

Part VI

Conservation Case Studies

Chapter

38

Landscape Genetics Applied to the Conservation of Primates in Flooded Forests

A Case Study of Orangutans in the Lower Kinabatangan Wildlife Sanctuary

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Introduction

Two major threats to biodiversity worldwide are habitat loss and fragmentation caused by human exploitation of environmental resources. One example is Singapore where over 70% of biodiversity has been lost because of large-scale deforestation and habitat modification (Brook *et al.* 2003; Brooks *et al.* 2002, 2006; Chapman & Peres 2001; Fahrig 2003; Fischer & Lindenmayer 2007; Myers *et al.* 2000; Pimm & Raven 2000; Segelbacher *et al.* 2010). In the matrix newly created by land-use changes and transformation, populations of wild animal species often become reduced and isolated. As a consequence, natural processes can be jeopardized and can induce a significant reduction in gene flow, increased genetic drift (and hence differentiation between populations that were once connected). Moreover, the fitness of the surviving and isolated populations can be reduced by inbreeding depression when closely related individuals mate. Combined, these processes reduce genetic diversity and can eventually lead to extinction (Amos & Balmford 2001; Caballero Rodríguez-Ramilo *et al.* 2009; Fernández *et al.* 2008; Frankham 2006; Hager 2003; Holderegger & Wagner 2008; O'Grady *et al.* 2006; Segelbacher *et al.* 2010; Toro & Caballero 2005). In addition to these genetic changes, demographic processes also play a major role in extinction rates. In particular, local extinction of small fragmented populations is increasing and becoming relatively common (Fahrig 2002; Kattan *et al.* 1994; Matthies *et al.* 2004; Michalski & Peres 2005; Tscharrntke 1992). Thus, species survival will increasingly rely on the ability of individuals to disperse and move across heterogeneous landscapes between surviving populations or re-colonize empty fragments, and on our ability to maintain connectivity between remaining populations (Fahrig & Merriam 1994). Therefore, habitat connectivity (Lindenmayer & Fischer 2006) is a key issue for the management of endangered species located in multiple-use landscapes (Lindenmayer *et al.* 2008; Taylor *et al.* 2006).

Primates have been greatly threatened by habitat conversion and fragmentation (Arroyo-Rodríguez & Mandujano 2009; Marsh & Chapman 2013). According to Chapman

and Peres (2001), primate habitat countries are losing annually *c.* 125 000 km² of forest resulting in remnant primate populations being increasingly isolated in highly fragmented and low-quality habitats; this has resulted in the extinction of several populations and species and this extinction process is anticipated to worsen within this decade and the next (Cowlshaw 1999; Cowlshaw & Dunbar 2000). Extensive literature reviews fail to detect clear patterns of the effects of habitat fragmentation and disturbance on primates, probably because the responses to habitat modification depend, among other factors, on the biological characteristics of each taxon and also on the highly variable ways of conceptualizing and measuring fragmentation effects (Arroyo-Rodríguez & Dias 2010; Arroyo-Rodríguez *et al.* 2013). Primates live in fragments throughout the globe (Marsh & Chapman 2013), including gibbons in Java (Nijman 2013), red howlers in Brazil (Boyle *et al.* 2013), howler and spider monkeys in Mexico (Cristóbal-Azkarate & Dunn 2013), red and black-and-white colobus in Uganda (Chapman *et al.* 2013), macaques in Thailand (Aggimarangsee 2013), capuchins in Venezuela (Ceballos-Mago & Chivers 2013) and orangutans throughout their range (Wich *et al.* 2009), to name just a few.

The Lower Kinabatangan Wildlife Sanctuary (LKWS) in Sabah, Malaysia, is an important site for primatology and primate conservation as ten sympatric non-human primate species (including the proboscis monkey (*Nasalis larvatus*), the Bornean gibbon (*Hylobates muelleri*) and the Bornean orangutan (*Pongo pygmaeus*) all endemic to Borneo) can be found at relatively high densities (Ancrenaz 2007). This protected area is located within the Kinabatangan River catchment, an important wetland in Malaysia, consisting of a variety of habitats including seasonally flooded, riverine and swamp forests, dry dipterocarp and mangrove (including nipah palm) forests (Azmi 1998). Most of the area has been extensively logged in the past (McMorrow & Talip 2001) and today only about 65 000 ha of highly degraded forests remain along the Kinabatangan River. The remaining matrix is primarily industrial oil palm monoculture surrounding different sized patches of forest of different quality which are poorly connected or

