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RESEARCH ARTICLE

# Using genetic analysis to estimate population size, sex ratio, and social organization in an Asian elephant population in conflict with humans in Alur, southern India

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**Abstract** With growing human and, possibly, elephant populations and a drastic increase in anthropogenic activities, human–elephant conflict in Asia has been on the rise. The Alur area in Karnataka state, southern India, is one such case in point, which has witnessed increasing levels of human–elephant conflict over the last two decades. The tiny, moderately protected habitat available for elephants in this human-dominated landscape does not appear to be able to support elephants over the long term. Options to deal with the escalating conflict include translocation of elephants, bringing elephants into captivity, and culling. We carried out a molecular genetic study of elephants in the Alur area to estimate the minimum number of elephants using the area, the sex ratio, genetic relatedness between individuals, and genetic structure with regard to the larger population in the landscape, so that informed management decisions could be made. Fresh dung samples were collected from the field and genotyped using 12 microsatellite loci. We found 29 unique individuals in the population, comprising 17 females and 12 males of different age classes. Relatedness between females suggested independent colonisations by discrete, small groups rather than by one cohesive clan of related females. This obviates the need for a single solution for dealing with all the females in the area in order to maintain social integrity, and has implications in terms how these elephants can be dealt with. We demonstrate how social organization inferred through molecular data from non-invasive sampling can inform management decisions.

**Keywords** Asian elephant · Genetic relatedness · Hassan district · Human–elephant conflict · Non-invasive sampling · Social organization

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## Introduction

Growing human populations and shrinking habitats have resulted in increased contact between wildlife and humans, leading to conflict of various kinds (damage/loss of crops, livestock, and property, loss of human and animal lives, disease transmission, increased stress, and changes in animal home ranges) (see Thirgood et al. 2005; Woodroffe et al. 2005; Sillero-Zubiri et al. 2007, and references therein). Conflict from large mammals such as elephants, bears, and wolves can be high (for example, Barnes 1996; Madel 1996; Bist 2002; Treves et al. 2002) or perceived to be high because of potentially catastrophic conflict events that they can cause (Naughton-Treves 1997). In either case,

addressing human–wildlife conflict is important in these species' conservation (Sillero-Zubiri et al. 2007). The Asian elephant (*Elephas maximus*) is one such large mammal, distributed across South and Southeast Asia (Hedges et al. 2008), that enters into conflict with humans and is of immediate conservation concern. Although the species has been an integral part of the human cultural milieu, increasing anthropogenic activities and expansion of human settlements have led to extensive fragmentation of forests and increasing elephant–human conflict in many parts of the Asian elephant's range (Leimgruber et al. 2003).

While the Asian elephant is endangered globally and has a global population size an order of magnitude smaller than those of its African cousins, a few populations of the Asian elephant have been growing over the last few decades (Bist 2002). However, these populations too are not immune from elephant–human conflict. One such growing population is the world's single largest Asian elephant population, in the Nilgiris-Eastern Ghats Reserve in southern India, which is estimated to hold over 8,500 elephants (Project Elephant data shown in Rangarajan et al. 2010). Censuses in the key conservation areas of this landscape have shown a significant increase in elephant population (Appayya 1995; Baskaran and Desai 2000), but there has also been a simultaneous decline in forest cover in parts of this landscape (Elouard 2000), and an increase in infestation by weeds such as *Lantana camara*, which may negatively affect elephants (Wilson et al. 2013). Since less than a quarter of the elephant habitat in southern India falls under Protected Areas, which provide strict legal protection for wildlife (Bist 2002), elephant population growth, along with habitat loss, fragmentation, and degradation, result in elephants dispersing out of Protected Areas. This combination of factors has exacerbated human–elephant conflict in southern India over the past few decades.

Dispersal of Asian elephants into new habitats, resulting in human–elephant conflict, has been dealt with by the construction of trenches/fences at forest edges, elephant drives to herd elephants back towards older habitat, capture and maintenance in captivity, translocation, and rare killing of rogue elephants (Appayya 1995, Baskaran et al. 2011). However, there has often not been much scientific data available before and after the management intervention. Public hearings and even opinions within the scientific community consequently reveal polarized views about the management intervention that should be followed. While the decision to cull would additionally depend on cultural acceptance, scientific data on population number and structure, habitat availability and connectivity, and the origin of small populations are required to decide between other management options such as capture and bringing into captivity, translocation, and maintenance of the population in situ with some conflict mitigation measures.

We carried out the present study in the Alur and Yesalur Forest Ranges in Hassan Forest Division and Shanivarasanthe Forest Range of Madikeri Forest Division (henceforth, collectively referred to as the Alur area) in Karnataka state, southern India, in order to obtain data on population number and structure that could be used to weigh management options. This area has a small, isolated elephant population in the midst of agriculture, coffee plantations, and human settlements, resulting in high levels of human–elephant conflict (Karnataka Elephant Task Force 2012). Our specific objectives were to (1) determine the minimum number of individuals in the area so that the desirability of keeping the population in the area, impact of removal on the remaining total population, and feasibility of removal could be assessed, (2) determine the age-sex structure of the population because subadult and adult males can be removed singly while females and dependent offspring would have to be removed as social groups, (3) identify the social structure of female groups, which would tell us whether there were multiple colonizations by females, and whether the females would have to be all managed as a single unit or multiple units in the case of removal/translocation, (4) identify the relatedness between the Alur area population and elephants in the larger landscape, which may influence decisions about where they can be translocated.

