Comparative spatial genetic structure of two rodent species in an agro-ecological landscape in southern Africa

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Abstract:

Determining the scale of genetic variation informs studies of dispersal, connectivity, and population dynamics particularly in heterogeneous landscapes. *Mastomys natalensis* and *Mus minutoides* are generalist rodents that utilize multiple habitat types within the mosaic of the agro-ecological landscape of southern African savannas. To study the comparative spatial genetic structure of these species we developed *9* new microsatellites for *Mus* and used 14 microsatellite loci previously developed for *Mastomys*, to genotype rodents sampled across an agro-ecological landscape (~200 km^2). Spatial genetic structure was measured using spatial autocorrelation and Moran's Eigenvector Maps analysis. In both species, non-random genetic similarity was limited to only the smallest spatial scales (<600m), and at that scale, it was significantly greater in *Mastomys* than in *Mus*. Only a small proportion of the genetic signal across the landscape was due to spatial signal in *Mastomys*, and there was no spatial signal detected for *Mus*. The lack of spatial autocorrelation beyond the first six hundred meters for both species illustrated that they are capable of high rates of dispersal, while the observed patterns of genetic panmixia found for both species is the predicted genetic outcome for species with omnivorous habits and plasticity in habitat selection. These findings have implications for both pest management and rodent-borne disease control.

Keywords: agro-ecology, comparative genetic structure, heterogeneity, neutral genetic structure, spatial auto-correlation, rodent

Introduction

All organisms display genetic structure at some spatial scale (Wright 1969). This is a result of some individuals being more closely related than others, a pattern that is expected to decrease with geographic distance (i.e. isolation-by-distance, Malécot 1967). In addition to distance, extrinsic (e.g. topography, habitat variability) and intrinsic features (e.g., ecological preferences, social structure) can play a role in determining the spatial structure of organisms (Petkova et al. 2016). Knowledge of the

spatial scale of genetic variation within a population is a prerequisite for understanding how connectivity might be affected in heterogeneous landscapes (Anderson et al. 2010), which in turn is essential for developing effective population management plans. These plans could include maintenance of connectivity and population viability for conservation purposes (Shirk et al. 2010) or for effective control of pest species (Richardson et al. 2017) and disease vectors (Davis et al. 2005).

Rodents of the genus *Mastomys* and *Mus* are ecological generalists and considered important reservoirs for zoonotic infections (Gratz 1997) and as agricultural pests (Mwanjabe et al. 2002). In southern Africa, two of the most common rodent species include the multimammate mouse (*Mastomys natalensis*) and the pygmy mouse (*Mus minutoides*), both of which occur in grasslands and bushy areas, cultivated land, and human habitations (Delany 1986, Monadjem et al. 2015). Both species are reservoir hosts to human zoonoses: *Mastomys* is the host for plague (*Yersinia pestis*) (Leirs et al. 1996) and Lassa Fever in West Africa (Lalis et al. 2012), and *Mus* is the likely host to Kokodo virus (Castiglia et al. 2006).

The multimammate mouse is relatively large-bodied (50-80 g) and highly fecund, producing litters as large as 23 (Leirs and Verheyen 1995). Dispersal in this species is likely to be high, with rapid local turnover due to emigration (Leirs et al. 1993) following disturbance (Monadjem and Perrin 2003), which has been shown to correlate with little to no detectable genetic structure over fine (< 300 m; Van Hooft et al. 2008) and large spatial scales (100s km; Lalis et al. 2012, Russo et al. 2018). In contrast, *Mus* spp. are small bodied (4-12 g) and produce litters of up to 8 pups every 8 weeks (De Graaff 1981). Relatively little is known about the movement and genetic differentiation in *Mus*. Furthermore, there have been no studies on genetic structuring in any African *Mus* spp.

Both species are also commonly detected in sugarcane plantations (Hurst et al. 2013; Monadjem 1997) which is a highly dynamic habitat that undergoes 11-month burn and harvest cycles in the Eswatini lowveld. The effect of this temporal heterogeneity in habitat may have differing impacts on the spatial structuring of these two species. For example, *Mastomys* is considered a pioneer species, able to rapidly colonize areas that are recovering from habitat destruction (Meester et al. 1979), such as recently harvested sugar cane fields. Such rapid, localized mass colonization is predicted to result in high diversity and limited spatial differentiation (Banks et al. 2013). For example, a typical 8-10 ha area of harvested sugar cane is expected to be quickly colonized from the surrounding sugar cane within days or weeks of sugar cane regrowth. This is supported by the observation that the abundance of *Mastomys* in emergent ($\leq$ 0.29 m) sugarcane does not differ significantly from larger growth stages in this system (A. Monadjem, unpublished data). Alternatively, *Mus* does not appear to make long distance movements and stays close to cover and burrow entrances (Long et al. 2013). However, both species are dietary generalists (Monadjem 1997, 1999). Whether similar patterns of genetic structuring would be observed in both these species across heterogenous agro-ecological landscapes dominated by patches of sugarcane monoculture remains unknown.

Certain species attributes such as dispersal ability, reproductive rate, and perceptual range, are correlated with how well a species is able to move through different habitats and its sensitivity to the negative impacts of habitat conversion and fragmentation (e.g., Swihart et al. 2003; Kierepka et al. 2016). Our main objective was to compare the genetic structure of these two terrestrial small mammal species at a fine spatial scale to address whether *Mastomys* and *Mus* have similar spatial structuring

across a common landscape. Additionally, we hoped to obtain insights into species-specific effects of an agro-ecosystem on dispersal.

