## POPULATION, AGE AND SEX ASSESSMENT OF FREE-RANGING ELEPHANTS OF PERIYAR TIGER RESERVE

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# Thesis submitted in partial fulfilment of the requirement for the degree of

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#### DECLARATION

I hereby declare that this thesis, entitled "POPULATION, AGE AND SEX ASSESSMENT OF FREE-RANGING ELEPHANTS OF PERIYAR TIGER RESERVE" is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society

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#### CERTIFICATE

Certified that this thesis, entitled "POPULATION, AGE AND SEX ASSESSMENT OF FREE-RANGING ELEPHANTS OF PERIYAR TIGER RESERVE" is a record of research work done independently by Subhash .C.K, under my guidance and supervision and it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to him.

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## Introduction

#### 1. INTRODUCTION

Once common throughout its historic range, from the Tigris- Euphrates in West Asia eastward through Persia into the Indian subcontinent, South and South -east Asia including the islands of Sri Lanka, Java, Sumatra and Borneo and into China northwards up to the Yangtze-Kiang, the Asian elephant (*Elephas maximus*) is now confined to about 439,000 sq.km, in 13 countries which is approximately six percentage of their original historical range. In India, the total elephant habitat is approximately 1,10,000 sq.km, confined to 18 states and the islands of Andaman and Nicobar, distributed in five distinct zones: Northeastern, Eastern, Northern, Southern and the feral population found on the islands (Bist, 2002).

Current estimates of 50,000 Asian elephants cannot be regarded as reliable as much of the information recorded on elephant numbers in many parts of Asia are entirely qualitative observations, such as sightings of elephants, their dung or tracks, and information gathered from field protection staff, that can only be classified as " informed guesses". India, holding an estimated 24,000-28,500 free-ranging elephants, is showing a negative trend in the populations in Northeastern and Eastern zones (Bist, 2002).

Elephants in southern India distributed along the Western and Eastern Ghats mountain ranges, covering an area of  $39,500 \text{ km}^2$  with a population size of 12,500-14,500 accounts for 22 per cent of India's free-ranging elephant population (Sukumar, 2003). Periyar Tiger Reserve, one of the well known wildlife reserves in southern India has been in the midst of a lot of speculations and concern due primarily to highly skewed adult sex ratio and adverse demographic parameters observed in its elephant population (Uma Ramakrishnan *et al.*, 1998), obviously consequent to heavy poaching during 1970-1990 period (Sukumar, 2003).

There have been a number of studies on the ecology, population dynamics and habitat utilization as early as 1969 (Kurup, 1971) in Periyar Tiger Reserve. The recorded data from recent studies on the above parameters on elephants is so unbelievably different from earlier surveys and a sure case of conservation concern that warrants further investigations, especially with regards to population assessment and herd composition using reliable tools and modern techniques, to predict the prospects of Periyar elephants.

The present study combines conventional methods with more recent and developing molecular scatology (using dung extracted DNA to sex individuals) to monitor free-ranging elephant population of Periyar Tiger Reserve, keeping in mind the following objectives:

- To study the population, age groups and sex ratio of free-ranging elephants in Periyar tiger reserve.
- 2) To assess viability of the population based on the sex ratio.
- To study the suitability of bio-molecular tools for identification of sex of free ranging animals

# **Review of Literature**

#### 2. REVIEW OF LITERATURE

Literature on population dynamics and demography of Asian elephants is less compared to that on African elephants. Hence the review in this thesis is built mainly from the population studies on free ranging African species. Published studies on the population density estimation of a wide variety of wild animals are available in plenty, but only significant literature necessary for the present study are reviewed. Molecular scatology, being a fast growing branch of genetic differentiation/ characterization studies on wild animal species is reviewed from the available literature that has special emphasis on elephants.

The review of literature on this study is presented under the following subtitles:

- 2.1 Population dynamics and Demography
- 2.2 Methods of Population Assessment
- 2.3 Morphometric Age Determination
- 2.4 Sex Determination
- 2.5 Conservation strategies

#### 2.1 POPULATION DYNAMICS AND DEMOGRAPHY

#### 2.1.1 Studies on African Elephants

Buss and Smith (1966) observed the breeding behaviour of the African elephant (*Loxodonta africana*) in Uganda and reported that some wild females begin to breed at approximately seven years and all females breed by approximately 11 years of age. They concluded that the declining populations and lower annual increment in elephants south of the Victoria Nile River in Murchison Falls park were

associated with an average postpartum preconception interval more than three times longer than was reported during 1947-1950, and that the lengthening of this period possibly functions to self-regulate elephant population numbers. Their observations revealed that elephant cows have multiple mates, there is no prolonged male-female relationship, and by bulls over females were rare.

Barnes (1982) described the mate searching behaviour of elephant bulls in Ruaha National park, Tanzania that has a semi-arid environment. The study describes how bull elephants maximize their mating opportunities in a highly seasonal environment with mating activity between the mid wet season and early dry season (February to July to August), with a peak in the mid wet season. There was also a remarkable change in the behaviour of bulls, appearing to be concentrating on feeding and showing no interest in the opposite sex in the early wet season, but later towards the end of January they started to travel long distances, feed for shorter periods and moved towards family units in search of oestrus females. Barnes also stated that large body sized bulls get the twin advantage of a higher probability of finding an oestrus cow and a higher probability of mating with her.

Ecology and behaviour of the Amboseli elephant population in southern Kenya was studied by Western and Lindsay (1984) to explain natural seasonal cycles in movement, habitat selectivity and herd dynamics. The Amboseli population is unique in that it is largely unaffected by human pressures, allowing observations on elephant's natural ecology and behaviour in an undisturbed ecosystem. Sample counts, total counts and radio tracking of individual elephants was used to estimate the numbers and distribution of elephants. It was found that the 500 elephants in the 3500 km<sup>2</sup> occur at an overall density of 0.14 elephants per km<sup>2</sup> and the total range of the population based on aerial surveys was 3042 km<sup>2</sup>, the range based on radio collared animals was 2756 km<sup>2</sup> and the combined range from both sources was 3588 sq.km. Herds ranged more widely during the rains using the bushed-grasslands and

concentrated in the basin during the dry season, using the denser woodlands and the swamps.

Calef (1988) theoretically calculated the maximum rate of population increase for the African elephant at seven percent and an elephant population increasing at this maximum rate will soon reach a stable age distribution consisting of 48 per cent animals older than 11 years of age (the age of first reproduction), and six to seven percentage calves less than one year old. It was worked out that at this rate of increase the population doubles every 11 years, and the two major factors that influence the rate of increase are the age of sexual maturity and the mean calving interval.

Poole (1989) studied male guarding of females, male mating success and female choice for eight years among a population of African elephants. It was observed that males were not able to compete successfully for access to oestrus females until approximately 25 years of age. Males between 25 and 35 years of age obtained mating during early and late oestrus, but rarely in mid oestrus. Large musth males over 35 years old guarded females in mid oestrus. Body size and longevity are considered important factors in determining the lifetime reproductive success of male elephants.

Barnes and Kapela (1991) studied the changes in the Ruaha elephant population, Tanzania to assess the impact of intense poaching in the late 1970s and early 1980s, which reduced the numbers of elephants by about 60 per cent. They also observed that poaching had distorted the age structure of the survivors, altered grouping patterns, higher proportion of tuskless cows indicating a change in gene frequencies and affected every aspect of population biology. There were few cows above 34 years, few bulls above 16 years and a lack of calves aged between two to nine years.

The age structure of the elephant population in Liwonde National Park, Malawi was determined for the first time in 1993 and, again in 1995 using photogrammetric method by Bhima and Bothma (1997) and it was observed that the population consisted of mostly young animals less than five years old (52.6 per cent and 44.8 per cent in 1993 and 1995 respectively). The other age cohorts were: 6-10 years old – 16.1 per cent and 21.7 per cent; 11-15 years old – 7.8 per cent and 9.2 per cent; 16-20 years old – 5.2 per cent and 4.7 per cent; and above 20 years old – 18.3 per cent and 20.5 per cent, suggesting a young and growing population, that was not affected by the severe drought that occurred from 1991/92 to 1994/95.

#### 2.1.2 Studies of Asian Elephants

Kurup (1971) conducted a preliminary ecological survey of Periyar wildlife sanctuary, Kerala state in 1969 and among other parameters studied, he recorded that in all the observed elephant herds the proportion of immature individuals to that of adults was very low, a ratio of about eight immature ones to every 10 cows indicating a significantly declining population. He inferred that this low juvenile proportion indicated a natural state of reduced natality, which is a manifestation of the working of the innate self-regulatory mechanism of population control. Assuming a potential pubertal age of 12 years, parturition- conception interval of about three years, gestation period of two years and allowing for a calf mortality of 30 per cent to include all possible hazards in Periyar elephants, Kurup calculated that the optimum mature female to immature ratio should be 1:2.7 in a herd of 20-25 years age structure. As against this, the ratio was 1:0.8 among Periyar elephants.

Santiapillai *et al.* (1984) conducted an ecological survey of the Asian elephant (*Elephas maximus*) in the Ruhuna National Park, Sri Lanka from May 1978 to June 1980 and recorded the population structure, social organization, spatial distribution, diurnal activity, reproduction and breeding and estimates of density and biomass. According to their observations the most stable organization was that of the

adult females and their attendant young, adult males leading a solitary or semisolitary life joining the herds mainly for breeding only. The average herd size was 5.9 and within the herds the adult sex ratio was in favour of the females. Sexual activity and breeding were observed in the dry season, while newborn calves were seen in the wet season.

The population of Asian elephants in Parambikulam wildlife sanctuary, Kerala was studied during 1981-1983 to record the number, herd composition and size and population structure. A total count indicated about 114 elephants with an ecological density of about 0.5 animals per square kilometer with no significant seasonal or monthly differences in herd size. The herd size of eleven was more frequent. Forty three per cent of the herds was without bulls and about 66 per cent of adult males observed were solitary. It was suggested that the adult sex ratio of 1:6.8 in the present study indicated a slight increase in the male mortality and, the percentage of juveniles and calves (21 per cent) indicated a high percentage of breeding females in the population (Easa and Balakrishnan, 1995).

Uma Ramakrishnan *et al.* (1998) carried out rapid surveys on the elephant population in Periyar Tiger Reserve during March-May 1994, June 1994 to June 1995 and during February-April 1997 observing population structure and demography. The total number of elephants in the sanctuary was found to be 1166 during 1994 survey and, using direct elephant sightings the adult male to female ratio was found to be 1:101. The study also indicated that the average fecundity was only 0.075/adult female/year in 1994-95, but showed an increase in births with a fecundity rate of 0.16/adult female/year in 1997, which was still lower than the potential rate. The authors have discussed the problems of skewed sex ratio and suggested methods to improve the population.

#### 2.2 METHODS OF POPULATION ASSESSMENT

There are two methods of estimating elephant numbers, viz; Direct and Indirect method.

#### 2.2.1 Direct Methods of Estimating Elephant Numbers

Direct methods record actual sightings of elephant and it is broadly divided into Total count (eg. Block count, Roadside count, Waterhole count, Fixed point count and Aerial count) and Sampling (Line transect method).

Aerial counts of elephants on a 1,500 sq. mile area was conducted in Uganda in different seasons over a period of July 1963 to May 1964 by Buss and Savidge (1966) and observed seasonal movements and different distribution patterns in different areas. The annual increment varied inversely with the population density.

Population estimates and distribution of the African elephant (*Loxodonta africana*) in the entire Kenya rangelands was conducted by Ottichilo *et al.* (1987) by aerial counts using light aircrafts, during the period 1977-1981.Systematic reconnaissance flights along parallel transects spaced either five or ten kilometer apart were carried out with three observers recording vegetation habitats, other environmental attributes, live animal numbers and carcasses, including fresh bones. The population of elephants for the whole of Kenya was calculated at 75,300  $\pm$  5000 in 1978 and 51,200  $\pm$  5000 in 1980-1981. The ratio of live to dead elephants decreased from 4.0:1 in 1977 to 2.0:1 in 1978 and 1.27:1 in 1980-1981; an indication of rapid rise in elephant mortality.