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Figure 38.1 The Kinabatangan floodplain. Forest fragments interspersed with oil palm plantations, human settlements and local agriculture. Photo: HUTAN. (A black and white version of this figure appears in some formats. For the colour version, please refer to the plate section.)

completely isolated (Figure 38.1). In 2005, the LKWS was officially gazetted by the Sabah's State Government comprising ten forest lots (about 26 000 ha) attempting to create a riparian corridor to link seven Virgin Jungle Forest Reserves (about 15 000 ha) and 10 000 ha of forested state and private lands (Ancrenaz *et al.* 2004; Figure 38.2). Due to its characteristics, the LKWS provides an ideal study site in which to assess the impact of forest fragmentation on primate population structure. Additionally, the Kinabatangan River bisects the sanctuary lengthwise (approximately 200 m in width), potentially acting as a natural barrier to primate dispersal.

Orangutans (*P. pygmaeus*) are one of ten primate species that can be found in the LKWS. They present extreme sexual dimorphism in body size and appearance, and also a pronounced bimaturism among sexually mature males (Delgado & Van Schaik 2000; Utami *et al.* 2002). The lifespan of this species in the wild is estimated to be at least 50 years for both sexes (Wich *et al.* 2004) with slow growth and development rates contributing to this trait. This ape is considered to have a 'semi-solitary' social system but van Schaik (1999) described an individual fission–fusion system for the Sumatran orangutan (*P. abelii*) in the swamp forest of Suaq Balimbing, Indonesia. In studies elsewhere, consortships, travel bands and temporary foraging parties at fruiting trees have also been described (Utami Atmoko *et al.*

2009). The mating system seems to be a combination of female choice and male harassment and coercion, with both morphs (flanged and unflanged males) being reproductively successful in the populations (Delgado & van Schaik 2000; Goossens *et al.* 2006a; Utami Atmoko *et al.* 2009; Utami *et al.* 2002). Maturing females tend to remain near the natal area (philopatry), while males disperse (Galdikas 1995; Singleton *et al.* 2009; Singleton & van Schaik 2001; van Noordwijk *et al.* 2009). However, there is evidence from one of the forest fragments in the LKWS's orangutan population for male and female philopatry (or dispersal) (Goossens *et al.* 2006, see section below on population structure, gene flow and dispersal). Additionally, orangutans occur at low densities with natural fluctuations in population parameters depending on forest type (Husson *et al.* 2009; Marshall *et al.* 2009), however, historically they might have occurred at higher densities (Meijaard *et al.* 2010). As the world's largest arboreal mammal, it has been suggested that terrestrial locomotion is part of the Bornean orangutan's natural behavioural repertoire to a much greater extent than previously thought, and is only modified by habitat disturbance (Ancrenaz *et al.* 2014a, b). Currently, most orangutans are increasingly restricted to small forest fragments (Wich *et al.* 2008, 2009).

The orangutan population of the LKWS is one of the best documented in the world, mainly due to efforts of the