Materials and methods

Rodent sampling was conducted in a 200 km^2 area located in the northeastern Lowveld of Eswatini during May-June, 2014 (Fig. 1). The sampled area consisted of sugar cane (*Saccharum* spp.) monoculture that was bisected by roads and the Mbuluzi River with remnants of savanna patches surrounded by sugarcane. Surrounding the sugarcane fields were three savanna protected areas: Mbuluzi Game Reserve (MGR) and Mlawula Nature Reserve (MNR) to the east and Hlane Royal National Park (HRNP) to the west and south . The habitat in these areas consisted of acacia savanna, woodland and riparian areas. Sampling was conducted in accordance with the UF Institute of Animal Care and Compliance project #201307772. Sampling was permitted by the Royal Swaziland Sugar Association, Mbuluzi Game Reserve, Hlane Royal National Park and Mlawula Nature Reserve.

Live-trapping of rodents was done either within long-term grids (20 traps, 40 m x 50 m) located in MGR, MNR and HRNP (McCleery et al. 2018), or sampled within the sugarcane plantations with traps placed along sugarcane edges. For the latter, grids were separated by > 1 km. Trapping was carried out for three consecutive nights using Sherman traps baited with oats and peanut butter. Captured rodents were sexed, weighed, and had a distal portion (~ 1mm) of one pinna removed using sterile micro-scissors and preserved in Longmire's solution. Genomic DNA was extracted using QIAGEN DNeasy Tissue Kit (QIAGEN, Valencia, CA) and standardized to 20 ng/µl. We screened 20 microsatellite loci developed for *Mastomys* (Galan et al. 2004; Loiseau et al. 2007), and retained 14 loci (MH5, MH51, MH206, MH30, MH174, MH52, M59,

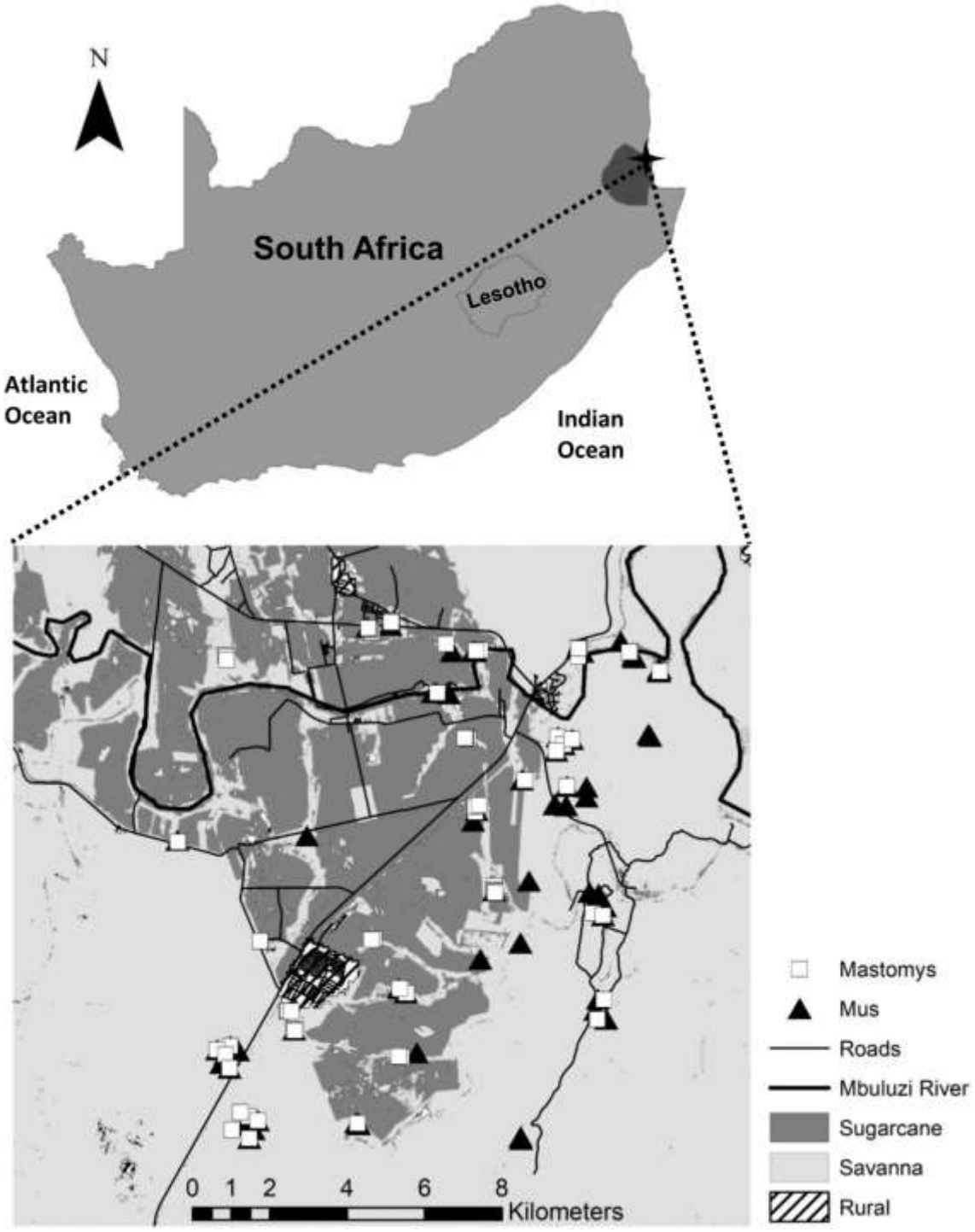

Fig. 1. Location of study and sample for *Mus* minutoides and Mastomys natalensis in NE Eswatini.

