Evaluating the short-term success of a reintroduction of the Critically Endangered Ploughshare tortoise, *Astrochelys yniphora*.

David Wallis

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“Cupped in our hands, these funny little pie-crust babies represented the future of their race. Guarded from harm, we knew that, ultimately, they would grow into those ponderous adults that, heavy and clumsy as knights in armour, would joust each season for their ladies, so that these extraordinary antediluvian creatures could breed and go lumbering on into new centuries to remind us how the world began and to delight us with their unique shape and habits.”

Gerald Durrell

Abstract

The translocation and reintroduction of animals in the wild is an important tool in modern conservation management. It may, however, take many years before projects of this sort can be determined a success, especially when they involve species with long generation times. In such cases, it is essential that regular assessments of short-term success are included as part of any long-term monitoring strategy.

The ploughshare tortoise (*Astrochelys yniphora*) of Madagascar is critically endangered with extinction. The Durrell Wildlife Conservation Trust (DWCT) oversee the in-situ captive breeding programme for this species and assessing the suitability of reintroduction from captivity as a management strategy is of great importance. Following a trial release of 5 individuals, 20 captive bred sub-adult juvenile tortoises were released into the Beaboaly region of Baly Bay National Park, Madagascar in January 2006. All animals were radio-tagged and monitored from 2006 – 2008. Short-term success was assessed by investigating survival and post-release response in body mass, movement and habitat use. Movement was analyzed using GIS and habitat use was analyzed using compositional analysis. All 20 released tortoises survived the duration of this study, remained in the area of release and used habitat and resources in a similar way to wild tortoises. The findings of this study confirm that reintroduction is a promising tool for ploughshare tortoise conservation.

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List of abbreviations

BM – Body mass

CA – Compositional analysis

DWCT – Durrell Wildlife Conservation Trust

GIS – Geographic Information System

GPS – Global Positioning System

LME – Linear mixed-effects model

LSCV – Least squares cross-validation

MCP – Minimum Convex Polygon

PVA – Population Viability Analysis

RRT – Reintroduction, reinforcement and translocation

Word count: 12,630

1. Introduction

1.1. Preface

The translocation and reintroduction of animals is an increasingly used tool in modern conservation management (Griffith et al, 1989). Such methods may be particularly useful in conserving tortoises due to certain aspects of their biology, such as innate behavior, lack of parental care and sedentary nature (Germano & Bishop, 2008). In the past few decades, numerous conservation interventions have been carried out involving the translocation of wild populations of tortoises although comparatively few have been attempted with animals raised in captivity. A notable example of a successful reintroduction from captivity is the long running release of captive-bred Galapagos tortoises, *Geochelone hoodensis,* onto their native Island of Espanola. Although there have been other successes, there are numerous projects that have been less successful and only a small proportion of tortoise reintroductions include any in-depth post-release evaluation (Dodd & Seigel 1991, Germano & Bishop 2008)

A translocation or reintroduction can only claim success if it leads to the establishment of a self-sustaining population (Griffith et al, 1989). As tortoises are slow-growing and have long generation times, it may take years or even decades before a project reaches this stage (Condon et al, 1993). An efficient long-term post-release monitoring strategy is therefore fundamental to any evaluation of a tortoise translocation or reintroduction. However, in reality, the success of such interventions is mainly only ever evaluated in the short-term. Post-release survival, movement, fidelity to release site, resource use, fitness, reproduction, behaviour and social interactions are all variables that might be investigated in an assessment of short-term success (Griffith et al 1989, Kahn 2006, Germano & Bishop 2008). It is, however, a common failing of conservation interventions involving the movement and release of animals that post-release monitoring is often neglected or the results are never made available. As a result, progress in this field of conservation management is hindered and avoidable mistakes are often repeated.

The Critically Endangered (IUCN 2009) ploughshare tortoise (*Astrocheylis yniphora*) is the rarest chelonian species in the world and, with as few as 600 remaining in the wild (O'Brian et al 2005) the conservation of this species is of great global importance. Since the mid 1980s, the Durrell Wildlife Conservation (DWCT) Trust have run a captive breeding programme for the Ploughshare tortoise in its native Madagascar. Following an encouraging trial release of 5 tortoises in 1998, a further 20 sub-adult juveniles were released into Baly Bay National Park in January 2006. Determining if reintroduction is an effective conservation tool for this species is critical in designing future conservation efforts and, ultimately, to its long-term survival.

1.2. Study objectives

The aim of this study is to investigate how 20 captive-bred ploughshare tortoises, released in 2006, have responded to life in the wild, and to provide an evaluation of the short-term success of this reintroduction project. The findings of this study will be used to make recommendations for future post-release monitoring strategies. Short-term success will be evaluated through the investigation of the following post-release responses of the released tortoises:

- Do the released tortoises survive release in the wild?
- Do released tortoises gain or lose condition following release into the wild?
- Do released tortoises establish home ranges and remain in the area into which they have been released or do they disperse out of range of effective management?
- Do released tortoises exploit resources in a similar way to wild tortoises?

1.3. Overview of thesis structure

Chapter 2 explores the field of animal reintroduction and translocation and discusses how success in projects of this sort might be assessed and the issues that arise in the process. The application of reintroduction and translocation as a tool for tortoise conservation is also explored and a summary of previous case studies is provided. Finally, this chapter includes a brief introduction to the species investigated in this study, the critically endangered ploughshare tortoise, *A. yniphora*, and gives details of its biology and conservation history.

Chapter 3 describes the methods used in this study and details the post-release monitoring protocol employed in the field in Madagascar and the analytical methods used.

Chapter 4 is the results section and data analysis findings, together with supporting figures, are collected here.

Chapter 5 consists of a discussion of the findings listed in Chapter 4 and places them in the context of previous work and wider conservation biology theory. The short-term success of the reintroduction is appraised and recommendations for future post-release monitoring strategies are made.

2. Background

2.1. Reintroduction and translocation as a tool for tortoise conservation

The movement and release of animals, both from the wild and captivity, is becoming a more and more common technique in conservation programmes around the world and across a wide range of taxa (Griffith et al 1989, Fischer & Lindenmayer 2000). Exactly how this movement and release is defined is context dependent. Reintroduction is defined by the IUCN (Soorae & Seddon 1998) as “an attempt to establish a species in an area which was once part of its historical range, but from which it has been extirpated or become extinct”; whilst translocation is defined as a “deliberate and mediated movement of wild individuals or populations from one part of their range to another”. Finally, re-inforcement/supplementation describes “addition of individuals to an existing population of conspecifics”. The difficulties, complexities and expense associated with the movement and release of animals are well known (Griffith 1989, Dodd & Seigel 1991, Germano & Bishop 2008) but, as exploitation and loss of habitat continue on a large scale, such methods are likely to play an increasingly important role in conservation management (Griffith 1989, Tuberville et al 2008).

In terms of tortoise conservation, the Reintroduction, reinforcement and translocation (RRT) of animals might be carried out for a number of reasons, such as mitigating habitat loss, increasing population size, increasing genetic diversity and establishing new populations (Rittenhouse et al 2007). To date, efforts to evaluate the efficacy of these types of projects for tortoises and other reptiles have been few and somewhat inadequate, especially when compared to similar studies of birds and mammals (Tuberville et al 2005). Indeed, Millspaugh and Marzluff (2001) reported that only 0.5% of the published studies in the Journal of Wildlife Management over a 25 year period investigated reptiles or amphibians. A review of reptile and amphibian translocations by Dodd and Seigel (1991) found that only 19% of projects were successful and suggested that such methods should not be advocated as acceptable management and mitigation practices. A more recent review by Germano and Bishop (2008), however, reported an increased success rate of 41%, but this is still a suitably low figure to suggest these strategies are far from ideal. Despite this uncertainty, RRT projects may often be the only option for re-establishing extirpated populations and reconnecting fragmented ones (Germano & Bishop 2008). One of the major problems with determining the success of RRTs, especially when involving long-lived species such as tortoises, is that many years of monitoring are required before a clear picture can be established (Griffith et al 1989, Dodd & Seigel 1991, Germano & Bishop 2008).

2.2. Evaluating tortoise reintroduction and translocation projects

Ultimately, a successful project is one that results in the establishment of a viable self-sustaining population in the wild (Griffith et al 1989, Dodd & Seigel 1991, Soorae & Seddon (1998), Germano & Bishop 2008). However, with long-lived species such as tortoises, it may take many years, if not decades, before such an outcome is apparent (Condon et al 1993). Such a potentially long time-scale highlights the necessity for detailed, long-term post-release monitoring. Indeed, the success of a project is directly related to the quality of its monitoring strategy (Durrell & Mallinson 1987), which should be a complex processes involving short and long-term measures of mortality, fidelity to release site, disease, fitness, reproduction and social interactions (Griffith et al 1989, Kahn 2006, Germano & Bishop 2008). A common failing of many past projects has been that long-term monitoring strategies have been inadequate or virtually non-existent. This may be because the primary goal of many projects has been the welfare of individual animals, as opposed to entire populations and, as a result, subsequent monitoring does not include an evaluation of overall project success (Tuberville et al 2007). Projects that actually set out to explicitly evaluate success are rare. A large proportion of previous RRTs involving tortoises have only consisted of a post-release monitoring period of three years or less and have not included any in-depth evaluation of subsequent reproduction and recruitment (Germano & Bishop 2008). As stated by Dodd and Seigel (1991), monitoring a population for only 10% of the time it takes to reach maturity is inadequate in terms of measuring success. Although a project may be reported as successful, often what is reported is, in fact, only short-term success (Germano & Bishop 2008). However, short-term evaluation is a vital part of any ongoing assessment of the effectiveness of this kind of management strategy, and must be incorporated as part of a committed wider long-term monitoring strategy.

Short-term success of an RRT may be evaluated in a number of ways, with assessment of post-release survival being primary (Cook et al 2004, Bertolero et al 2007, Tuberville et al 2008). Studies may also include evaluation of post-release changes in size and body mass as a reflection of condition and fitness (Pedrono & Sarovy 2000, Field et al 2007). The majority of studies involving the release of animals also incorporate an investigation of post-release movement and home range establishment and, in recent years, the most widely used method in this regard has been the use of radio-tagging, GPS (Global Positioning System) and GIS (Geographic Information System). The fidelity of released individuals to the release site is another key concern in this type of project (Tuberville et al 2005), and knowledge of exactly where and how far released animals range should be a central concern of any ongoing monitoring strategy. This sort of information can be especially useful when comparing different groups of released animals (Tuberville et al 2005, Field et al 2007) or when comparing a released population with a wild or already-established one (See Pedrono & Sarovy 2000, Rittenhouse et al 2007). If released animals fail to remain in the release are and move away over large distances, they may be at greater risk to mortality or poaching and may no longer be within range of effective management (Griffith et al 1989). Indeed, large movements and migration away from the release site are the most common cause of failure in the majority of RRTs (Griffith 1989, Germano & Bishop 2008).