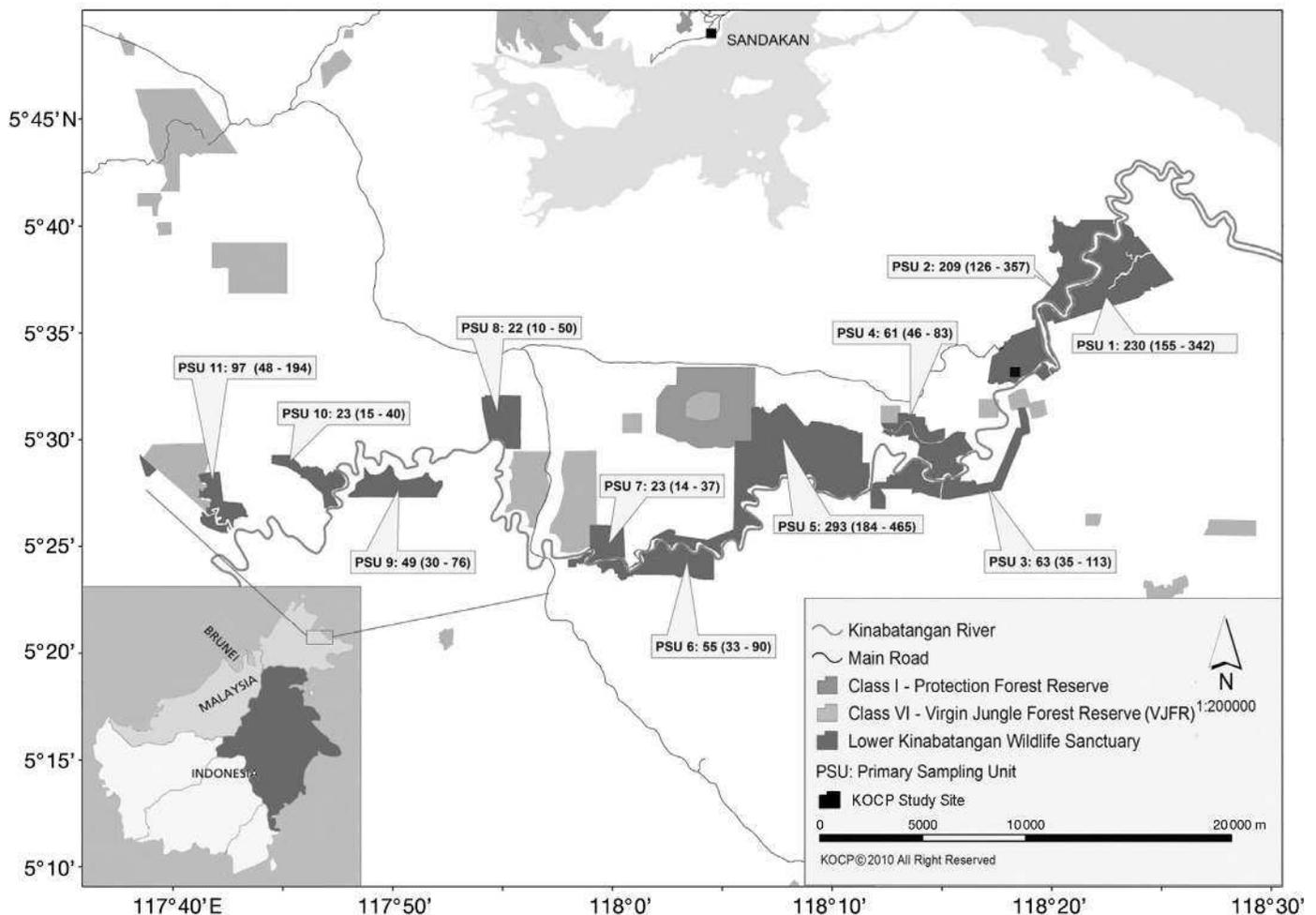


Figure 38.2 Kinabatangan region and the 11 PSUs. Values indicate mean number of orangutans estimated to occur in the respective PSUs in 2004 (range in parentheses). Modified from Ancrenaz *et al.* (2004), with permission from the author.

Kinabatangan Orangutan Conservation Programme (KOCP) by the French NGO, HUTAN. Among other activities, KOCP studies the impact of habitat alteration on orangutan socioecology, and aims to find ways to achieve long-term survival of orangutan populations in exploited areas, especially within and around the Kinabatangan Wildlife Sanctuary. Thus, studies on this population range from systematic estimates of population size and density (Ancrenaz 2007, 2008; Ancrenaz *et al.* 2004, 2005) to endocrine stress responses to habituation and tourism (Muehlenbein *et al.* 2012) and adaptation to newly built environments (Ancrenaz *et al.* 2014a, b). This chapter reviews several genetic studies that were conducted in the area on this population and how they have contributed to the local conservation of the orangutan.

Orangutan Population Structure, Genetic Diversity and Dispersal

Genetic studies on wild orangutans have bloomed in this and the previous decade (Arora *et al.* 2010; Bruford *et al.* 2010; Goossens *et al.* 2004, 2006; Greminger *et al.* 2014; Jalil *et al.* 2008; Morrogh-Bernard *et al.* 2010; Nater *et al.* 2011, 2012; Nietlisbach *et al.* 2012; Sharma *et al.* 2012; Utami *et al.* 2002). This chapter describes, to our knowledge, the

first comprehensive population genetic study using samples collected in the wild at a small spatial scale.