MH60, MH10, MH1, MH146, MH216, MH141, and MH146) based on their amplification and scoring success (Table 1).

Table 1. Microsatellite loci summary statistics and loci information for Mus minutoides and Mastomys natalensis sampled across the study area. Number of alleles (A), size range of alleles detected, observed H_O and expected H_E heterozygosity are given for each locus. The microsatellite motif from the original sequence data, and PCR primers are given for Mus. Sources for locus information are listed for Mastomys.

Species	*A*	Amplicon Size Range	H_O	H_E	Motif	Primer info (5′–3′)
			Mus minutoides			
Mus12	12	185–217	0.679	0.722	AAC_7	F – TGTGTGAAATCCATGCCTTG R – GCATTGGTTGTTGGTTCCTT
Mus29	16	231–262	0.738	0.900	GT_{26}	F – GGTATGGGCCACACAACTTT R – CAAAGGGAAGGACACATGCT
Mus18	14	132–195	0.667	0.899	$AGAT_8$	F – GCCCATCAACATTTGACCTT R – ATGACCCTGGCAGTTTTGTT
Mus14	11	191–229	0.959	0.874	AAT_9	F – GTCGTTGGAGGGGTCTGTAG R – ATTCCTGACCTTGGCTTTGA
Mus17	10	204–240	0.759	0.778	$AGAT_{10}$	F – CGGGTTCCTATGCCTGTATG R – CCACCGAGGATTGGTATTCT
Mus23	17	177–213	0.740	0.916	TG_{21}	F – GTGTCAAACTGCACAGCTT R – TCCATGCCAGCCTGTACTAA
Mus21	31	203–275	0.960	0.956	$[TA]_3T[TA]_{12}$	F – CCTCACTGGGTACCTGCATT R – CCATGTCTACCACCTTCAAACA
Mus24	16	168–204	0.738	0.851	CA_{30}	F – CCTGACAACTTCCCCTCTCA R – GCCAGGCGTAATAGAAACCA
Mus16	13	224–272	0.766	0.868	$AAAT_8$	F – CCTCCTGCTTAGGACACATGA R – GAAATTTGAAAGGGGGCTTC
			Mastomys natalensis			
MH51	16	123–155	0.895	0.897	TG_{27}	Galan et al. (2004)
MH30	15	254–288	0.781	0.893	TG_{21}	Loiseau et al. (2007)
MH5	18	105–135	0.838	0.898	GT_{18}	Loiseau et al. (2007)
MH206	14	179–211	0.743	0.795	TG_{24}	Galan et al. (2004)
MH174	14	331–357	0.867	0.894	TG_{17}	Galan et al. (2004)
MH52	14	104–132	0.899	0.876	TG_{20}	Galan et al. (2004)
MH60	40	160–196	0.904	0.959	TC_{28}	Galan et al. (2004)
MH1	17	359–389	0.909	0.902	$CA_{15}GA(CA)_7$	Loiseau et al. (2007)
MH39	6	143–153	0.514	0.560	GT_{39}	Loiseau et al. (2007)
MH10	4	332–338	0.198	0.306	$CA_7TA(CA)_8$	Loiseau et al. (2007)
MH146	20	125–169	0.941	0.936	TG_{13}	Galan et al. (2004)
MH216	16	179–215	0.897	0.891	TG_{13}	Galan et al. (2004)
MH141	15	254–284	0.863	0.862	TG_{22}	Galan et al. (2004)
MH133	19	334–378	0.899	0.931	TC_{35}	Galan et al. (2004)

Microsatellite markers were developed *de novo* for *Mus minotoides*. Genomic DNA was isolated from a single individual using the Qiagen DNeasy tissue protocol

(Qiagen, Valencia, California, USA). We used single molecule real-time sequencing (SMRT) cell technology on the PacBio RS II platform (Pacific Biosciences, California). In brief, we sheared DNA (140ng/uL), and annealed primers in accordance with PacBio protocols, generating fragment libraries with average fragment size of 2 Kb. We sequenced on a single SMRT cell using P4/XL chemistry. The ≥2x circular reads gave 46,023 and 54,900 unfiltered reads, and average raw read lengths of 10,687 and 10,342 bp, respectively. All analyses were performed on the consensus FASTA files. Low-quality (Q<20) sequence reads were trimmed prior to microsatellite identification. Resulting sequences were screened for di, tri and tetranucleotides with > 8 repeats using the program Msatcommander 0.8.2 (Faircloth 2008) and primers designed using PRIMER 3 (Rozen and Skaletsky 2000). Twenty-five *Mus* primers were initially tested on 3 to 8 individuals to determine amplification success and to evaluate allele morphology. We retained nine *Mus* loci that amplified consistently and produced allele morphology that was easy to score (Table 1; Genbank accession: TBA). All PCR products were run on an ABI Automated Sequencer 3130xl and analyzed using GeneMarker software (Softgenetics, State College, Pennsylvania). We re-genotyped 10% of samples from both species to confirm genotyping consistency and we used Micro-Checker (Van Oosterhout et al. 2004) to screen for patterns reflecting broad genotyping errors (allele dropout, null alleles and stuttering leading to scoring error).