Small populations of forest dwelling animals are difficult to count reliably (Barnes 2002), even if they are elephants, and indirect methods including genetic techniques often prove to be useful in such efforts (for example, Hedrick 1995; Taberlet et al. 1997; Eggert et al. 2003; Vidya et al. 2007; Hedges et al. 2013; Gray et al. 2014). Genetic methods can also be used to sex elephants (for example, Fernando and Melnick 2001; Vidya et al. 2003; Ahlering et al. 2011) and glean some information about their social structure (Fernando and Lande 2000; Nyakaana et al. 2001; Vidya et al. 2005, 2007; Ahlering et al. 2011) in the absence of significant amounts of behavioural data. We, therefore, used molecular genetic techniques in concert with non-invasive sampling of dung to carry out this study.

## Study area

The Alur area (Fig. 1) lies to the northwest of the Nilgiris-Eastern Ghats Reserve and comprises Alur and Yesalur Forest Ranges in Hassan Forest Division, Hassan district, and Shanivarasanthe Forest Range of Madikeri Forest Division, Kodagu district. The Alur area is a mosaic of largely coffee estates, rice, maize, banana, coconut, and arecanut fields, and fallow land. Forest patches exist inside

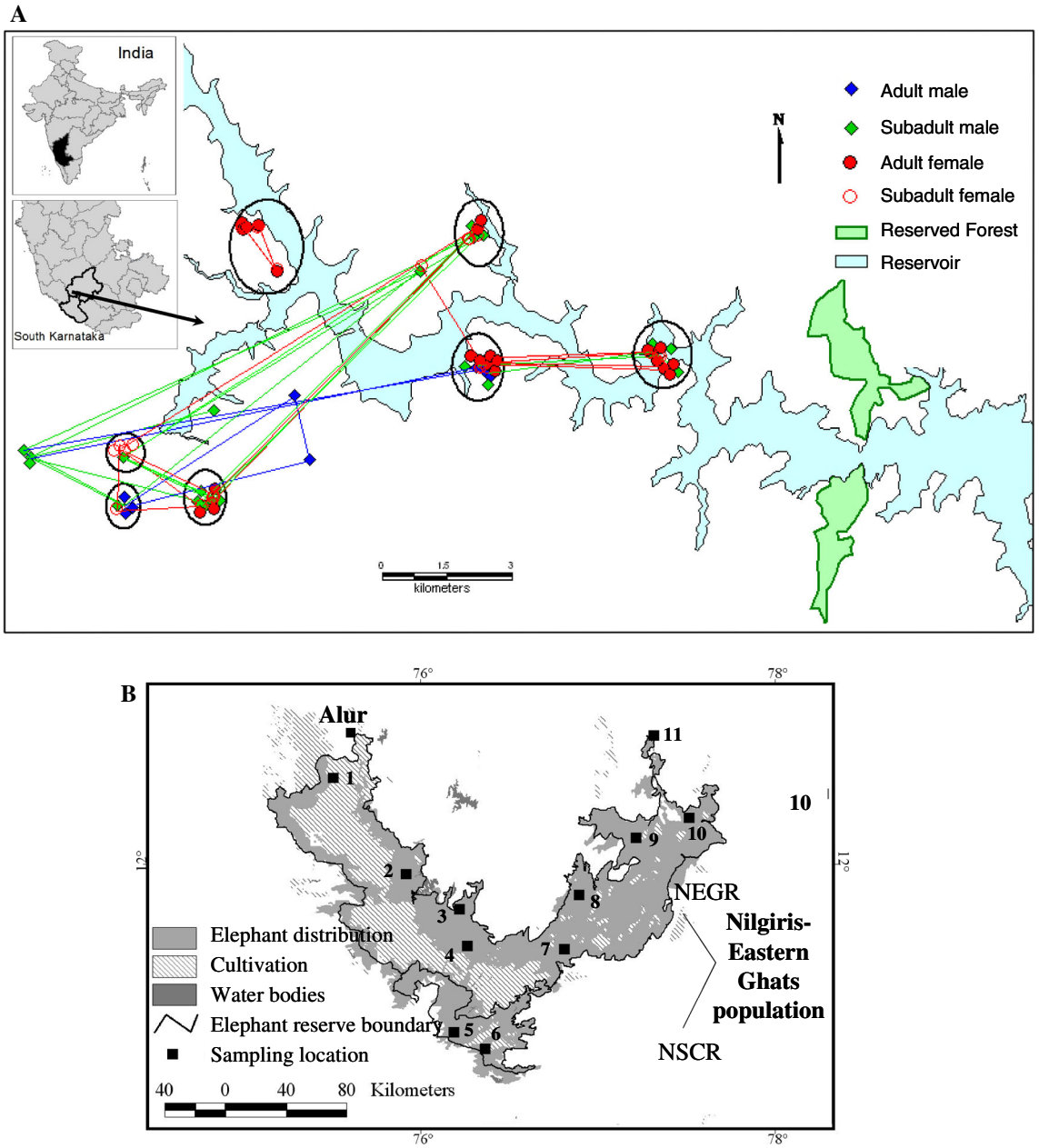


**Fig. 1** **A** Study area showing the locations of dung samples collected, the Hemavathi Reservoir, and the two largest forest fragments in the Alur area. The rest of the area is largely under coffee and agriculture. The reservoir boundary applies to the wet season and, therefore, there are dung sample locations in the area, which was dry during our field sampling season. Clusters of spatially close dung samples from subadult and adult females which were used for examining genetic relatedness are encircled. Lines connect the same individuals moving across the area within the sampling period. Inset: India map and map of south Karnataka with Hassan (*above*) and Kodagu (*below*) districts outlined in bold. **B** Map of the Nilgiris-Eastern Ghats landscape with the location of Alur, and the other locations that were compared with Alur. The location numbers on the map of the Nilgiris-Eastern Ghats landscape correspond to the locations in Table 5

coffee estates, largely along hill slopes and ridges: the two largest forest patches (Kattepura and Doddabetta Reserve Forests) together cover only 6 km$^2$ and are managed by the State Forest Department (Karnataka Elephant Task Force 2012). The Alur area is isolated from other forest areas: there are larger forest patches 9–10 km to the west, in Sakleshpur, but this area too is fragmented and has sub-optimal habitat for elephants. To the south, approximately 20 km away, are the fragmented and highly degraded forest patches of Madikeri Forest Division, which are a source of severe human–elephant conflict. Good elephant habitat is found even further south of the Alur area, approximately 40 km away, where contiguous forest habitat (moist deciduous forest) begins.