In 2004, Ancrenaz *et al.* (2004) divided the Lower Kinabatangan area into 11 'primary sampling units' (PSUs, equivalent to each lot of the LKWS, with lot 10 divided into two distinct parts; Figure 38.2) to estimate orangutan distribution, density and population size. These surveys were conducted by counting orangutan nests along the ground and via aerial transects (Ancrenaz *et al.* 2004). The census estimated a total of 1125 (95% CI 691–1807) individuals, across the 11 PSUs with variable figures ranging from as few as 22 individuals (PSU 8) to as many as 293 (PSU 5) (Figure 38.2). Goossens *et al.* (2005) grouped the 11 PSU into nine sampling units (S1–S9, PSUs 5 and 7 were fused, as well as PSUs 10 and 11) from where faecal samples of 200 wild individuals were collected, representing the largest ever genetic sample from a wild orangutan population. The patterns of genetic diversity and structure within the Lower Kinabatangan orangutan population were then investigated by analysing the genotypes of 14 microsatellite loci. Despite the fragmentation of their habitat, the orangutan population exhibited a high level of genetic variability (e.g. an expected heterozygosity – H_E of 0.74 (Goossens *et al.* 2005). However, this genetic diversity seemed to be the remnant of

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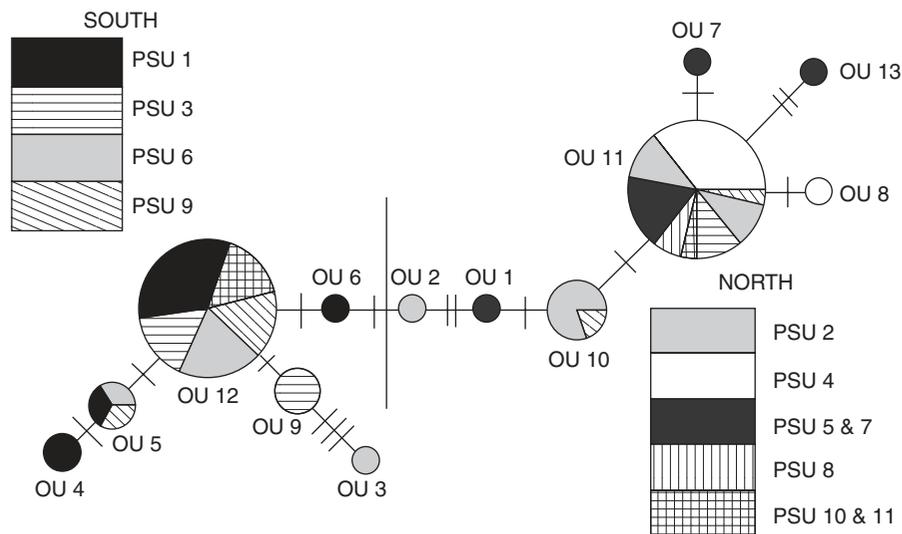


Figure 38.3 Minimum spanning network (MSN) of the Kinabatangan orangutan sequences. Each circle represents a haplotype and the diameter scales to haplotype frequency. The smallest circles represent singletons. Mutational steps are represented by solid black bars on lines connecting haplotypes. Dotted line represents the apparent partition between the populations on both banks of the Kinabatangan River.

an ancient significantly larger population that inhabited the whole region (Goossens *et al.* 2006). Significant genetic differentiation was found between most sampling units but the absolute level of genetic differentiation was limited (average $F_{ST} = 0.04$, $p < 0.001$). This difference was higher between samples separated by the Kinabatangan River than between samples from the same river side ($F_{ST} = 0.06$ versus $F_{ST} = 0.02$, $p < 0.01$). These results indicated the role played by the Kinabatangan River as a natural barrier for orangutan dispersal. To explore the effect of the river on gene flow, Bayesian migration estimation and assignment tests were performed by Goossens *et al.* (2005). The authors found that there was a high frequency of individuals moving between PSUs on the same side of the river. They also found that migration across the river was close to zero. Due to the necessarily limited number of genetic markers and the low level of genetic differentiation, it could not be completely ruled out. Thus, the results of Goossens *et al.* (2005) indicated that orangutans used to move relatively freely between neighbouring PSUs of the LKWS until recently, and that there was a need to maintain migration between isolated forest fragments on the same side of the Kinabatangan River in order to facilitate gene flow (Goossens *et al.* 2005).

