = Canopy

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
<i>Accacia</i>	A	0.1±0.5	0	0	0-4	0.1±0.4	0	0	0-3	0.2±0.6	0	0	0-4	0.1±0.4	0	0	0-2.4
	H	0.1±0.5	0	0	0-3	0.3±0.8	0	0	0-3	0±0	0	0	0-0	0.1±0.5	0	0	0-2.4
	L	0.1±0.3	0	0	0-2.2	0±0	0	0	0-0	0±0	0	0	0-0	0.2±0.6	0	0	0-2.2
	C	0.2±0.5	0	0	0-2.2	0±0	0	0	0-0	0.3±0.6	0	0.2	0-1.7	0.2±0.6	0	0	0-2.2
<i>Alyxia</i>	A	0.2±1	0	0	0-8.8	0±0	0	0	0-0	0.4±1.2	0	0	0-7.5	0.3±1.2	0	0	0-8.8
	H	0.2±1	0	0	0-6	0±0	0	0	0-0	0.6±1.7	0	0	0-6	0±0	0	0	0-0
	L	0.4±1.5	0	0	0-8.8	0±0	0	0	0-0	0.6±1.7	0	0.8	0-7.5	0.6±1.9	0	0	0-8.8
	C	0.1±0.7	0	0	0-4	0±0	0	0	0-0	0.1±0.2	0	0	0-0.7	0.3±1.2	0	0	0-4
<i>Atriplex</i>	A	0±0	0	0	0-0.4	0±0	0	0	0-0	0±0	0	0	0-0.4	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	C	0±0.1	0	0	0-0.4	0±0	0	0	0-0	0±0.1	0	0	0-0.4	0±0	0	0	0-0
<i>Beyeria</i>	A	0.1±0.7	0	0	0-5.4	0.2±0.7	0	0	0-4	0.2±0.8	0	0	0-5.4	0.1±0.5	0	0	0-4.4
	H	0.1±0.8	0	0	0-5.4	0±0	0	0	0-0	0.4±1.3	0	0	0-5.4	0±0	0	0	0-0
	L	0.1±0.5	0	0	0-3.9	0.2±0.9	0	0	0-3.9	0.1±0.4	0	0	0-1.8	0±0	0	0	0-0
	C	0.2±0.5	0	0	0-2.1	0.3±0.6	0	0	0-1.8	0.3±0.7	0	0	0-2.1	0±0.2	0	0	0-0.6
<i>Callitris</i>	A	0.1±0.6	0	0	0-8	0.1±0.5	0	0	0-4.8	0.1±1	0	0	0-8	0±0	0	0	0-0
	H	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
	L	0.1±0.6	0	0	0-5	0±0	0	0	0-0	0.2±1.1	0	0	0-5	0±0	0	0	0-0
	C	0.2±1.4	0	0	0-8	0±0	0	0	0-0	0.7±2.3	0	0	0-8	0±0	0	0	0-0
<i>Cassyltha</i>	A	0±0.3	0	0	0-2.5	0±0.2	0	0	0-1.3	0.1±0.4	0	0	0-2.5	0±0	0	0	0-0
	H	0±0	0	0	0-0.2	0±0	0	0	0-0	0±0	0	0	0-0.2	0±0	0	0	0-0
	L	0.1±0.4	0	0	0-2.5	0±0	0	0	0-0	0.2±0.7	0	0	0-2.5	0±0	0	0	0-0
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
<i>Cratystylis</i>	A	0±0.3	0	0	0-3.6	0±0	0	0	0-0	0±0.2	0	0	0-2	0.1±0.6	0	0	0-3.6
	H	0±0.4	0	0	0-2.9	0±0	0	0	0-0	0±0	0	0	0-0	0.1±0.6	0	0	0-2.9
	L	0.1±0.4	0	0	0-3	0±0	0	0	0-0	0±0.2	0	0	0-0.8	0.1±0.7	0	0	0-3
	C	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0	0±0	0	0	0-0
<i>Dodonea</i>	A	0.1±0.4	0	0	0-4	0±0	0	0	0-0	0.2±0.7	0	0	0-4	0±0.3	0	0	0-3.3
	H	0.1±0.5	0	0	0-4	0±0	0	0	0-0	0.3±0.9	0	0	0-4	0±0	0	0	0-0
	L	0±0.1	0	0	0-0.8	0±0	0	0	0-0	0.1±0.2	0	0	0-0.8	0±0	0	0	0-0
	C	0.1±0.7	0	0	0-4	0±0	0	0	0-0	0.4±1.2	0	0	0-4	0±0	0	0	0-0
<i>Eremophila</i>	A	0.4±1.4	0	0	0-15.4	0±0.1	0	0	0-0.7	0.1±0.3	0	0	0-2	1±2.2	0	1	0-15.4
	H	0.8±2.3	0	0	0-15.4	0±0.2	0	0	0-0.7	0.1±0.3	0	0	0-1.6	2.4±3.6	1	4	0-15.4
	L	0.1±0.4	0	0	0-2.4	0±0	0	0	0-0	0±0	0	0	0-0	0.3±0.6	0	0.4	0-2.4
	C	0.3±1.8	0	0	0-10.5	0±0	0	0	0-0	0±0	0	0	0-0	0.9±3	0	0	0-10.5
<i>Eucalyptus</i>	A	3.1±6.1	1.2	3.5	0-60	3.3±3.2	2.2	3.7	0-15.4	4.8±9.5	1.4	3.9	0-60	1.2±1.8	0.2	1.6	0-9
	H	3.5±7.9	2	3.4	0-60	3±3.2	2	2.5	0-12.8	5.9±12.8	2.1	3.9	0-60	1.5±1.8	1	2.5	0-6
	L	2.7±5.8	0.6	2.5	0-37.6	3.9±3.7	2.8	3.5	0-12.5	3.3±8.9	0.5	1.5	0-37.6	1±2	0.2	0.6	0-6.8
	C	3.4±6.6	1.6	4.5	0-36	4.2±4.5	2.4	4.4	0-15.4	4.6±10.2	0	4	0-36	1.4±2	0.2	2	0-6
<i>Exocarpos</i>	A	0.1±0.7	0	0	0-8	0±0	0	0	0-0	0.3±1.3	0	0	0-8	0±0	0	0	0-0
	H	0±0.2	0	0	0-1.7	0±0	0	0	0-0	0.1±0.4	0	0	0-1.7	0±0	0	0	0-0
	L	0.2±0.9	0	0	0-6	0±0	0	0	0-0	0.5±1.5	0	0	0-6	0±0	0	0	0-0
	C	0±0.1	0	0	0-0.6	0±0	0	0	0-0	0±0.2	0	0	0-0.6	0±0	0	0	0-0