Although we tested for similar patterns of spatial genetic structure between the two species using methods free from assumptions of Hardy-Weinberg equilibrium (HWE), we tested for HWE and non-random associations of alleles [Linkage disequilibrium (LD)] in order to evaluate the performance of the newly developed *Mus* SSR loci for subsequent use in substructured populations. Exact tests for HWE and LD were performed using Arlequin vers. 3.5 (Excoffier and Lischer 2010). Because non-

random mating can be confounded by population substructure and other biological factors (Waples 2015), we tested for HWE and LD using only the adult *Mus* collected from Mbuluzi Game Reserve (n = 35). We also used Arlequin to calculate observed (H_O) and expected heterozygosity (H_E) and allelic diversity (A) across all samples for each species.

We used STRUCTURE 2.3.4 (Pritchard et al. 2000) to evaluate how many discrete population clusters (*K*) were present in the data. We tested *K* at 1 to 10 for each data set, using the admixture model with correlated allele frequencies. Each value of *K* was run 20 times, using a parameter set of 50,000 burn-in and 150,000 MCMC replications after the burn-in. We calculated the average log probability of the data (L(*K*)) and standard deviation across the 20 runs for each *K* and then calculated the ln Pr(X|K) using the ad hoc delta K value (Evanno et al., 2005) to help infer which K was the most likely where different K values had similar likelihoods.

We used a multivariate approach implemented in Genalex ver. 6.0 (Smouse and Peakall 1999; Peakall et al. 2003), to compare the spatial signal of each species. Genalex calculates an autocorrelation coefficient (*r*) of genotypic similarity between pairs of individuals across a series of *a priori* defined distance classes using pairwise geographic distances (estimated in Genalex from x- and y-coordinates of trap locations) and pairwise squared genetic distance matrices (Peakall et al. 1995; Smouse and Peakall 1999). The resulting correlograms represent *r,* plotted as a function of distance class. Comparisons at distance classes, particularly at shorter distance intervals, where *r* is significant are considered spatially autocorrelated, or more genetically similar ($r > 0$) or dissimilar ($r < 0$) than expected if genetic similarity were random. Where estimates of *r* first intercept 0 defines the spatial scale of nonrandom positive genetic structure; however, this can vary depending on the choice of *a priori* interval class sizes. In

addition, power to detect structure will increase with sample size per interval (Peakall et al. 2003).

When conducting spatial autocorrelation analysis an important consideration is the selection of distance classes, as this can influence the interpretation (Vekemans and Hardy 2004). We first selected distance classes that produced even sample size (approximately 100) per distance class. This resulted in distance classes that varied because sampling was not uniform across the study area (see Fig. 1). For *Mastomys* there were 45 distance classes beginning with class 1 (0-145 m), and ranging to class 45 (12,882-13,438 m). For *Mus* there were 30 distance classes beginning with class 1 (0-553 m) and ending with 13,447-14,762 m; see results). Based on these analyses we then asked whether spatial heterogeneity existed in the scale of spatial autocorrelation between the two species, particularly at the smallest distances which was where spatial autocorrelation was pronounced in both species. In order to test this we needed identical distance interval class sizes, allowing us to specifically test for heterogeneity in *r* between *Mastomys* and *Mus* using the nonparametric test of Smouse et al. (2008). Briefly, identical distance classes across both species are used to calculate a pooled autocorrelation, representing the base autocorrelation levels under the null hypothesis of no difference between the species. We used fixed variable distance classes of: class 1 (0-600 m), class 2 (601-1769 m), class 3 (1770-2200 m), class 4 (2201-2500 m), class 5 (2501-3000 m), class 6 (3001-3500 m), class 7 (3501-4000), class 8 (4001-4500m), and class 9 (4501-5000 m). These distance classes represented a balance between maintaining adequate sample sizes per distance class (≥ 90 per species), while minimizing the skew in sample sizes among distance classes. Although the fixed variable distances classes represent non-standardized distance classes, they were necessary due to the patchy distribution of samples. We then evaluated the distribution

of random departure from this average by bootstrap resampling (Smouse et al. 2008). A squared paired-sample t-test statistic (t^2) was used for each distance class to test for deviations from the null hypothesis. Finally, the two species' correlograms were compared, drawing on the *P* values for the t^2 statistic at each distance class to compute the correlogram-wide 'Omega' (ω). We then determined the probability that the observed ω was larger than expected under the null hypothesis of homogeneous correlograms. Significance for spatial autocorrelation was determined at $\alpha \leq 0.01$ (Banks and Peakall 2012). All tests for statistical significance were performed using 999 random permutations to estimate distributions of *r* under the null hypothesis of no spatial autocorrelation ($r = 0$) with a 95% CI, and 999 bootstraps to estimate 95% confidence around *r* (i.e., 95% CI > 0).

We also explored spatial genetic patterns among individuals with the R package MEMGENE (Galpern et al. 2014). MEMGENE combines Moran's Eigenvector Maps (MEM) with a regression framework where genetic distance matrices are regressed against raw predictors (McArdle and Anderson, 2001). Eigenvectors were created from principal coordinate analysis of distance matrices, producing orthonormal variables that described both positive and negative spatial autocorrelation (Dray et al. 2006, Galpren et al. 2014). This analysis produced new spatially independent MEM scores that summarized the spatial relationship of genetic differences between sampled individuals (Galpern et al. 2014). The shared allele distance (Bowcock et al. 1994) was then estimated among pairwise individuals and the matrix was regressed against the predictor variables to identify MEM eigenvectors that described significant patterns of positive or negative spatial autocorrelation (Galpren et al. 2014).