The Alur area does not seem to have had any sizeable elephant population historically, and satellite imagery before the construction of the Hemavathi Reservoir in 1973 in the area does not show much forest even then (Karnataka Elephant Task Force 2012). The elephants in the Alur area seemed to have originated from Kodagu in the south, which had experienced an 18 % loss in overall forest cover (originally 2,566 $km^2$) and 46 % loss of dry and moist deciduous forests (which are preferred by elephants), largely to coffee plantations, within two decades (Elouard 2000). The initial population comprised only subadult and adult males in the 1980s. These males caused severe conflict around the Kattepura and Doddabetta Reserve Forest areas and several of them were subsequently captured and translocated, while some were brought into captivity, and most of the remaining males were poached for ivory. New males from the south moved into this area, but it was only from 1993 that the area began to be colonised by females. Elephant deaths due to retaliatory killings ensured that the population remained low, making it a sink population for dispersing elephants from the south (Karnataka Elephant Task Force 2012).

Despite the presence of an estimated small number of elephants, human–elephant conflict in this area is high, with 276 reported elephant attacks on people between 1986 and 2006, resulting in 33 human deaths and 243 humans injured (Karnataka Elephant Task Force 2012). Human–elephant conflict here has also been increasing spatially, with 38 villages being affected by elephants in 2007 and 79 villages being affected by 2012, and in intensity, with 13 people being killed between 2007 and 2011 and nine elephants being killed in a 3-year period around the same time (Karnataka Elephant Task Force 2012). Therefore, the Alur area was cited as a removal zone, from which it was necessary to remove elephants because of the unsustainability of the present situation (Karnataka Elephant Task Force 2012).

## Methods

### Sample collection

Fresh elephant dung samples were collected during May 2012 from Alur, Yesalur, and Shanivarasanthe Ranges (see Fig. 1 for locations of dung samples). Sampling was based on information about the presence of elephants in various areas and covered the entire area under human–elephant conflict. Sampling was carried out in collaboration with the Forest Department and a network of contacts with at least one informer per village was set up in all villages in the area used by elephants. Contacts were selected based on their familiarity with elephants, their contacts with other informers in the village and also those who regularly reported elephants to the Forest Department. During the dung collection period, all these contacts were called daily to ascertain the presence of elephants near villages, and villages that reported elephants were visited for dung sample collection. Most of the samples collected were associated with direct sightings of elephants and were, therefore, fresh. GPS locations of sample sites were recorded. The outermost layer of dung, which is rich in endothelial cells, was collected into 95 % ethanol and stored at ambient temperature. The maximum circumferences of the intact dung boli were also measured and mean circumferences calculated for each set of boli (each animal). Based on Tyson et al. (2002), we assigned age classes to individuals based on mean circumference as <30 cm being juveniles or calves, 30–42 cm being subadults, and >42 cm being adults.