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
<i>Geijera</i>	A	0.5 ± 1.8	0	0	0 - 13.5	0 ± 0	0	0	0 - 0	1.1 ± 2.3	0	1.3	0 - 12	0.4 ± 1.9	0	0	0 - 13.5
	H	0.2 ± 0.9	0	0	0 - 6	0 ± 0	0	0	0 - 0	0.6 ± 1.4	0	0.8	0 - 6	0 ± 0	0	0	0 - 0
	L	1.1 ± 2.5	0	0.8	0 - 12	0 ± 0	0	0	0 - 0	2.1 ± 3.1	0	3	0 - 12	1.3 ± 2.7	0	0.8	0 - 11
	C	0.3 ± 1.1	0	0	0 - 5.4	0 ± 0	0	0	0 - 0	0.8 ± 1.8	0	0.3	0 - 5.4	0 ± 0	0	0	0 - 0
<i>Grevillea</i>	A	0 ± 0	0	0	0 - 0.6	0 ± 0	0	0	0 - 0	0 ± 0.1	0	0	0 - 0.6	0 ± 0.1	0	0	0 - 0.5
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Lasiopetalum</i>	A	0 ± 0	0	0	0 - 0.2	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.2	0 ± 0	0	0	0 - 0
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Melaleuca</i>	A	2.7 ± 4.7	0	3.8	0 - 30	6.4 ± 6.2	4.8	5.9	0 - 30	0.2 ± 0.8	0	0	0 - 4.8	1.6 ± 2.9	0	3	0 - 18
	H	2.2 ± 3.9	0	2.7	0 - 18	6.1 ± 5	6	5.8	0 - 18	0.1 ± 0.3	0	0	0 - 1.2	0.8 ± 1.2	0	1	0 - 3.2
	L	2.8 ± 4.9	0	3.5	0 - 22.5	8 ± 5.9	6.6	8.3	0 - 22.5	0 ± 0	0	0	0 - 0	0.9 ± 1.7	0	0	0 - 6.3
	C	2.9 ± 3.9	1.8	4.8	0 - 18	5.2 ± 3	4.8	2.5	0 - 10.5	0.6 ± 1.4	0	0.1	0 - 4.8	3.1 ± 5.1	1.4	3.6	0 - 18
<i>Microcybe</i>	A	0 ± 0.1	0	0	0 - 2.2	0 ± 0.1	0	0	0 - 0.7	0 ± 0	0	0	0 - 0	0 ± 0.2	0	0	0 - 2.2
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0.1 ± 0.4	0	0	0 - 2.2	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.2 ± 0.6	0	0	0 - 2.2
<i>Myoporium</i>	A	0.1 ± 0.5	0	0	0 - 4.8	0 ± 0	0	0	0 - 0	0.1 ± 0.5	0	0	0 - 4.5	0.2 ± 0.7	0	0	0 - 4.8
	H	0.1 ± 0.3	0	0	0 - 2.1	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.2 ± 0.6	0	0	0 - 2.1
	L	0.1 ± 0.5	0	0	0 - 3.6	0 ± 0	0	0	0 - 0	0.1 ± 0.4	0	0	0 - 2	0.2 ± 0.8	0	0	0 - 3.6
	C	0 ± 0.3	0	0	0 - 1.6	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.1 ± 0.5	0	0	0 - 1.6
<i>Olearia</i>	A	0 ± 0.1	0	0	0 - 2.5	0 ± 0	0	0	0 - 0.3	0 ± 0.3	0	0	0 - 2.5	0 ± 0	0	0	0 - 0
	H	0 ± 0	0	0	0 - 0.3	0 ± 0.1	0	0	0 - 0.3	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0 ± 0.3	0	0	0 - 2.5	0 ± 0	0	0	0 - 0	0.1 ± 0.5	0	0	0 - 2.5	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Pittosporum</i>	A	0 ± 0.2	0	0	0 - 2.4	0 ± 0.3	0	0	0 - 2.4	0 ± 0.1	0	0	0 - 1	0 ± 0.2	0	0	0 - 2.2