Lahiri Choudhury (1991) summarized the direct count of elephants in different states of North-east India from 1966 to 1986 and concluded that sampling as a method of estimating wild populations is not inherently superior to total count, but is only more suitable, cost and time-wise and the additional advantage of being amenable to statistical treatment. He also stated that sampling might not be the best way to enumerate small populations consisting of non-homogeneously distributed mobile units, but would be adequate for determining densities of large populations such as Axis deer.

Krishnamurthy (1991) described elephant census in Tamil Nadu using total count method in 1979 and 1983 by dividing elephant habitats in each of the forest divisions into convenient blocks of 5 to 10 sq. km. extent, so that an enumeration party can cover each area in a single day. Actual sightings and group composition of elephants were recorded on data sheets provided. It was concluded that the total count method needs a lot of ground work, more man power with adequate skills to classify herds and, in difficult terrain having dense undergrowth with poor visibility, counting all the animals were not possible, hence estimate was less than the actual number present.

Varman and Sukumar (1995) evaluated techniques of estimating animal density through direct counts using line transects during 1988-1992 in the tropical deciduous forests of Mudumalai wildlife sanctuary in southern India, for four species of large herbivorous animals, namely, chital (*Axis axis*), sambar (*Cervus unicolor*), Asian elephant (*Elephas maximus*) and gaur (*Bos gaurus*). It was reported that on the basis of suggesting a model with the greatest precision, the Fourier Series model gave estimates with the lowest coefficient of variation followed closely by the Half-Normal model. Density estimates along roads were generally significantly different from those in the interior of the forest, indicating that roadside counts may not be appropriate for most species.

Varman *et al.* (1995) conducted a study comparing density estimates of elephants in Mudumalai wildlife sanctuary from direct and indirect (dung) counts during 1991-1992. For the study, the sanctuary was divided into several zones based on location and habitat type, and for the direct count four permanent transects of 3-4

km each were cut and walked twice a month, whereas for the indirect count, transects of 1-4 km were cut and enumerated during each of the three seasons. The estimate of mean density from the direct count was higher (3.09 elephants per sq.km) than that obtained by the indirect count (1.54 elephants per sq. km). The reasons for the differences and the strengths and weaknesses of both the methods was also analyzed.

#### 2.2.2 Indirect Methods of Estimating Elephant Numbers

Indirect method of estimation involves deducing numbers from indirect evidence rather than from direct sighting by counting the evidence left behind by the animal, such as dung or footprint.

Jachmann and Bell (1984) estimated elephant numbers in Kasungu National Park, Malawi by means of dropping counts and it appeared to be heavily biased in the direction of over-estimation according to a series of aerial surveys. Over estimation errors were primarily due to the boli of one dropping being spread over a considerable distance, and to double counting of one defecation by scouts counting next to each other. The dry season defecation rate for the Kasungu elephants was estimated to be 15.7 droppings per elephant per day, and applying correction factors, the number of elephants estimated was 1189 in 1978. The authors also described an alternate method for determining the age structure of an elephant population by means of circumference measurements of the individual boli from a sample of droppings.

Studies on the distribution, ecological requirements and resource availability to selected mammals of Periyar wildlife sanctuary was carried out for a period of five years from 1977 to 1982 by recording population parameters, activities of animals sighted and collection of indirect evidences of animals from systematically laid out sample plots by Ramachandran *et al.* (1986). A total number of 800 elephants was estimated based on the density of dung heaps counted from the sample plots, giving an overall density of one elephant per sq.km. The proportion of individuals in various classes and their sex ratio was comparable to healthy elephant populations elsewhere, except in the proportion of adult male elephants. Based on sightings of tuskers from 1979 to 1982, it was concluded that there were only about nine tuskers in the study area.

Dekker *et al.* (1991) elaborated an indirect method for counting Asian elephants in forests, a method first developed by Barnes and Jensen (1987) and used in the Central African forests. The method involved measuring three distinct parameters to arrive at an estimate of elephant density. They are: 1) Density of dung (number of piles per sq.km.) = Y; 2) Rate of dung decay (per day) = r; 3) Defecation rate (number of dung piles produced per elephant per day) = D. From these parameters the elephant density (E) could be estimated using the formula: E x D = Y x r.

Sukumar *et al.* (1991) reviewed the various methods used to estimate elephant numbers. It was concluded that there is no single method that can be termed as the 'best method' for estimating elephant numbers under all conditions, and the choice of a method depended on the density of elephants in an area, the various attributes of the habitat, the level of skilled personnel available and the requirements of the researcher or manager. It was suggested that indirect method through dropping counts is the most appropriate for rain forest habitats with very low elephant densities and the direct sample count methods are appropriate in drier forests where elephants are found at higher densities.

#### 2.3 MORPHOMETRIC AGE DETERMINATION

Rensch and Harde (1966) measured the body parts of captive Indian elephants of different ages and body size to study the postnatal growth gradients in

an effort to comprehend the correlations connecting the single parts of the body with one another and the body as a whole. The study confirmed that Indian elephants are not fully grown, before they have reached an age of 25 years, and afterwards grow very slowly until a high age suggesting that the oldest specimens are the largest ones. Measurements also revealed that the height of shoulder and back, length of foreleg, circumference of forefoot and length of ear grow with negative allometry during postnatal development, whereas widths of ear grow with positive allometry. The tail and the hind legs grow isometrically.

Western and Moss (1983) developed an inexpensive, simple technique to conduct rapid age estimates of elephants in Amboseli National Park, Kenya. They related the length of hind foot prints to the age of animals ranging to 14.5 years from 17 herds, a sample which included a total number of 143 animals. A highly significant correlation was found between foot print length (f) and shoulder height (h): f = 0.158h + 3.008, and in conditions where length measurements are difficult the hind print width (w) was used to calculate the length (f) by the equation: w = 0.810f + 5.551.

Pilgram and Western (1986) showed that the sex of African elephants can be inferred from tusk measurements by considering total length and circumference in combination, and their age can be estimated from tusk weight or circumference. They also suggested that this information could be used to assess mortality patterns, which in turn could help to study the impact of hunting on African elephant populations.

Sukumar *et al.* (1988) studied the data available on body measurements of captive Asian elephants in southern India to derive equations for growth in body length, body weight and tusk circumference at lip line with age. The study revealed evidence for a post-pubertal secondary growth spurt in both male and female elephants. Domestic elephants born in captivity or captured at a young age showed

reduced growth in height in both the sexes and, in body weight in males compared to wild elephants. The height was twice the circumference of front foot throughout the life span, indicating an isometric relationship.

Watve and Sukumar (1997) showed that the length of the tusks of male Asian elephants, corrected for differences due to age, is significantly negatively correlated with intestinal parasite loads based on a study conducted in Mudumalai wildlife sanctuary, Tamil Nadu state. They also suggested that the less aggregated distribution of parasites in this elephant population, as compared to other mammalian species indicate that ivory poaching may have already selectively removed a significant proportion of parasite-resistant individuals.

Reilly (2002) investigated age-related growth in Sumatran elephants (*Elephas* maximus sumatranus) and the relationship of a number of measures (shoulder height, fore foot circumference and dung bolus size) to age was explored using the Von Bertalanffy growth function. It was found that all length measurements were highly correlated with age in the Sumatran elephants and provided growth models for determining the age structure of wild populations. The growth model relating dung diameter to age was then used to predict the age structure of wild elephant population in Way Kambas National Park, Sumatra. The use of dung diameter to predict age offered a robust field technique for use in situations where direct observations are limited and the use of other age estimation methods are impractical.

#### 2.4 SEX DETERMINATION

#### 2.4.1 Conventional Method

Traditional method of sex determination was by using demographic data from direct field observations. This was feasible as Asian elephants show sexual dimorphism, the chief difference being that females do not carry tusks while males may carry tusks, and differences in behaviour. Other methods include observations of genital organs, mammary development, body structure etc.

Easa (2001) carried out demographic study of elephants in Periyar Tiger Reserve and adjacent areas including Ranni, Konni and Achenkovil Forest Divisions in Kerala state during 1997 to collect information on the group composition, sex ratio, density and population especially of tuskers. During the period of study, a total of 379 elephants were sighted in 63 herds and herd size was found to range from 1 to 19 elephants. Total number of adult males was found to be 28 with tusker to makhna proportion of 1:1.Adult females constituted 58.6 per cent of the population, and the overall male to female sex ratio was 1:87 and adult male to female 1:7.9. The overall density was 0.5/sq.km. in the study area and the density was higher in grasslands (0.9/sq.km) compared to other habitats.

#### 2.4.2 Molecular Genetic Studies

Fernando *et al.* (2000) conducted genetic analysis of free-ranging Asian elephants (*Elephas maximus*) from Sri Lanka, Bhutan/North India and Laos/Vietnam by extracting DNA from dung, PCR amplifying and sequencing 630 nucleotides of mitochondrial DNA. Seventeen haplotypes were identified within Asian elephants, which clustered into two well differentiated assemblages with an estimated Pliocene divergence of 2.5-3.5 million years ago. Significant genetic differentiation was observed between the mainland and Sri Lanka, and between northern, mid-latitude and southern regions in Sri Lanka.On the mainland, little genetic differentiation was observed between elephant populations of Bhutan and India or Laos and Vietnam.

Fernando *et al.* (2001) described the isolation and characterization of five triand tetranucleotide markers in the Asian elephant in an attempt to gain in-depth knowledge on particular populations. All the five loci were found to be polymorphic in Asian elephants. Fernando and Melnick (2001) reported molecular sexing of mammals by polymerase chain reaction (PCR) amplification of Y chromosome fragments or co amplification of homologous fragments from both sex chromosomes, which are discriminated by size polymorphism or Y- specific restriction digestion. Here they described a simple approach, using 'double peaks' in the chromatogram upon direct sequencing of PCR products from males, to identify Y-specific restriction sites, and demonstrated its utility by application to a range of mammals such as Asian elephants, Indian rhinoceros, domestic dog and Tonkean monkey.

Fleischer *et al.* (2001) sequenced mitochondrial DNA (mtDNA) to assay genetic variation and phylogeography of Asian elephants using samples collected from captive animals in work camps and zoos across the world. The results indicate that Asian elephants have typical levels of mitochondrial DNA variation and there is no evidence to suggest that historically recent range fragmentation and population decreases have impacted within-population genetic variability to any great extent. Also, reconstructed phylogenies revealed two highly differentiated clades (A and B) that differ in their representation across localities, and to some extent, subspecies. Individuals of both major clades exist in all locations, except Malaysia and Indonesia. Most elephants from Malaysia and all from Indonesia were in well-supported basal clades within clade A, thus supporting their status as evolutionary significant units.

Eggert *et al.* (2002) studied the phylogeography of western and Central African forest and savannah elephant populations with mtDNA sequences from eastern and southern African savannah elephant populations to infer the patterns and processes of the evolution of today's African elephant. They examined mitochondrial cytochrome b control region sequences and four microsatellite loci to investigate the genetic differences and revealed several deeply divergent lineages that do not correspond with the currently recognized taxonomy: (1) the forest elephants of

Central Africa; (2) the forest and savannah elephants of West Africa; and (3) the savannah elephants of eastern, southern and Central Africa. It was proposed that the complex phylogeographic patterns detected in African elephants resulted from repeated continental-scale climatic changes over their five-to-six million year evolutionary history.

Fernando *et al.* (2003) compared DNA of Borneo elephants to that of elephants from across the range of Asian elephant, using a fragment of mitochondrial DNA, and five autosomal microsatellite loci and found that Borneo's elephants are genetically distinct, with molecular divergence indicative of a Pleistocene colonization of Borneo and subsequent isolation, which warrants their recognition as a separate evolutionary significant unit.