### Genetic analysis

DNA extraction from dung samples followed Fernando et al. (2003) and consisted of digesting approximately 0.5 g of the sample in a digestion buffer with SDS and Proteinase K overnight, extracting DNA with phenol:chloroform:isoamyl alcohol, and purifying it using QIAGEN gel extraction kit with the manufacturer's protocol. DNA extracts were stored at −20 °C. A set of 12 microsatellite markers isolated from elephants, LafMS03, LafMS02, LafMS05 (Nyakaana and Arctander 1998), EMX-2 (Fernando et al. 2001), Emu03, Emu04, Emu12, Emu14, Emu15, Emu17 (Kongrit et al. 2008), Fh60, and Fh94 (Comstock et al. 2000), were used to genotype samples. PCRs was carried out in 12.5 µl volumes, using 2 µl DNA, 0.25 µl of each primer (10 µM; the forward primer was fluorescently labelled), 0.1 µl Taq DNA polymerase, and 9.9 µl master mix (containing $MgCl_2$, Tris, KCl, dNTPs, and BSA, adjusted to pH 8.4). The annealing temperatures used for the 12 loci were as follows: Emu17, LafMS05: 58 °C; Emu15, Emu04, Fh94, Emu03: 63 °C; Emu14, Fh60: 65 °C; LafMS03: 54 °C; EMX-2, LafMS02: 62 °C; Emu12: 61 °C. For all loci, a 93 °C denaturation for 1 min, annealing for 1 min, and a 72 °C extension for 1 min were employed, followed by an extension at 72 °C for 15 min after the completion of all 40–42 cycles. Samples that did not show high (>4,000 in electropherograms) or clear peaks or showed mismatches were repeated. Two matching genotypes of heterozygotes or homozygotes (if the peak sizes were high) were considered to be the true genotype. Because of the good quality of samples collected and standardized protocols (see Fernando et al. 2003), we did not have a problem with low amplification or many mismatches (see "Results" section). Dedicated areas and instruments were used for low-copy number DNA and amplified products in order to prevent cross-contamination between samples. Aerosol resistant filter tips were also always used while

pipetting. The microsatellite loci were electrophoresed in an ABI 3730 DNA Analyzer and scored using the ABI Gene Scan analysis software v.3.1.2 (Applied Biosystems Inc.).

Molecular sexing was carried out using primers ZF79F 5′-AAATGCACAAGTGTAAATTCT-3′, ZF324R 5′-GAA TGGCATCTCTTTACTATG-3′, and ZFY161R 5′-TACTG GGGAGAAACCCA-3′ (Prithiviraj Fernando, pers. comm.) to amplify a ~265 bp fragment from the X chromosome and two bands from the Y chromosome. The internal primer, ZFY161R, was used so that the Y-specific band would be amplified separately. Thus, females exhibited a single band and males exhibited two bands, owing to the presence of Y-chromosome in males. Such molecular sexing using ZFX–ZFY polymorphism can reliably identify males and females (Fernando and Melnick 2001) present in a study area from non-invasively collected faecal samples (Vidya et al. 2003). Once the number of unique individuals was found based on 12 microsatellite loci, molecular sexing was carried out on samples belonging to the same individual so that we could confirm the sex of the individual and rule out allelic dropout (that could potentially lead to males being misidentified as females).

### Data analysis

Microsatellite loci were checked for Hardy–Weinberg equilibrium, linkage equilibrium, and genetic diversity using GenePop v.4.2 (Rousset 2008), and for null alleles using Micro-Checker v.2.2.3 (Van Oosterhout et al. 2004). The probability of identity ($P_{ID}$) (Paetkau and Strobeck 1994), which is the probability of any two individuals in the population showing the same genotype, and $P_{ID}$(sib) (see Evett and Weir 1998), the probability of two siblings showing the same genotype, were also calculated. These measures become smaller with increasing number of loci used and should be sufficiently small to reliably differentiate between individuals. The software IDENTITY (Wagner and Sefc 1999) was used to calculate $P_{ID}$ and $P_{ID}$(sib). Genetic relatedness between individuals was calculated using Queller and Goodnight's (1989) relatedness estimator in the software COANCESTRY v.1.0.1.1 (Wang 2011). Pedigrees were constructed using COLONY 2.0.5.0 (Wang 2008; Jones and Wang 2010), which allows for inference of parentage and sibships by taking the likelihood of the entire pedigree into account.

$F_{ST}$ values between the Alur elephants and elephants previously sampled from the larger Nilgiris-Eastern Ghats landscape were calculated using Arlequin v.3.1 (Excoffier et al. 2005). $F_{ST}$s were calculated using loci common to the present study and that of Vidya et al. (2005), which were LafMS03, LafMS02, and EMX-2. The first two were the most variable loci of the previous study. The sizes of PCR products scored from the ABI 3730 DNA Analyzer in the current study were 1–3 bp shorter (depending on the locus) than those from the ABI 377 DNA Sequencer used in Vidya et al. (2005). The alleles were matched before analysis based on the genotypes of individually identified elephants from southern India that had been genotyped on both sequencers. We also looked for signs of fine-scale population genetic structure that could be used to assign the Alur area samples to one of the locations in the larger Nilgiris-Eastern Ghats landscape using STRUCTURE v.2.3.4 (Pritchard et al. 2000, Hubisz et al. 2009). We used 10 independent runs, each with 20,000 steps as the burnin period and 200,000 MCMC iterations. Sample group information was used but ancestry of the Alur population was set as unknown (POPFLAG = 0), and admixture was allowed. The values of K (number of populations) used ranged from one to nine and the ΔK method of Evanno et al. (2005) was used to find the optimal K. Mitochondrial haplotypes were not examined in this study because of the absence of mitochondrial DNA variability across the entire Nilgiris-Eastern Ghats landscape (Vidya et al. 2005).

