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Estimation of leopard population size in a secondary forest within Malaysia's capital agglomeration using unsupervised classification of pugmarks

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Abstract: The objective of this study was to estimate the population size of common leopard (*Panthera pardus*) in Ayer Hitam Forest Reserve in Selangor, Malaysia. Long term survival of leopard population in this area is threatened due to small size of the forest, isolation and presence of various disturbances in the habitat. However, no estimates are available on the number of leopard in the study area. We used unsupervised classification of pugmarks that allows clustering of the data sets based on their inherent similarities. Study was conducted during February to November 2008. Linear measurements of front and hind tracks and strides (n = 124) were classified using hierarchical cluster and discriminant analysis which indicated that at least four individuals of leopard were present in the study area of 1,411 ha. Despite the small size of the forest, the area is still rich in both fauna and flora. The leopard being the predator, has an important role in maintaining the health of the ecosystem. Therefore, the population size of the leopard could be used as indicator of sustainable conservation and management of the species in the area.

Resumen: El objetivo de este estudio fue estimar el tamaño poblacional del leopardo común (*Panthera pardus*) en la Reserva Forestal Ayer Hitam in Selangor, Malasia. La supervivencia a largo plazo de la población de leopardos en esta área está amenazada debido al tamaño pequeño del bosque, al aislamiento y a la presencia de varios disturbios en el hábitat. Sin embargo, no hay estimaciones disponibles de leopardos en el área de estudio. Usamos una clasificación no supervisada de las huellas, la cual permite la agrupación de los conjuntos de datos con base en sus similitudes inherentes. El estudio fue realizado de febrero a noviembre de 2008. Se hizo una clasificación de las medidas lineales de las huellas delanteras y traseras y de las zancadas (n = 124) por medio de un análisis discriminante jerárquico de conglomerados, el cual indicó que al menos hay cuatro individuos de leopardos presentes en el área de estudio de 1,411 ha. A pesar del pequeño tamaño del bosque, el área sigue siendo rica en fauna y flora. Tratándose de un depredador, el leopardo juega un papel importante en el mantenimiento de la salud del ecosistema. Por lo tanto, el tamaño poblacional del leopardo podría ser usado como un indicador de la conservación y el manejo sustentables de la especie en el área.

Resumo: O objectivo deste estudo foi o de estimar a população do leopardo comum (*Panthera pardus*) na Reserva Florestal de Ayer Hitam em Selangor, Malásia. A sobrevivência a longo prazo da população de leopardos nesta área está ameaçada devido à pequena dimensão da floresta, isolamento e presença de vários distúrbios do habitat. Contudo, não estão disponíveis estimas sobre o número de leopardos na área estudada. Usou-se uma classificação não supervisionada de pegadas que permitiram o agrupamento das séries de dados baseados nas suas semelhanças inerentes. O estudo foi conduzido de Fevereiro a Novembro de 2008. As medidas lineares das distâncias entre os sinais dos membros anteriores e posteriores e da

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passada (n = 124) foram classificados usando uma análise de agrupamento e análise discriminante o que indicou que pelo menos existiam quatro leopardos na área de estudo (1,411 ha). Não obstante a pequena dimensão da floresta, a área é ainda rica quer quanto a fauna quer quanto a flora. Sendo o leopardo um predador, ele tem um papel importante na manutenção da saúde do ecossistema. Sendo assim, a dimensão da população de leopardos pode ser utilizada como indicador da conservação e da gestão sustentada da espécie na área.

Key words: Discriminant analysis, fragmented habitat, Malaysian tropical rain forest, *Panthera pardus*, population size, track measurements.