	H	0 ± 0.2	0	0	0 - 1	0.1 ± 0.2	0	0	0 - 1	0 ± 0.2	0	0	0 - 1	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0.1 ± 0.4	0	0	0 - 2.2	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.2 ± 0.6	0	0	0 - 2.2
Raised litter	A	0.3 ± 0.8	0	0.3	0 - 9.2	0.3 ± 0.7	0	0.4	0 - 3	0.3 ± 1	0	0.2	0 - 9.2	0.3 ± 0.7	0	0.3	0 - 4
	H	0.4 ± 1.3	0	0.3	0 - 9.2	0.3 ± 0.8	0	0.1	0 - 2.5	0.5 ± 2	0	0	0 - 9.2	0.4 ± 0.7	0	0.6	0 - 2.5
	L	0.2 ± 0.3	0	0.1	0 - 1.5	0.1 ± 0.3	0	0	0 - 1	0.2 ± 0.4	0	0.2	0 - 1.5	0.1 ± 0.3	0	0	0 - 0.8
	C	0.3 ± 0.6	0	0.2	0 - 3	0.4 ± 0.5	0	0.6	0 - 1.4	0.5 ± 0.9	0	0.3	0 - 3	0 ± 0	0	0	0 - 0
<i>Rhagodia</i>	A	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.1
	H	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0.1
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Santalum</i>	A	0.1 ± 0.9	0	0	0 - 15	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0	0.2 ± 1.6	0	0	0 - 15
	H	0.1 ± 0.5	0	0	0 - 3.8	0 ± 0	0	0	0 - 0.1	0 ± 0	0	0	0 - 0	0.2 ± 0.8	0	0	0 - 3.8
	L	0.3 ± 1.9	0	0	0 - 15	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.9 ± 3.3	0	0	0 - 15
	C	0 ± 0.3	0	0	0 - 1.6	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.1 ± 0.5	0	0	0 - 1.6
<i>Senna</i>	A	0.1 ± 0.5	0	0	0 - 4.8	0 ± 0	0	0	0 - 0	0.4 ± 0.9	0	0	0 - 4.8	0 ± 0.2	0	0	0 - 2
	H	0 ± 0.4	0	0	0 - 3	0 ± 0	0	0	0 - 0	0.1 ± 0.7	0	0	0 - 3	0 ± 0	0	0	0 - 0
	L	0.2 ± 0.5	0	0	0 - 2.1	0 ± 0	0	0	0 - 0	0.4 ± 0.7	0	0.3	0 - 2.1	0.1 ± 0.4	0	0	0 - 2
	C	0.1 ± 0.5	0	0	0 - 2.7	0 ± 0	0	0	0 - 0	0.3 ± 0.8	0	0.1	0 - 2.7	0 ± 0	0	0	0 - 0
<i>Templetonia</i>	A	0.1 ± 0.5	0	0	0 - 5	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.2 ± 0.8	0	0	0 - 5
	H	0.2 ± 0.9	0	0	0 - 5	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.7 ± 1.5	0	0	0 - 5
	L	0.1 ± 0.3	0	0	0 - 1.6	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.2 ± 0.5	0	0	0 - 1.6
	C	0 ± 0.2	0	0	0 - 1	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.1 ± 0.3	0	0	0 - 1