2.3. Translocation of wild tortoises

To date, the greatest number of tortoise RRT projects have been carried out in the United States with *Gopherus* species (Berry 1986, Burke 1989, Eubanks et al 2003, Field et al 2007, Ashton & Burke 2007, Riedle et al 2008, Tuberville et al 2005, 2008). Few of these studies have clearly demonstrated success in terms of the establishment of stable or self-sustaining populations (Kahn 2006) and are generally characterised by a short-duration monitoring phase (see Burke 1989, Duda & Krysik 1998, Tuberville et al 2005, Field et al 2007). The majority of these projects have been carried out in mitigation of the development and mining of tortoise habitat, and the goal of any follow up monitoring has generally assessed immediate, short-term success through the study of post-release survival, movement and site fidelity (Berry 1986, Duda & Krysik 1998, Eubanks et al 2003, Field et al 2007, Tuberville et al 2008). A number of recent studies, however, have adopted a more long-term perspective, albeit retrospectively (Ashton & Burke 2007, Tuberville et al 2008). A study by Ashton and Burke (2007) involved an evaluation of historical data focussing on individual and population level retention, growth and reproduction of a translocated population of gopher tortoises 17 yrs after relocation. Tuberville et al (2008), meanwhile, also carried out a review of capture history data for a population of translocated gopher tortoises, gathered over 18 years, in an assessment of post-release survival. The fact that these kinds of studies are rare, retrospective in nature and involve the review of previous studies by other authors highlights that thorough, pre-planned long-term monitoring strategies are lacking and that the majority of studies are experimental in nature rather than evaluations of success.

2.4. Reintroduction of captive-bred tortoises

Reintroduction is the primary goal of keeping animals in captivity and animals bred and released within their own countries can excite local and national interest and pride (Durrell & Mallinson 1987). To date, however, reintroductions of entirely captive-bred animals have been less successful than projects involving entirely wild-caught subjects (Griffith et al 1989). Tortoises, though, may be better suited to this kind of intervention than other taxa as they have simple social structures, are not wide ranging, lack parental care and are less susceptible to the negative effects of captivity, such as loss of natural behaviour, that are associated with birds and mammals (Pedrono & Sarovy 2000, Germano & Bishop 2008). An example of a long-running and successful programme is the reintroduction of captive bred giant Galapagos tortoises, *Geochelone hoodensis,* on to Espanola Island, Galapagos (MacFarland 1974). By 1964 the population of this species on Espanola had been reduced to 14 animals as a result of over exploitation and habitat loss (Milinkovitch et al 2004). The resulting recovery plan for the species involved the removal of the remaining tortoises to a breeding centre on Isla Santa Cruz and the ecological restoration of Espanola Island (MacFarland et al 1974). As of 2002 the original Espanola population, together with a single male from San Diego Zoo, have produced 1200 offspring in captivity that have been successfully repatriated. In 1994, the first wild offspring from repatriated parents were recorded (Milinkovitch et al 2004). Long running programmes like this, however, are rare and other reintroductions from captivity have been less successful. The release of captive-bred Aldabran giant tortoises, *Dipsochelys dussumieri*, onto Curieuse Island in the Seychelles in the late 1970s and early 1980s, for example, suffered from a lack of post-release management and monitoring. By 1990, over half of the 250 tortoises released had been lost; primarily as a result of poaching, disease and predation by invasive species (Samour 1987, Hambler 1994, Gerlach 2007). There have also been attempts to establish additional populations of this species on other Seychelles Islands, but these have mostly been carried out by private landowners who have ambitions in the international pet trade (Gerlach 2007). As a result, very little post-release monitoring and research has taken place and very little published material is available in the literature. Further releases of other giant tortoise species have also occurred on other Seychelles Islands, including the release of 5 adult Arnold's giant tortoises, *Dipsocheylis arnoldi*, onto Silhouette Island in 2006 and 2007 (Gerlach 2008). There are also plans for the future reintroduction of 20 native Seychelles giant tortoises, *Dipsochelys hoolissa*, in 2009 (Gerlach 2007). As yet, however, no post-release studies have been made available. In the last twenty years, translocations and reintroductions of wild (Guyot & Clobert 1997) and captive-bred Hermann's tortoise (*Testudo hermanni*) have also been carried out, in Southern France and wider Western Mediterranean region but, again, very few studies have explicitly assessed the viability of these methods (Bertolero et al 2007).

An emerging motivation behind the release of captive-bred tortoises into the wild is as a part of wider ecosystem restoration projects (Gibbs et al 2008). Endangered species of reptiles can be introduced into areas where they once may have played an important role in the ecosystem as grazers and dispersers of native plants (Gibbs et al 2008). An example of this form of management is the release of non-native species of giant tortoises on to Round Island in the Mauritius. Here, released tortoises act as analogues (ecological replacements) of extinct native grazing tortoise species (Muller 2006).

2.5.1. Ploughshare tortoise - Status, habitat and distribution

The ploughshare tortoise, *Astrochelys yniphora* (Angonoka in Malagasy), is critically endangered with extinction (IUCN 2009) and is considered to be the rarest tortoise species in the world (Pedrono & Sarovy 2000). Endemic to the Baly Bay National Park in north-west Madagascar (Pedrono & Sarovy 2000, O'Brian et al 2005), the species has undergone severe historical decline through habitat loss and over-exploitation and is now restricted to six isolated populations which total as few as 600 adult and sub-adult individuals (DWCT unpublished data). A survey of Baly Bay in 2000 estimated that around 8000 ha of suitable habitat remains, comprising of mosaics of bamboo scrub, deciduous forest and grassland (Fig. 2.1) (Andriandrasana 2000), surrounded by increasingly large areas of fire-derived palm savannah (Pedrono 2000, Pedrono et al 2001, Bourou et al 2001). Due to the effects of recent poaching, only around 6600 ha of this habitat is thought to be occupied by tortoises. The climate of the Baly Bay region is harsh and arid, characterised by a prolonged and severe dry season with zero precipitation, and rains only occurring from late November through until April (O'Brian et al 2005, Lewis et al 2009).

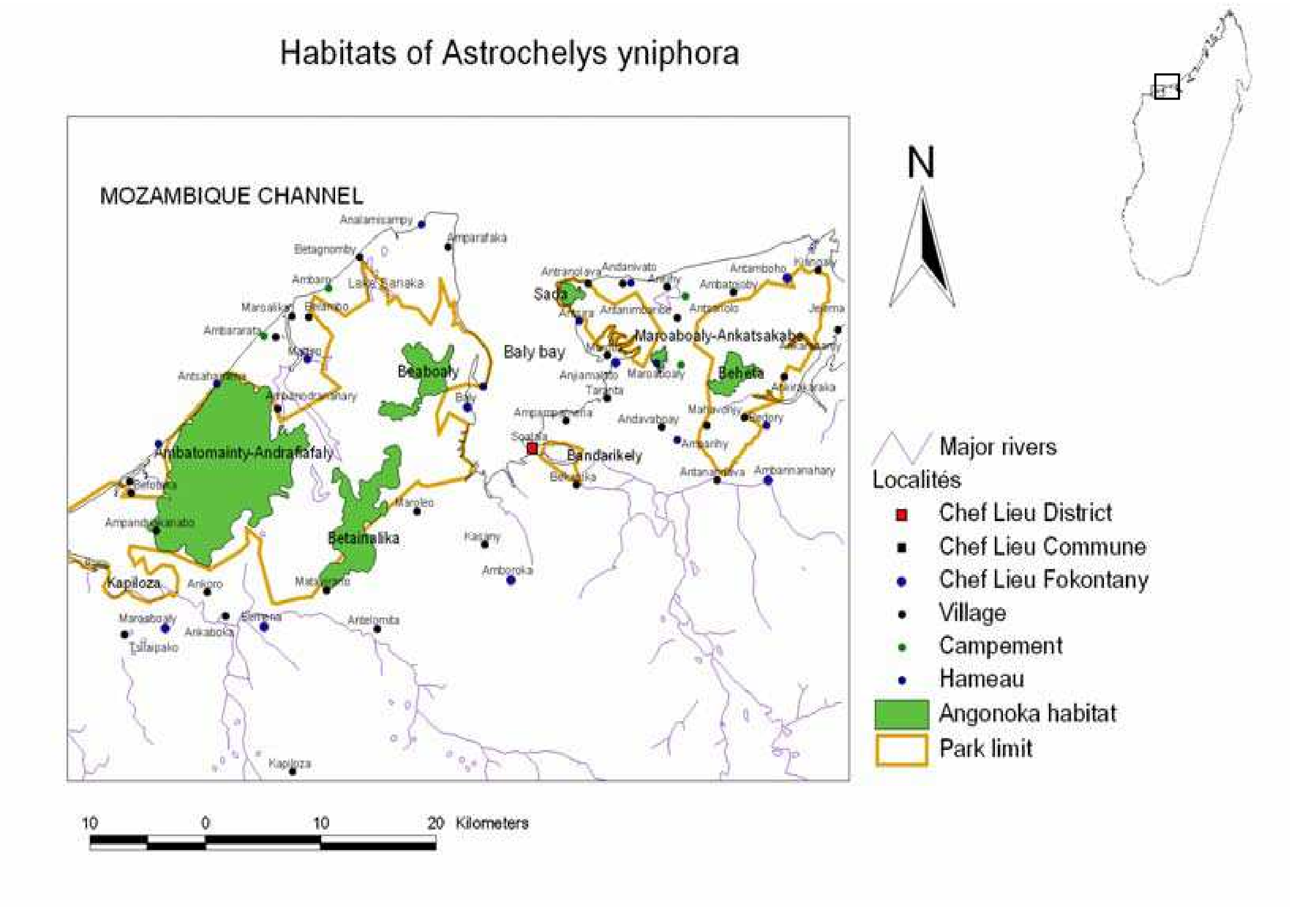

Figure 2.1. Map of existing ploughshare tortoise habitat in Baly Bay National Park, north-west Madagascar (Source: Lewis et al 2009).

2.5.2. Ploughshare tortoise morphology

The ploughshare tortoise (Fig 2.2) is the largest extant Malagasy tortoise (Pedrono & Sarovy 2000) and can weigh up to 20 kg and reach a carapace length of 50cm (Bourou et al 2001). The species is characterised by an elongated projection of shell under the head, known as a gular scute, which is used by males in inter-male conflict and in courtship of females (Smith et al 1999). Adults are sexually dimorphic, with males being larger and having longer gular scutes than the females (Juvik et al 1981, Curl et al 1985). Males also have concave plastrons (action plan 2008). Ploughshares have a long juvenile phase of around 20 yrs and reach maturity at a given body size (at around 30 cm carapace length) rather than a specific age (Kuchling & Lopez 2000, Pedrono & Sarovy 2000, O'Brian et al 2005).