Results

The average number of alleles from all samples of *Mus* (N = 80) and *Mastomys* (N = 97) were 17.4 and 15.7 respectively. The number of alleles per locus ranged from 10 to 31 in *Mus*, and 4–38 in *Mastomys*. Mean observed and expected heterozygosity across all samples were 0.778 and 0.863 for *Mus,* and 0.803 and 0.823 for *Mastomys.* Considering only the three loci (MH30, MH60, MH133) in common with *Mastomys* in Brouat et al. (2007) and Russo et al. (2018), observed and expected heterozygosity was higher in the present study. Mean expected heterozygosity for these 3 loci ranged from 0.89 to 0.95. Test for HWE for samples from Mbuluzi identified three *Mus* loci deviated from HWE following Bonferroni correction (Mus21 $P < 0.000$, Mus18 $P = 0.0006$, and Mus16 $P = 0.002$). Deviations were due to excess homozygous genotypes. The same three loci were significant when tested across the entire study area. No loci were out of HWE for 14 *Mastomys* collected at the same location, however, across the entire study area 4 loci had an excess of homozygosity (MH10, MH141, MH 206, MH133, $P < 0.003$), and 1 had an excess of heterozygosity (MH1 $P < 0.000$).

Clustering analysis from STRUCTURE runs on *Mus* found the highest P(*K*) and smallest variance for K = 1. Increases in K were uniformly lower in mean L(*K*) and high variance across replicates (Fig. 2). For *Mastomys*, mean L(*K*) increased slightly from K = 1 to K = 2 and K = 3, although the variance increased considerably from 0.92 to 38.49 and 19.28 respectively. Examining K = 3 for *Mastomys,* there was little evidence of geographic structuring (Fig. 2).

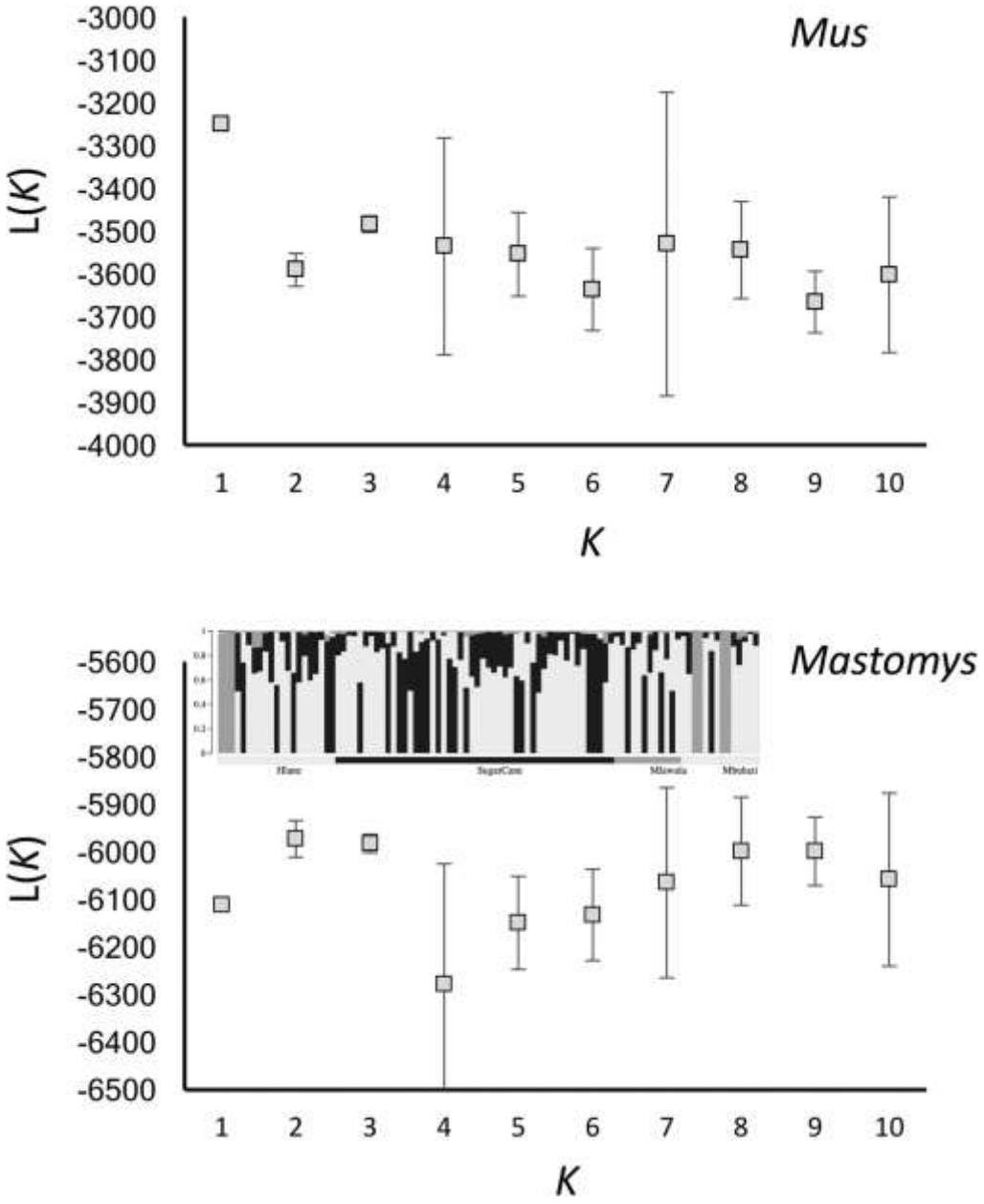

Fig. 2. Structure results for *Mus* minutoides and Mastomys natalensis. Plotted are mean (± S.D.) log probabilities of the data for estimates of the number of clusters (K) tested from K = 1 to K = 10. (Top) Results for *Mus* indicating that K = 1 has the highest log probability. (Bottom) Results for *Mastomys* higher support at K = 2 and K = 3. Results for K = 3 are shown in inset, and indicate no discernable geographic pattern of genetic structure. Results at K = 2 were ambiguous (see text).