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
<i>Thryptomene</i>	A	0 ± 0.1	0	0	0 - 2.2	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0.2	0	0	0 - 2.2
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0.1 ± 0.4	0	0	0 - 2.2	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.2 ± 0.6	0	0	0 - 2.2

Table S 5-8 Summary statistics of explanatory variables (Canopy). We present the percentage cover mean with 1 standard deviation (mean), the median (med), the interquartile range (IQR) and the percentage cover range (range). Rows with *All* visits show statistics of the complete dataset with all visits taken into account. Rows with *High/Low/Ctrl* visits show the statistics of the reduced subset of the data with high (top 25% visited) and low use (bottom 25% visited) sites as well as controls (not visited) which were used for the decision tree models. V = Visits, A = All, H/L = High/Low use, C = Control, G = Ground cover, M = Midstorey, C = Canopy

	V	All				Lock				SR159				SR260			
		Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range	Mean	Med	IQR	Range
<i>Acacia</i>	A	0 ± 0.2	0	0	0 - 2.5	0 ± 0	0	0	0 - 0	0 ± 0.3	0	0	0 - 2.5	0 ± 0	0	0	0 - 0
	H	0 ± 0.3	0	0	0 - 2.5	0 ± 0	0	0	0 - 0	0.1 ± 0.5	0	0	0 - 2.5	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Callitris</i>	A	0.1 ± 1.3	0	0	0 - 22.5	0 ± 0	0	0	0 - 0	0.2 ± 2.3	0	0	0 - 22.5	0 ± 0	0	0	0 - 0
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0.6 ± 3.8	0	0	0 - 22.5	0 ± 0	0	0	0 - 0	1.9 ± 6.5	0	0	0 - 22.5	0 ± 0	0	0	0 - 0
<i>Cassyntha</i>	A	0 ± 0.1	0	0	0 - 0.8	0 ± 0	0	0	0 - 0	0 ± 0.1	0	0	0 - 0.8	0 ± 0	0	0	0 - 0
	H	0 ± 0.1	0	0	0 - 0.8	0 ± 0	0	0	0 - 0	0.1 ± 0.2	0	0	0 - 0.8	0 ± 0	0	0	0 - 0
	L	0 ± 0.1	0	0	0 - 0.6	0 ± 0	0	0	0 - 0	0 ± 0.2	0	0	0 - 0.6	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Eucalyptus</i>	A	16.8 ± 13.4	15	19	0 - 60	14.1 ± 9.1	12	13	0 - 40	17.4 ± 15.7	15	23.5	0 - 60	18.7 ± 14.1	17	18	0 - 55
	H	17.1 ± 13.7	16	19	0 - 60	17.3 ± 7.5	17	13.5	5 - 30	16.8 ± 18.4	10	22	0 - 60	17.1 ± 13.3	16	19	0 - 50
	L	16.8 ± 14.6	13	20	0 - 50	15.3 ± 12.2	13	17	0 - 40	15.3 ± 16.6	10	23	0 - 50	19.8 ± 14.8	18	19	0 - 50
	C	16.1 ± 15.1	10	18.2	0 - 55	14.2 ± 8.6	10	12	5 - 30	18.8 ± 20	14	26	0 - 55	15.3 ± 15.1	10.5	11.2	0 - 50
<i>Melaleuca</i>	A	0.1 ± 0.6	0	0	0 - 9	0 ± 0	0	0	0 - 0	0.1 ± 0.6	0	0	0 - 5	0.1 ± 0.9	0	0	0 - 9
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0.1 ± 0.7	0	0	0 - 5	0 ± 0	0	0	0 - 0	0.4 ± 1.2	0	0	0 - 5	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Myoporium</i>	A	0.1 ± 1.2	0	0	0 - 16	0 ± 0	0	0	0 - 0	0.4 ± 2	0	0	0 - 16	0 ± 0	0	0	0 - 0
	H	0.3 ± 2	0	0	0 - 16	0 ± 0	0	0	0 - 0	0.8 ± 3.5	0	0	0 - 16	0 ± 0	0	0	0 - 0
	L	0.1 ± 0.5	0	0	0 - 4	0 ± 0	0	0	0 - 0	0.2 ± 0.9	0	0	0 - 4	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
Raised litter	A	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	C	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
<i>Santalum</i>	A	0 ± 0.1	0	0	0 - 2	0 ± 0	0	0	0 - 0	0 ± 0.2	0	0	0 - 1.6	0 ± 0.2	0	0	0 - 2
	H	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0
	L	0 ± 0.2	0	0	0 - 1.6	0 ± 0	0	0	0 - 0	0.1 ± 0.3	0	0	0 - 1.6	0 ± 0	0	0	0 - 0
	C	0.1 ± 0.3	0	0	0 - 2	0 ± 0	0	0	0 - 0	0 ± 0	0	0	0 - 0	0.2 ± 0.6	0	0	0 - 2

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Chapter 6:

General discussion



Figure on previous page: Malleefowl tracks in the sand. Credit: Peri Stenhouse

In this thesis, I primarily aimed to enhance our understanding of how historic and recent anthropogenic change has influenced Malleefowl on the Eyre Peninsula. Using a multifaceted approach, I studied Malleefowl population trends, landscape genetics, movement patterns and habitat preferences within a fragmented landscape undergoing climate change.