Figure 2.2. Adult male (right) and juvenile (left) Ploughshare tortoises (*Astrochelys yniphora*). Note the elongated gular scute of the adult. Sex of juveniles can only be determined endoscopically (Kuchling & Lopez 2000). (Image Source: Male- Arkive 2009; Juvenile- BCG 2006)

2.5.3. Ploughshare tortoise biology

Ploughshare tortoises have a generalist diet and eat leaves and other plant material, including dry leaves from shrubs and grasses (Bourou et al 2001). Feeding on the faeces of other species has also been observed in the wild (DWCT unpublished data). The species is diurnal (Juvik et al 1981) and has defined active and inactive phases, with the active phase occurring during the hot, wet season from October to May (Bourou et al 2001). During the inactive period, tortoises burrow into fallen leaves within patches of bamboo and other dense vegetation, where they remain relatively inactive but do not enter complete aestivation (Lewis et al 2009). When mature, the ploughshare tortoise has no natural predators, however, juveniles may be predated by a number of species including the Indian palm civet (*Vierricula indica*) and Madagascar buzzard *(Buteo brachyphterus*) (Pedrono & Sarovy 2000). Juveniles are also known to be predated by the introduced African bush pig (*Potamochoerus larvatus*), which is also a confirmed nest predator (Pedrono et al 2001).

2.5.4. Ploughshare tortoise reproduction

Mating occurs at the start of the wet season (Pedrono et al 2001) and courtship is preceded by a period of inter-male combat (Reid et al 1989). Nesting begins one to three months after mating ends (Reid et al 1989), with the number of eggs per nest ranging from 1-6 (Bourou et al 2001). Emergence from the buried nests is triggered by the first rains that follow the dry season, when the ground is softened, and enabling newly hatched tortoises to dig their way to the surface (Reid et al 1989). Sex of juveniles is likely influenced by incubation temperatures in the nest (Kuchling & Lopez 2000) and first year survival in the wild has been recorded at 42% (Bourou et al 2001).

2.6.1. Ploughshare tortoise conservation - Threats

The current status of the ploughshare tortoise is as a result of many centuries of commercial trade (Pedrono & Sarovy 2000) and the loss of habitat and direct mortality from fire (Lewis et al 2009). The species was once also regularly collected for food by locals, however this is now rare and eating tortoise is considered “Fady” or taboo (Curl 1986). Today, all trade in the ploughshare tortoise is illegal and the species is listed under Appendix I of CITES (CITES 2009). However, despite conservation success in the 1990s, this century has seen an increased threat from illegal trafficking and, at the current rate of extraction, extinction in the wild could occur within a few years (Lewis et al 2009). Typical of a chelonian species, the ploughshare tortoise is characterised by slow growth rate, delayed maturity and low fecundity and, as a result, populations are highly sensitive to loss at the adult stage (Pedrono et al 2004). For terrestrial tortoise species, maintenance of stable populations is dependent on high adult survival (Tuberville et al 2008). There is a high demand for the species from the international pet trade and, in 2001, an individual was offered for sale on the Internet at over $10000 (Lewis et al 2009), and tortoises have been known to fetch a street value of £35000 on the black market (BBC 2001). Such demand, coupled with the high level of poverty in local human communities, means that all tortoises in the wild are at risk (Lewis et al 2009). There is also concern that the species is being targeted for its meat by traders from China (Chiew 2009). Theft of individuals from captivity has also occurred on two occasions. Firstly, in 1996, when 73 juveniles and 2 adult females were stolen from the captive breeding centre in Ampijoroa and again, in May 2009, when 4 sub-adult juveniles were stolen from pre-release enclosures inside Baly Bay National Park (BBC 2009).

The other major threat currently facing the species is from habitat loss and alteration that occurs as a result of bushfires (Pedrono et al 2004). The entire region in which the tortoises occur is subject to frequent bushfires (Bourou et al 2001), which are started deliberately each dry season by local farmers to encourage the growth of new grass for cattle (Curl 1986). The result of this burning is the fragmentation of the tortoise's habitat and the gradual conversion of bamboo scrub and forest to barren savannah (Pedrono & Sarovy 2000). Fires are also known to kill tortoises directly (Pedrono et al 2004).

2.6.2. Ploughshare tortoise conservation - Project Angonoka

In 1985, with the backing of the IUCN/SSC Tortoise and Freshwater Turtle Specialist Group, the DWCT developed and began implementing a conservation management plan for the ploughshare tortoise in Madagascar (Pedrono & Sarovy 2000). Named Project Angonoka, after the species' Malagasy name, the project represented collaboration between the DWCT and the Malagasy Department of Water and Forests (now the Direction des Forets) (Pedrono & Sarovy 2000, Lewis et al 2009). In 1986, a captive breeding and head-starting program was started at Ampijoroa Forestry Station in Ankarafantsika National Park, c150km east of the species' present natural range (Curl 1986, Pedrono & Sarovy 2000). The station represents the World's only captive breeding colony of ploughshare tortoises (Pedrono & Sarovy 2000) and has produced more than 200 captive born animals, 25 of which have so far been released into natural habitat at Beaboaly, and are the focus of this study (Lewis et al 2009). Project Angonoka is a multi-disciplinary project, and has also included detailed field studies of wild populations as well as captivity based research at Ampijoroa (Smith et al 99, Pedrono et al 2000, Kuchling & Lopez 2000, Bourou et al 2001). Another vital aspect of the Project has been the inclusion of the local Sakalava communities, whose support and consultation has aided in the tackling of sources of decline in the species, principally in the reduction of bushfire occurrence (Pedrono & Sarovy 2000). As a result, since 1995, uncontrolled burning has been greatly reduced through the creation of a series of fire-breaks around key areas of tortoise habitat (Lewis et al 2009). Also, in December 1997, with the support of the surrounding local communities, the Baly Bay National Park was created with the specific purpose of protecting the remaining wild ploughshare tortoise populations (Lewis et al 2009). Despite this effort, however, fires do still occur within the park and continue to be a major threat to the long-term survival of the species (Lewis et al 2009).

2.6.3. Ploughshare tortoise conservation - Reintroduction from captivity

The main goal of the Project's captive-breeding programme has been to increase the ploughshare tortoise population to a size that will ensure its long-term survival (Pedrono & Sarovy 2000). Ploughshare tortoises may be considered particularly suitable candidates for programmes involving captive-breeding with reintroduction on account of the delayed maturity, low mortality and innate behaviour associated with the species (Pedrono 2000, Pedrono & Sarovy 2000). As part of a model of interactive management, captive-bred juveniles are used to create an artificial metapopulation structure for the species and so reduce the risk to the wild population (Pedrono et al 2004). Also, individuals bred in captivity may be released into the wild to supplement existing wild populations or to create new-populations in areas of its former range (Pedrono & Sarovy 2000). A habitat survey of the Baly Bay National Park identified the Beaboaly area in the west as the most ecologically favourable site for the release of captive-bred individuals back into the wild (Andrianandrasana 2000). Beoboaly has been known to contain ploughshare tortoises in the past; however, they have been locally extirpated for a number of years (Curl et al 1985, Pedrono & Sarovy 2000). Population modelling has shown that a self-sufficient population could be established in this area based on 50 individuals (Pedrono *et al* 2004). In 1998, a trial release of five captive-bred juveniles was carried out in the site at Beaboaly (Pedrono & Sarovy 2000). The juveniles were of unknown sex but of similar size, selected based on their being large enough to be relatively safe from predation (Pedrono & Sarovy 2000). The number of suitable candidates for release had been greatly reduced by the theft from the breeding centre in 1996 (Pedrono & Sarovy 2000). The results of the release were positive and demonstrated that captive raised juveniles are able to adapt and survive in their natural habitat (Pedrono & Sarovy 2000). The results of the release suggested that reintroduction of captive-bred juveniles is a viable conservation strategy for the species and allowed for the refinement of techniques that could maximise the success of future releases (Pedrono & Sarovy 2000). Due to the success of the trial release, a further 20 tortoises were released into Beaboaly in January 2006 following a period of pre-release acclimatisation.

3. Methods

3.1. Study site and subjects

The tortoises released in this study were all bred at the specialist captive breeding facility at Ampijoroa, 150km east of the species' natural range in northwest Madagascar (Reid et al 1989). All tortoises experienced the same captive conditions and husbandry regime. After emergence, hatchlings were kept by cohort in sheltered 1m^2 rearing enclosures until 4 years of age were they were then transferred to a larger 20m^2 compound with natural plants, logs and stones (Pedrono & Sarovy 2000). Feeding occurred twice a day in the morning and evening and drinking water was made available at all times throughout the year (Pedrono & Sarovy 2000).

Tortoise ID	Sex	Date of Birth	Age at release (Years)	Transfer Date	Enclosure ID	Release Date
T3020	M	11/12/1990	15	07/12/2005	2	22/01/2006
T3095	M	19/11/1989	16	07/12/2005	1	22/01/2006
T3096	F	16/11/1989	16	07/12/2005	3	22/01/2006
T3102	F	01/12/1988	17	07/12/2005	4	22/01/2006
T3106	F	03/11/1989	16	07/12/2005	3	22/01/2006
T3112	F	16/11/1991	14	07/12/2005	1	22/01/2006
T3117	F	16/11/1991	14	07/12/2005	3	22/01/2006
T3119	F	22/11/1990	15	07/12/2005	4	22/01/2006
T3123	F	14/11/1991	14	07/12/2005	1	22/01/2006
T3124	F	17/11/1991	14	07/12/2005	2	22/01/2006
T3125	F	24/11/1992	13	07/12/2005	4	22/01/2006
T3128	F	27/11/1993	12	07/12/2005	3	22/01/2006
T3130	F	19/11/1990	15	07/12/2005	4	22/01/2006
T3134	M	19/11/1993	12	07/12/2005	3	22/01/2006
T3146	F	05/12/1994	11	07/12/2005	2	22/01/2006
T3160	?	19/11/1995	10	07/12/2005	1	22/01/2006
T3196	?	28/11/1996	9	07/12/2005	1	22/01/2006
T3207	M	28/11/1996	9	07/12/2005	2	22/01/2006
T3332	?	13/11/1993	12	07/12/2005	4	22/01/2006
T3409	F	19/11/1991	14	07/12/2005	2	22/01/2006

Table 3.1. Age, sex and release details of the captive-reared tortoises used in this study.

Individuals selected for release had to be of a size that would ensure that risk from predation was minimal and at a developmental stage that would optimise resistance to environmental factors (e.g. climate and food availability) without compromising their potential to settle within the area of release (Pedrono & Sarovy 2000). It was also preferable that the released tortoises should be at the same stage of development as the trial release tortoises already established at Beaboaly. First generation offspring were therefore used. All individuals were at the juvenile/sub-adult juvenile stage of development. Details of all tortoises sampled in this study are shown in Table 3.1. Sex was known for 17 of the 20 tortoises and the ratio of females to males was 13:4. As with the trial release, all individuals were siblings of unknown paternity (Pedrono & Sarovy 2000).

The release site was the Beaboaly region of Baly Bay National Park (16,_040S, 45_150E), the same area in which the trial release was carried out in 1998 (Pedrono & Sarovy 2000). This site is located to the west of Baly Bay (Fig. 2.1) and is several square kilometres in size (Pedrono & Sarovy 2000). Beaboaly was selected for its remote location and ecological suitability (Andrianandrasana 2000). Pedrono and Sarovy (2000) stated that it was important for the release site to fulfill the following criteria:

- Must be able to provide suitable habitat for the species
- Must be large enough to allow free movement of individuals
- Must contains no wild ploughshare tortoise population
- Must contain habitat isolated or unconnected to that containing wild ploughshare tortoise populations.