Spatial correlograms indicated significant ω values for both species, indicating an overall departure from random (*Mastomys*, ω = 241.20, P = 0.001; *Mus* Omega = 117.28, P = 0.001). For *Mastomys* there were an average of 101.3 observations (range 99 -131) across 45 distance intervals (Fig. 3). The *r* values were positive and significant

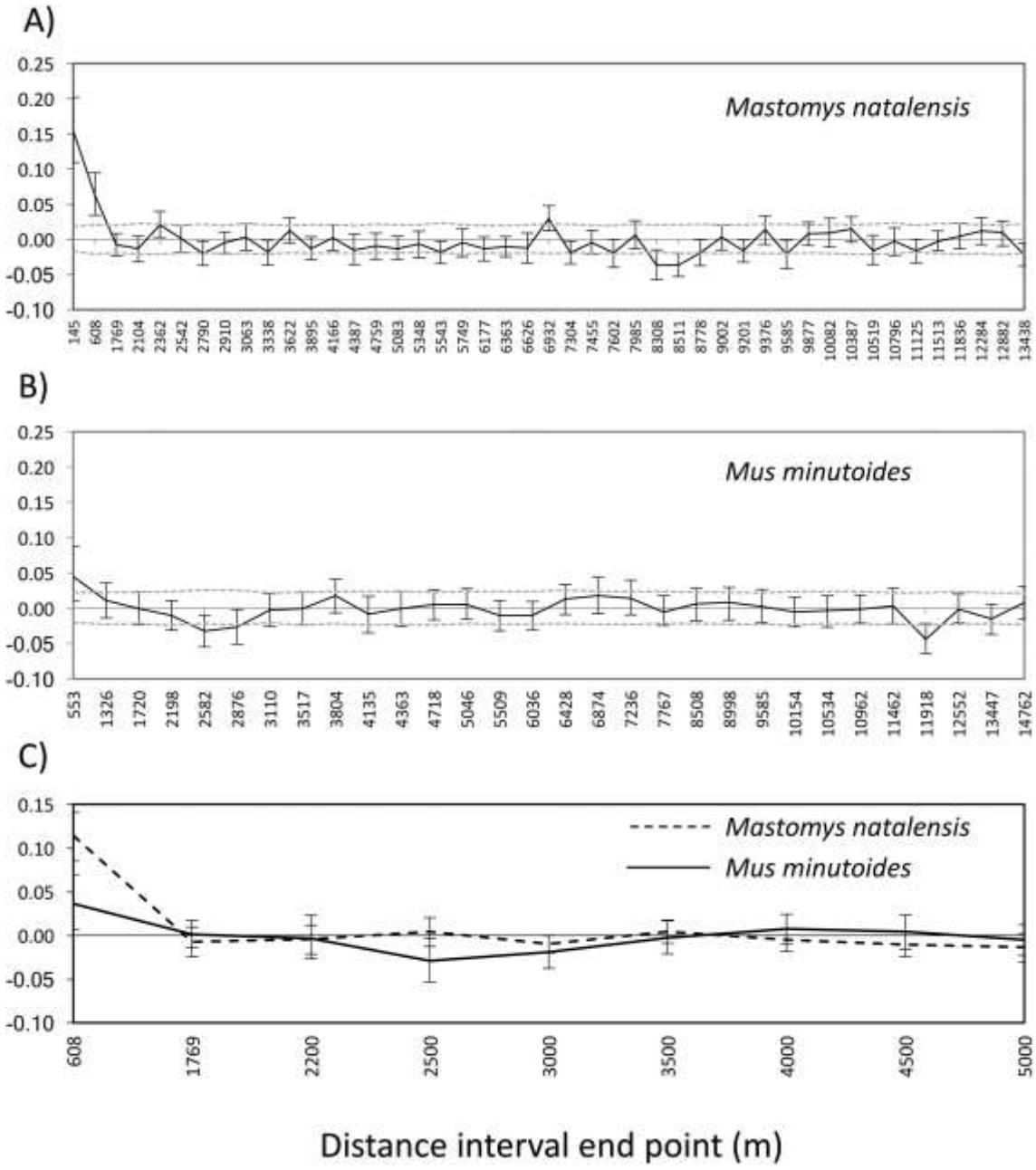

Fig. 3. Correlograms of genotypic similarity (*r*) across geographic distance intervals. Results from (A) Mastomys natalensis, and (B) *Mus* minutoides. The solid line tracks genotypic similarity, dashed lines represent the upper (U) and lower (L) 95% CI around random expectations, whereas bars around *r* show the 95% CI determined by bootstrapping. (C) Results from a test of *r* heterogeneity between *Mastomys* and *Mus*. There was significantly greater spatial autocorrelation in *Mastomys* at the 0–608 m distance class.

at the first two distance classes in *Mastomys* (145 m and 608 m, both P = 0.001) with an x-intercept at 1647 m. There were also significant positive oscillations (i.e. deviations from 0) at 6900 m (P = 0.007), and negative oscillations at 8308 m (*P* = 0.001) and 8511 m (*P* = 0.001). For *Mus*, with an average number of observations 102.9 (range 101-124) across 30 distance intervals, the smallest distance class of *r* was significant (553 m, *P* = 0.001), with an x-intercept of 1706 m (Fig. 2). There were two significant negative values at 2582 m (P = 0.004), and again at 11,918 m (P = 0.002).