6.1 Trends

Chapter 2 showed that Malleefowl on the Eyre Peninsula have indeed declined over the last few decades and that human-driven environmental factors appear to be the contributing to this. My research showed that bare ground (i.e. lack of vegetation) and environmental variables related to soil moisture (e.g. Southern Oscillation Index and rain) were the driving forces behind declining numbers and that breeding activity, a proxy for population numbers, decreased significantly over the study period (2001 to 2018). Factors influencing ground cover include land clearing for agriculture or infrastructure, overgrazing by introduced and overabundant native herbivores and wildfires.

This chapter supports previous findings of declining Malleefowl numbers (Benshemesh, Southwell *et al.* 2020) on a local scale and highlights the adverse effects of habitat loss and fragmentation on this iconic species in degraded and fragmented agricultural landscapes (Benshemesh 1992; Benshemesh, Barker *et al.* 2007; Frith 1962). As anthropogenic driven changes to weather patterns are expected to lead to more frequent heat and drought periods and more intense and frequent wildfires (Dey, Lewis *et al.* 2019; Dowdy, Ye *et al.* 2019; Guerin, O'Connor *et al.* 2018), avian species are expected to decline (Chambers, Hughes *et al.* 2005; Mac Nally, Bennett *et al.* 2009) thus I expect conditions to further deteriorate for Malleefowl, particularly in the semi-arid zones like the Eyre Peninsula, which were too dry to be cleared for agriculture. Further, this chapter shows the importance of long-term data collection in understanding responses to climatic changes.

6.2 Landscape genetics

Chapter 3 highlights the potential problems caused by anthropogenic land cover changes for threatened Malleefowl populations on the Eyre Peninsula from a genetic perspective. Using high-throughput DNA sequencing, I showed that Malleefowl population structure on the Eyre Peninsula was not homogenous and that anthropogenic habitat fragmentation with subsequent isolation has reduced gene flow between Malleefowl living in isolated remnant vegetation patches causing the differentiation of at least two distinct populations. Furthermore, this chapter showed preliminary genetic evidence of female dispersal, which is consistent with avian dispersal ecology (Greenwood 1980). As little research has been

conducted on Malleefowl genetics to date, both are significant findings. Two populations west and east of the Eyrean Barrier had been suggested in the past (Cope, Mulder *et al.* 2014) but no sub-populations were previously identified.

This chapter highlighted how modern sequencing technologies can benefit ecological studies, as they can improve our understanding of past movements and dispersal patterns using non-intrusive sample collection. Additionally, it shows the importance of both large continuous native vegetation patches and small connecting habitat fragments in highly fragmented landscapes. While there was evidence of differentiation, this appears to be a relatively recent development and thus probably means that some dispersal has been happening between disconnected habitat patches, possibly through female dispersal. Alternatively, some of the mallee habitat on the Eastern Eyre Peninsula was only cleared in the last 30-40 years and so the longer term genetic effects may not yet be realised.

6.3 Movement

Modern telemetry can improve our understanding of animal movement without exposing the animal to undue stress through repeated trapping. I used solar-powered GPS trackers to investigate different aspects and temporal scales of Malleefowl movement and dispersal. The tracking data was used in chapters 4 and 5. In chapter 4, I investigated individual movement patterns of Malleefowl as well as seasonal and gender-based differences. I showed that Malleefowl movement was tightly associated with breeding status, with breeding birds staying close to the mound and non-breeding birds disassociating from the mound and moving long distances. A significant finding in this chapter was that Malleefowl movement was influenced by patch size and fragmentation. I showed that non-breeding birds in larger, contiguous patches of native habitat moved further than birds living in smaller patches. However, Malleefowl were able to persist in small patches of native vegetation too, highlighting the conservation value of small remnants (Fahrig, Arroyo-Rodríguez *et al.* 2019; Tulloch, Barnes *et al.* 2016). Another interesting finding was that Malleefowl moved less with increasing temperature. With climate models projecting steady increases in temperatures over the coming years (CSIRO and BOM 2015), this may have severe impacts on the persistence of the species. Further, the mortality of the Eyre Peninsula Malleefowl was very high, with over 66% of tracked individuals dying within a year of trapping. This shows the impact of introduced predators on native Fauna and highlights the need for continued and improved predator control. I found Malleefowl rarely visited or moved across cleared agricultural land but could survive and breed in very small remnants suggesting that connectivity is a key requirement for future conservation efforts. Some small remnants are likely highly productive, as the most fertile land was usually cleared for

agriculture. Protecting and enhancing these remnants may be the key to future survival of the species.