Vidya *et al.* (2003) demonstrated the application of molecular sexing of freeranging elephants for the first time in India from DNA extracted from dung, based on ZFX-ZFY fragment amplification and ZFY-specific Bam HI site restriction to differentiate between the sexes. Molecular sexing was suggested to be a useful tool to sex juveniles that lack dimorphism, or to estimate population sex ratios by carrying out noninvasive sampling. By application of this technique the adult sex ratios in Nagerhole and Mudumalai-Bandipur reserves in southern India during May 2001 were 1:3.1, matching the demography-based sex ratio for the same month, and 1:9.4, respectively.

Vidya *et al.* (2005) conducted the first population genetic study of freeranging Asian elephants, examining within- and among-population differentiation by analyzing mitochondrial DNA (mt DNA) and nuclear microsatellite DNA differentiation across the Nilgris-Eastern Ghats, Anamalai and Periyar elephant reserves of southern India. It was observed that the Nilgiri population had only one mtDNA haplotype and lower microsatellite diversity and almost no mtDNA or microsatellite differentiation among localities within the Nilgris suggesting extensive gene flow in the past. It was also found that the Nilgiri population was genetically distinct at both mitochondrial and microsatellite markers from the Anamalai and Periyar populations, which in turn were not genetically differentiated from each other.

#### 2.5 CONSERVATION STRATEGIES

Hanks and McIntosh (1973) constructed models of elephant populations to see how variations in the reproductive performance and mortality schedules influenced the rate of population growth and to examine the factors responsible for the natural regulation of elephant populations. The models indicated that a change in the duration of the calving interval is more important as a population regulating mechanism than a change in the age at puberty, and that the proportion of old reproductively inactive females is of little significance. A variation in mortality, especially neonatal mortality is of far greater importance as a population controlling mechanism and an annual population increase of 4 per cent would probably be the maximum rate of increase that an elephant population could be expected to attain under ideal biological conditions.

Shaffer (1981) proposed tentative criteria for successful species conservation at the population level, discussed the various methods available for determining the population sizes and their area requirements to meet these criteria, and related both to overall conservation strategy. He put forth the following definition of minimum viable population size: A minimum viable population for any given species in any given habitat is the smallest isolated population having a 99 per cent chance of remaining extant for 1000 years despite the foreseeable effects of demographic, environmental, and genetic stochasticity, and natural catastrophes. Also explained five possible approaches to determining minimum viable population sizes and their area requirements such as experiments, biogeographic patterns, theoretical models, simulation models and genetic considerations. Lande (1988) discussed the role of genetics and demography in biological conservation and suggested that demography is usually of more immediate importance than population genetics in determining the minimum viable sizes of wild populations. To justify this, several population genetic factors and demographic factors crucial to the persistence of populations are examined in detail in the paper.

Recent long term studies of population ecology of large herbivorous mammals suggest that survival of prime aged females varies little from year to year and across populations. Juvenile survival, on the other hand, varies considerably from year to year. The pattern of high and stable adult survival and variable juvenile survival is observed in contrasting environments, independently of the main proximal sources of mortality and regardless of whether mortality is stochastic or density-dependant. High yearly variability of juvenile survival may play a dominant role in population dynamics, as reported by Gaillard *et al.* (1998).

Sukumar *et al.* (1998) estimated the minimum number of elephants poached and the quantities of ivory harvested over a 20 year period (1974-1994) in the Periyar reserve of southern India using a simulation modeling approach from the observed elephant population structure. It was estimated that 336-388 tuskers have been poached and 3256-3334 kg of ivory harvested by poachers over the 20-year period with the maximum harvest from the 10-20 year age class. Trends in the various demographic parameters such as population numbers, tusked male to tuskless male ratio and female fecundity are also discussed using the model.

An overview of elephant conservation in India by Bist (2002) provides an intense insight into the present distribution of elephants in India, designated elephant reserves and their managemental issues, significance of elephant corridors, humanelephant conflict and related damages, annual pattern of elephant mortality due to human factors and the status of captive elephants with their legal implications.

## Materials and Methods

#### 3. MATERIALS AND METHODS

#### 3.1 STUDY AREA

#### 3.1.1 Location and Boundaries

Periyar Tiger Reserve (PTR), situated in the Cardamom Hills and Pandalam Hills between latitudes 9° 15' and 9° 40' N and longitudes 76° 55' and 77° 25' E., one of the single largest compact forest blocks in the Western Ghats, contiguous with the forest areas of the proposed Meghamalai wildlife sanctuary, Srivilliputhur Grizzled Giant Squirrel sanctuary and Tirunelvely Division in Tamil Nadu, and Kottayam and Ranni Forest Divisions in Kerala was the location of the study. The Periyar Conservation Unit extends right upto the Shengotta Gap and have tenuous linkages with the Agasthyamalai Conservation Unit comprising of Kalakkad-Mundanthurai Tiger Reserve in Tamil Nadu and Neyyar, Peppara and Shenduruni Sanctuaries in Kerala.

PTR is divided into two territorial divisions, Periyar West and Periyar East with headquarters at Thekkady and Peerumedu respectively, and further divided into five ranges, viz; Periyar, Thekkady, Vallakadavu, Azhutha and Pamba ranges. Periyar Tiger Reserve (PTR) forms a significant part of the physical and cultural catchments of two major rivers of Kerala namely, Periyar and Pamba. Apart from its ecological significance as a man-made but unique aquatic system, the 26 km<sup>2</sup> Periyar Lake is an important water source to Tamil Nadu for irrigation, drinking and power generation purposes.

The total area of 777 sq.km consisting of core 350 sq.km, buffer 427 sq km and tourism zones 50 sqkm (within buffer zone) was brought under Project Tiger in 1978, the 10<sup>th</sup> Tiger Reserve in the country. The core area of the Tiger Reserve has been declared as a National Park during 1982. In 1991, the area was also declared as part of Project Elephant Reserve No.10, a contiguous forest tract

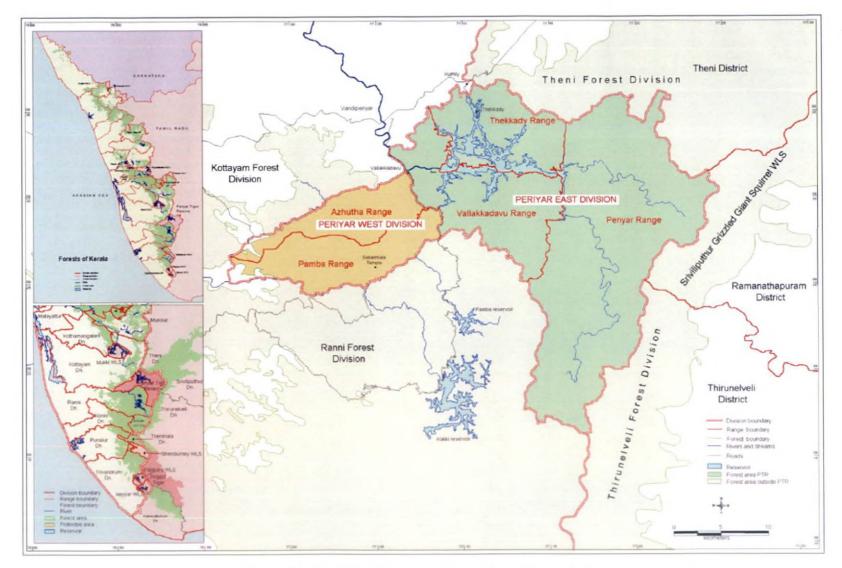


Fig. 1. Periyar Tiger Reserve: Location and boundaries

extending over an area of 3800 km<sup>2</sup> in Kerala and Tamil Nadu. The location map of Periyar Tiger Reserve with boundaries is given in figure 1.

#### 3.1.2 Terrain and Vegetation

The terrain is undulating in many areas and rugged with lofty peaks and precipitous slopes in other areas. The elevation ranges from 100 m (Pambavalley) to 2019 m (Kottamala) above sea level with an average altitude of 800 m. The average rainfall is 2000 mm annually. Thekkady has a rather cool climate; the temperature varies between 15°C and 31°C. The average humidity varies between 60 % and 85 % round the year.

Major vegetation types in Periyar Tiger Reserve comprise of Tropical evergreen and semi evergreen forests (579.64 km<sup>2</sup>), moist deciduous forests (104.54 km<sup>2</sup>), shoal/grasslands (11.66 km<sup>2</sup>) and Eucalyptus plantations (55.17km<sup>2</sup>) Periyar Lake forms an important aquatic ecosystem, which occupies about 26 km<sup>2</sup> of the area. The vegetation map of Periyar Tiger Reserve is given in figure 2 A., and the percentage of vegetation types/ecosystems in PTR is shown in figure 2 B.

#### 3.1.3 Animal Diversity

In Periyar Tiger Reserve (PTR), the wealth of vertebrate diversity is very rich. Of the 63 species of mammals recorded, Elephant, Gaur, Sambar deer, Wild boar and Barking deer are the common herbivorous species. Kurup (1971) has reported that elephant is the dominant species of Periyar sanctuary. Apart from these, Tiger, Leopard, Wild dog, Sloth bear, Nilgiri langur, Lion-tailed macaque, Nilgiri tahr, Small travancore flying squirrel, Nilgiri marten and Strip necked mongoose are frequently sighted. 41 species of fish has been reported including the famous game fish Mahaseer and seven other species of fish, which are endemic to PTR. 27 species of amphibians have been identified in the Park, of which 10 are endemic to Western Ghats. There are 45 species of reptiles including two species of testudines, 11 species of lizards and 30 species of

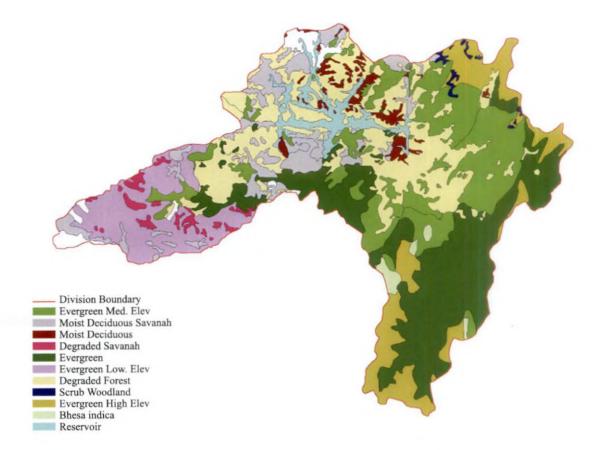


Fig. 2. Periyar Tiger Reserve: Vegetation Types

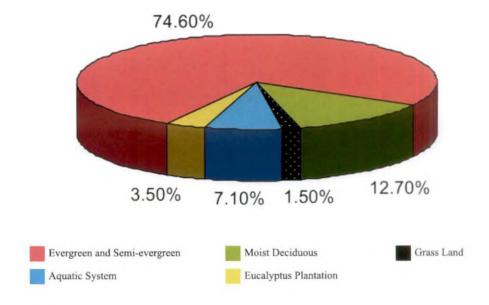


Fig. 3. Percentage of vegetation types in Periyar Tiger Reserve

snakes; of which 13 snake species are endemic to Western Ghats. There are over 323 species of birds and about 94 species of moths and butterflies so far recorded from Periyar.

## 3.2 MONITORING ELEPHANTS IN THE RESERVE

The entire exercise of collecting information on free ranging elephants in the reserve was done primarily on foot, and at times in jeep, perambulating higher density areas more frequently and taking care to visit every forest section in the reserve at least once.

The period of study was from March to July 2005, taking advantage of the better visibility factor available during the summer months in the reserve. The main parameters observed for the study were herd composition, age estimation, sex ratio in different age groups and population density. During the study, attempts were made to identify different herds separately based on the physical characters of the matriarch or the 'target animal' to exclude duplication while assessing sex ratio.