Our test of heterogeneity between *Mastomys* and *Mus*, did not find significant combined spatial structure over the entire correlogram (nine distance classes $\omega = 15.85$, P = 0.024). However, the two species were significantly different at the first distance class (0-608 m, n = 385, $\omega = 45.08$, *P* = 0.001), with *Mastomys* reflecting significantly greater spatial autocorrelation than *Mus*; (*Mastomys r* = 0.114, 95% CI 0.087-0.143; *Mus r* = 0.036, 95% CI 0.003-0.066) ($t^2 = 9.990$, *P* = 0.002). There was no further detected heterogeneity across larger distance classes.

MEMGENE results highlighted the limited spatial structure of genetic information in both species. The amount of genetic variation that could be explained by spatial pattern was low ($R^2_{adj} = 0.0536$) in *Mastomys* and effectively 0 ($R^2_{adj} = -0.0001$) in *Mus*. The visualization of the first MEM variable score for *Mastomys* indicated that the central and southern portion of the sugarcane were differentiated from the eastern protected areas, whereas interior Hlane and northern sugarcane areas were genetically intermediate, with Hlane MEM scores being closer to the sugarcane, and most northern mice samples (north of the Mbuluzi River) with MEM scores closer to eastern protected areas of Mbuluzi and Mlawula (Fig. 4). The first MEM variable, representing the principal coordinate eigenvector, explained 31.2% of the spatial variation. Visualizing the MEM variable scores for *Mus* revealed genetic similarity between Hlane and

northeastern sugarcane *Mus*, and greater similarity for Mus in other parts of the sugarcane and eastern savanna locations (Fig. 4). The first MEM variable for *Mus* explained 54.8% of the variation.

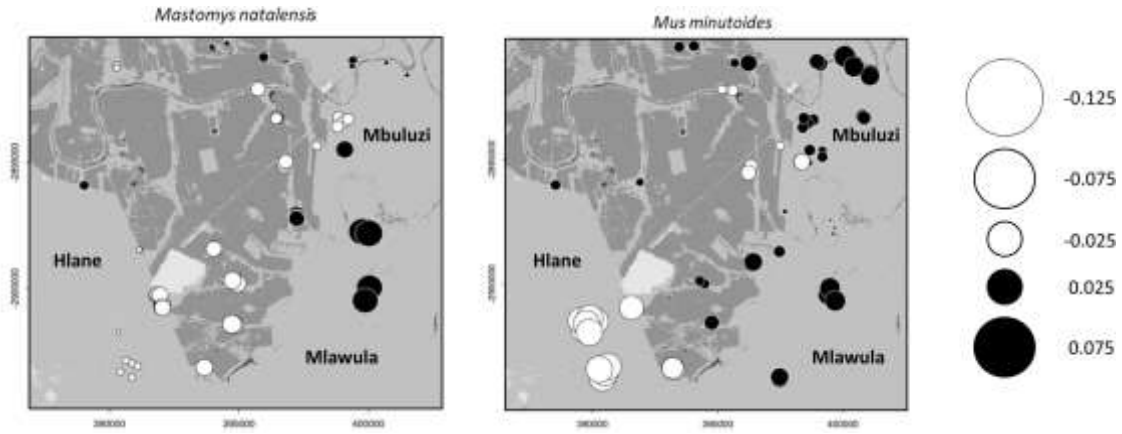

Fig. 4. Visualization of the first MEMGENE variables with scores superimposed over the landscape surface. Circles of similar size indicate rodents with similar MEME scores, and black and white reflect positive or negative (respectively) axis score values. Additional MEMGENE variables explained no spatial genetic pattern and were not presented.

Discussion

Overall, our spatial genetic analyses revealed limited spatial structure for both species in this agro-ecological landscape. For both species, spatial genetic structure was limited to only the smallest distance classes. The greater genetic similarity among individual Mastomys relative to *Mus* sampled at the shortest distance class of ≤600 m could be the result of differences in juvenile social structure represented by greater association of litter-mates or parent-offspring in *Mastomys* prior to dispersal, which would have the effect of inflating the relatedness (r) at this spatial scale (Van Hooft et al., 2008). While trapping *Mastomys* in this landscape, we observed multiple instances where individuals (often juveniles or subadults) were captured together (˜20 trap occurrences out of 670 trap nights, compared to 3 occurrences for *Mus*). Previous studies on *Mastomys* suggested that the species exhibits kin clustering, which would also result in fine

spatial-scale structuring (Van Hooft et al., 2008; Russo et al., 2018); however, Russo et al. (2018) found greater spatial structure, including spatial autocorrelation, at up to 10 km, which far exceeded what we observed here. Unlike our study area, their study encompassed a large (960 km^2) heterogeneous savanna (Hluhluwe-iMfolozi Park, South Africa), that lacked agricultural cultivation and any major areas of human-altered landscape, other than small tourist rest camps.