The influence of the Kinabatangan River on the population genetic structure of the LKWS orangutans was further confirmed in a study by Jalil *et al.* (2008). In that study, sequences of the mitochondrial DNA (mtDNA) control region were examined for genetic variability and structuring. About 7% (73 individuals from Goossens *et al.* 2005) of the total Kinabatangan population was analysed and 13 haplotypes were identified. A population bottleneck followed by rapid growth and accumulation of mutations was suggested based on the overall high haplotype (0.734 ± 0.035) and low nucleotide diversity (0.008 ± 0.005) found for the whole sanctuary. In addition, the samples on either side of the river were strongly differentiated ($\Phi_{ST} = 0.404$, $p < 0.001$), and a minimum spanning tree analysis on gene genealogies indicated a separation of the haplotypes into two groups, one on the north and one on the south riverbank (Minimum Spanning Network, Figure 38.3), reinforcing the previous inference that the Kinabatangan is

(and has been for long periods) a major barrier to gene flow, and this is congruent with evidence elsewhere in Borneo on rivers acting as barriers to the dispersal of this species (Arora *et al.* 2010; Jalil *et al.* 2008).

In addition to the genetic diversity analyses, population structure was further investigated by examining patterns of relatedness and parentage (Goossens *et al.* 2006). Thirteen microsatellite loci (from the 14 used by Goossens *et al.* 2005) were used to genotype 32 identified individuals residing in the KOCP intensive study area located in PSU 2 (north riverbank) of the LKWS (Ancrenaz *et al.* 2004; Goossens *et al.* 2006). The genotypes of 95 individuals from the 200 identified in Goossens *et al.* (2005), but which resided elsewhere in the north bank of the Kinabatangan River, were added to guarantee an unbiased relatedness analysis. The results from the study indicate philopatric behaviour of both male and female orangutans, contrasting with the often reported male-biased dispersal behaviour of this primate (Galdikas 1985, 2008; Houston 2000; MacKinnon 1974; Mitani 1989; Nater *et al.* 2011; Nietlisbach *et al.* 2012; Rijksen 1978; Rodman 1973; Singleton & van Schaik 2002; van Schaik & van Hooff 1996). To be precise, all individuals resident in the KOCP site were on average more related to one another than individuals outside the core area but still within PSU 2 (Goossens *et al.* 2006). This unexpected dispersal pattern, and the high density of males observed in the area, suggest that more work should be carried out to determine whether this is due to the fragmented state of the Kinabatangan forest and its recent reduction in size (Goossens *et al.* 2006b).

Population Decline and Viability

The results presented in the studies by Goossens *et al.* (2005, 2006) and Jalil *et al.* (2008) demonstrated the role played by the Kinabatangan River as a barrier to orangutan movement and gene flow. Additional results of these same studies also suggested that fragments had not yet drifted significantly from each other and were still little differentiated. At the same time, the high densities and the lack of clear differences between males and

females in terms of philopatry suggested that some effects on the genetic structure and gene flow were potentially starting to appear. In fact, given the long generation time of orangutans, it is not necessarily surprising that there is a lag in genetic effects brought on by fragmentation. To investigate whether the genetic patterns were showing long-term past or recent events, three different but complementary approaches were used to detect, quantify, and date a putative decline in orangutan populations (Goossens *et al.* 2006b). These methods are described in detail in this study and were named the EWCL (for Ewens-Watterson-Cornuet-Luikart), the Beaumont (Beaumont 1999), and the Storz and Beaumont (Storz & Beaumont 2002) methods.