#### 3.2.1 Observation of Herd Composition

The parameters observed were herd size, composition and proportion of different age and sex classes in the population based on direct sightings obtained while perambulating the reserve during the study period. Care was taken to spend sufficient time in all the habitat types proportionate to their extent. The elephant herds were classified into type I, type II and type III depending on the nature and extend of age and sex assessment possible during observations. Type I indicates that all elephants in the group has been accurately classified; type II indicates that not all elephants were classified but, the presence or absence of adult males in the group has been confirmed, and type III indicates that not all elephants were classified or absence of adult males in the group is also not confirmed.

During observations, necessary steps were taken to identify each elephant herd/group separately to avoid duplication. This was achieved by recording moving visuals of every individual in the herd/group using video camera. Herd identification was also achieved by noting conspicuous body features of the matriarch and other members if possible, such as torn ears, disproportionate ears, holes on ear flaps, cut tail, stumpy tail, other anatomical peculiarities and deformities. The visuals taken and peculiarities noted of every observed herd/group were compared regularly to differentiate herds.

Using the herd composition data, the fecundity rate and calving interval was worked out from the number of calves and adult females in the observed population. The methods of age assessment and sex determination are explained under the respective headings below.

#### 3.2.2 Estimation of Age of Free Ranging Elephants

The study entails classification of free ranging elephants in the Reserve into four categories based on age. The elephants were broadly classified into four age categories described by Sukumar (1989).

1. Calf	less than one year (Height, 3-4 ft)					
2. Juveniles	1-5 years	(Height, 4-5 ft)				
3. Sub adults	5-15 years	(Height 5 ft for females; 5-8 ft for males)				
4. Adults	Above 15 years	(Height above than 7 ft for females, above 8 ft for male)				

Age assessment of the elephants in groups and of loners sighted in the reserve during the survey period from March to July 2005 was performed by observing a combination of several criteria such as shoulder height, areas of depigmentation on body parts (blotches), size of ears and folds on upper margin of ears, hollows/depression on forehead, temples and buccal region, mammary development in case of females, size of tusks in males etc. The shoulder height of the matriarch ('target animal/oldest female') was first taken using photogrammetric method described by Jachmann (1980), with minor practical

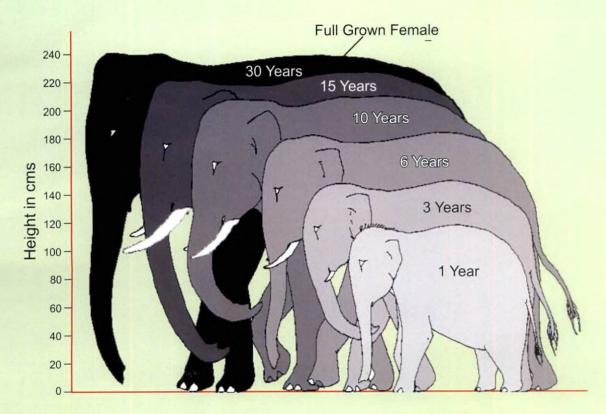


Fig 3 A: Field key for ageing young male elephants

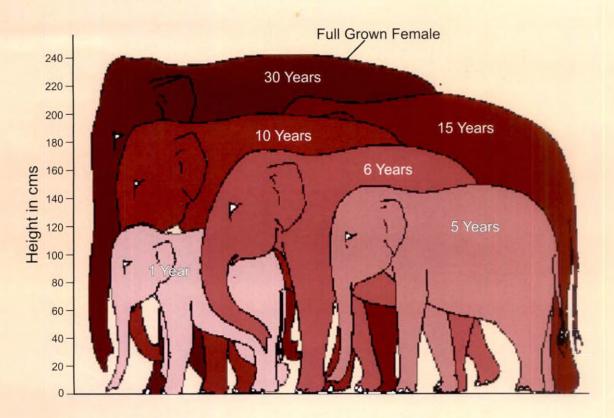


Fig 3 B: Field key for ageing young female elephants

modifications and the shoulder height of other members of the herd was assessed by comparing with the matriarch's height. The photogrammetric method involves taking photographs (visuals in this study) of the elephant close to and against a natural object such as a tree, and measuring the height from the natural object after the elephant has moved away.

From the assessed shoulder height, the age of all sex/age categories were calculated from the charts prepared by Sukumar (1989). The charts are presented in figures 3A and 3B. Other physical characteristics mentioned above were also taken into consideration before the age of the members was recorded finally.

#### 3.2.3 Sex Assessment of Free Ranging Elephants

Two methods were employed to assess the sex of elephants in the reserve (observed sex ratio).

#### 3.2.3.1 Physical Verification

The elephants were physically verified by observing from a safe distance, preferably using a binocular primarily relying on observation of genital organs, supported by accessory sexual characters such as mammary development, presence of tusks etc.to determine the sex.

Any elephant, which cannot be readily categorized as bull or Cow, was recorded as Unsexed

## 3.2.3.2 Molecular Sexing

In rare instances it may not be possible to accurately identify the sex of certain elephants especially those belonging to sub adult, juvenile and calf categories by physical verification because of inconspicuous genitalia and other accessory sex characters. In such cases, the dung samples of those animals were taken for sexing using molecular technique.

The DNA extraction from dung and sexing procedure adopted by Vidya et al. (2003) was followed in this study.

## 3.2.3.2.1 DNA Extraction Using Qiagen Gel Purification Kit

- 1. Approximately 10g of the outermost layer of dung that is rich in endothelial cells were collected in a sterile vial containing 95% ethanol.
- 2. The collected samples were stored at room temperature for laboratory analysis
- 3. 0.25 g of each dung sample was transferred into 2 ml microcentrifuge tubes, pippetting out excess alcohol.
- 4. The sample was incubated at 70 °C till the remaining alcohol evaporates and the sample is dry.
- 5. To the dried sample 1 ml of Digestion buffer was added, mixing it well with the pipette tip while transferring.
- 6. 10-20 µl of Proteinase K (20mg/ml) was then added to the sample.
- A little more of the digestion buffer (0.5 ml approximately) was added to the sample, homogenized and allowed to incubate overnight in a shaker at 55 °C.
- 8. After overnight incubation the sample was homogenized and centrifuged at 12,000 rpm for two minutes.
- 9. Another set of tubes were taken and labelled; transferred 400  $\mu$ l of the supernatant to the tubes.
- Added 800 µl of phenol-chloroform-isoamylalcohol (25:24:1) to each tube, vortexed and incubated for a minimum of 20 minutes.

- 11. After incubation the samples were vortexed and centrifuged at 12,000 rpm for two minutes.
- The upper aqueous layer containing the DNA (approximately 350 μl) was separated into fresh set of labelled tubes.
- 13. One ml of solubilization buffer (QIAGEN gel purification kit), approximately thrice the volume was added to the separated aqueous layer, homogenized and transferred to QIAGEN gel purification spin columns and centrifuged for a minute at 12,000 rpm. The DNA will bind to the silica column in the presence of salt in the buffer, while the impurities filter down.
- 14. The columns were washed using wash buffer (Wash buffer PE-QIAGEN), 650 μl, centrifuged for four minutes at 12,000 rpm. The impurities filtered down were removed and again centrifuged for a minute at 12,000 rpm.
- 15. The columns were removed and placed in fresh set of marked 1.5 ml tubes.
- Added 50 μl of 1/10 elution buffer (Buffer EB—QIAGEN) warmed to 80 °C to the filter directly in each column.
- Centrifuged for two minutes at 12,000 rpm to elute the DNA into the
   1.5 ml tubes.
- 18. The extract (DNA) obtained was then stored at -20 °C

(The composition and methods of preparation of the reagents and buffers are presented in the Annexure I)

#### 3.2.3.2.2 PCR Amplification of the Extracted DNA

Polymerase chain reaction (PCR) was then performed to amplify a ~300 bp segment of ZFX-ZFY locus of the DNA samples.

#### 3.2.3.2.3 Primers Used

The primers P1-5EZ: 5' – ATAATCACATGGAGAGCCACAAGCT- 3', and P2 –3EZ: - 5'- GCACTTCTTTGGTATCTGAGAAAGT 3' (Operon Technologies Inc., USA) were used for the PCR amplification reactions.

# 3.2.3.2.4 Preparation of Samples for PCR

PCR amplification reactions were carried out with 50  $\mu$ l volumes using, 8  $\mu$ l of extracted DNA, 1  $\mu$ l each (10 pica mols) of P1 –5EZ and P2-3EZ, 0.6  $\mu$ l of Taq DNA polymerase (MBI Fermentas 5 U/ $\mu$ l), 36  $\mu$ l of Master (1 ml master mix containing 13.1 x 4 $\mu$ l (dATP, dCTP, dGTP, dTTP) of 10 mM dNTPmix, 17.34  $\mu$ l of 100 mg/ml BSA, 16.37  $\mu$ l of 4M KCl and 13.09  $\mu$ l of 1M Tris Ph 8.4., 1.96  $\mu$ l of 1M MgCl<sub>2</sub> and 858.8  $\mu$ l water).

A negative control was also set up using water instead of DNA to minimize experimental error.

## 3.2.3.2.5 PCR Amplification Conditions

The PCR cycles comprised of an initial denaturation at 93°C for 3 minutes, followed by 39 cycles each of 1 minute denaturation at 93°C, annealing at 51°C for 1 minute, extension at 72°C for 1 minute to complete a total of 40 cycles, and final extension at 72°C for 5-7 minutes. The PCR amplified products were stored at -20°C till gel analysis was performed.

## 3.2.3.2.6 Analysis of Amplified Products

The PCR products thus obtained were then subjected to electrophoresis on 1% agarose in 0.5 X TBE buffer to detect the presence of amplified DNA.

#### 3.2.3.2.7 Agarose Gel Preparation

Thirty millilitre of 1% agarose gel was prepared by dissolving 0.3g in 30 ml of 0.5 X TBE buffer of pH 8.0. The prepared gel was then sterilized at 360°C for one minute, cooled to around 60°C and added a tinge of ethidium bromide. The gel was then poured into the gel tray avoiding air bubbles, and the gel tray was immersed in the electrophoresis chamber containing buffer.

## 3.2.3.2.8 Sample Loading and Electrophoresis

Five  $\mu$ I of amplified PCR products were loaded into the wells in the gel tray after mixing with the loading dye. The electrophoresis was carried out in room temperature with negative control at 70 Volts and 300mA current for 20 minutes or until the bromphenol blue reaches the two-third length of the gel.

## 3.2.3.2.9 Analysis of Gels

The gels were visualized under a UV transilluminator and subsequently photographed. The presence of band at ~300 bp was recorded. The positive PCR samples were then subjected to restriction digestion.

#### 3.2.3.2.10 Restriction Digestion (RFLP- Restriction site on ZFY Fragment)

The PCR products were then subjected to restriction digestion with 5  $\mu$ l of *Bam* HI (MBI Fermentas 5U/  $\mu$ l), 5.5  $\mu$ l of 10X buffer with BSA, 20  $\mu$ l of PCR product and 19.5  $\mu$ l of water at 37 °C in a water bath for three hours.

## 3.2.3.2.11 Analysis of Restriction Digests

Electrophoresis was then conducted on a 2% 1 agarose: 1 low melt agar gel using the restriction digested products with a positive control to minimize experimental error under the same conditions described above for analysis of amplified products. The gel was then examined under a UV transilluminator to differentiate the male and female from the bands obtained on the gels. The gels were subsequently photographed.

## 3.2.4 Estimation of elephant numbers

Density of elephants was estimated using a direct sample count method (Block Count). The data for population number estimates was collected during the elephant census conducted by Project elephant, Ministry of Environment and Forests, GOI, in association with the respective state Forest Departments on the 6'th of May 2005.

## 3.2.4.1 Block count method of density estimation

The total park area of 777sq.km in the five ranges namely Thekkady, Periyar, Vallakkadavu, Azhutha and Pamba was divided into 61 blocks based on vegetation, geographic land marks, natural boundaries and elephant usage. The map of Periyar Tiger Reserve with the blocks divided is shown in figure 4. Out of these, 23 blocks were selected for the sample count. The list of the selected blocks in each range with their area in square kilometer is given in Annexure II. Each block was perambulated on foot by a trained team from morning till evening and information collected regarding the number of elephants sighted, age and sex composition. The data thus collected was recorded on a data sheet supplied to every team. (Annexure III).