6.4 Habitat use

In chapter 5, using tracking data and traditional ground-based vegetation surveys, I was able to show that fine-scale Malleefowl habitat selection is mainly driven by canopy height and to a lesser degree by a preference for certain plant species. Several important predictors, such as a tall canopy, a reduced understorey, raised litter and reduced *Triodia* cover are defining features of mallee eucalypts that are 35+ years post-fire (Gosper, Prober *et al.* 2013). While this may suggest that Malleefowl favour long unburnt habitat as previously suggested (Benshemesh 1992), all my sites were in the same fire age of at least 28+ post-fire, with similarly mature trees and conditions. However, as fires burn patchily, and the vegetation survey quadrats were relatively small - making a generalisation on a landscape scale difficult - this chapter was focussed more on the importance of micropatches of tall mallee stands within a mature *Eucalyptus* matrix – likely as refuges from heat and predators. Additionally, I observed individual variations in the birds' habitat preferences, which were likely driven by the presence of different vegetation communities in each birds' home range. Against expectations, the influence of food plants was small and possibly reflects the fact that Malleefowl have a highly variable diet and opportunistically feed on what is available in their surroundings (Harlen and Priddel 1996).

6.5 Limitations

Like many other studies of threatened species, this PhD project was challenged by low sample sizes of the study species. The trapping process was very time intensive - it took up to eleven days to trap a Malleefowl on its mound and these were not consecutive days, as the trap had to be removed latest after four days to allow the birds access to the mound to regulate the temperature to ensure successful hatching. Further, one of the trackers did not reliably record fix locations through the winter months, resulting in a lack of data for this individual.

Tracked individuals had a high mortality rate (six out of nine tracked Malleefowl were preyed on by feral cats or foxes within a year), highlighting the catastrophic effects introduced predators have on native Australian fauna. It would be valuable to perform similar studies in pest-free sanctuaries to gather more robust long-term data unaffected by predation. Such a study may highlight seasonal behavioural differences between individuals or genders without feral predator disturbance. It might also enable us to judge better if mortality is connected to long-term tag attachment. While solar-charged GPS telemetry has the

advantage of reducing undue stress through repeated trapping, it does not allow us to check for negative health impacts of wearing tags (e.g. chafing harness). Malleefowl resuming breeding (some over multiple seasons) and camera trap images where the birds are displaying with raised wings and otherwise appear to be behaving normally suggest they are in good health, but detrimental tag effects over the long term cannot be 100% dismissed.

The DNA sample pool I used was very localised and relatively small. After extraction and sequencing, some samples proved to be duplicates and further extraction was not possible due to cost and time restraints. Further, because many samples were exposed to the environment for extended periods before collection (e.g. feather in the sun), few of them produced DNA and even less were of sufficient quality for SNP genotyping. Tissue samples provided the best results, and Malleefowl conservation would benefit from an Australia wide study using samples from tissue collections in the future. Generally, it would be highly beneficial to continue monitoring Malleefowl populations and considering genetic aspects to improve conservation outcomes of threatened species.

The small sample size also means that movement data analysis had to be interpreted with caution. I only tracked one female, therefore whether the movement patterns of this individual were different because it was a female or other reasons is open for discussion. However, presenting the female's data separately to the males' still has merit, as it is possible that this female is representative of a larger number of females. Also, the female was one of only two Malleefowl that dispersed. The female's dispersal was likely triggered by capture, while the male dispersed many months after capture. Again, it is hard to interpret these findings for only two birds and more research needs to be done in this area to enable more definitive conclusions. Similarly, all three birds that displayed long-range movement and never resumed breeding were all in the largest continuous native vegetation patch, which may have confounded the results.

Vegetation surveys were also very time intensive and required local plant knowledge or lengthy follow-ups at the SA herbarium, somewhat limiting the number of my vegetation sites. In the future, this could perhaps be remedied by using satellite imagery (e.g. NDVI) which would provide vegetation data on a much larger scale, albeit this would not be helpful on a finer scale as I have studied. Localised, high-resolution remote sensing using drone imagery would provide faster coverage but also with limitations around habitat structural complexity (Egli and Höpke 2020). Perhaps other imaging technology, such as LIDAR (Reutebuch, Andersen *et al.* 2005), could be used to provide this structural information in a faster and more cost-effective manner (Bradbury, Hill *et al.* 2005; de Vries, Koma *et al.* 2021; Gray, Ridge *et al.* 2018).