Introduction

Reliable estimation of population size is a key component in wildlife ecology, conservation and management (Alibhai *et al.* 2008; Caughley 1977; Seber 1992; Stander 1998). This is also required to identify the priorities in allocation of limited resources, formulate conservation strategies, evaluate conservation programs and develop approaches of effective management (Karanth 2003; Nowell & Jackson 1996; Stander 1998). Use of indices, particularly those based on counts and measurements of presence signs (tracks and scats), have received much attention in recent years to describe the status and trends of wildlife populations (Pollock *et al.* 2002). This approach is promising and practical in studies of large predators, e.g. wild cats (family *Felidae*), which are generally sparsely distributed, secretive, nocturnal and mostly solitary (Beier *et al.* 1995; Eltringham 1979; Gusset & Burgner 2005; Sargeant *et al.* 2003; Seidensticker *et al.* 1973). Track measurements can be efficiently used to recognize individual felids as their shapes and sizes vary between ages and sexes across various taxa e.g., *Panthera tigris* (Panwar 1979a, b; Sankhla 1978; Sharma *et al.* 2003, 2005), *Panthera parduus* (Gusset & Burgener 2005), *Panthera uncia* (Riordan 1998) and *Puma concolor* (Beier & Cunningham 1996; Fjelline & Mansfield 1989; Grigione *et al.* 1999; Lewison *et al.* 2001). As track measurements depend on soil types, slopes and even involved personnel, this methodology needs strict standardization (Fjelline & Mansfield 1989; Grigione *et al.* 1999; Lewison *et al.* 2001; Sharma *et al.* 2005).

High leopard densities have been recorded previously in a number of sites such as Rhodes Matopos National Park, Zimbabwe (23.6 leopards/ 100 km^2), Londolozi Game Reserve, South Africa (23.8 leopards/100 km^2), Sanjay Gandhi National Park, India (41.7 leopards/100 km^2) (Edgaonkar & Chellam 1998; Marker & Dickman 2005; Stander *et al.* 1997). However, in South-East Asia, Simcharoen *et al.* (2008) estimated a density of 7 adult females/100 km^2 in Huai Kha Khaeng Wildlife Sanctuary, Thailand. In general, leopard home range size could be as small as 8.8 km^2 in a prey rich habitat (Grassman 1999). Even though availability of medium-sized ungulates is the principal determinant of leopard densities (Hayward *et al.* 2006), it is not an obligatory requirement as in Rhodes Matopos local leopards prey mainly on the rock hyrax (*Procavia capensis*) and bush hyrax (*Heterohyrax brucei*) (Grobler & Wilson 1972; Smith 1977).

In this study, we used track measurements to estimate the leopard population size in Ayer Hitam Forest Reserve, Malaysia, which is located within the capital agglomeration of Kuala Lumpur. Very little is known about this predator in Malaysia and elsewhere in South-East Asia, particularly about its living in completely human-dominated environments where only small patches of its original habitats have remained intact (Azlan & Sharma 2006; Grassman 1999; Kawanishi 2002; Rabinowitz 1989; Simcharoen *et al.* 2008). In this paper we present the results of leopard population estimation conducted in an isolated forest patch in Selangor, Malaysia.

Study site

Ayer Hitam Forest Reserve (AHFR) is located in Puchong, state of Selangor in Malaysia, about 10 km south west of Kuala Lumpur. AHFR is considered a lowland tropical rain forest having steep slopes with highest peak reaching 645 m above sea level (Khairil 2001; Noor *et al.* 2007a). The mean monthly temperature ranges from 22.7 - 32.1 °C and the daily average humidity is about 83 %