The data obtained from sample blocks was pooled and analysed statistically using the calculations suggested by Sukumar *et al.* (1991) to compute the total number of elephants in Periyar Tiger Reserve.

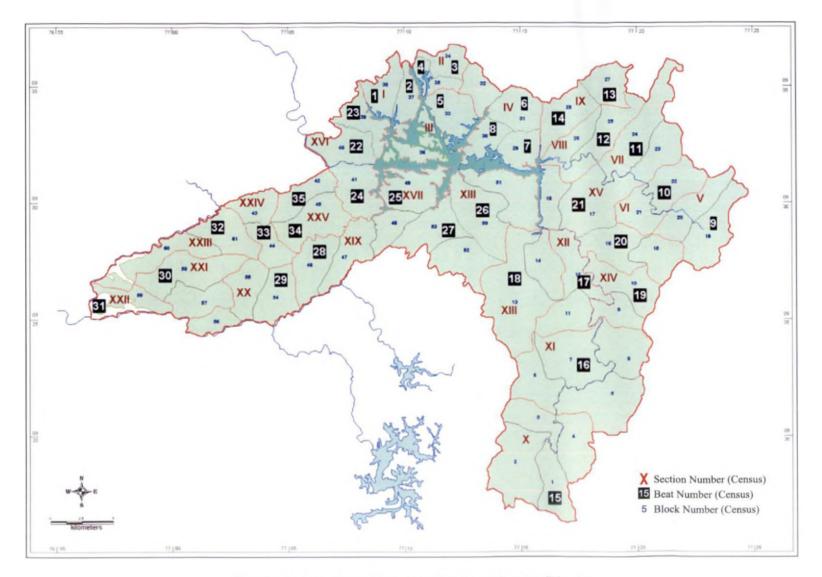


Fig. 4. Periyar Tiger Reserve: Elephant Census Blocks

Results

## 4. RESULTS

The area of study was surveyed on alternate weeks starting from March 2005 to May 2005 for a total period of 45 days, perambulating approximately 30-40 per cent of Periyar Tiger Reserve. Majority of the area was covered on foot, and at times jeep and boat also formed the mode of conveyance with repeated visits to high density elephant areas based on information collected from the forest authorities.

## 4.1 TOTAL ELEPHANT SIGHTINGS/CLASSIFIED

A total of 239 elephants were sighted during the study period. Fifteen animals in three herds were observed on more than one occasion, 17 animals observed were classified as type 11, and 14 animals sighted were classified into type 111. A total of 193 elephants were classified based on age and sex.

## 4.2 HABITAT PREFERRED BY ELEPHANTS

The vegetation of Periyar Tiger Reserve comprised mainly of tropical evergreen and semi-evergreen forest (74.6%), moist deciduous forest (12.7%), eucalyptus plantations (7.1%), grasslands (1.5%) and the reservoir (3.5%). During the period of study, high percentage of animals observed were in grasslands and adjoining sholas followed by the moist deciduous forests adjacent to the Periyar reservoir (Plate 1). The density was least in the evergreen patches.

#### 4.3 HERD COMPOSITION

#### 4.3.1 Herd Size

Forty three herds were observed in the reserve during the period of study and the herd size varied from 1 to 16, with an average of 5.5. Six solitary elephants were sighted, of which one elephant was identified as 'already recorded'. All solitary elephants recorded were very old females (Plate 2). Two adult males aged between 15 -20 years only were recorded during the entire period of the study and at the time of observation, they were attached to groups (Plate 3). Figure 5 shows the number of animals in different groups observed.

#### 4.3.2 Social Structure

The social organization and nature of interactions among the social groups as recorded by earlier investigators on Asian elephants were observed in PTR also, exhibiting a matriarchal order with family units comprising of adult cows and their offsprings, including daughters of all ages and sons of prepubertal age (Plate 4). Such units are stable and characterized by strong social bonds.

#### 4.3.3 Age structure

Age wise distribution of males and females in the population is given in figure 7 and 8. Adult females formed a major portion of the population (52.41%), followed by sub adult females (17.12%) and juvenile males (10.90%). Adult males formed 0.889 per cent and sub adult males, 3.63 per cent of the population. Among adult females, the frequently sighted elephants were in the age group 20-30 years (16.086%) and 30-40 year age category (14.53%). Surprisingly, no adult male above 20 years was recorded in the reserve. A total of ten calves were recorded in nine different herds.

## 4.3.4 Sex Composition

A schematic presentation of the proportion of males and females in different age classes of elephants in the population is given in figure 6. It was observed that sex can be identified by physical verification alone in majority of the animals by observing the genitalia (Plate 5). Identification of sex of animals in the calf category was very difficult and it was not possible to collect dung

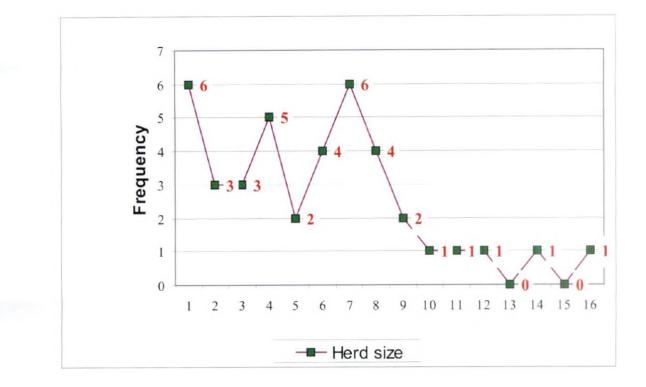


Fig. 5. Herd size frequency of elephants in the observed population

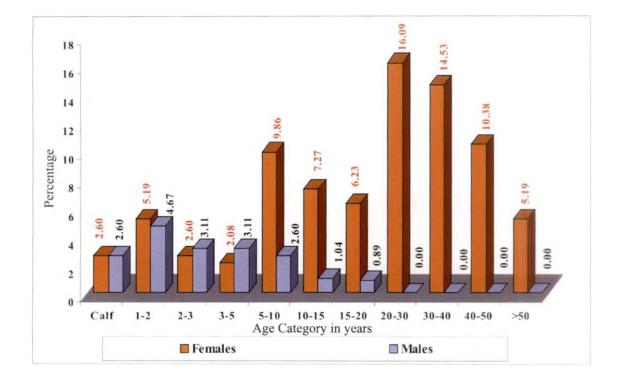


Fig. 6. Proportion of age-sex classes in the observed population

samples of such calves because the dung was even difficult to spot probably because of trampling by other members of the group. Out of the ten calves recorded four were confirmed to be males based on physical verification and the others were unsexed (Plate 7). But considering the constraints in sexing calves, the proportion of male to female calves was taken as equal in the present study. Females formed 81.99 per cent and males formed only 18.01 per cent of the population, but the proportion of males and females in the calf and juvenile category was almost same, with a shift biased towards the females from the sub adult category onwards. The photograph of some of the sub adult males recorded in the reserve is shown in Plate 6.

## 4.3.4.1 Molecular Sexing

Extraction of DNA from dung sample was carried out using QIAGEN Gel Purification Kit. PCR conditions were standardized with cycles that comprised of an initial denaturation at 93°C for 3 minutes, followed by 39 cycles each of 1 minute denaturation at 93°C, annealing at 51°C for 1 minute, extension at 72°C for 1 minute to complete a total of 40 cycles, and final extension at 72°C for 5-7 minutes. The PCR products were subjected to restriction digestion with 5  $\mu$ l of *Bam* HI (MBI Fermentas 5U/  $\mu$ l) and electrophoresed on a 2 per cent 1 agarose: 1 low melt agar gel to observe for bands that differentiate males and females.

Sixteen dung samples of animals of unidentified sex were collected from the reserve. They belonged mainly to juveniles, a group apparently difficult to sex by physical verification. After the DNA extraction, subsequent restriction digestion and analysis of the samples by electrophoresis, it was found that six samples produced three bands (males) and eight samples exhibited single band each (females). Two of the samples did not give any results. The photographs of gels corresponding to the presence of DNA at ~300 bp, and gels showing the bands of male and female are shown in Plates 8 and 9.

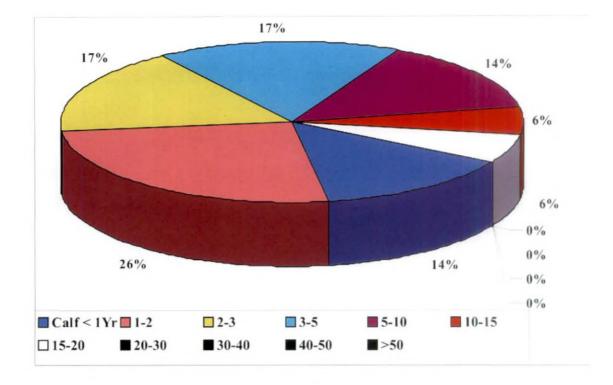


Fig. 7. Age-wise distribution of male elephants in the observed population

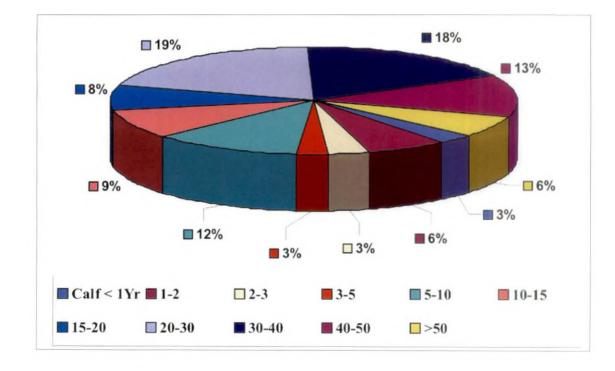


Fig. 8. Age-wise distribution of female elephants in the observed population

#### 4.3.5 Sex Ratio

The sex ratio was calculated based on the observed proportion of males and females in the population. The adult female to male ratio was found to be 1: 58.95; in sub-adults 1: 4.71, and in the juvenile category 1: 0.9.

#### 4.3.6 Fecundity Rate

The fecundity rate in the observed population was calculated by dividing the number of calves with the total number of adult females. The rate was worked out to be 0.099.

## 4.3.7 Intercalving Interval

The intercalving interval was calculated by dividing the number of adult females with the total number of calves observed in the population. The intercalving interval was worked out to be 10.1 years.

## 4.3.7 Population Number

Using the method of block count the total number of elephants counted by direct sighting on a single day in 23 selected blocks was 241 in different age groups. From the number of elephants recorded in the selected blocks, the total number of elephants in Periyar Tiger Reserve was calculated to be 644 with an R value of 0.85. The calculations are presented in Table 1 and 2. In the calculations, Periyar, Vallakkadavu, Thekkady, Pamba and Azhutha ranges are designated as Stratum A, B, C, D and E respectively. The total effective area for calculating elephant population in Periyar Tiger Reserve was taken as 760 km<sup>2</sup>, not taking into account the reservoir area during summer.