Ploughshare tortoises were known to occur in Beaboaly in the past, however, the last known specimen was found burned in the early 1980s (Curl et al 1985). The site at Beaboaly, shown in Figure 3.1, is typical of extant ploughshare tortoise habitat and is characterised by areas of bamboo-scrub and bamboo-forest surrounded by areas of open and palm-savannah. There are also some patches of deciduous forest, palm plantation and an area of marshland to the east.

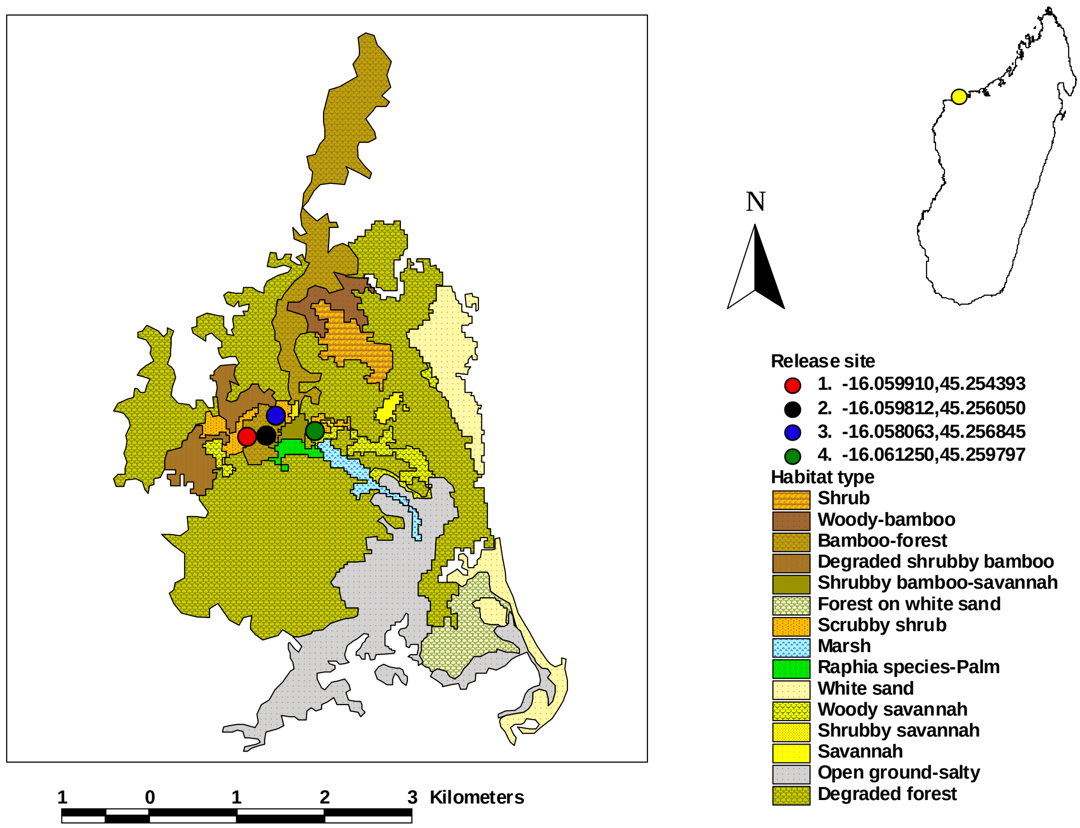

Figure 3.1. Map of Beaboaly release site showing site of pre-release pens and habitat types.

3.2. Release procedure and monitoring

Prior to release, tortoises were transferred from the breeding centre at Ampijoroa to four pre-release pens within Beaboaly. Tortoises were held here from 7 December 2005 until release on 22 January 2006, a duration of 36 days. Tortoises were held in groups of 5 and were provided food and constant access to drinking water. Tortoises were weighed 10-16 days after transfer to the release pans and then at roughly monthly intervals until the end of the 2006 sampling period. Tortoises were released during the wet season. The tortoises were fitted with AVM transmitters (AVM Instrument Company Ltd), which were glued to the final vertebral scutes with epoxy resin. Radio-tagged tortoises were then followed using a LA12Q receiver with an H-shaped antenna. Tortoises were located at uneven intervals over a period of 122 days from release until 24-25 May. When a tortoise was located, its co-ordinates were recorded using GPS and the habitat type in which it was found was also recorded. Tortoises were again monitored in 2007 and 2008. A summary of sampling regimes is shown in Table 3.2.

Tortoise I.D.	Release Group	Number of observations 2006	Number of observations 2007	Number of observations 2008
T3020	2006	35	5	18
T3095	2006	36	11	20
T3096	2006	36	17	29
T3097	Trial	36	14	16
T3098	Trial	33	23	15
T3101	Trial	31	11	20
T3102	2006	34	0	22
T3104	Trial	37	17	28
T3106	2006	35	13	24
T3112	2006	37	16	20
T3117	2006	37	0	0
T3119	2006	42	0	0
T3123	2006	35	10	16
T3124	2006	36	0	12
T3125	2006	36	19	21
T3128	2006	35	16	30
T3130	2006	42	6	30
T3134	2006	36	13	24
T3146	2006	40	20	25
T3160	2006	37	12	28
T3196	2006	36	12	26
T3207	2006	36	6	29
T3332	2006	38	18	19
T3409	2006	35	13	23

Table 3.2. Number of observations for each individual tortoise during 2006 – 2008 sampling periods

3.3. Data analysis

Radio tracking data was mapped in GIS (ArcView 3.3, Esri, California, USA), and the Animal Movement Extension (Hooge & Eichenlaub 1997) was used to calculate movement data and produce home range estimates. 100% Minimum Convex Polygon (MCP) and 95% Kernel with least squares cross-validation (LSCV) smoothing factor (as recommended by Seaman et al 1999) were used to estimate home range size. The 100% MCP is the most commonly used method in studies of this kind (O'Connor et al 1994, Duda et al 1999, Tuberville et al 2005, Field et al 2007, Hester et al 2008) but fixed kernel estimators are considered to be more robust than MCP methods and their use has increased in recent years (Kernohan et al 2001, Hemson 2005). 100% MCPs were used in this study to define home range as parameters of post-release movement and habitat availability and 95% Kernels with LSCV were used to provide an alternative estimation of home range as a parameter for post-release movement only. No removal of outlying data was carried out on account of the small sample size and because a full representation of the extent of post-release movement was required. The use of two home range estimation methods is recommended by Harris (1990) and Kernohan et al (2001). In keeping with the reporting in Pedrono and Sarovy (2000), home range estimates presented in the results section are those calculated using the 95% Kernel method.

Data were tested for normality using the Shapiro-Wilk test and for homogeneity of variance with the Fligner-Killeen test. Data analysis was carried out using R 2.9.1. (R Development Core Team, 2008) unless stated otherwise. To avoid bias, comparisons between years were only calculated for tortoises with greater than or equal to 14 observations in all 3 years (see Table 3.2.). Post-release change in body mass was investigated as a potential reflection of body condition and fitness. Difference in mean pre-release body mass (BM) between groups was analysed using a Wilcoxon Rank Sum Test. Differences in overall mean percentage BM change and mean successive percentage BM change between groups was analysed using Two Sample t-tests. Change in body mass was calculated as percentage change so as to reinstate heterogeneity of variance. A linear mixed effects model (LME) was used to analyse difference between BM at pre-release and end of sampling, with time period (start or end) as fixed effect and individual tortoise I.D as random effect. Prior to analysis, BM was Log to the base 10 transformed in order to reinstate normal distribution.

Due to the non-daily sampling regime and inconsistent time between observations, daily movement data was standardised by dividing the straight-line distance between observations by the number of days elapsed between observations. All distance data were Log to the base 10 transformed to reinstate normal distribution. Differences in daily movement between groups was analysed using LME with release group as fixed effect and days since release and individual tortoise I.D. as random effects. Differences in mean daily movement after 30, 60 and 120 days was analysed using LME with days since release as fixed effect and individual tortoise I.D. as random effect. Difference in mean daily movement between years was analysed with LME with year as fixed effect and individual tortoise I.D. as random effect. Difference in mean distance from release site of 2006 release tortoises after 30, 60 and 120 days was analysed using LME with days since release as fixed effect and individual tortoise I.D. as random effect. Difference in mean distance from release site between years was also analysed using LME with year as fixed effect and individual tortoise I.D. as random effect.

Differences in mean home range size between groups was analysed with Wilcoxon Rank Sum Tests. Difference in home range size between years was analysed using LME with year as fixed effect and individual tortoise I.D. as random effect. Home range size was Log10 transformed prior to analysis to reinstate normal distribution of residuals. Directed movement was tested for with the Rayleigh test using the Animal Movement Extension (Hooge & Eichenlaub, 2000) in Arc View 3.3.

3.3.1. Analysis of habitat and resource use

Movement and location data can be used in the evaluation of resource use by animals after release (Pace III 2001). The habitat use of a released individual is non-random (O'Connor et al 94) and understanding this use is crucial in assessing the adaptation of these individuals to their new environment (Dickinson et al 2001). A common approach to this sort of evaluation using radiotelemetry data is the comparison of resource use to resource availability (Erikson et al 2001). Compositional analysis (CA) (Aebischer et al 1993) has become a popular method for analysing habitat use data (Erikson et al 2001) and has been used in recent studies involving the Texas tortoise, *Gopherus berlandier,* (Kazmeier et al 2001) and Desert tortoise, *Gopherus agassizii,* (Riedle et al 2008). This technique uses log-ratios of use and availability in a multivariate analysis to evaluate selection among habitat types (Kazmaier et al 2001). The animal is the experimental unit (Aebishcer et al 1993).

CA was carried out with radiotelemetry data from the released tortoises during the 2006 sampling period. Individual 100% MCPs were merged with a habitat map of Beaboaly using GIS and the proportions of each habitat type within each individual polygon were calculated, as were the proportion of individual observations occurring within each habitat type. The proportion of habitat types within the overall study area (defined as 100% MCP calculated for all observations of all released tortoises in 2006) was also calculated. A classical CA (Aebischer et al 1993) was then carried out and second and third order habitat selection calculated. Second order habitat selection describes an individual's selection of home range within the wider study area and third order selection describes the location of observations within an individual's home range (Johnson 1980). This was calculated using the Adehabitat package 1.8.3 and the Compana function (Calenge 2006) in R 2.9.1 (R Development Core Team 2009). Zero values for habitat use were replaced by a small default value of 0.001 and missing values for habitat availability were replaced using the weighted mean of all non-missing values for that log-ratio difference (Weighted mean lambda; see Aebischer et al 1993). A matrix of log-ratio differences in use and availability between each habitat pair was produced and habitats ranked according to preference (Aebischer et al 1993). The matrix shows either positive (selected for) or negative (selected against) values for a certain habitat (Riedle et al 2008). If use and availability differ for a habitat, selection is considered to have occurred when use is greater than availability, the opposite being the case for avoidance (Kazmeier et al 2001).

4. Results

4.1. Survival

All tortoises released in 2006 survived and had no apparent serious negative reaction to release in the wild. Tortoises foraged and fed successfully and no health problems were encountered. The trial release tortoises released in 1998 (Pedrono & Sarovy, 2000) also continued to survive without incident throughout the duration of this study.