6.6 Management implications

In this thesis, I demonstrated that small, isolated patches can be important for Malleefowl conservation and suggested several management actions that may reverse the current decline and strengthen the populations for the future. Some conservation actions can have immediate effects by reducing current pressures, but most will have also long-lasting effects on Malleefowl population health and help the species to persist:

One of the most important findings of this study was the high mortality rate of adult birds from predators and the ongoing decline in breeding success across the Eyre Peninsula. These findings are concerning and suggest more work is needed to control introduced predators in Malleefowl habitat. This is particularly relevant for feral cats which are difficult to control using baiting (Comer, Clausen *et al.* 2020; Hohnen, Murphy *et al.* 2020) and have severely impacted Australian fauna (Hamer, Gardiner *et al.* 2021; Woinarski, Murphy *et al.* 2017; Woolley, Murphy *et al.* 2020). Feral cats are especially good hunters in habitats disturbed by fire or intense grazing, i.e. with an open understorey (McGregor, Legge *et al.* 2015; McGregor, Legge *et al.* 2014) and compound the effects of fox predation (Stobo-Wilson, Murphy *et al.* 2021). In this study, three of six killed Malleefowl were killed by feral cats and at least two by foxes. Cessation of habitat clearing and controlling herbivores through fencing or culling to improve understorey regeneration, as well as continued integrated predator control through baiting but also target specific control of feral cats through grooming traps (Moseby, McGregor *et al.* 2020) might be very beneficial in the short term.

Another important finding was that patch size restricted movement, but that even small patches of 100 ha could support breeding pairs. The limited movement of Malleefowl across open cropping ground suggested that patch connectivity and the quality of the connecting matrix will be critical to enabling Malleefowl to travel between and utilise small patches of habitat. Genetic data suggested that the connectivity of patches will also help ensure there is no continued decline in genetic diversity over time. Protecting and creating habitat corridors between small habitat patches in fragmented agricultural landscapes will likely help improve the long term conservation prospects of the Malleefowl and enable high-quality fragmented habitat to be used for breeding. Malleefowl have been observed in roadside vegetation and roadside strips have been shown to have high conservation value (Carthew, Garrett *et al.* 2013; Hall, Nimmo *et al.* 2016; New, Sands *et al.* 2021). Therefore, I suggest that corridors should be at least 20 m wide with patches of native vegetation no more than 250 m apart.

A further finding was that Malleefowl prefer microhabitats with tall canopies which could potentially be created through more fire management to create patchy fires. Additionally, modern remote-sensing methods (e.g. LIDAR) could possibly be used to pinpoint areas with pockets of trees with the preferred height of approximately 6 to 8 meters which could be the focus of conservation efforts.

While these measures might slow the decline of Malleefowl in the interim, the biggest challenge for Malleefowl in the future is climate change. The Malleefowl's decline, alongside other Australian species, is concerning. I have shown here that anthropological habitat destruction, predation and drying conditions have negatively affected the Eyre Peninsula Malleefowl. As the most productive habitat in the highest rainfall areas has been disproportionately cleared for agriculture, Malleefowl are pushed to the drier areas where rainfall is most variable and droughts are common. As climate change exacerbates these pressures, we need to address this directly by reducing emissions and habitat destruction as well as scaling up revegetation efforts to avoid catastrophic failure of climate systems and flora and fauna communities in the future.

6.7 Future directions

This PhD has uncovered several areas that would benefit from more in-depth research. My sample size of nine tagged birds was low and Malleefowl research would benefit from continued tracking of a larger number of individuals in a variety of habitats and seasons and a better representation of the two sexes. I have used tracking data of two male Malleefowl over the whole year (they did not breed in the summer) but of only one female (who did breed in the summer). More tracked individuals would allow us to focus on non-breeding birds in the winter, for example. Trapping efficiency could be improved by using different kinds of traps, such as soft-sided Thomas traps. These have been successfully used for other animals many times and tested on Malleefowl recently and - provided the Malleefowl is given enough time to acclimatise to being fed at or in the trap - would speed up the trapping process.

An Australia-wide study on Malleefowl genetics would benefit Malleefowl research. This could lead to a better understanding of population structure and range contraction since European settlement and the long-term effects of habitat loss and fragmentation. This could be achieved by accessing all samples available through various Museums. Studies have shown that feather condition and exposure to sunlight, heat and humidity can be detrimental to DNA quality (Hogan, Cooke *et al.* 2008; Vili, Nemesházi *et al.* 2013). Sample quality and lab processing time could be improved by using techniques such as feather snares (i.e. sticky tapes) to ensure timely collection of samples in the field and/or focusing on samples

from active mounds during the breeding period that are less likely to have been exposed to the environment for long periods. I also encourage collectors to store samples in the freezer or in ethanol to maximise DNA quality.

Fine-scale habitat preferences for tall canopies may be caused by a need for shelter from the heat or as protection from aerial predation. Another possibility is that early morning and late afternoon fixes were influenced by the Malleefowl's proximity to roosting sites. I have not investigated the night-time use of sites in this thesis, and it would be very interesting to see if tall canopies play a role at night-time too and thus possibly influenced the results of this study.