Sample	e Periyar (A)			Vallakkadavu (B)			Thekkady (C)			Pamba (D)			Azhutha (E)		
unit	Z	a	Za	Z	a	Za	Z	a	Za	Z·	а	Za	Z	а	Za
1	8.73	0	0	9.35	23	215.05	11.38	4	45.52	11.27	5	56.35	10.16	32	325.12
2	9.62	0	0	13.09	9	117.81	7.98	7	55.86	10.75	8	86	8.56	5	42.8
3	14.72	0	0	7.74	6	46.44	.14.59	22	320.98				13.45	26	349.7
4	13.65	12	163.8	<u>13.9</u>	29	403.1	7.92	17	134.64						
5	11.94	13	155.22				-								
6	11.56	12	138.72				,								
7	9.7	11	106.7												
8	12.71	0	0	Í				-	-		_				
9	19.75	0	0	·						·					
10	15.35	0	0			[				{			_		
TOTAL	127.73	48	564.44	44.08	67	782.4	41.87	.50	557	22.02	13	142.35	32.17	63	717.62
MEAN	12.77	4.8	56.44	11.02	16.75	195.6	10.47	12.5	139.25	11.01	6.5	. 71.18	10.723	21	239.21
Vz	9.78			6,53			7.63			0.067			4.144	<u> </u>	
Va		34.76			91.19			53.25			2.25			134	

Table 1A. Block Census Data

Z = Area of sampled unit

a = Number of animals in that unit

Vz = Variance in the area of units within each stratum

Va = Variance in the number of elephants recorded in each stratum

33

STRATUM	N	n	N(N-n)/n	Mean a	Va	Vz	Na	Nz
Α	362.26	10	12760.97	4.8	34.76	9.78	1738.85	4627.15
В	137.93	4	4618.24	16.75	91.19	6.53	2310.33	1519.99
С	94.97	4	2159.86	12.5	53.25	7.63	1187.13	994.05
D	78.16	2	2976.33	6.5	2.25	0.068	508.04	860.54
E	86.64	3	2415.52	21	134	4.14	1819.44	929.04
Total	759.96	23	24930.92	61.55	315.45	28.148	7563.79	8930.77

Table 1B. Computation of Block Census Data

= Total number of sampling units Ν

= Number of units sampled n

= Variance of numbers within units of a strata V

Nz is computed by multiplying N and the mean z for each stratum

The total number of elephants

 $X = Na \times Z$ 

Nz

644

X

8930.77

= X/Z = 644 / 760R 0.85 =

= 7563.79 X 760

Therefore the total number of elephants in Periyar Tiger reserve with an effective area for calculating elephant population is 644 with an R value of 0.85



Α



В

PLATE - 1: A - Grass lands in Azhutha Range of Periyar Tiger Reserve are the most preferred habitats by elephants.

B - The Periyar Reservoir covering an area of 26 sq. km. is a perennial source of water and the grass patches along the lake shore are very much preferred by elephant herds.





В

PLATE - 2: Solitary old females sighted in different parts of the reserve. A- In Thondiyar B - Near Thannikudy





В

PLATE - 3: A - Adult tusker aged between 15-20 years sighted in Nellikkampatty.

B- A group of elephants near a shola in Uppupara. The marked elephant was identified to be a 'makhna' aged approximately 15 - 20 years.

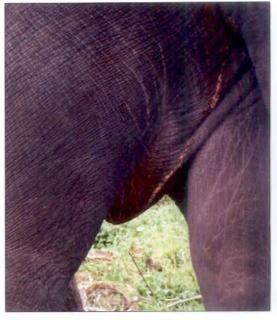




В

- PLATE 4: Elephant herds comprising of members in different age groups exhibiting matriarchial order.'
  - A Elephant herd in Anchuruli
  - B In Thannikudy





В



С

- PLATE 5: Observation of genitalia is the main method of sex determination by physical verification.
  - A Appearance of female genitalia when viewed from the rear.
  - B Appearance of male genitalia when viewed from the rear.
  - C- The circled part shows female genitalia.







В



С



D

- PLATE 6 : Sub adult tuskers sighted in different parts of the Reserve during the study period.
  - A In Thondiyar (10-15 years) B Near Edapalayam (5-10 years)
  - C Near Pachakkad (10-15 years) D- In Anchuruli (5-10 years)





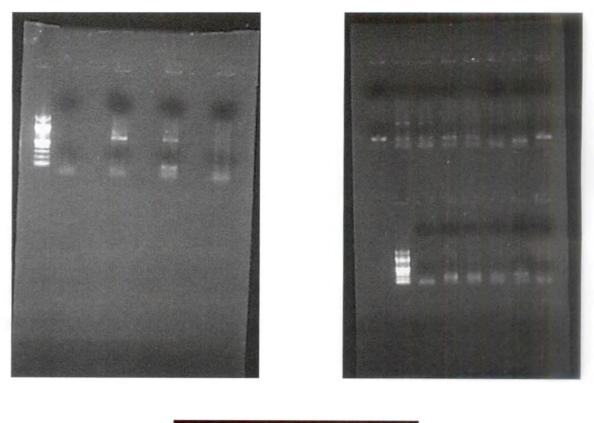
В



С

D

- PLATE 7 : During the study period, a number of calves below 1 year in most groups was sighted.
  - A Sighted in Anchuruli. B Pachakkadu
  - C Thannikkudi.
- D Uppupara.



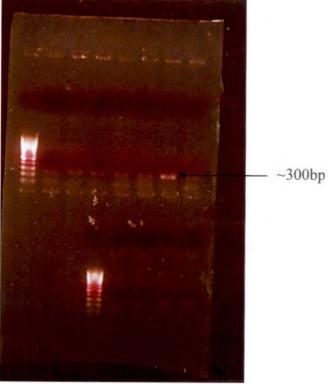
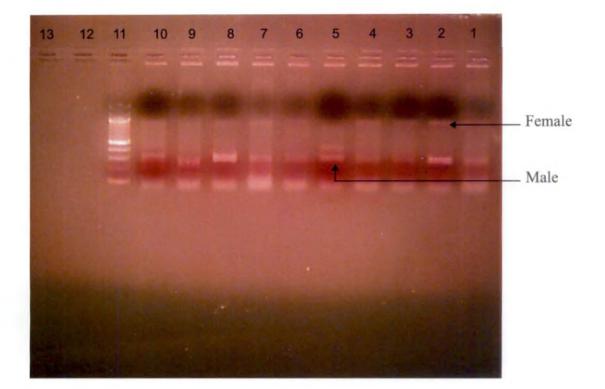


PLATE - 8 : Results of PCR amplification bands at ~300bp is positive.



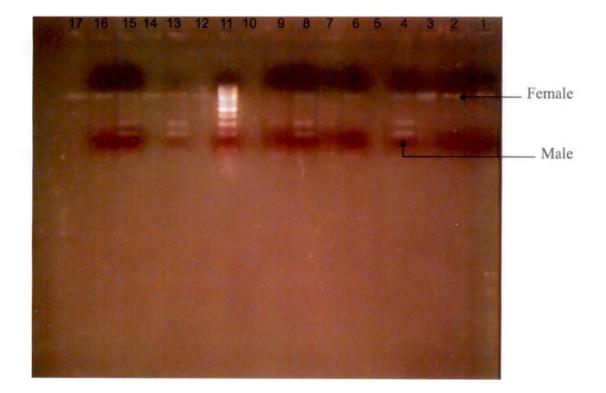


PLATE - 9 : Restriction digests of ZFX-ZFY PCR products with BamH1 electrophoresed on a 2% agarose gel. Arrow points to lines with a female and a male, the band of the female (intact ZFX fragment) corresponding to about 280 bp. A 100 bp lader is shown in lane 11.



PLATE - 10: A view of Sabarimala Temple located in Periyar Tiger Reserve.



PLATE -11: An elephant carcass floating in the reservoir.





В

PLATE -12: The absence of adult dominant breeding bulls in the population could possibly give chance for younger bulls or sub adult males to mate with oestrus females.

- A Sub adult male located with a group near Thondiyar
- B Sub adult male sighted along with an adult female near Edapalayam.



Α



В

С

PLATE -13: A - An adult female with deformity in the left hind limb. Such animals can be identified for marking particular groups.

B - Depigmentation or blotches on skin are taken in to account when age is determined.

C - Hollows or deep depressions on skull are indications of very old age.





В

PLATE - 14: A - Adult female with a juvenile female aged 3-5 years. B - A 20 year old adult female with a juvenile tusker.



PLATE - 15:Elephants and gaur sharing the same habitat for feed requirements. A scene from Manakkavala.



PLATE - 16: Elephants feeding on grass in Eucalyptus Plantation in Azhutha Range.

# Discussion

# 5. DISCUSSION

A number of studies on the population dynamics and demography of free ranging elephants of Periyar Tiger reserve has been conducted by different research agencies till date (Kurup, 1971; Vijayan *et al.*, 1979; Nair *et al.*, 1985). The published results by different investigators varied widely with regards to the structure and pattern of the population, especially the sex ratio in different age groups. But everyone in unison indicated a declining trend in the population and biased sex ratios towards females giving the elephants in Periyar Tiger reserve a high priority conservation status. This information formed the basis for the study.

#### 5.1 HERD COMPOSITION

#### 5.1.1 Age Structure

The age structure of a population, expressed as the distribution of the number of individuals in each age group, reflects the net result of fecundity and mortality schedules of that particular population. As such, age class structure is an important way of measuring demographical changes over time and for comparing different populations (Lindeque, 1991).

During the present study, a total of 239 elephants were sighted in 43 herds Among the different age groups adult females formed more than 50 per cent of the population, adult males constituting only 0.889 per cent, juveniles between 1-5 years, 20.76 per cent, subadults between 5-15 years, 20.76 per cent and calves below one year, 5.19 per cent of the total observed population. This indicates that there is a further decrease in the births in the Periyar population suggesting a deviation from the normal reproductive parameters in elephants observed elsewhere in Asian elephant populations. Only 10 calves were observed in the 43 herds that comprised of 101 adult females above 15 years of age suggesting an alteration in the reproductive pattern of adult females due to some stress, such as absence or limited presence of breeding males in the population. This is more evident by the mere presence of elephant groups comprising of only members from the sub adult category onwards and many adult females appear to be underproductive without any mammary development suggesting that they have not reproduced or in any stage of lactation or late pregnancy. In the observed agesex classes, adult females and sub adult females formed the major portion (69.53%) of the population, a shift towards the older age class.

The present study recorded the proportion of calves in the observed population to be only 5.19 per cent. This did not agree with the demographic study on elephants in Periyar and adjacent areas by Easa (2001), who concluded that the proportion of calves formed 12.40 per cent of the population which indicated a good proportion of breeding females and after comparing the results of the study with earlier studies, he came to the conclusion that Periyar population indicated a stable age distribution, pointing to a steady environment regardless of the fluctuations in the population.

During the ecological survey conducted in Periyar sanctuary in 1969, it was seen that the proportions of immature individuals to that of adults were very low, and thus taking all herds together, the proportion of immature individuals in the population was worked out to be 29 per cent and a ratio of about eight immature animals to every 10 cows indicating a significantly declining population. The low juvenile proportion indicates a natural state of reduced natality, which is a manifestation of the innate self- regulatory mechanism of population control in a given species In the African elephant this mechanism was working through the immediate physiological lowering of fertility level and a relatively slower process of reduced growth and delayed sexual maturity, indicated visibly in the population by longer mean calving interval (Kurup, 1971). The present study also observed a low proportion of immature individuals in the population.

Bhima and Bothma (1997) determined the age structure of the elephant population in Liwonde National Park, Malawi in 1993 and 1995 and it was observed that the population consisted of mostly young animals less than five years old (52.6 % and 44.8 % in 1993 and 1995 respectively). The sub adults aged between 5 to 15 years formed 23.9 per cent and 30.9 per cent and adults above 20 years constituted 23.5 per cent and 25.2 per cent in the respective years, suggesting a young and growing population. In comparison, the observed elephant population in Periyar Tiger Reserve constituted only 25.95 per cent young animals below five years, 20.56 per cent sub adults between 5-15 years and 53.3 per cent adults above 15 years. This suggests a population trend that is certainly declining indicated by a low proportion of young animals in the population.

Lindeque (1991) opined that if the rapid changes in population size were mostly due to sudden changes in recruitment or mortality, the population age structure would show changes in the relative proportion of individuals in each age category. The only known mortality factor that could possibly affect all age categories is indiscriminate poaching.

#### 5.1.2 Sex Categories and Sex Ratios

Periyar Tiger Reserve came into the limelight when research articles published on population study of elephants declared a progressively skewed adult sex ratio from about 1:6 (1969) to 1:19 (1977-'79) and then skewed substantially to 1:71 (1980-1982) reaching a peak of 1:122 in 1987-'89 (Uma Ramakrishnan, 1998). Skewing of the adult sex ratio will proceed at a geometric rate with the progressive death of one sex, thus the death of one or a few adult males will skew the ratio substantially as their numbers reduce in the population.