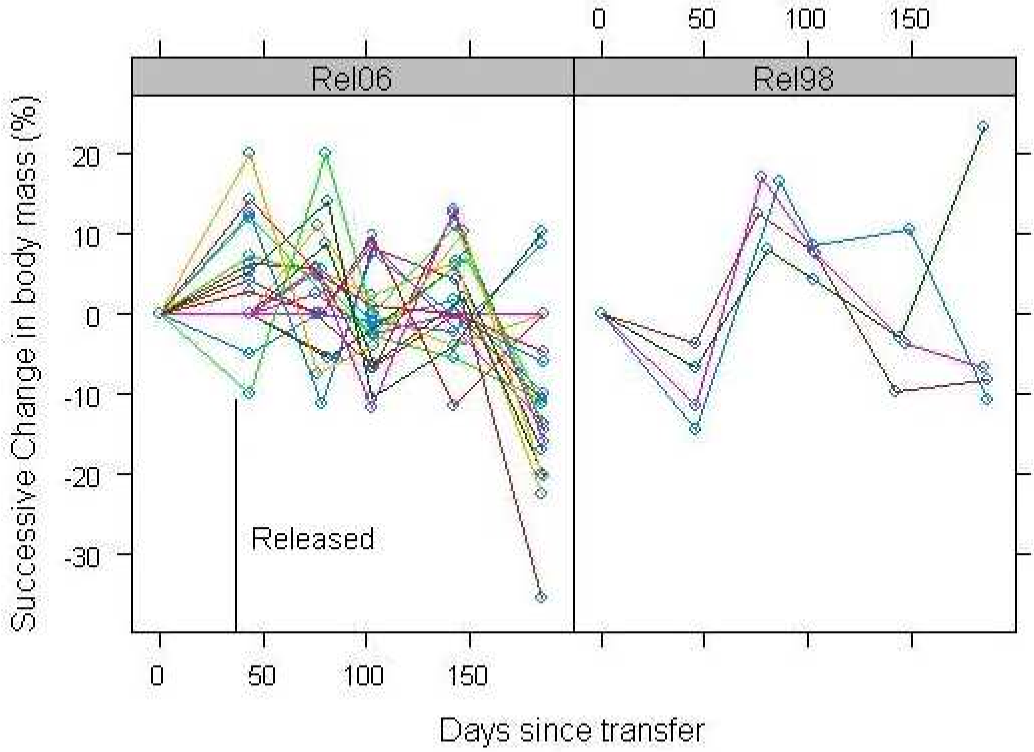

Figure 4.1. Successive changes in body mass (%) for 2006 release (Rel06) and trial release (Rel98) tortoises from date of 2006 release group's pre-release transfer on 7 December 2005 until end of sampling 10-12 June 2006. The 2006 group were released 46 days after pre-release transfer.

4.2. Body mass

Mean body mass (BM) at time of pre-release of the 2006 release group (4.44 ± 0.99kg, range 2.38 – 7.04kg) was lower than that of the trial release group (7.18 ± 0.11kg, range 7.06 – 7.29kg; t = -11.9275, df 20.837, p <0.001). Figure 4.1 shows that BM fluctuated for individuals of both release groups during the first year sampling period. However, BM at the end of sampling (Table A.1) did not differ from that recorded at the time of pre-release for the

2006 (LME with log10 transformation: t = 1.7261, df 19, p 0.1) or the trial release tortoises over the same period (LME with log10 transformation: t = -1.355, df 3, p 0.27). Mean percentage change in BM from pre-release captivity to the end of sampling in 2006 was -3.95 $\pm$ 11.21 % (range -31.11 – 13.68 %) for the 2006 release group, whilst the mean percentage BM change of the trial release group was 7.55 $\pm$ 12.95 % (range -2.857 – 25.926%) over the same period. As can be seen in Table A.1, only 5 of the 20 tortoises released in 2006 gained in BM, whilst 4 showed no net gain and 10 declined in BM. Whereas, no discernable pattern in BM change can be observed for the 2006 release group, the trial release tortoises all show a gain in BM during the first two months of the study followed by declines in 3 of the 4 subjects (Fig. 4.1.).

4.3. Dispersal and movement

Released individuals settled in areas near to their release sites (Fig. 4.3), displaying strong site fidelity. Figure 4.2. shows that tortoises moved away from their release pens in all directions and did not display homing behaviour towards the centre where they were bred (due east). Only one individual displayed statistically significant directed movement (T3146).

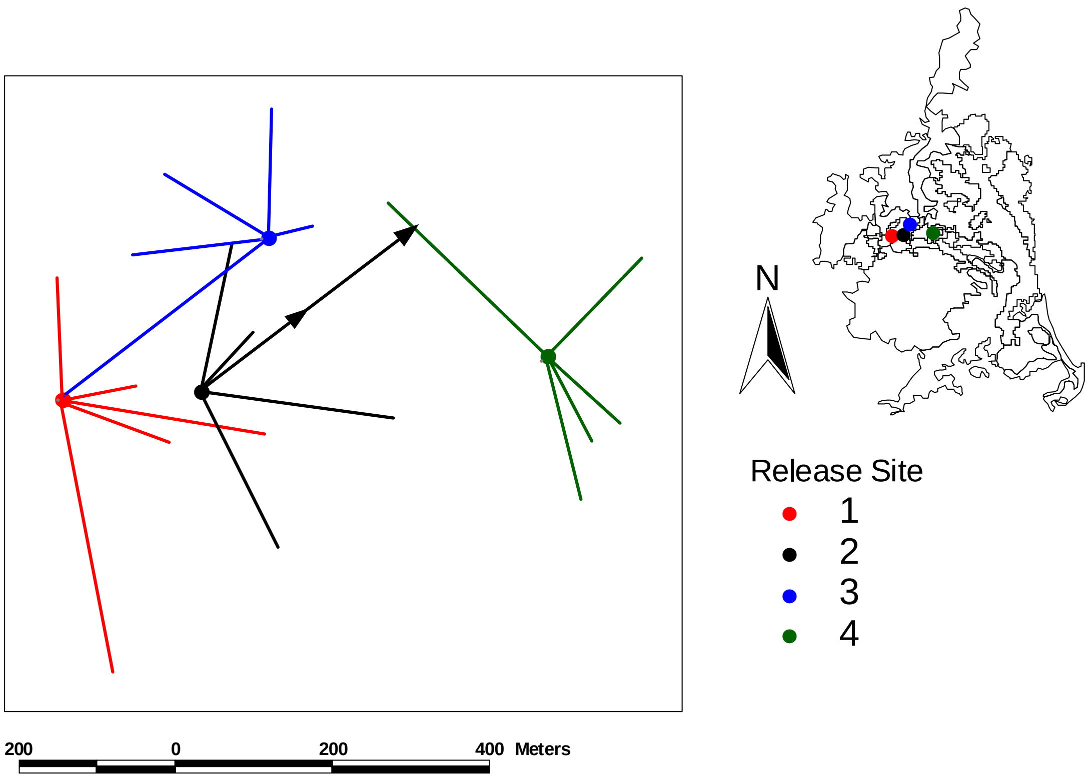

Figure 4.2. Azimuths showing mean bearing and mean distance (m) from release site for 2006 release tortoises during 2006 sampling period. The arrows indicate directed movement (z=3.023, p <0.01) by T3146.

The mean straight-line distance from release site during the 2006 study period was 192.28 ± 129.99m (range 4.77 - 1066.06m), with only one individual (T3146), on one occasion, being recorded at a distance over 1 km from its release site (Fig A.1. shows 2006 movement polyline for T3146). The next greatest distance recorded from release site was 669.66 m (T3106).

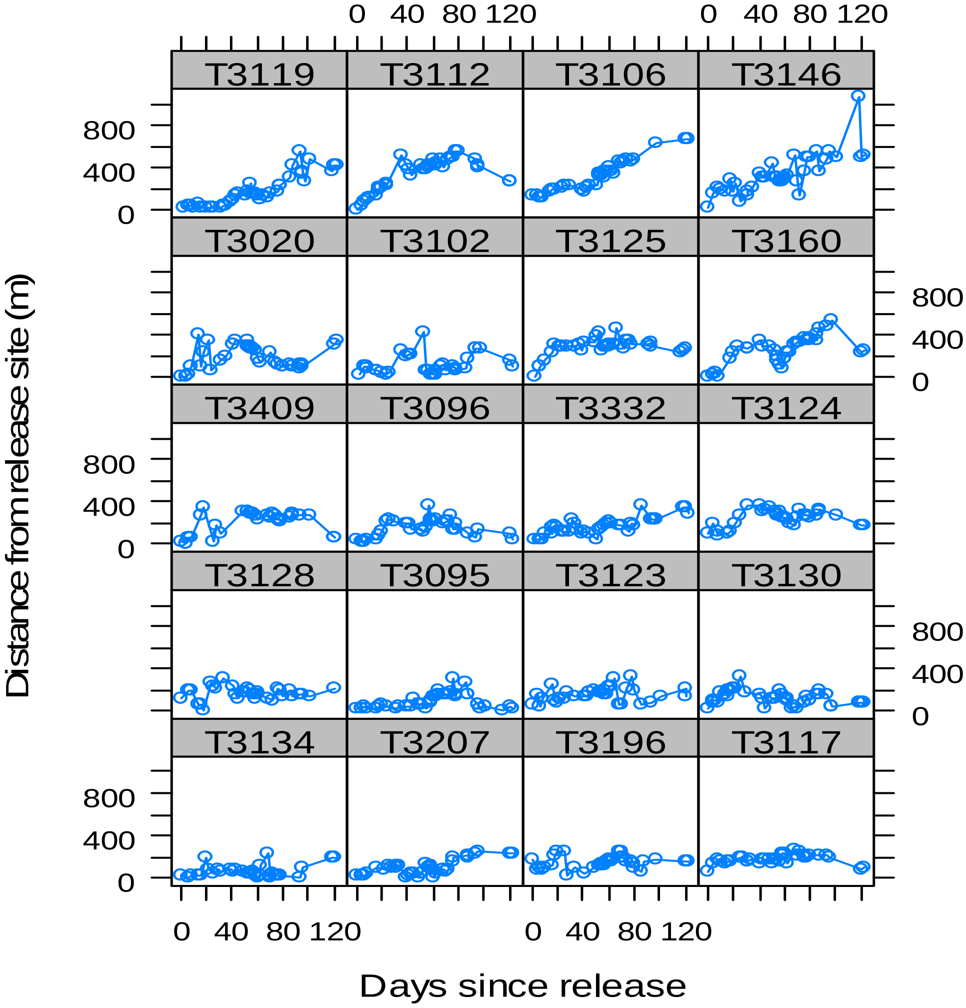

Figure 4.3. Distance from release site of 2006 release tortoises during 2006 sampling period. Tortoises are displayed in order of degree of distance from release site with smallest bottom left and greatest movement top right.