## Results

A total of 101 fresh dung samples were collected and DNA extracted from them. The average amplification success across the 12 microsatellite loci was 98 %. All the loci used were polymorphic, with 2–6 alleles per locus, showed moderate to high heterozygosity, and were in Hardy–Weinberg equilibrium ($P > 0.01$, Bonferroni corrected $P$ value cutoff = 0.004) (Table 1). There was no evidence for null alleles at any locus. No significant linkage disequilibrium was observed between any pair of loci after Bonferroni correction (Supplementary Material 1). The combined $P_{ID}$ across 12 loci was $1.24 \times 10^{-8}$ and combined $P_{ID}$(sib) was $1.13 \times 10^{-3}$. Therefore, the probability of wrongly identifying two different individuals as the same individual was miniscule.

From the 101 dung samples collected, we found 29 unique individuals. This did not change even when allowances for genotyping error, of up to three alleles, were made, indicating the robustness of this result. Molecular sexing showed 17 females and 12 males (Table 2). The genetic relatedness (mean ± 1.96 SE) between adult females ($n = 10$ comparisons) was found to be 0.012 ± 0.157 and that between adult and subadult females ($n = 45$), 0.016 ± 0.079, indicating that they were not related to one another overall. The relatedness between (I) adult females and adult males ($n = 20$), (II) adult females and subadult males ($n = 35$), (III) adult males ($n = 6$), (IV) subadult males ($n = 21$), and (V) adult and subadult males ($n = 28$) were all also not significantly greater than zero (Table 3). We also examined the

**Table 1** Number of alleles, observed heterozygosity, *P* value for the Hardy–Weinberg equilibrium test (HWE P), and allele sizes and frequencies for the 12 loci used

| Locus | No. of alleles | $H_{obs}$ | HWE P | Allele size/frequency | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| EMX-2 | 2 | 0.379 | 0.425 | 219 | 225 | | | | |
| | | | | 0.328 | 0.672 | | | | |
| Emu04 | 3 | 0.310 | 0.394 | 97 | 99 | 103 | | | |
| | | | | 0.121 | 0.793 | 0.086 | | | |
| LafMS02 | 3 | 0.759 | 0.337 | 135 | 137 | 141 | | | |
| | | | | 0.224 | 0.517 | 0.259 | | | |
| Emu12 | 4 | 0.793 | 0.839 | 139 | 141 | 148 | 152 | | |
| | | | | 0.483 | 0.259 | 0.121 | 0.138 | | |
| LafMS05 | 4 | 0.517 | 0.735 | 144 | 150 | 152 | 156 | | |
| | | | | 0.052 | 0.069 | 0.672 | 0.207 | | |
| FH94 | 4 | 0.552 | 0.776 | 215 | 217 | 221 | 229 | | |
| | | | | 0.293 | 0.621 | 0.052 | 0.034 | | |
| LafMS03 | 4 | 0.690 | 1.000 | 137 | 139 | 149 | 155 | | |
| | | | | 0.517 | 0.293 | 0.155 | 0.034 | | |
| Emu03 | 4 | 0.655 | 0.497 | 134 | 136 | 138 | 140 | | |
| | | | | 0.259 | 0.552 | 0.086 | 0.103 | | |
| Emu15 | 4 | 0.759 | 0.255 | 144 | 152 | 154 | 156 | | |
| | | | | 0.362 | 0.293 | 0.138 | 0.207 | | |
| Emu17 | 5 | 0.621 | 0.942 | 120 | 122 | 124 | 126 | 128 | |
| | | | | 0.069 | 0.276 | 0.552 | 0.052 | 0.052 | |
| Emu14 | 5 | 0.630 | 0.016 | 129 | 131 | 133 | 137 | 145 | |
| | | | | 0.056 | 0.019 | 0.537 | 0.019 | 0.370 | |
| FH60 | 6 | 0.821 | 0.490 | 148 | 152 | 154 | 156 | 158 | 162 |
| | | | | 0.179 | 0.107 | 0.268 | 0.250 | 0.125 | 0.071 |

For EMX-2, LafMS02 and LafMS03, the allele sizes written here correspond to the sizes in Vidya et al. (2005)

relatedness within groups of adult and subadult females, whose dung samples were spatially close to one another. Among seven such spatial clusters (Fig. 1), only two showed high relatedness (Table 4). When we examined individuals that were sampled from the same locations (either when elephants were directly sighted or when dung piles were collected from the same resting spot) on the same day, the average (±1.96 SE) relatedness between adult and subadult females was 0.105 (±0.107). This was found to be low because of two females that associated with several unrelated females. Upon excluding these two females (F_16 and F_26), the relatedness between adult and subadult females was 0.220 ± 0.103 (Table 3). Thus, there seemed to be at least some family groups in the area (see Fig. 2). The adult and subadult males sampled within a day of each other in the same vicinity were unrelated (−0.034 ± 0.164). However, while males were unrelated overall, two pairs of males were highly related and identified as sibships (using COLONY software) (R = 0.329, 0.482; Fig. 2). A third pair of males had a high R value (R = 0.386) but was not identified as a sibship. Construction of a pedigree showed related groups of individuals, but, more interestingly, that there were sets of offspring sired by the same males (Fig. 2). For example, three of the four juveniles in the area were sired by a single adult male (M_12), currently present in the Alur area. Therefore, there were paternal relatives that spanned different female groups (Fig. 2).