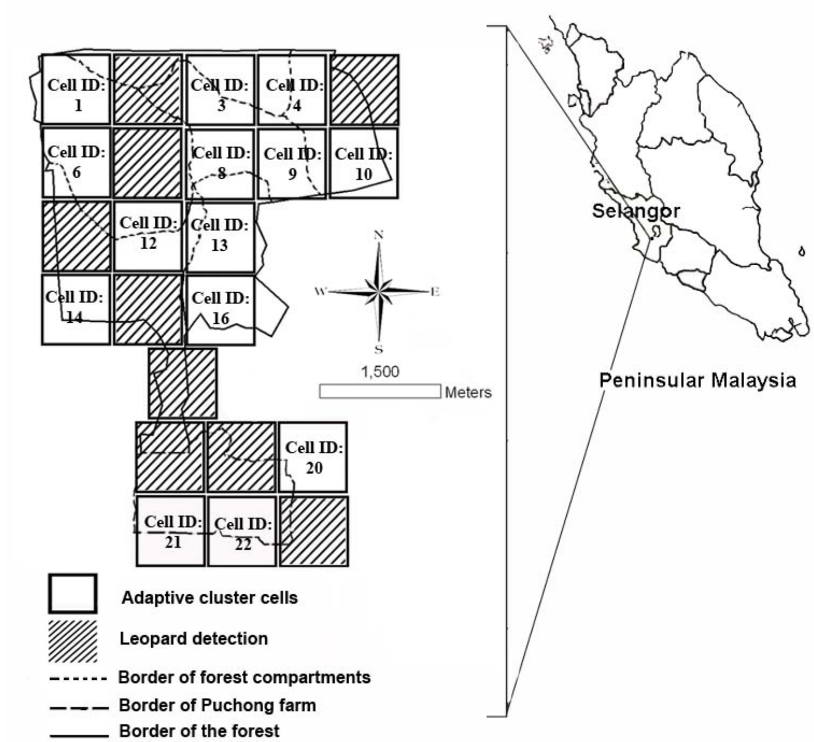

Fig. 1. Location of the study area and adaptive cluster sampling design in the site.

(Bawon & Amat Ramsa 2005). The forest was selectively logged between 1936 and 1965 (Zakaria & Topani 1999). The original size of AHFR shrank from 4,266.23 ha in 1965 to 1,262.33 ha in 1997 because of road construction, agriculture and housing projects (Noor *et al.* 2007b). Current size of AHFR is only 1,248 ha (Ainuddin *et al.* 2007). However, the reserve is still rich in floral and faunal diversity. A total of 430 species of fruit trees, 127 timber species and 98 species of medical plants are reported from AHFR (Faridah Hanum 1999). In addition, a total of 232 species of vertebrates including mammals, birds, reptiles, amphibians and fishes have been recorded (Norini *et al.* 2003; Zakaria & Rahim 1999; Zakaria & Topani 1999; Zakaria *et al.* 2001). Furthermore, AHFR represents the only lowland tract left within the metropolitan area of Kuala Lumpur (Faridah Hanum & Khamis 2004).

In the south, AHFR is connected with the Puchong Farm (PF, 163 ha) which includes the agricultural fields, fishery, buffalo and chicken farms (TPU 2009). The farm was originally a part of the forested area of AHFR. Therefore, this study was conducted both in AHFR and PF, covering the entire area of 1,411 ha (Fig. 1).

Methods

Data collection

The collection of data was carried out periodically on the monthly basis between February - November 2008 using the leopard track surveys across the 23 1x1 km grid cells that covered the whole study area (Fig. 1; Burdett *et al.* 2006; Karanth & Nichols 2002). The size of the grid cells was smaller than the minimum female leopard home range known in literature (Grassman 1999). Thus, the area was covered uniformly without gaps (Henschel & Ray 2003). Prior to cell-to-cell surveys, we obtained preliminary information about leopard occurrence from indigenous and knowledgeable people that live close to the forest. All leopard individuals sighted in the study area and those reported from other studies were melanic and individual identification using camera traps or direct sightings would not have given desirable results (Kawanishi 2002; Mark Rayan, Pers. Comm.). Therefore, use of track measurements became an indispensable tool. Further, this method met the following assumptions (i) Study period had to be short as total length of footprints

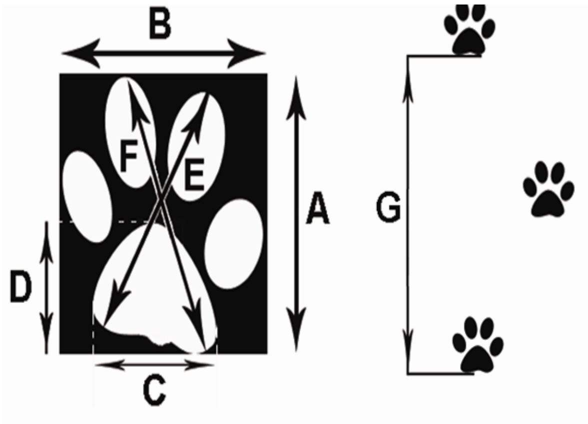

Fig. 2. Linear measurements of leopard (*Panthera pardus*) tracks taken in this study. A: Track length, B: Track width, C: Pad width, D: Pad length, E: Diameter distance between top of second (left) toe and right pad curve, F: Diameter distance between top of third toe and left pad curve, G: Stride size.

and stride sizes may vary significantly over a long study span; (ii) Estimating population using track measurements could be practical when target population is closed to immigration of non-resident individuals to the site and/or emigration of existing individuals to another area; (iii) This is a reliable method for population estimation in a small and fragmented area.