Tortoises moved away from release pens following release. Mean distance from release site after 60 days (199.21 ± 97.13m, range 49.34 – 409.87m) increased significantly (LME with log10 transformation: t = 3.666, df 32, p <0.001) from that at 30 days after release (125.59 ± 51.19m, range 32.68 – 207.70m). Mean distance from release site after 120 days (235.12 ± 125.28m, range 70.02 – 501.70m), was also significantly greater than that at 30 days (LME with log 10 transformation: t = 5.012, df 32, p <0.001). Figure 4.3 shows that released tortoises stayed near to the area in which they were released and consistently returned to this area, even following large movements away. Only 4 of the 20 released tortoises (T3119, T3106 and T3146) appear to show progressive movement away from the release site during the 2006 sampling period and only T3106 does not show any returning movement towards its area of release.

As can be seen from Figure 4.4., daily linear movements over 100m (forays) were rare for both release groups (3% of all 2006 release and 7% of all trial release movements). The largest recorded daily linear movement by a 2006 release tortoise was 554.75m, again by T3146. The largest movement by a trial release individual was 915.20m (T3104). Forays of around 200m were not uncommon for individuals of either group.

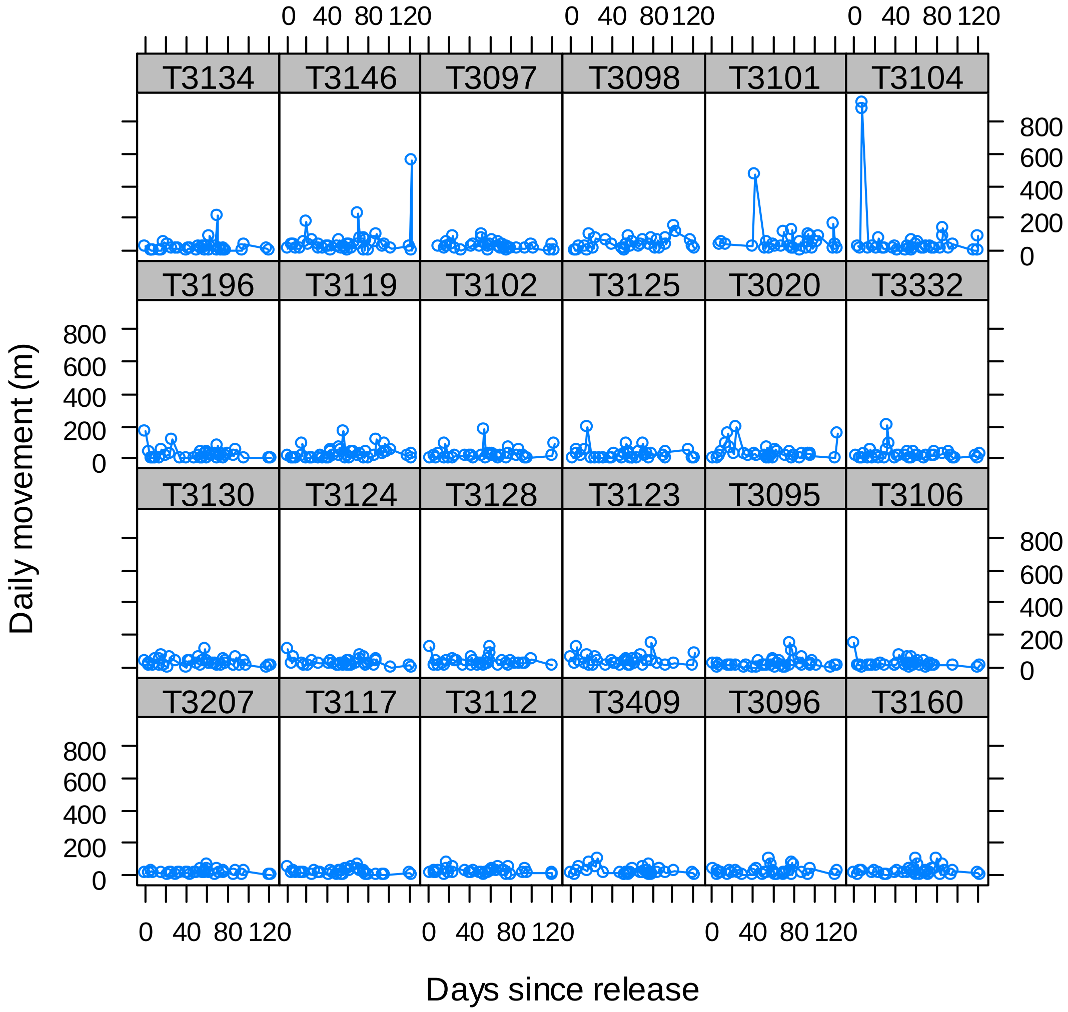

Figure 4.4. Daily movement (m) of 2006 release and trial release tortoises during 2006 sampling period. Trial release tortoises are T3097, T3098, T3101 and T3104 located in the top right of the figure.

As can be seen from figures 4.3 and 4.4, forays were immediately followed by a return to the area near to where they had set off. The 2006 release group (28.68 $\pm$ 37.11m, range 0.47 – 554.75m) exhibited less daily movement during the 2006 sampling period (LME with log10 transformation: t = 3.116, df 22, *p* 0.005) than the trial release group (55.14 $\pm$ 116.92m, range 1.6 – 915.20m). Mean daily movement of the newly released tortoises remained constant with that at 30 days after 60 (LME with log 10 transformation: t = -0.199, df 32, *p* 0.844) and 120 days (LME with log10 transformation: t = 0.515, df 32, *p* 0.610). The same was also true for the trial release group (LME with log10 transformation : 60 days:- t = -0.450, df 6, p 0.635; 120 days:- t = -0.707, df 6, *p* 0.506).

Although mean distance of 2006 release tortoises from release site increased significantly in 2007 (LME log10 transformation: t = 2.943, df 10, *p* 0.015), mean distance from release site in 2008 did not differ significantly to that recorded in 2006 (LME with log 10 transformation: t = 1.903, df 10, *p* 0.086).

Daily linear movement in 2007 (LME with log 10 transformation: t = 1.856, df 10, *p* 0.09) and 2008 (LME with log 10 transformation: t = -0.289, df 10, *p* 0.78) did not vary significantly to that in 2006 for 2006 release tortoises. The same was also true in respect to the trial release group (LME with log 10 transformation: 2007: t = 0.398, df 4, *p* 0.711; 2008: t = 0.568, df 4, *p* 0.568). Daily linear movements of over 100m remained rare in 2007 (0.9 %) and 2008 (2%). The largest daily movement recorded in 2007 was 562.83m (T3125), however, T3146 remained the 2006 release tortoise with greatest movement (mean 77.51 $\pm$ 106.53m, range 9.83 – 453.73m). No individuals carried out large forays in all 3 years or repeated forays in a single given year.

4.4. Home range establishment

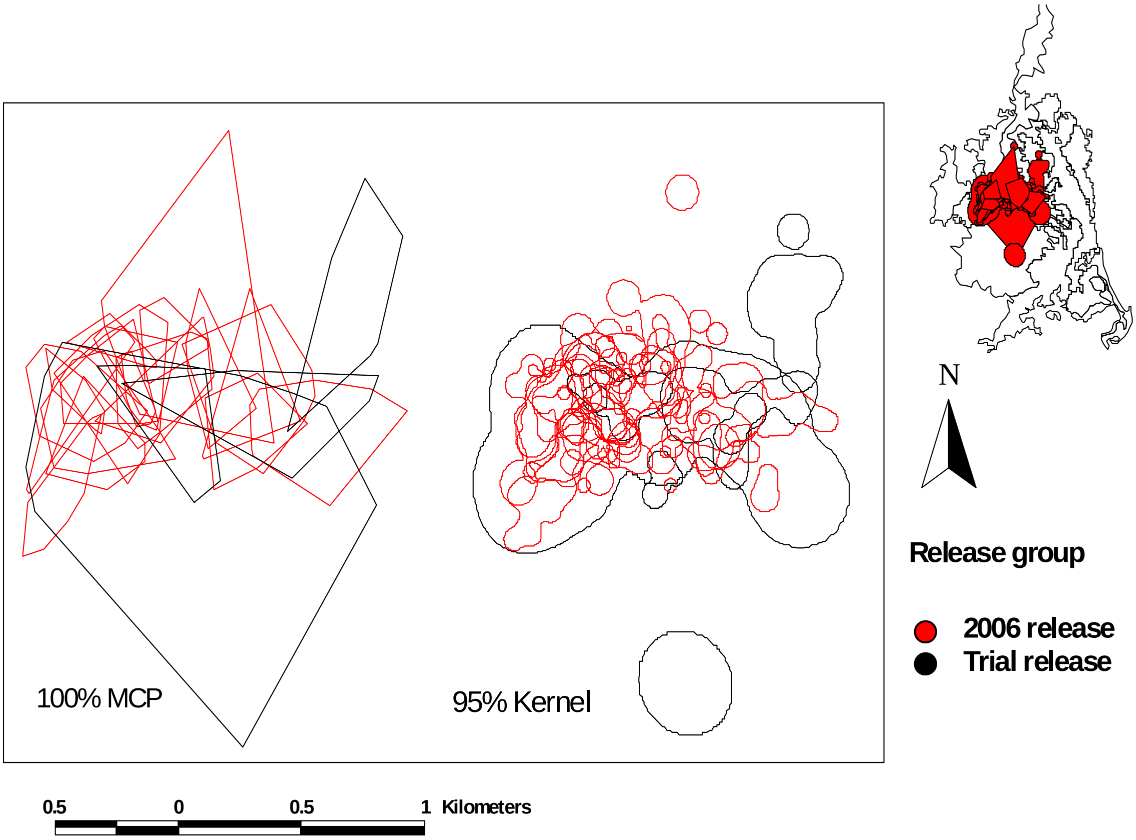

Figure 4.5. 100% MCP and 95% Kernel (with LSCV) home range estimates for 2006 release and trial release tortoises during the 2006 sampling period.

Both groups of tortoises exhibited considerable individual differences in size of cumulative home use area (Fig. 4.5). There was no difference in the respective size of home range estimates calculated using the 100% MCP and 95% kernel methods for either group (2006 release: w = 196, *p* 0.665; trial release: w = 10, *p* 0.6857). Also, there was considerable overlap between individual home ranges for both groups (Fig. 4.5). The 2006 mean home range area of the established trial release tortoises (462,627.00 $\pm$ 559,522.01m^2, range = 116,169.7 – 1,298,712.80m) was larger (w = 11, *p* 0.02) than that of the newly released group (110,839.20 $\pm$ 74,000.15m^2, range 17,676 – 316,263m^2). Mean home range did not differ significantly between groups in 2007 (w = 6, *p* 0.54) or 2008 (w = 21, *p* 0.275).