**Table 2** Number of males and females of different age classes

| Age class | Male | Female |
|---|---|---|
| Adult | 4 | 5 |
| Subadult | 7 | 9 |
| Juvenile/calf | 1 | 3 |

Pairwise $F_{ST}$s between Alur and the other locations in the Nilgiris-Eastern Ghats landscape [comprising the Nilgiris-Eastern Ghats Reserve (NEGR) and the more southern Nilambur-Silent Valley-Coimbatore Reserve (NSCR)] were not significant except for the two locations sampled in NSCR (Table 5). There was hardly any division in the larger population as revealed by STRUCTURE either. The value of ΔK was highest for K = 2, but the

**Table 3** Mean, standard error, and 95 % confidence intervals of relatedness between different categories of animals

| Category | *N* | Mean | SE | Lower CI | Upper CI |
|---|---|---|---|---|---|
| Adult females | 10 | 0.01 | 0.080 | −0.145 | 0.169 |
| Subadult females | 36 | −0.01 | 0.031 | −0.067 | 0.055 |
| Adult males | 6 | −0.11 | 0.121 | −0.351 | 0.124 |
| Subadult males | 21 | −0.12 | 0.046 | −0.211 | −0.030 |
| Adult females and subadult females | 45 | 0.02 | 0.040 | −0.063 | 0.095 |
| Adult females and adult males | 20 | −0.09 | 0.044 | −0.173 | −0.002 |
| Adult females and subadult males | 35 | 0.02 | 0.047 | −0.076 | 0.106 |
| Subadult females and adult males | 36 | −0.07 | 0.030 | −0.127 | −0.009 |
| Subadult females and subadult males | 63 | −0.03 | 0.032 | −0.097 | 0.027 |
| Adult males and subadult males | 28 | −0.07 | 0.040 | −0.149 | 0.007 |
| Adult and subadult females at the same location, sampled on the same day | 22 | 0.11 | 0.054 | −0.002 | 0.212 |
| Adult and subadult females at the same location sampled on the same day, excluding F_16 and F_26 | 15 | 0.22 | 0.052 | 0.117 | 0.322 |
| Adult and subadult males at the same location, sampled on the same day | 12 | −0.03 | 0.084 | −0.198 | 0.130 |

**Table 4** Mean and variance of the Queller and Goodnight relatedness estimate within spatial clusters of subadult and adult females without using a time criterion

| Groups | Mean relatedness | Variance |
|---|---|---|
| G1 | 0.355 | 0.047 |
| G2 | −0.044 | 0.053 |
| G3 | 0.192 | 0.034 |
| G4 | 0.008 | 0.094 |
| G5 | −0.055 | 0.034 |
| G6 | −0.092 | 0.001 |
| G7 | 0.034 | 0.000 |

value itself was small (Supplementary Material 2), indicating low magnitude of structuring, and ΔK cannot be calculated for K = 1 (Evanno et al. 2005). A plot of the posterior probabilities based on K = 2 showed that NSCR was differentiated from NEGR (Supplementary Material 2). The Alur samples clustered with other locations in NEGR but could not be assigned to any location within NEGR due to the lack of further structure. There were no unique alleles in the Alur samples at the three loci that were used for comparison with the other locations.

## Discussion

We found 29 individual elephants in the Alur area, which is slightly higher than the previous minimum estimate. The population was estimated at over 23–24 (Appayya and Desai 2007) or at least 26 (Srinivasaiah and Sinha 2012) individuals through rapid field surveys. Because of elephants being scattered across the area, it was not possible to implement regular sampling methods and sampling was based on prior knowledge about elephant presence in different areas. Therefore, our estimate must be considered a minimum estimate. However, since the sampling was carried out widely across the area, this is not likely to be very different from the actual number of elephants using the area. Given the $P_{ID}$ of the microsatellite loci we used, we are confident about not having misidentified any two individuals as the same individual from the 101 dung samples we collected. Removal of the Alur area elephants had been suggested by the Karnataka Elephant Task Force in the light of the tiny habitat available under the Forest Department, impracticality of expanding the elephant habitat in the area, contiguous forest being available 40 km away, the absence of elephants in the area historically, and the escalating human–elephant conflict (Karnataka Elephant Task Force 2012). The small population size and genetic structure we found in this study supports the Karnataka Elephant Task Force's (2012) proposal to remove these elephants in the light of high conflict and low habitat availability as the population does not offer significant future conservation potential in terms of genetic variability or uniqueness. The removal of these 29 elephants is not likely to adversely impact the remaining total population of over 8,500 elephants in the larger landscape genetically. However, this should not be used as a precedent for indiscriminate capture of elephants from other areas in the future. In addition, removal will have to be carried out in conjunction with erecting physical barriers so that new animals from the south or the west are not able to disperse into the Alur area. While logistics and lack of experience may preclude large scale operations with simultaneous removal of about 30 elephants from this area, smaller numbers of animals may be removed at a time (see below).