Regardless of the mutation and demographic models used, the molecular analysis of the microsatellite genotypes from the 200 individuals sampled by Goossens *et al.* (2005) showed strong evidence for a recent and dramatic population decline. Precisely, a particular signature of a population collapse of more than 95% was detected and dated to recent times, and excluding times older than a couple millennia. Thus, the dating strongly suggested that the cause of that decline was unlikely explained by prehistoric hunting and Pleistocene climatic events, nor could it be explained by the arrival of the first farmers in the area. The recent anthropogenic fragmentation of the habitat, namely the exploitation of Sabah's forests which started in 1890, was found to be the only major event that might have significantly influenced orangutan populations in the last decades or centuries. Moreover, the migration patterns reported by Goossens *et al.* (2005) were consistent, as we noted above, with the recent history of logging (1950s) and subsequent oil palm agriculture since the 1970s and 1980s. However, the role of recent forest exploitation in generating bottlenecks in orangutan populations should not be interpolated to every region in Borneo and more theoretical work to understand the multiple demographic events impacting the genome of this species must be encouraged (Arora *et al.* 2010; Meijaard *et al.* 2010; Sharma *et al.* 2012). In addition to the evidence of recent population collapse, Goossens *et al.* (2006b) found extremely low current population size estimates, which were in close agreement with the census estimates of Ancrenaz *et al.* (2004). These two lines of evidence implied the need for immediate conservation efforts to halt genetic drift from quickly eliminating the remaining genetic diversity in the fragmented forests of the Kinabatangan floodplain.

To further assess orangutan population viability in the LKWS, genetic data were incorporated into a stochastic population modelling program under different management strategies to predict the evolution of genetic diversity and demography at different times in the future (Bruford *et al.* 2010). The parameters of the model were based on previous PHVA (Singleton *et al.* 2004), and research and observation in the LKWS (Ancrenaz *et al.* 2004). Different models were designed to test the genetic and demographic consequences of: (1) no intervention, (2) translocations, (3) establishment of forest corridors and (4) a mixed approach combining translocations and corridors. The possible outcomes for the LKWS population are not optimistic under a non-intervention policy where high extinction probabilities ($\geq 5\%$) are expected for six of

the PSUs either including (PSU 4, 6, 7, 8, 9, 10) or excluding (PSU 7, 8, 9, 10) inbreeding depression in the model. These extinctions are predicted to occur within the next 250 years, and even within the next 100 years in the case of PSU 10 (< 5 generations). Furthermore, mean final population size was predicted to decrease in seven of the 11 PSUs (i.e. 64%) when inbreeding depression was not included and in nine of 11 PSUs (i.e. 82%) when inbreeding was included (see Table 1 in Bruford *et al.* 2010). When the model incorporated the translocation of a single adult female every 50 years from PSU 2 to both PSUs 4 and 7 and from PSU 5 to PSUs 8, 10 and 11 (north of the river) and from the south side of the river, from PSU 1 to PSUs 3, 6 and 9, the accumulation of significant amounts of inbreeding within the PSUs having the smallest carrying capacity was not prevented. However, scenarios with more frequent translocations (10 and 20 years) were more successful at controlling inbreeding coefficients in these populations.

As there exist only one or two large source populations donating to several or many small sink populations, the donor populations could become demographically unstable, as was found for PSU 1 if the translocation was conducted every 10 years. In a conservative approach, the third model simulated the establishment of corridors over 100 years (PSUs 4 to 5 and 5 to 7 and PSUs 1 to 3) or 250 years (the remainder). Under this model, corridor reconnection seemed unlikely to occur rapidly enough for the most isolated PSUs, therefore this measure alone might not be able to prevent large-scale genetic and demographic losses nor to prevent extinction in these areas. In contrast to the sole use of translocations or corridors, demographic stability and an inbreeding threshold below 10% were achieved by the mixed approach model where the translocation of one adult female every 20 years was simulated along with the corridor establishment (Figure 38.4). This mixed management approach seemed to be a pragmatic and realistic solution to the current orangutan demographic problem.

Conservation Measures

The Orangutan Action Plan (Sabah Wildlife Department 2011) for the state of Sabah, Malaysia, was the outcome of an extensive consultation process and embodies a consensus of recommendations from relevant stakeholders involved in the management of orangutan populations in Sabah. The conclusions from the studies reviewed in this chapter were incorporated into the actions for the forests of the Lower Kinabatangan floodplain, hence emphasizing the urgent need for habitat restoration and connectivity. However, the creation of habitat corridors for sustainable conservation management can be extremely difficult to achieve due to many factors that can influence the timescale and demographic gains associated with forest corridor establishment, such as financial constraints (land purchase), forest reestablishment rates (which are slow for high canopy dipterocarp forest but which can be much quicker for riparian and seasonally inundated forest) and habitat occupancy and corridor usage dynamics by the faunal community, which could in principle be almost instantaneous or could be a protracted process (Bruford *et al.* 2010). Therefore, as a