A good number of juvenile tuskers (more than 15) were observed in PTR during the study period. This is a very good indication of the improving number of tuskers in the population, but a lot depends on how they fare in the coming

years to become potential breeding bulls given the intense pressure Periyar Tiger Reserve is facing at present.

The observed sex ratio in different age groups in the present study also revealed a skewed sex ratio from the sub adult category onwards with more disparate ratio in the adults (1:59). The skewed adult sex ratio at Periyar is the most disparate recorded for any major Asian elephant population or African population. This disparity has been mainly attributed to severe poaching of males from the sub adult category onwards for ivory until early 1990's resulting in selective removal of one sex from the population (Uma Ramakrishnan *et al.*, 1998). This is one factor that has been agreed upon by researchers and forest authorities alike, although in earlier times skewed sex ratios were possibly prevalent due to capture of ideal tuskers by South Indian Kings for their army. But, at present the situation has worsened due to unscrupulous killing for ivory and weak protection measures.

With an adult sex ratio of 1:101 and a total population of 1166 (605 adult females, 6 adult males) recorded by Sukumar *et al.* (1998), the genetically effective population size (Ne) considering only the female biased sex ratio, was calculated to be 24. Thus, Ne is at best only 2 per cent of the population and 4 per cent of the breeding population, assuming that all adults are potentially capable of breeding. Compared with this; Ne may be 40 per cent and 77 per cent of the total and adult populations, respectively, in Sri Lanka. Heavily skewed sex ratios could make the genetically effective population size much less than the actual population size in a polygynous species such as the elephant (Katugaha *et al.*, 1999).

Sukumar (1989) discussed in detail the possible influence of disparate sex ratio on the fertility of the elephant population. Most mammalian populations are reported to have an adult sex ratio biased towards the females. He suggested that the operational adult sex ratio need not be as disparate as observed sex ratio in the population. While ivory poaching has been a problem in most southern Indian states, reports suggest that Periyar Tiger is the worst affected. The highly skewed or disparate sex ratio in age groups above sub adult stage has been attributed mainly to severe poaching during the period prior to 1987, when even juveniles have not been spared (Uma Ramakrishnan *et al.*, 1998). Computer simulation modeling to deduce the number of male elephants killed and the quantities of ivory harvested over a 20 year period in Periyar Tiger Reserve conservatively estimated that 336-388 tuskers have been poached and 3256-3334 kg ivory harvested by poachers over the 20 year period from 1974-1994, and the maximum harvest came from the 10-20 year old class (Sukumar *et al.*, 1998). Easa (2001) also interpreted that another factor responsible for the disparate sex ratio was due to the official capture of elephants from the Reserve since 1810 that lasted till the 1970's.

It is reported that poaching for ivory began assuming threatening proportions during the 1970's, the average number of elephants poached over the last decade in India being approximately 87 per year (data compiled by Asian Elephant Research and Conservation Centre, Bangalore and Wildlife Trust of India, New-Delhi). The increasingly female biased sex ratios could affect populations seriously, by lowering the effective population size, lowering birth rates and decreasing genetic viability due to inbreeding. It is thus imperative to monitor sex ratios on a regular basis in addition to population sizes (Vidya, *et al.* 2003).

#### 5.2 THE CALVING INTERVAL

The mean calving interval has been considered to be the single most important parameter influencing the growth rate of an elephant population, the effect being slightly more pronounced as mortality increases (Hanks and McIntosh, 1973). Of all the parameters worked out during the present study, the calving interval of the elephants in Periyar is a matter of great concern, because the value calculated is above 10 years. Majority of the elephants observed were adult females and a great proportion of these animals were without calves and also did not apparently show signs of late pregnancy or signs of mammary development. It is possible that the alteration of reproductive physiology in female elephants could be related to the near absence of adult males in the population over an extended period of time (Uma Ramakrishnan *et al.*, 1998).

The present study reveals that only one in 10 adult females conceive every year, which again is suggestive of severe shortage of adult males in the population. The absence of older dominant males in the population gives opportunity for the simple question about the presence of 10 calves below one year in the population. It is suggested that with the absence of older males, the younger age groups successfully mate with the oestrus females. But more investigations are necessary to prove this possibility (Plate 12).

#### 5.3 FECUNDITY RATE

An important demographic effect of high rates of poaching and extreme skew in adult sex ratios is a decline in the birth rate of the population. The present study calculated the fecundity rate as 0.099, drastically low compared to other Asian elephant populations. With an extremely low number of adult males in the population, a substantial number of females coming into oestrus would not be detected and mated. Another possibility reason for the low number of calves and juveniles seen at Periyar is that females may have stopped reproducing under conditions of extreme stress (poaching), or infant mortality may have been very high, though accurate records on calf mortality is unavailable.

Lande (1988) had suggested that the most likely cause of fecundity decline is an allele effect; which means that individuals in population declining to low numbers experience low viability and reproduction from nongenetic reasons, and there may be a threshold density or number of individuals from below which the population cannot recover. Uma Ramakrishnan *et al.* (1998) reported that the average fecundity was only 0.075/adult female/year in Periyar Tiger Reserve as compared to between 0.20 and 0.025 for the more productive populations in southern India.

#### 5.4 MOLECULAR SEXING

Extracting DNA from dung adopted the modern technique of molecular scatology in this study to sex animals difficult to identify by physical verification. This is the first study to combine reliable conventional tools and molecular methods to assess sex of free-ranging elephant population of Periyar Tiger Reserve. It is found that molecular sexing is a very reliable tool for assessing the sex of individuals of free-ranging wild animal populations.

Vidya *et al.* (2003) demonstrated the application of molecular sexing of free-ranging elephants for the first time in India in Nagerhole and Mudumalai-Bandipur Reserves, from DNA extracted from dung, based on ZFX-ZFY fragment amplification and ZFY-specific *Bam* HI site restriction to differentiate between the sexes. Molecular sexing was suggested to be a useful tool to sex juveniles that lack dimorphism, or to estimate population sex ratios by carrying out noninvasive sampling.

#### 5.5 NUMBER OF ELEPHANTS IN PERIYAR TIGER RESERVE

As per reports on the census figures, the year 1978 recorded 588 elephants, 950 elephants in 1987, 980 in 1989, 615 in 1993, and 559  $(0.72/\text{km}^2)$  in 1997.

The present study included a block count method for estimating the number of elephants in the reserve and it gave a total of 241 elephants in all age groups from selected 23 blocks across the reserve. After computing this value to the entire area, the total number of elephants in Periyar Tiger Reserve was calculated as 644 with an R value of 0.85.

Official records on the census conducted this year using an indirect dung count method is yet to be released. The population numbers obtained by the block count method in this study indicate an elephant population that is not growing and a figure that is widely accepted by many research agencies. In addition, the present study on the herd composition of the observed population points to declining trends because of highly disparate demographic parameters. It must also be noted that for a large mammal like the elephant with very long generation interval, routine monitoring for an extended period of time is essential to identify trends in the population. Careful attention should be paid to the demography, age structure, sex ratio, mortality, fecundity and trends in each population to avoid demographic catastrophe or accelerated loss of genetic variability.

Kurup (1971) had reported that elephant was the dominant species of wildlife in Periyar sanctuary and their dominance was easily seen in the by-lake sector where large herds comprising of several family units seemed to be ubiquitous, especially near the grassy peripheral belt around the lake. He also suggested that this has led the forest officials to assume that the elephant population in the sanctuary was increasing at a faster rate, but a closer look at the herd composition and specifically the immature- adult ratio indicated a population crash rather than an explosion.

Ramachandran *et al.* (1986) estimated about 800 elephants in the reserve during 1977-'78 giving an overall density of approximately one elephant per square kilometer by the indirect dung count method. At the same time, they reported an ecological density of about three elephants per square kilometre in certain areas. Uma Ramakrishnan *et al.* (1998) conducted rapid surveys on the elephant population in Periyar Tiger Reserve during March- May 1994, June 1994 to June 1995 and during February-April 1997 and the total number of elephants in the sanctuary was found to be 1166 during 1994 survey using line transect, indirect count method.

Considering the population numbers worked out for Periyar Tiger Reserve during the present study, it will be beneficial to find the effective population size or the minimum viable population size after long-term monitoring of the population

The effective population size can also be roughly calculated as four times the number of breeding males multiplied by the number of breeding females, divided by the sum of breeding males and breeding females. Populations of less than 500 animals will always have an effective population size of less than half that number and this will be further reduced if the sex ratio becomes distorted or if the contribution of adults to breeding is very unequal. Removal of adult males from the population will result in reduced effective population size and increased rate of genetic drift. These two factors threaten the long term survival of the Asian elephant as a species by gradually losing the ability to respond to environment changes by adaptation.

Uma Ramakrishnan *et al.* (1998) has stated that the elephant population in Periyar Tiger Reserve can possibly withstand the drastic reduction in the genetically effective population size (Ne), because this reduction has occurred over a single elephant generation (about 20 years). In theory, a one generation reduction of Ne to 20+ still retains about 98 per cent of the original (prior to the poaching spate) heterozygosity, assuming random sampling of individuals. The authors also state that the allelic diversity could, however be expected to suffer greater decline, with rare alleles having a high probability of being lost during a population bottleneck.

But, this may not be true unless we have sufficient reason to believe that poaching has been totally controlled in Periyar region. The recorded number of ivory poaching in Periyar in recent years is very low and attributing this to 'efficient protection' may not be entirely true because of the simple logic that there are no more good sized ivory to be poached from Periyar at present. It must also be noted that tiger and leopard skin, and other valuable wildlife goods were recently seized from different parts of Kerala, especially Idukki district indicating that poachers are still very much active and waiting. So the population bottleneck is likely to continue affecting future elephant generations also.

Shaffer (1981) opined that the concept of minimum viable population for any given species in any given habitat is the smallest isolated population having a 99 percent chance of remaining extant for 1000 years despite the foreseeable effects of demographic, environmental, and genetic stochasticity, and natural catastrophes.

Populations of 500-2000 animals will require minimal genetic intervention in the next 100 years (about five elephant generations). Every effort should be made to maintain or allow increase in these populations, and considerations should be given to the introduction of new genetic material (one breeding bull per generation is considered adequate) to achieve the goal of a naturally reproducing population of 2000+ animals.

#### 5.6 PTR, AN IDEAL HABITAT FOR ELEPHANTS ?

Periyar Tiger reserve, one of the biggest wildlife reserves in southern India, part of the designated Elephant reserve No. 10 under Project elephant, forms a vast contiguous compact block that extends up to Shengotta and eastwards into the state of Tamil Nadu. The tropical warm climate, high elevation (about 1000 to 2000 m), heavy rainfall (2000 mm), undulating terrain, extensive grasslands, marshes, river and dense forests are highly favourable for wild animals.

During the period of the present study, it was observed that the Reserve was less disturbed compared to most protected areas in Kerala with respect to the number of settlements, enclosures and estates. But it was observed that many of the areas around the Periyar lake are temporarily colonized by tribal fisher folk

granted permission to harvest freshwater fish from the reservoir as part of the eco-development programme implemented since 1996-97. The ever increasing influx of devotees to the Sabarimala temple situated in the core area of the reserve is certainly taking its toll on the pristine jungles by way of degradation of forests, human disturbance and extensive temple development projects (Plate 10). This reserve and adjoining contiguous forests still remain a wonderful mosaic of ecosystems that can harbour and maintain good elephant population if degradation processes cease to continue and protection measures rise to the occasion to save the remaining population, especially the immature animals.