According to adaptive cluster sampling method, neighboring cells are added to the sampling procedure whenever the variable of interest was found in a cell (Roesch 1993; Thompson 1996). This builds on a simple or stratified random sampling design by sampling all the cells bordering those where leopard presence was recorded in the initial survey, and continuing to do so until the cluster is surrounded by cells that fail to detect leopard presence. Consequently, transect lines were selected in the cells with confirmed leopard detections and based on main riversides and indigenous people trails as leopards are known to show propensity for existing trails or riversides rather than dense vegetation (Kawanishi 2002; Stander 1998). Each transect was continued until the cell was completed or topographical prevention was met. In order to estimate the number of surveys required in the study area, leopard detection/non-detection data were obtained in 24 surveys conducted in the first month of the study. The detection probabilities of leopards were estimated for each survey using PRESENCE software version 2.4 (Hines 2006). The lowest detection probability (i.e. p = 0.38) was used to determine the overall number of surveys required to maximize the probability of detection in the study area using the equations 1 and 2 (Stauffer *et al.* 2002):

$$Power_{unit} = 1 - (1 - p)^n \quad \text{(Eq. 1)}$$

$$n_{power} = \log(1 - power_{unit}) / \log(1 - p) \quad \text{(Eq. 2)}$$

where, p is detection probability in a survey when species is present, n is the number of surveys, $Power_{unit}$ is the probability of detection of the species at least once and n_{power} is the number of surveys required. Therefore, 10 surveys were conducted monthly in the study area to cover the total length of 11685 m of transect lines per month.

Measurement of tracks

For each encountered track, seven linear measurements were taken and following points were noted: A: track length. B: track width, C: pad width, D: pad length, E: diameter distance between top of second (left) toe and right pad curve, F: diameter distance between top of third toe and left pad curve, G: Stride size (in mm) (Fig. 2). The aim of this study was not to find the effect of substrate and slopes on track width, track length and stride. Therefore, both the tracks and strides found on similar soil and slope conditions were included in sampling procedures. All linear measurements were taken from hard substrates covered by soil, loam, cinder or sand with slope less than 35° in the cell numbers 2, 5, 7, 11, 15, 17, 18, 19 and 23 (Fig. 1). This criterion in sampling process was performed for consistency in data collection and to obtain only comparable and reliable records (Grigione *et al.* 1999; Karanth *et al.* 2003; Riordan 1998). In addition, Spearman's correlation coefficient (ρ) did not show any significant correlation among linear measurements and substrate types (*P* value varying from 0.26 to 0.99; ρ value varying from -0.11 to 0.15). All the measurements were taken by the same person (AS) to exclude the observer bias. Linear factors of pugmarks were measured directly in the field using digital caliper to minimize associated errors with indirect methodologies such as photography of tracks and plaster casts (see also Lewison *et al.* 2001; Riordan 1998; Sharma *et al.* 2005). The slopes were recorded as a percentage of slope declivity using a clinometer. Owing to the ground conditions in the study site (tropical rain forest ground), only three out of seven measurements (Fig. 2; track length: A, track width: B and

Table 1. Statistical summary of four groups (=individuals) determined by hierarchical cluster analysis from track width, track length and stride in the leopard with their relative standard errors (mean ± SE).

Groups	Hind leg footprints		Front leg footprints		Stride size
	Width mean (cm)	Length mean (cm)	Width mean (cm)	Length mean (cm)	Mean (cm)
Group 1 (n = 31)	5.50 ± 0.00	6.56 ± 0.47	7.25 ± 0.98	6.17 ± 0.30	29.50 ± 1.16
Group 2 (n = 44)	7.20 ± 0.11	6.96 ± 0.12	8.80 ± 0.16	9.62 ± 0.16	46.26 ± 0.95
Group 3 (n = 31)	8.52 ± 0.11	8.52 ± 0.03	10.34 ± 0.15	9.72 ± 0.24	61.75 ± 1.25
Group 4 (n = 18)	9.56 ± 0.21	9.60 ± 0.20	12.02 ± 0.41	12.94 ± 0.35	70.62 ± 1.58
Misclassification rate	1.16 ± 0.15		1.27 ± 0.17		0.72 ± 0.18

Table 2. Statistical summary of four groups (=individuals) determined by discriminant analysis from track width, track length and stride in the leopard.