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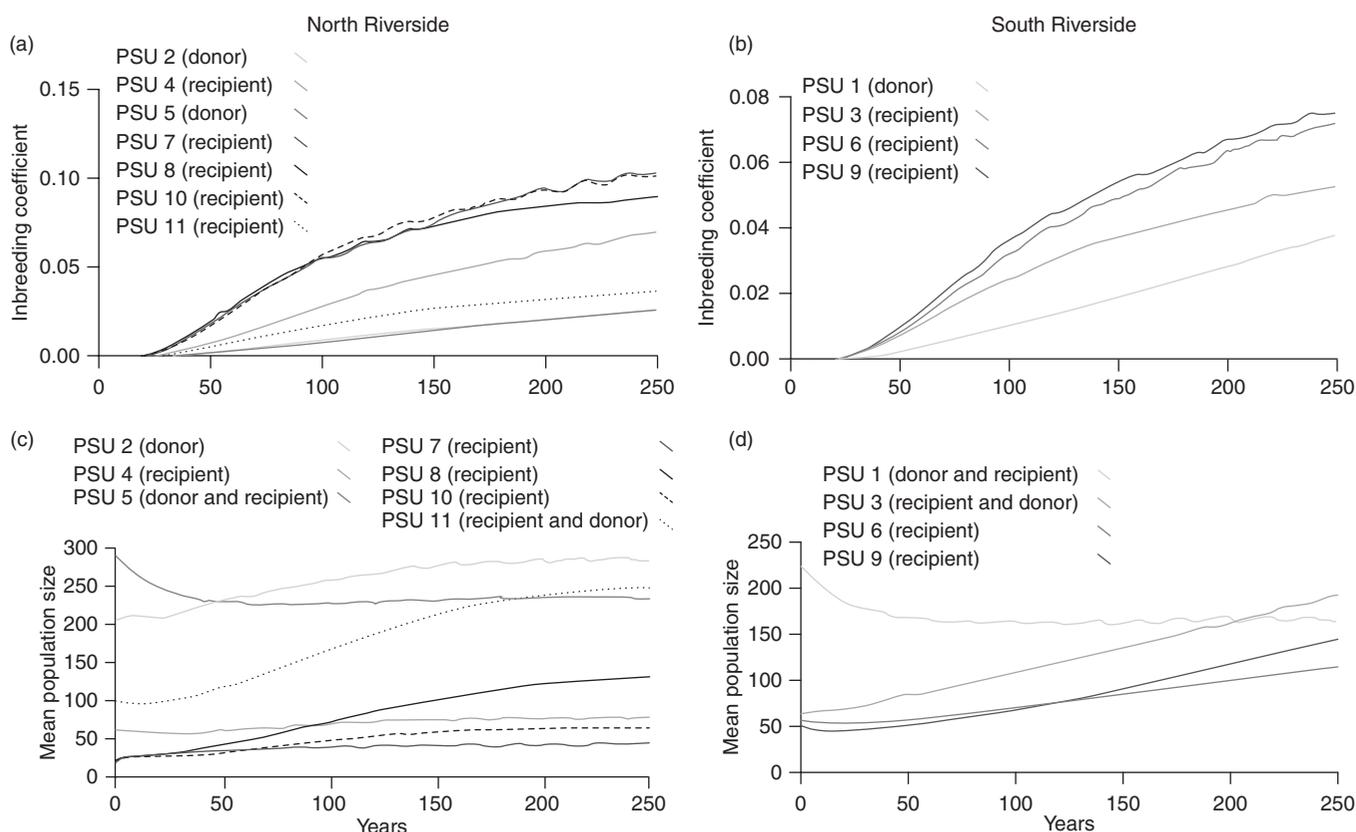


Figure 38.4 Effect of corridor connection and translocations on within-PSU inbreeding coefficients and mean population sizes. (a) Effect of the mixed approach on the inbreeding coefficients of the PSUs on the north riverbank; (b) effect of the mixed approach on the inbreeding coefficients of the PSUs on the south riverbank; (c) effect of the mixed approach on the population sizes of the PSUs on the north riverbank; (d) effect of the mixed approach on the population sizes of the PSUs on the south riverbank. (A black and white version of this figure appears in some formats. For the colour version, please refer to the plate section.)