In elephant reserve No. 10, most of the evergreen forests are confined to Periyar Tiger reserve and Goodrickal range of Ranni forest Division. Extensive reed areas are located in Goodrickal, Kallar and Vadasserikkara Ranges regularly exploited for industrial purpose. However, the eastern part of this Reserve bordering TamilNadu is still intact. Major portions of the evergreen forests are infested with the weed, *Mikania*. The forests are more or less contiguous with a few enclosures, settlements and estates in Vallakkadavu Range, but Sabarimala areas of Vallakkadavu are degraded due to pressure from pilgrims. The fewer number of settlements and enclosures in the Reserve make it an ideal habitat for elephants (Easa, 2001).

The prospects of Periyar Tiger Reserve in the years to come will be looked upon with concern unless stringent managemental measures are adopted to protect its forests and wildlife. The extensive developmental projects initiated at Sabarimala will in due course certainly affect the ecosystem drastically through progressive and rapid forest degradation. It is highly imperative that a clean, upto-date mortality recording system for elephants (Plate 11) be followed in the reserve which at present is lacking. Adopting a regular population and health monitoring programme at least on annual basis can provide important information on the status of elephants to help implement strategies in crisis situations.

Identification of individual elephant herds/groups in the reserve area should be taken up as part of the monitoring programme (Plate 13 and 14) to predict the trends in the population and changes in demographic parameters for an extended period of time, like the Amboseli Elephant Research Project in Amboseli National Park, Kenya which maintains the only continuous, long-term data set for an individually known free-ranging elephant population (Moss, 2001).

Periyar Tiger reserve is an excellent habitat for elephants and they coexist well with other larger mammals found in the reserve (Plate 15 and 16).

#### 5.7 VIEWS PROVOKE THOUGHTS...

There are many options and alternatives suggested to resolve the Periyar crisis. Many are aimed at improving the habitat conditions, stepping up protection activities, boosting eco-development programmes and educating the local mass.

Uma Ramakrishnan *et al.* (1998) suggested the translocation of adult male elephants from other populations with surpluses, in order to improve the genetically effective population size to 50 or above. This would also serve to stimulate the birth rate of the population. But it is a known fact that all other south Indian elephant populations are also facing the problem of skewed sex ratios and poaching. In such a situation, it was suggested that translocation of makhnas from the North-east Indian population is a possibility, provided population genetic studies are conducted in both the populations. North-east population is believed to have good sex ratios and good number of adult makhnas.

Sukumar *et al.* (1998) reported that 336-388 tuskers were removed from the Periyar population during the 20 year period from 1974-1994 by computer simulation modelling studies. If that is true, there should have been a proportionate increase in the makhna population in Periyar tiger reserve. But, during the present study, only very few makhnas were sighted in different age categories in the observed population. This observation prompts me to suggest the existence of natural selection against makhnas in Periyar. In such a situation, translocation of makhnas from the North-east population should be undertaken with caution and after thorough investigations, as their survivability in Periyar could be under question.

It is also interesting to look at the situation prevailing in Kerala regarding the free ranging and captive elephant population. Studies have proved beyond doubt that the free ranging population has very low number of adult males, but the captive population of elephants (approximately 1000 numbers) in the State is comprised of more than 95 percent adult massive tuskers, many of which are ageing and under utilized for breeding purposes. If this wide gene pool can be utilized in some feasible way to improve the status of wild populations, such as captive breeding programmes followed for other endangered species, then the question of viability of free ranging populations will not be a problem in the near future. In Kerala, there are many captive male elephants under different ownerships that are considered "rouge" and chained for life. An attempt can be made to release these elephants into the wild after radio collaring and subsequent close monitoring to observe ranging pattern, breeding behaviour and above all survival from poaching and human-elephant conflict.

Kerala state has the expertise, infrastructure and resource to preserve this fantastic gene pool through cryopreservation of semen. But where are the will power, moral support and funds to encourage enthusiastic professionals to take up the responsibility of conserving this magnificent animal. In my opinion, these attempts and many others like radio-collaring of suitable elephants are long overdue in the state of Kerala.



#### 6. SUMMARY

Conservation of biodiversity is one of the top priority issues in the present context. Elephants act as flagship species in conserving the diversity of our tropical forests and hence information on different aspects of their ecology is essential. Periyar Tiger Reserve in Southern India, where once elephants roamed freely and in abundance is now cause for concern because of decline in population trends and disparity in sex ratios in different age groups, especially in adults. Several field researchers attributed the phenomenon primarily to severe poaching that took place in the Reserve since the 1970's. As a result of the selective removal of one sex from the population, severe detectable demographic changes in the population were observed such as skewed sex ratios, lengthy calving interval and poor fecundity rates by earlier workers.

A significant amount of variations have been observed regarding the results of demographic studies conducted in Periyar Tiger Reserve by different agencies. In this study, an attempt was made to assess the population, sex and age of the elephant population using a combination of reliable conventional tools and modern molecular techniques. The study was conducted during the summer months from March to May 2005. The important findings of the study are summarized as follows:

- A total of 239 elephants were sighted in 43 herds, with a mean herd size of 5.5 with sex ratio in calf and juvenile category remaining almost same as observed with Asian elephant populations elsewhere.
- The observed elephant population constituted of more than 50 per cent adult females above 15 years of age.

- 3) Only two adult males aged between 15-20 years were observed during the entire period of study, and the proportion of adult males in the population was only 0.889 per cent indicating a highly skewed adult sex ratio.
- 4) The fecundity rate calculated for the elephant population based on the observations was 0.099, compared to 0.20- 0.25 in similar populations across the country.
- 5) The intercalving interval in the population was more than 10 years suggesting low number of adult bulls.
- 6) Elephants preferred the grasslands and adjoining sholas more during the summer months followed by the moist deciduous forests along the Periyar reservoir.
- 7) There is a decline in the population numbers evident from the direct block count census during May 2005.
- 8) There is a paradigm shift in sex ratio from the subadult category biased towards the females and this could be attributed to severe poaching that took place till the early 1990's.
- 9) The sex ratio calculated in Periyar is 1:58.95 in adults, 1:4.71 in subadults and 1:0.9 in juveniles.
- Molecular scatology is an important tool to assess the sex ratio of free-ranging elephant populations where other conventional methods are not feasible.

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# POPULATION, AGE AND SEX ASSESSMENT OF FREE-RANGING ELEPHANTS OF PERIYAR TIGER RESERVE

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Abstract of the thesis submitted in partial fulfilment of the requirement for the degree of

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### ABSTRACT

A demographic study on the free ranging elephants of Periyar Tiger Reserve was conducted from March to May 2005. Herd composition, age structure and sex assessment were the main parameters recorded. The technique of molecular sexing using dung extracted DNA was incorporated into the study to reduce error in determining sex by physical verification. The published results by different investigators on the structure and pattern of elephant population in Periyar Tiger Reserve varied widely, especially on the age structure and biased sex ratios towards females giving the elephants in Periyar Tiger reserve a high priority conservation status. This information formed the basis for the study.

Of the total 239 elephants observed in 43 herds, 193 elephants were classified based on age and sex. It was found that more than 50 per cent of the recorded elephants were adult females. The calves formed 5.19 per cent, juveniles 20.76 per cent, sub adults 20.76 per cent and adult males only 0.889 per cent of the observed population. Females formed 81.99 per cent and males formed only 18.01% of the population, but the proportion of males and females in the calf and juvenile category was almost same, with a shift biased towards the females from the sub adult categories were 1:0.9, 1:4.71 and 1:58.95 respectively. In the observed age-sex classes, adult females and sub adult females formed the major portion (69.53 %) of the population, a shift towards the older age class.

The calving interval of the Periyar elephants was above 10 years and the fecundity rate 0.099 indicating a population suggestive of acute shortage of breeding bulls and approximately only one in ten adult females conceive every year. This is the first study to combine reliable conventional tools and molecular methods to assess sex of free-ranging elephant population of Periyar Tiger

Reserve. It is found that molecular sexing is a very reliable tool for assessing the sex of individuals of free-ranging wild animal populations.

The study infers that the present situation of the Periyar elephants is crucial and calls for further research and immediate active population management strategies.

Appendix

## **ANNEXURE 1**

Composition of reagents and buffers used in the study

1. Composition of Digestion buffer (100 ml)

a.	100 mM Nacl	2 ml

Ъ.	10 mM	Tris pH	8.0	1 ml
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- c. 25mM EDTA -- 5 ml
- d. 2 % SDS -- 20 ml
- e. Water a.a --100ml

2. 5x TBE buffer (1000 ml)

Tris buffer	54g
Boric acid	27.5g
EDTA	3.722g
Water a.a	1000ml

3. Sodium chloride (5 M)

For 100 ml, dissolved 29.22 g of sodium chloride in 80 ml of distilled water, make up the volume to 100 ml, filtered and stored at room temperature.

4. Phenol: Chloroform: Isoamyl alcohol mixture

Phenol: chloroform: isoamyl alcohol mixture was prepared by mixing 25 parts phenol; 24 parts chloroform and 1 part isoamyl alcohol.

5. EDTA (0.5 M, pH 8.0)

Dissolved 18.61 g of EDTA (disodium, dihydrate) in 80 ml of distilled water. Adjusted Ph to 8.0 with NaOH and made the volume to 100 ml. The solution was filtered, autoclaved and stored at room temperature.

6. Tris Buffered Saline (pH)

Sodium chloride	140 mM	8.18 g
Potassium chloride	0.5 mM	0.0373 g
Tris base	0.25 Mm	0.0303 g

Dissolved in 900 ml distilled water and adjusted the pH to 8.0. Made up the volume to 1000 ml, filtered, autoclaved and stored at room temperature.

7. Ethidium Bromide (10 mg/ ml)

Dissolved 100 mg of ethidium bromide in 10 ml distilled water and stored in a dark bottle at 4 degree Celsius.

8. Gel Loading Buffer (6X)

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Bromphenol blue	0.25%	50 mg
Xylene cyanol	0.25%	50 mg
Sucrose	40%	8 g

Stirred well in 20 ml distilled water and stored at 4 degree Celsius.

## ANNEXURE II KERALA FOREST DEPARTMENT ELEPHANT CENSUS - 2005

## **BLOCK COUNT RECORDS – DIRECT SIGHTINGS**

Forest Division	:	Vegetation Types
Forest Range	:	(in percentage)
		Evergreen :
Date	:	Semi evergreen :
Weather	:	Shola forest :
Name of Camp	:	Moist deciduous:
No. & Name of Bloc	: <b>k</b> :	Grassland :
Block size	:	Vayals :
Starting Time	:	Plantation :
Finishing Time	:	Others (specify):

Time	No. of animals	M	ale	Fer	nale	Juve nile	Calf	Un known	Type of habitat	Place of sighting	Remarks
		Adult	S.adult	Adult	S.adult			,			

Investigator's Name Address & Signature

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Name, designation & Signature of the staff

## Annexure : III Selected Census Blocks

## Periyar Range

Block Number	Block Name	Block Size
1	Chokkampatty	8.73
2	Mukkar	9.62
3	Moolavaiga	14.72
4	Lakshmipara	13.65
5	Mlappara	11.94
6	Ummikuppan	11.56
7	Kumarikulam	9.70
8	Kottamalapallam	12.71
9	Randattinkara	19.75
10	Uppermanalar	15.35
Total Area	· · · · · · · · · · · · · · · · · · ·	127.73

## Vallakkadavu Range

Block Number	Block Name	Block Size
1	Thondiyar	9.35
2	Dam site	13.09
3	Kozhikanam	7.74
4	Aruvipalam	13.90
Total	· · · ·	44.08

## Thekkadi Range

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Block Number	Block Name	Block Size	
1	Methakanam	11.38	
2	Brandipara	7.98	
3	Manakavala	14.59	
4	Edapalayam	7.92	
Total		41.87	

## Azhutha Range

Block Number	Block Name	Block Size	
1	Sathram	10.16	
2	Undamedu	8.56	
3	Uppuppara	13.45	
Total		32.17	

## Pamba Range

Block Number	Block Name	Block Size	
1	Sabarimala	11.27	
2	Pudhusery	10.75	
Total		22.02	