The lack of spatial autocorrelation beyond the first six hundred meters for both species suggests that they are capable of high rates of dispersal, which would result in a lack of isolation by distance across the spatial scale examined in our study. Rodent dispersal is likely a response to a number of variables including mate availability, habitat quality and intraspecific competition. *Mastomys* is an opportunistic generalist that has been characterized as somewhat dependent on water resources (Chimimba et al. 2005), and capable of moving over large distances (e.g. 400 m, Leirs et al. 1996a and over 100 m in agricultural landscapes Monadjem et al. 2011). In this area of sugarcane agriculture, irrigation is used extensively, which combined with abundant food resources may increase rodent abundance and dispersal capability.

The phenotypic plasticity displayed by *Mastomys* has previously been shown to translate to population genetic differences within the genus. In Senegal Mastomys natalensis sampled from peri-urban environments displayed vastly different spatial genetic structure compared to *Mastomys erythroleucus* which was sampled at the same scale from native savanna vegetation. In this instance, peri-urban mice had significant spatial structure, whereas those in native habitats displayed panmixia (Brouat et al., 2007). In our study system both *Mastomys* and *Mus* displayed patterns of panmixia at most spatial scales. The similar lack of genetic spatial structure in *Mus* as in *Mastomys* may reflect a common ability to respond quickly following regular turnover in

sugarcane agricultural blocks. *Mastomys* has been characterized as a pioneer species, able to colonize areas that are recovering from habitat destruction (Meester et al., 1979; Monadjem and Perrin, 2003), including recently harvested sugar cane fields. For both *Mus* and *Mastomys*, the regular irrigation, abundance of insects and other resources combined with annual massive disturbance may result in conditions leading to mass re-colonizations following burns from surrounding sugar cane. This is predicted to result in high diversity and limited differentiation at the landscape scale (Banks et al., 2013).

The patterns reflecting genetic panmixia found for both species illustrate the genetic outcomes for species with omnivorous habits and plasticity in habitat selection. Stomach content analysis has confirmed that *Mastomys* has a generalist diet with some seasonal variation suggesting opportunism (Leirs and Verheyen, 1994; Monadjem, 1997). Both of these species have been found throughout the habitat mosaic of this agro-ecological system (Hurst et al., 2013) and the most likely scenario for the observed genetic structure across the landscape is the ability of each species to utilize both edge and the interior of patches. Both species have been found utilizing the edge of the savanna-sugarcane interface as well as hundreds of meters within the interior of these habitat patch types (Hurst et al., 2013, this study). Also, sugar cane harvesting may serve as a mechanism for enhancing dispersal in small mammal populations by continuously disturbing habitat, resulting re-colonization. Spatio-temporal variation in habitat quality across the study site as a result of different land-use types seems to favor dispersal and might be a crucial determinant of such extended populations. However, sugarcane at every stage of growth appears to be highly suitable for both species.

Representatives from both species were also captured in savanna habitat, peripheral to the main area of sugar cane. However, there was no suggestion that habitat type had an impact on spatial structuring in either species. For example, There was no

clear pattern resulting from the STRUCTURE analyses to suggest savanna or sugarcane represented separate source populations. Similarly, MEMGENE analysis is expected to be useful in identifying landscape features that may be influencing gene flow, even when levels of gene flow are high, and spatial patterns are cryptic (Galpern et al. 2014). A small proportion ($R^2 = 0.05$) of the genetic variation was explained by spatial pattern in *Mastomys*, and our visualization suggests that this variation may be largely associated with an east-west pattern linked with the eastern boundary between sugarcane and savanna. However, we did not find a similar relationship with the western boundary. We found no spatial association in our *Mus* data set, although these results are likely affected by small sample sizes.

Our observation of genetic panmixia in these two species suggests that both have high dispersal capabilities through this dynamic and heterogeneous agro-ecological landscape. Rodent dispersal distances are poorly understood for most species; however, there is increasing evidence for relatively rare long-distance movements (Austin et al., 2015; Estes-Zumpf et al., 2010; Le Galliard et al., 2011). In some instances, agricultural landscapes appear to act as partial or complete barriers to dispersal (Estes-Zumpf et al., 2010), and have been associated with long-term declines of rodent populations with increasing area of crop production (Butet and Leroux, 2001). In the case of *Mus* and *Mastomys*, abundance has been shown to be higher in this sugarcane system than in adjacent savanna (Hurst et al., 2014) suggesting that this type of agriculture is favorable for these generalists. During a recent drought in this region, we found a greatly reduced abundance of *Mastomys* in savanna (Hlane, Mbuluzi and Mlawula) relative to sugarcane (unpublished data), suggesting that irrigated landscapes maintain a positive demographic effect on generalist rodent species.

These findings have implications for both pest management and rodent-borne disease control. Heterogeneous habitat composition has previously been suggested as a way to control outbreaks of cyclic microtine rodent pest species (Stenseth, 1977). Vegetation height, particularly the use of mowing and harvesting in agro-ecosystems has been used to control rodents locally (Jacob, 2008). Although we did not directly test for this observed pattern, our results suggest that different heights of sugarcane, from the 11-month harvest rotation, did not appear to create the genetic dissimilarities expected when dispersal and colonization were disrupted. The inferred high rates of dispersal suggested by this study illustrate how these two species could be efficient vectors of disease by connecting naïve with infected populations as a result of their high rates of dispersals, particularly in agriculturally intensive areas.

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