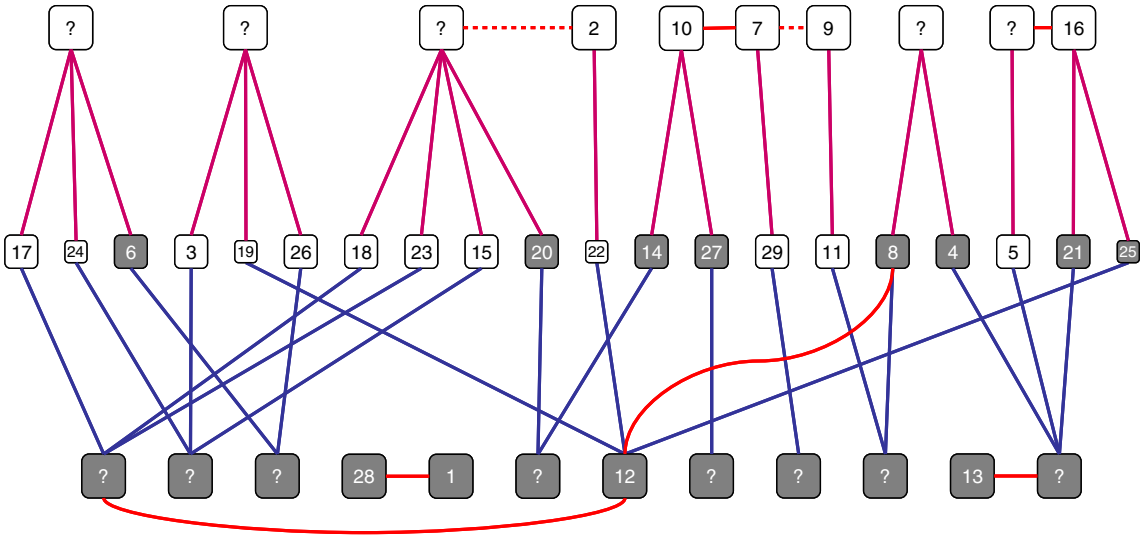


**Fig. 2** Parents (mothers *above*, fathers *below*) of subadults and juveniles (*middle row*) found in the Alur area. Females are shown as white squares and males as *grey squares*. The sampled individuals are numbered and inferred individuals are labeled with a question mark. The three sizes of *squares* correspond to adults, subadults, and juveniles, in decreasing order of size. *Straight lines* from adults to the subadults and juveniles in the *middle row* indicate parentage based on the output from COLONY. Relatedness between adult females and between adult males based on full/half sibships from COLONY are shown as *solid lines*. *Dotted lines* between adult females indicate possible relatedness, inferred from relatedness values ($\sim$0.2 and above) between adults and subadults/juveniles that are not their own offspring. If all such pairwise relatedness values above $\sim$0.125 are taken into account, the group with "10–7–9" may be related to group "?–2", giving one large and three small family groups ("8–4" are not considered since there are no surviving females in that group). However, the adult females may also be related to some subadults and juveniles through adult males rather than forming a large family group. Males 1 and 28, and 12 and 8 were identified as sibling pairs

**Table 5** $F_{ST}$ and associated P values between Alur and other locations in the Nilgiris-Eastern Ghats population

| No. | Location names | $F_{ST}$ | *P* value |
|---|---|---|---|
| 1. | Madikeri Forest Division | −0.008 | 0.654 |
| 2. | Nagarahole (Rajiv Gandhi) National Park and Tiger Reserve | −0.009 | 0.677 |
| 3. | Bandipur National Park and Tiger Reserve | 0.002 | 0.373 |
| 4. | Mudumalai Wildlife Sanctuary (now Tiger Reserve) | 0.016 | 0.065 |
| 5. | Silent Valley National Park | 0.258* | <0.0001 |
| 6. | Mannarkkad Forest Division | 0.225* | <0.0001 |
| 7. | Satyamangalam Forest Division (now Wildlife Sanctuary and Tiger Reserve) | 0.040 | 0.055 |
| 8. | BRT Wildlife Sanctuary (now Tiger Reserve) | 0.034 | 0.041 |
| 9. | Cauvery Wildlife Sanctuary | 0.005 | 0.422 |
| 10. | Hosur Forest Division | 0.087 | 0.005 |
| 11. | Bannerghatta National Park | −0.094 | 0.917 |

$F_{ST}$s that were significant after Bonferroni correction (Bonferroni corrected $P = 0.0008$ because of 66 pairwise comparisons between all location pairs) are marked with asterisks

The location names are from when the sampling was carried out (so that they correspond to the names in Vidya et al. 2005) as well as the current status. Mannarkkad and Silent Valley are in NSCR (Fig. 1)

It was surprising that there were more females than males in the area (11 adult and subadult males, 1 juvenile male, of the 29 individuals). One would expect males to explore new ranges as they disperse naturally when they reach puberty, and for females to show philopatry (Desai and Johnsingh 1995; Vidya and Sukumar 2005). The female herds here have probably dispersed out of their natal ranges because of high density and/or marginal habitat due to anthropogenic pressure. Neither the adult/subadult males nor adult/subadult females nor combinations of them were significantly related amongst themselves when all individuals in such categories were considered. Since adult/subadult males and females were not related to one another, males seemed to show locational dispersal based on the small sample size. Three pairs of males were also significantly related to one another,

suggesting that there might be a small level of biased locational dispersal (with related males dispersing to the same area) also (see Vidya and Sukumar 2005).