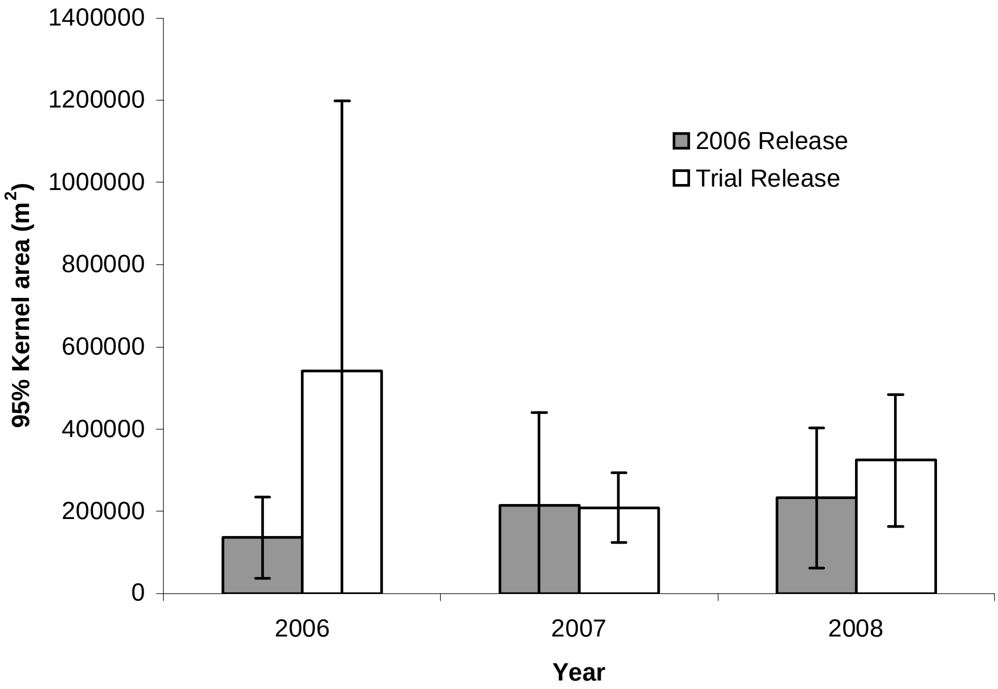

Figure 4.6. Mean 95% kernel areas (m^2) for 2006 release and trial release tortoises in 2006, 2007 and 2008. Only individuals with $\geq$14 observations per year were included.

Although mean home range of the 2006 release tortoises in 2007 (213,714.60 $\pm$ 226, 557.24m^2, range 40,777.49 – 531,317.95m^2) was almost double that of 2006 (Fig. 4.6), they did not differ statistically (LME with log 10 transformation: t = 0.516, df 21, *p* 0.516). Nor did mean home range in 2008 (182,286 $\pm$ 179,241m^2, range 20,685.64 – 604, 845.13 m^2) differ to that of 2006 (LME with log 10 transformation: t = 1.048, df 21, *p* 0.306). Mean home range of the trial release group in 2007 (209,337.90 $\pm$ 84,373.63 m^2, range 113, 237.10 – 271,258.80m^2) and 2008 (263,393.70 $\pm$ 177,186.80m^2, range 84,421.38 – 435,345.00m^2) also did not differ to that reported in 2006 (LME log 10 transformation: 06 vs 07 t = -1.150, df 5, *p* 0.302; 06 vs 08 t = -0.757, df 5, *p* 0.483). This is despite 2006 home range (540,464.12 $\pm$ 658,214.33m^2, range 116,169.68 – 1,298,712.78m^2) being nearly double that recorded in 2007 (209337.89 $\pm$ 84373.63m^2, range 113,237.12 – 271,258.80m^2).

4.5. Habitat use – Compositional Analysis

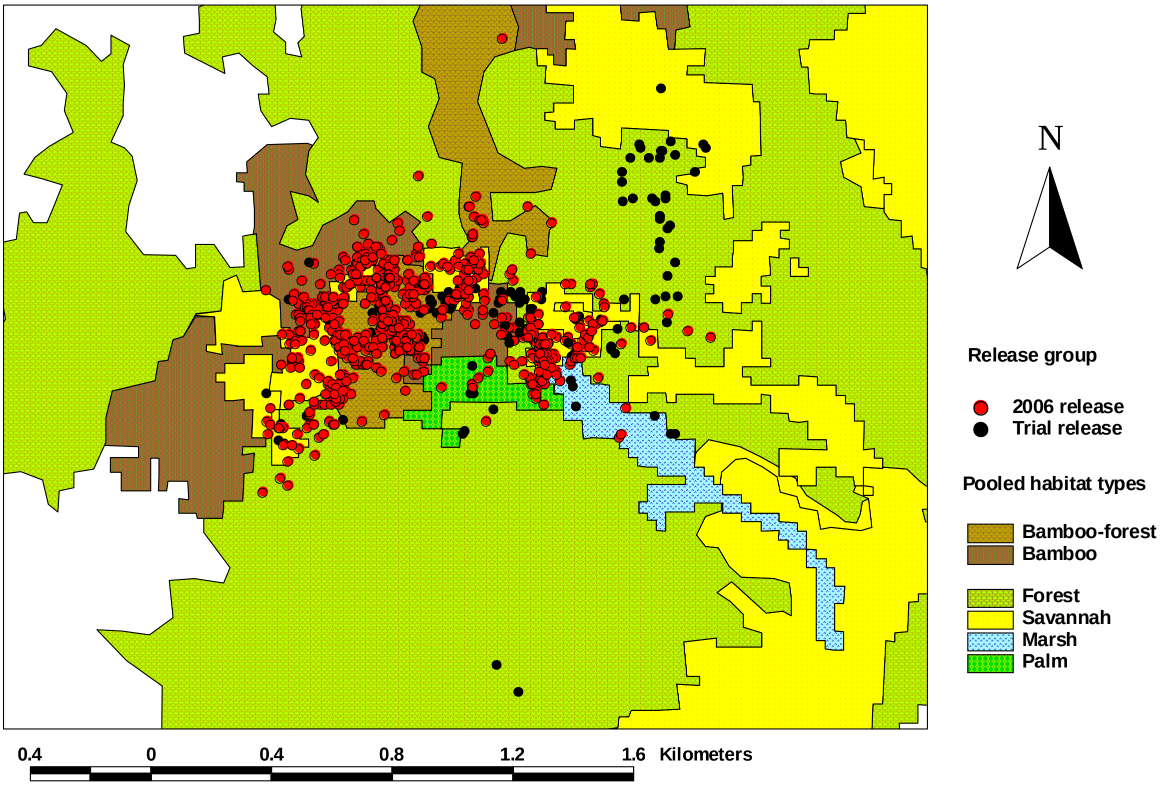

Figure 4.7. Locations of 2006 release and trial release tortoises during 2006 sampling period. M**ap shows pooled habitat types as used in compositional analysis.**

Figure 4.7. shows the location of all tortoises in this study during the 2006 sampling period and how they correspond to the pooled categories of habitat type, as used in the CA. Home range selection (2nd order habitat selection) after 5 months of release in the wild by tortoises released in 2006 was non-random and composition of habitats within home ranges differed from that in the study area (Wilks lambda = 0.228, df 5, p <0.001). Table 4.1 ranks the habitat types in the order: savannah>bamboo>bamboo-forest>>>forest>palm>marsh.

	bamboo	bamboo forest	forest	savannah	palm	marsh	Rank
bamboo		0.343	2.793	-0.823	4.220	4.843	**4**
		+	+++	-	+++	+++	
bamboo forest	-0.343		2.539	-1.003	3.032	3.660	**3**
	-		+++	-	+++	+++	
forest	-2.793	-2.539		-4.805	0.925	1.697	**2**
	---	---		---	+	+	
savannah	0.823	1.003	4.805		5.841	7.593	**5**
	+	+	+++		+++	+++	
palm	-4.220	-3.032	-0.925	-5.841		1.213	**1**
	---	---	-	---		+	
marsh	-4.843	-3.660	-1.697	-7.593	-1.213		**0**
	---	---	-	---	-		

Table 4.1. Matrix of habitat preferences in individual home range (2nd order) selection for 2006 release tortoises. Values are t values. "+++" and "---" indicate statistical significance.

	bamboo	bamboo forest	forest	savannah	palm	marsh	Rank
bamboo		3.258	-1.257	3.722	2.670	4.060	**4**
		+++	-	+++	+	+	
bamboo forest	-3.258		-3.103	1.831	4.325	6.023	**3**
	---		---	+	+++	+++	
forest	1.257	3.103		3.665	4.057	8.081	**5**
	+	+++		+++	+++	+++	
savannah	-3.722	-1.831	-3.665		1.772	8.014	**2**
	---	-	---		+	+++	
palm	-2.670	-4.325	-4.057	-1.772		6.142	**1**
	-	---	---	-		+	
marsh	-4.060	-6.023	-8.081	-8.014	-6.142		**0**
	-	---	---	---	-		

Table 4.2. Matrix of habitat preferences in individual habitat use (3rd order selection) for 2006 release tortoises. Values are mean differences between used and available log-ratios. "+++" and "---" indicate statistical significance.

Individual habitat use (3rd order habitat selection) was also non-random for tortoises released in 2006 after 5 months in the wild (Wilks lambda = 0.01339855, df 5, p 0.05). From Table 4.2, habitats were ranked in the following order:

Forest>bamboo>bamboo-forest>>>savannah>palm>marsh.

5. Discussion

5.1.1. Response of released tortoises- Survival

As with the trial release (Pedrono and Sarovy 2000), all tortoises released in this study survived their first year of release in the wild. There were also no losses during 2007 and 2008. The 0% mortality is highly encouraging and confirms that captive-breading and release is a viable management strategy for the ploughshare tortoise. 0% first year mortality in a project of this kind involving tortoises is unprecedented. A mortality rate of 40% was observed after 17 months of release with reintroduced captive-bred Galapagos giant tortoises, *G. hoodensis* (MacFarland et al 1974) and, in general for RRTs involving tortoises, mortality rates of between 20-30% are considered the norm (see Guyot & Clobert 1997). The tortoises released in this study and by Pedrono and Sarovy (2000) were able to survive the drought conditions characteristic of the region's dry season and independently find drinking water. In contrast, Field et al (2007) reported 21% first year mortality of translocated wild desert tortoises, *G. agassizii*, primarily as a result of drought conditions experienced after release. Also, further to what was observed by Pedrono and Sarovy (2000), this study has shown that captive-reared juvenile Ploughshare tortoises are able to locate and feed on suitable plant species in the wild after 12 – 20 years in captivity. The successful transition to a natural diet can partly be ascribed to the generalist nature of this species (Pedrono & Sarovy 2000). These findings confirm that captive-bred juvenile Ploughshare tortoises are able to survive in the wild and successfully adapt to conditions in their native habitat. The released tortoises also survived in spite of the presence of potential predators such as the African bush pig at the release site. During this study, tortoises were, and still are, also at very high risk of removal by theft or poaching and, although no tortoises involved in this study were lost, four tortoises were stolen from a pre-release enclosure within Beaboaly on 6 May 2009.