potentially quicker alternative, and in addition to the restoration and size augmentation of the LKWS, the establishment of orangutan bridges wherever necessary was also recommended (Sabah Wildlife Department 2011). These bridges would temporarily substitute the natural canopy coverage, which is currently lacking, for the orangutans to move freely above the small tributaries of the Kinabatangan River and the drains created by the oil palm plantations. To date, eight orangutan bridges have been set up over four small tributaries of the Kinabatangan River (Figure 38.5; Ancrenaz 2010; DGFC and Sabah Wildlife Department 2010; Lackman & Ancrenaz 2009), with documented success in all of them (Ancrenaz 2010).

The management of small populations of endangered species is complex by nature: in addition to the scientific aspects, we need to incorporate a welfare dimension that we cannot underestimate for iconic species such as the orangutan. As an example, a rescue operation was approved by the Sabah Wildlife Department on August 2012. A small forest on private land nearby PSU 2 (north riverbank) was cleared for oil palm plantation and a male and a female orangutan were urgently transferred to PSU 6 (south riverbank).

A conservative conservation management plan would normally seek to maintain the natural demographic isolation between populations to the north and south of the Kinabatangan River (Goossens *et al.* 2005; Jalil *et al.* 2008). Indeed, a recommendation by Bruford *et al.* (2010) was to maintain these

populations as separate Management Units. However, under time and logistical constraints, measures were taken for the sake of the individuals' welfare as well as for the species conservation. It was thus decided to translocate the animals to a PSU under high extinction risk and where continuous monitoring activities are undertaken. After that incident, effort was made to follow the recommendations from Bruford *et al.* (2010) and the subsequent case (September 2013) was that of a flanged male which was rescued in the area of Gomantong (on the north riverbank) and was then translocated to PSU5 (same riverbank); this individual has been observed frequently since, on camera traps (DGFC, camera traps database).

The importance of genetic diversity in population viability and management has been debated in the past (Asquith 2001) and it is not always clear whether demographic, environmental or genetic factors will be the first to impact threatened species. However, it is increasingly recognized that genetic data provide unique information. While the application of genetics (and genomics; Sharma *et al.* 2012) in the management of threatened species is increasing (DeSalle & Amato 2004), there seems to be a general failure to incorporate these types of data into concrete conservation actions. This failure may be due to two main factors: first, the difficulty of interpreting the results of genetic data for non-geneticists; and, second, the difficulty of becoming involved in policy and practical conservation decisions for conservation geneticists. There is also a need



Figure 38.5 Orangutan using the 'orangutan bridge' over the Rasang River, a tributary of the Kinabatangan River. Photo: Ajirun Osman/HUTAN-KOCP.

to develop tools that will help conservation biologists to use and master population genetics concepts (i.e. Conservation Genetic Resources for Effective Species Survival (CONGRESS), <http://www.congressgenetics.eu>). The incorporation of genetic data into species action plans has recently been advocated, but will require the above-mentioned difficulties to be overcome (Frankham 2009; Laikre 2010). Genetic data must be integrated with an understanding of landscape dynamics and area-based conservation actions to achieve successful decisions concerning areas, landscapes and species.

Conclusions

Nearly 15 years have passed since genetic samples were collected by Goossens *et al.* (2005) in 2001, and it has been more than half a decade since the publication of the Orangutan Action Plan (Sabah Wildlife Department 2011). During this time, much was achieved to advance the knowledge of the LKWS orangutan's population genetics. It certainly stands out as a major achievement to have the data incorporated into an official management plan. Nonetheless, the orangutan is a slow breeder and it will take time before deleterious genetic effects are detected in this population. Time is pressing, land conversion in the area is still ongoing and the population size is still declining (Alfred *et al.* 2010; Ancrenaz 2008). For some species on the brink of extinction (i.e. the Sumatran rhinoceros, Goossens *et al.* 2013) a 10-year wait might be an unaffordable luxury. Therefore, faster mechanisms for the incorporation of genetic data into management plans should be devised and scientists and policy makers should also make a compromise regarding the extent of genetic information really

needed before expediting the successful and urgent protection of a species like the orangutan through restoration of habitat connectivity and other means already known to us.

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