	Hind leg		Front hand		Stride
	Length (n=30)	Width (n=30)	Length (n=45)	Width (n=45)	(n=49)
Eigen value	16.52	0.41	5.36	0.33	7.37
% of variance	97.5	2.5	94.1	5.9	100
% Correctly classified cases	96.7		97.8		95.9

strides: G) were successfully taken in all tracks. In total, 30 sets of hind track measurements (i.e., 30 records of width and 30 records of length in the same 30 tracks), 45 sets of front track measurements (45 records of width and 45 records of length in 45 tracks) and 49 stride records were obtained and used in statistical analyses.

Analyses

Linear measurements were carried out to measure track width, length of front and hind legs and sizes of stride for grouping purposes using the hierarchical cluster and discriminant analyses (Grigione *et al.* 1999; Lewison *et al.* 2001; Rajapandian *et al.* 2005; Riordan 1998; Sharma *et al.* 2005). Hierarchical cluster analysis extracts inherent clusters within the data set without prior tagging the individual data into the known groups. However, this method would not provide any information on efficiency of classification processes. Therefore, discriminant analysis was used to describe the validity of obtained clusters from hierarchical cluster analysis. We combined the data from right leg tracks with the data from left leg tracks because previous studies demonstrated little variability among most linear measurements from left and right tracks (Riordan 1998). Analysis of variance and chi-square test were used to test the significance of differences between front and hind leg tracks in total samples and within an individual. Track length and track width were significantly different between hind and front tracks both in total (ANOVA, track width: $F_{1,\ 103}$ = 61.13, P = 0.000; track length: $F_{1,\ 103}$ = 56.88, P = 0.000) and within an individual (track width: χ^2 = 67.81, P = 0.000; track length: χ^2 =73.71, P= 0.000). Therefore, we have categorized the measurements into the "front tracks", "hind tracks" and "stride" *a priori* groups and subjected them to the mentioned analyses. Statistical significance of difference between *a posteriori* groups was measured at $P < 0.05$.

Results

A total of 168 tracks and strides of leopards (i.e. 54 hind leg tracks, 61 front leg tracks and 53 strides) were encountered in the field. However, only 124 tracks and strides were used for analysis because of pace pattern, unclear borders of the tracks or substrate and slope condition. Both hierarchical cluster analysis and discriminant analysis proved the existence of four significant groups of tracks ($P < 0.05$) (Tables 1 and 2, Figs. 3A and 3B). Each group is logically attributed to one individual leopard.