6.8 Wider application of results

My research contributes to evidence for preserving existing habitat patches and restoring connectivity between them. This is important for Malleefowl but is also likely to be critical for other species which are dependent on suitable habitat for dispersal (Stevens, Harrison *et al.* 2018; Stevens, Verkenne *et al.* 2006). My research highlights the value of small patches in agricultural matrices for the persistence of threatened species (Jones, Bain *et al.* 2021; Tulloch, Barnes *et al.* 2016; Volenec and Dobson 2020). In areas where few isolated and degraded patches remain, the protection of these patches and the improvement of the functional connectivity with habitat corridors would be the best strategy for biodiversity in general (Fahrig 2007; Fahrig, Arroyo-Rodríguez *et al.* 2019; Fahrig and Merriam 1985; Martensen, Pimentel *et al.* 2008).

Introduced predators have decimated other native species too, particularly mammals in the critical weight range (Woinarski, Burbidge *et al.* 2015) and they continue to exert unsustainable pressure on populations of small to medium sized vertebrates across most of Australia (Geyle, Woinarski *et al.* 2018). The fact that Malleefowl are mainly ground dwelling and breeding activities focus on the mound makes them particularly susceptible to predation. As such they are important indicators of feral predation pressure – a fact highlighted by the high mortality caused by cat and fox predation in this research. This supports other findings that highlight the need to control introduced predators (Wheeler and Priddel 2009) or protect and enhance habitat (Jones, Bain *et al.* 2021; McGregor, Legge *et al.* 2015; Stobo-Wilson, Stokeld *et al.* 2020) to safeguard native species, especially in fragmented landscapes where additional pressures exist.

Climate change is rapidly changing global weather patterns (Sippel, Meinshausen *et al.* 2020). My research illustrates some challenges faced by Malleefowl in the face of a changing climate and more extreme weather patterns. Many species are already

experiencing the impacts from rapidly changing environmental conditions caused by climate change such as higher temperatures, erratic rainfall and degraded habitat (Cooper, Withers *et al.* 2019; Iknayan and Beissinger 2018; Soroye, Newbold *et al.* 2020; Visser, Both *et al.* 2004). Many more are expected to be affected in the future and are likely to go extinct unless appropriate management actions are undertaken (Radchuk, Reed *et al.* 2019; Woinarski, Burbidge *et al.* 2015). My study contributes new knowledge on the impacts of climate change in semi-arid systems by highlighting the importance of several interacting climate-related factors that are predicted to worsen in the next decades (BOM and CSIRO 2020; CSIRO and BOM 2015). This again underlines the importance of providing functional connectivity between habitat patches to facilitate movement to thermal refuges, movement triggered by catastrophic events, such as wildfires or land clearing but also breeding dispersal. Increased connectivity may encourage gene flow between isolated patches and thus improve long-term genetic survival.

6.9 Conclusions

Malleefowl are an iconic Australian species and play an important ecological role in the mallee shrublands and woodlands in which they live. Considering the findings of this thesis, the long-term prognosis for Malleefowl is poor. This thesis showed that anthropogenic habitat alterations and changing environmental conditions since colonisation have restricted Malleefowl movement, dispersal and gene flow with long-term genetic effects visible today. I underline the significance of small native vegetation patches in an agricultural matrix (Fahrig, Arroyo-Rodríguez *et al.* 2019; Volenec and Dobson 2020) and the importance of protecting and creating habitat corridors between these, as well as continued and improved predator control efforts. Improving the quality of the connecting agricultural matrix through controlling herbivores would also improve biodiversity in a broader context (Franklin and Lindenmayer 2009; Prugh Laura, Hodges Karen *et al.* 2008). Further, my findings suggest that we can expect climate change to exacerbate all existing pressures on Malleefowl through more extreme temperatures, and increased risk of droughts and wildfires. A lack of winter rainfall causes the cessation of breeding attempts for the season. If this happens more frequently, this could be catastrophic for Malleefowl recruitment and combined with the high mortality of adults lead to population collapse. Increased ambient temperatures lead to reduced movement, which may reduce the Malleefowl's chances of finding a mate, tending the mound or dispersing, thus reducing its fitness. While smaller and patchier wildfires may be beneficial to Malleefowl, high-intensity wildfires are not (Benshemesh 1992; Frith 1962) and may lead to the destruction of large swathes of habitat for many years. The frequency of more intense wildfires is predicted to increase (CSIRO and BOM 2015).

While all these effects may not be immediately obvious, in the long term, the inability to disperse and pass on genetic material may result in the decline of the species' genetic health and ultimately cause extinction.

6.10 References

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