When we examined females from the same locations (irrespective of time of sampling), we found that unrelated females used the exact same areas and that there was movement of females and males across forest fragments even within the short sampling period. Except for two females that were seen to associate with unrelated females (these dung samples had been collected after directly sighting the elephants), it is not certain if unrelated female groups actually associate with one another over longer time periods, as seen in other disturbed populations (Nyakaana et al. 2001; Charif et al. 2005; Vidya et al. 2007), or are forced to share the same areas because of spatial restrictions. The relatedness between females sampled from the same locations on the same day (with the exception of the two unrelated females) was similar to that seen in family groups (confidence intervals overlapping with the average relatedness seen in the previous studies of Vidya and Sukumar 2005 and Ahlering et al. 2011). The genetic relationships reconstructed in Fig. 2 and the result that adult and subadult females in the area were overall not related significantly indicated the presence of family group-like social units, rather than one cohesive clan of related females operating in the area. Therefore, there seem to have been several independent dispersals of small female groups that were possibly forced out of their natal habitat. A cohesive clan dispersing to the Alur area, with subsequent deaths of several adult females, would also result in lowered genetic relatedness across females in the area. The pedigree in Fig. 2 shows that several adult females are missing and presumably have died. However, in the case of a single cohesive clan, one would expect adult females to be more related amongst themselves, and subadults and juveniles to be less related amongst themselves when compared to adult females. We do not find this pattern and, in fact, find more subadults and juveniles related to one another through their fathers.

The observed relatedness structure has important consequences for management of this population. Since there are discrete family group-like social units rather than one cohesive clan, it allows for these groups to be removed in smaller related sub-groups rather than being constrained to be removed all together. If there was one cohesive clan, since elephants are very social, separating females of the clan can be traumatic to the animals (Stüwe et al. 1998), and keeping the entire social group together in the removal practice followed would be the most ethical option (Lötter et al. 2008; Slotow et al. 2008).

Given the low practicality of culling in the Indian situation, the only options following removal are translocation and rearing in captivity. Rearing in captivity is logistically expensive over the elephants' lifespans, given the number of elephants, and has no direct conservation benefit as animals are removed from the gene pool. Translocation has thought to sometimes not solve elephant–human conflict, especially because of males homing back (Lahiri-Choudhury 1993; Appayya 1995; Barua 1995; Fernando et al. 2012), but home ranges of translocated animals prior to translocations have generally not been taken into consideration. There is no detailed information on translocating female Asian elephants. Translocations of African savannah elephants have, however, been fairly successful in terms of few problems with homing (Slotow and van Dyk 2004, Pinter-Wollman 2009), translocated animals acclimatizing to the release site (Pinter-Wollman et al. 2009), and population growth rate (Slotow et al. 2005). It would, therefore, appear that there are many factors that influence the success of a translocation operation, such as gender, local conditions, and boldness (see Pinter-Wollman 2009). In the Alur area, because there is no constraint to deal with all the animals uniformly, combinations of the two can possibly be attempted in the current situation, with some elephants being brought into captivity and others translocated to contiguous habitat as an experiment to examine interactions between resident groups and the new group(s). The latter will offer us insight into elephant behaviour and allow for more informed management in the future. Translocation, if undertaken, would have to be carried out in the season when resources are not very limiting and to areas that are not near human habitation and do not have high elephant density. It would possibly work in concert with the creation of new habitats (Pinter-Wollman 2012).

It had been found previously that there was only a single mitochondrial DNA haplotype within the Nilgiris-Eastern Ghats population, and little structuring based on nuclear microsatellite DNA, with only locus, LafMS03, showing limited structure (Vidya et al. 2005). Pairwise $F_{ST}$ and $R_{ST}$ values between locations within the landscape were largely not significant: those that were pertained to two locations in NSCR, possibly because of smaller sample sizes (Vidya et al. 2005). The Alur samples also showed a similar pattern, being differentiated at the loci examined only from the two locations in NSCR. The STRUCTURE results also supported this. Therefore, if translocation must be undertaken, elephants can be moved to any of the locations in NEGR or NSCR as far as the impact on population genetic structure is concerned. It would be more pertinent to consider social and demographic factors and the probability of homing back in the current situation in order to choose a release site. Elephant ranges in the Nilgiris-Eastern Ghats landscape have been found to be of the order of 600 $km^2$ (Baskaran et al. 1995), and a radiocollared male in the Alur area ranged across nearly 300 $km^2$ in a year through coffee estates, agricultural land, and human settlements (Desai et al. 2013). Because of the lack of population genetic structure in the larger landscape, it is possible to choose

release sites that are sufficiently far from Alur for homing to be unlikely and yet have little impact on the genetic structure of the larger population. Given the complexity of various situations, one would not expect one size to fit all in conservation biology. Fortuitously, the Alur elephant population allows us to not force all the animals into one removal strategy: different decisions can be taken regarding the females in different family groups and males of different ages. It is likely that such situations are not rare across populations of endangered species. We suggest that analysis of population genetic structure as well as social organization be taken into account before management decisions are made.

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