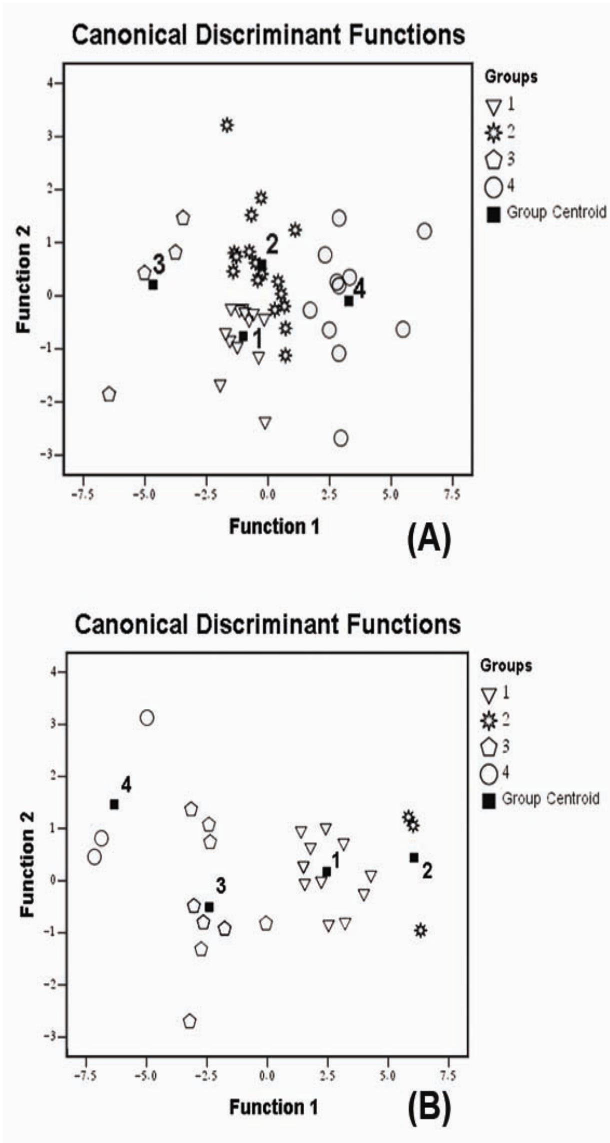

Fig. 3. Discrimination of four groups (=individuals) from front track measurements (A) and hind track measurements (B). Function 1= track length, function 2= track width.

Most variance between groups was caused by track length (front tracks: 94.1 % and hind tracks: 97.5 %), whereas the contribution of track width was negligible (front tracks: 5.9 % and hind tracks: 2.5) (Table 2). Figures 3A and 3B also prove that track length (Function 1) is the principal determinant of groupings, as the group centroids differ much more horizontally (across Function 1) than vertically (Function 2).

Discussion

Ayer Hitam Forest Reserve (AHFR) is located in a rapidly developing area. In the past 14 years (from 1983 to 1997), 68.49 % of the forest was converted to highways, housing areas and other land uses (Noor *et al.* 2007b). Therefore, leopards from the surrounding areas (3004 ha had been deforested since 1965) were pushed to this small remaining forest. The use of track measurements suggests that a minimum of four leopards survived in this 1411 ha protected area. Although, it is not possible to produce a definite density estimate of leopards due to small size of the study area, we suggest that leopard density at present site is unusually high due to rapid shrinking and fragmentation of the forested habitat where leopards have been trapped. Furthermore, our study on the leopard prey (*in prep.*) indicates that there is adequate prey base to support present leopard population.

Recommended methodologies in the previous studies were used to estimate small population size of leopard in a highly fragmented area. Data collection was conducted systematically and measurements were obtained from tracks on similar slope and substrate conditions to reduce the associated errors. Track measurements work well on estimating sizes of small populations of felids, so we believe that our leopard census in AHFR is accurate and reliable (Gusset & Burgener 2005; Lewison *et al.* 2001; Sharma *et al.* 2005). Track length has manifested itself as much more reliable character in individual recognition of leopards (Grigione *et al.* 1999; Riordan 1998; Sharma *et al.* 2005). From the measurement of the smallest tracks (group 1) it could be speculated that these tracks might be from a clouded leopard. However, due to small size of the forest it is unlikely that two top predators could use the study area. Moreover, clouded leopards are mostly found in dense primary evergreen tropical forests (Alderton 1993; Sunquist & Sunquist 2002), and never been reported in the study area. Sightings of leopards have frequently been reported by both local and indigenous settlers at the farm and the forest. In addition, two incidences of conflict between leopards and stray dogs were reported by the local communities in the study area.

AHFR represents the vestige of rich biodiversity within the densely populated metropolitan area which needs sustainable forest management practices using the leopard as a flagship species. Territorial nature of the species and requirement of relatively spacious habitat by each individual could be the main consideration of the species' viability at the study area. However, corridors can not be established as there is no other forest patch adjacent to the present study site. Therefore, we suggest that the boundary of the forest should be protected and any further destruction of the habitat should also be prevented. Moreover, the reproduction status of the leopards is uncertain and leopards may fail to raise their cubs because of the restricted size of the forest which serves as their habitat. Besides, genetic depression may happen in long term because of successive inbreeding. However, since sex composition of the population is unknown and population size is critically small, any fatality of leopard individuals may lead to population extinction in the study area. Therefore, more studies should be carried out to investigate the reproduction status and sex composition of the leopard population at AHFR. This would help the managers to control the population size over successive years using suitable management techniques.

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