5.1.2. Body mass and size

The body mass and size of an individual are important correlates of survival (O'Brian & Tiandray 2005) and, investigating how these change over the course of a reintroduction is a vital part of any ongoing appraisal. It is suggested by O'Brian et al (2005), that the success of a release programme for captive-bred tortoises is likely to depend on the size of juveniles released. The fact that all of the released tortoises survived shows that they were at least at a size where they were not at threat to predation, an important concern in determining the optimum size at which individuals should be considered ready for release (O'Brian & Tiandray

2005, Pedrono & Sarovy 2000). It is, therefore, also reasonable to think that none of the tortoises in this study are at any risk of future predation. The developmental stage of released animals is also an important factor in reintroductions (Germano & Bishop 2008). All of the tortoises released in this study were juvenile/sub-adult juveniles; a phase characterised by rapid growth. Captive bred individuals have been shown to have a higher growth rate than that of their wild counterparts (Pedrono and Sarovy 2000). However, the tortoises released in this study showed a mean decline in body mass in the six months from pre-release captivity to the end of the 2006 sampling period. In some chelonian species, an increase in individual body mass results in increased fitness (Pedrono et al 2001), however, the observed reduction in BM here may be as a result of the animals taking time to adapt to their new environment and is unlikely to be an indication of health problems or reduced fitness. In captivity prior to release, tortoises were fed twice a day and had access to drinking water at all times, even during the dry season (Pedrono & Sarovy 2000). Field et al 2007 found that desert tortoises, *G.agassizii*, experienced reduction in BM with reduced water availability and this may explain the post-release decline in BM in this study. Further, Field et al (2007) observed that subjects also experienced many periods where no growth or shrinking occurred.

BM of adult ploughshare tortoises is known to be affected by seasonality, with weights peaking in mid-winter (July - when tortoises are least active) before declining in the spring, prior to the onset of reproductive activity (Reid et al 1989). The decline in BM of the trial release tortoises during the second and third month of sampling fits with this observation and matches that observed for these same tortoises after their initial release in February 1998 (Pedrono & Sarovy 2000). In Pedrono and Sarovy (2000), the initial decline in mean BM is followed by a marked increase during March and early April. However, as this study was only carried out over a five month period and almost entirely during the wet season, it is not possible to conclude what affects seasonality may have had on BM. It is also not known whether or not a net gain of BM may have been observed after a full year, as was the case with the Pedrono and Sarovy study (2000).

The respective differences and changes in BM between the release groups may partially be explained by the larger sample size of the 2006 release group (n = 20, compared to n =4 for trial release group) as well as the greater range and lower mean age at release (2006 release: mean 13 $\pm$ 2 yrs, range 9 – 18 yrs; trial release: mean 16 $\pm$ 1 yr, range 16 - 17 yrs). BM appears to be highly variable at the juvenile stage of development and the difference between groups is likely due to a number of unknown factors. Again, the observed changes in BM in this study are unlikely to be a reflection of fitness and the mean decline in BM observed in the released tortoises does not indicate decline in health. The large difference in sample size between the groups presents a problem in terms of making comparisons and further highlights a need for caution in drawing too finely detailed conclusions from the observed results.

5.1.3. Home range establishment and post-release movement

As was the case with Pedrono & Sarovy (2000), but unlike in previous translocations of other tortoise species (Tuberville et al 2005, Field et al 2007), all individuals released in this study established home ranges near the release site, and displayed strong site fidelity. That home range area only differed between groups in 2006 is due to the extremely large home range of T3098 (1,298,712.78m^2) rather than any reflection in differences between them. Again, due to the uneven group sizes, the value of any comparison is limited and results should be viewed with caution. Also, the respective home range estimation methods used in this study both have their merits and their drawbacks. Whilst the use of MCPs may result in overestimation of home range size (O'Connor et al 1994, Laver & Kelly 2008), kernels may just as likely exclude areas tortoises may frequently pass through (Duda & Krysik 1999) (see Fig. 4.4). Also, kernels with the LSCV smoothing factor may be inappropriate for studies of sedentary species and can over emphasize the time an individual spends inactive (De Solla 1999). As to which estimation method is the best applied in future analyses is beyond the scope of this study.

Due to uneven sampling (Table 3.2), comparison of home range size between years is also of limited value and, again, should be viewed with caution. By only sampling individuals with greater than 14 observations, the sample size was greatly reduced. The number of observations taken per individual in a sampling regime is a critical consideration in any study of animal populations, especially those using radiotelemetry data, and is particularly influential in terms of home range estimation (Kernohan et al, 2001). In terms of achieving a representative sample, Seaman et al (1999) recommended a minimum of 30 observations per individual taken over a 70-90 day period. Following this, only the 2006 results should be used in comparative studies with future releases. Autocorrelation, which occurs when an animal has too little time between observations to fully exploit its range, is another common issue in home range estimation (Kernohan et al 2001). Animals typically move in a non-random manner and, as a result, autocorrelated data are often unavoidable and may actually provide a more accurate estimate of home range size (De Solla et al 1999). To minimise the negative effects of autocorrelation whilst avoiding loss of biological information, sampling regimes in future studies could be improved by extending the sampling season to include more of the active season.

The movement and settlement of released animals is central to the success of any RRT project (Rittenhouse et al 2007, Griffith et al 1989) and the results observed in this study are highly encouraging. Dispersal, the movement of an individual out of range of effective management, is a major cause of failure in RRT projects for all taxa (Griffith et al 1989, Tuberville et al 2005). Homing, the directed movement back towards where the animal was moved from, is a major driver of dispersal and has, in the past, been reported in ploughshare tortoises that have been kept as pets (Andrianarivo 1977). No homing behaviour was observed in this study and no individuals dispersed out of the area. It is extremely encouraging that directed movement does not occur in released individuals, even after up to 17 years in captivity. The site fidelity displayed by the released tortoises in this study may have been enhanced by their "soft-release" into the wild, as has been found in other tortoise species (Tuberville et al 2005) and other taxa (Grifith et al 1989). That tortoises released in 1998 (Pedrono and Sarovy 2000) also continued to exploit the same area is further evidence of the strong site fidelity shown by released captive-bred ploughshare tortoises.

The long-distance movements recorded in this study were generally short-duration forays that were followed by an immediate return to the area of commencement. Forays of this type were also observed by Pedrono and Sarovy (2000). Overall, forays were rare. Only 3 of the 24 tortoises studied made daily movements of over 400m and no individual made more than one foray in a given year. Whether or not certain individuals are more likely to undertake large forays than others is unclear. Tortoises of both sexes were recorded making large forays and, due to the small sample size and female-biased sex ratio (13:4) no relationship between sex and foray embarkation was detectable. Daily movements of over 1km only occurred twice in 3 years and, on both occasions, the individual immediately returned to the proximity of the release site. It appears that released captive-bred ploughshare tortoises do not exhibit a wide ranging exploratory phase, as has been observed in other tortoise species (Tuberville et al 2005, Field et al 2007). Distance from release site did, however, increase over time in 2006, but this may be explained as an expected increase in habitat exploitation. The movement behaviour of the released tortoises was highly encouraging. However, because daily movement data were standardised (as described in methods), some ecological information is invariably lost. This is especially true where there are a large number of days between successive observations (mean days between observations in 2006: 3 $\pm$ 4 days, range 1 – 26 days). Future sampling strategies should, therefore, avoid this problem by maintaining a consistent interval between observations.

The movement behaviour of released animals may be affected by a number of factors. One possible determinant of post-release movement is the nature and quality of habitat into which the animals have been released and increased movement may occur in areas of more heterogeneous habitat (Longpierre et al 2001, Arvisais et al 2004, Rittenhouse et al 2007), such as is found in Beaboaly. The tortoises released in this study, however, all remained in the area of release, thus suggesting that habitat had no influence on site fidelity. Ploughshare tortoise movement in the wild is also affected by seasonality (Lewis et al 2009). Increased activity occurs during the wet season from October to May and is followed by a period of near total inactivity during the cooler dry months (Juvik et al 1981, Smith et al 1999, Bourou et al 2001). Movement data was collected in this study during the “active” wet season and, as a result, no comparison can be made between seasons. However, released tortoises did indeed become inactive with the onset of the dry season (Richard Young, Pers. Comm. 5 May 2009). Pedrono and Sarovy (2000) also observed that tortoises became inactive during a few days of rainfall.

5.1.4. Habitat selection

Habitat composition and quality has a large effect on post-release home range establishment (Arvisais 2004) and understanding how captive-reared tortoises exploit their habitat is important in assessing their adaptation to their new environment (Dickinson et al 2001). Analysis of habitat use is also critical in assessing the suitability of the release site (Germano & Bishop 2008).

The primary habitat of wild ploughshare tortoises is thought to be deciduous bamboo scrub (Curl et al 1985, Pedrono et al 2001) although they are not confined to this habitat (Juvik et al 1997). Indeed, Ploughshare tortoises have been observed nesting in savannah (Pedrono et al 2001), and the ecological spectrum of this species may be greater than first thought. The results of the CA in this study are largely as one would expect for this species, with bamboo habitats being selected in both 2nd and 3rd order habitat selection. The fact that savannah is found to be the most selected habitat in terms of home range establishment, however, is perhaps surprising. Savannah habitats make up 16% of the total habitat area of Beabaoly, compared to a combined 30% for bamboo and bamboo-forest and over 40% forest. However, a band of savannah habitat runs east to west through the middle of the area into which the tortoises were actually released (see Fig 4.6), and this may explain why it occurred so prominently within individual home ranges compared to the wider study area.

That forest is the most selected habitat type in 3rd order selection is surprising considering that it does not appear to occur prominently in the area of release (Fig. 4.6). 3rd order habitat selection is of the most interest in terms of this study and the key finding from the analysis is that released tortoises clearly select bamboo and forest habitats in their fine-scale habitat use. This confirms that these habitats appear to be of ecological importance to this species, and future release sites should be selected that contain as large an area of these habitat types as possible. Conservation efforts should, therefore, also continue to focus on preserving these habitat types.

The high degree of selection for forest habitat types in this analysis may be explained as a reflection of fine-scale discrepancies between observations made on the ground and the overlayed habitat map used in GIS. The map onto which the individual home ranges were projected and the habitat availability calculated is an imperfect reflection of the mosaic of habitat types that actually occur in the release site. Also, CA requires that habitat types are pooled into a small number of categories (5 – 6 Aebischer et al 1993) and, as a result, information is inevitably lost. The accuracy of future analyses could be improved with the development of a finer resolution habitat map. Despite this drawback, the CA carried out this study can be considered robust and the key findings useful.

5.2. Evaluating reintroduction success

It is a failing of many previous RRT projects involving long-lived species that success has been claimed on the basis of a monitoring period that only encompasses a tiny fraction of the released animal's life history (Dodd & Seigel 1991). This reintroduction of Ploughshare tortoises is still very much in its infancy and, as such, it is impossible to claim "success" at this stage. At this moment in time it is only possible to assess short-term success, and even such an assessment should only really be interpreted as a "status update". Currently, therefore, the outcome of this project can only be defined as "uncertain" (Griffith et al 89, Dodd & Seigel 91, Germano & Bishop 08). It will only be known for certain whether or not this strategy has been a success, however, if and when the released individuals survive into adulthood, continue to remain within the area of release, mate successfully and produce offspring that themselves go on to reproduce. Obviously, it will not be possible to ascertain whether or not this will be the case for several decades and, as a result, effective short-term evaluation is crucial. However, the analysis of the post-release responses of captive-bred ploughshare tortoises carried out in this study have made it possible for an assessment of the short-term success of this reintroduction to be made. All the released tortoises survived and successfully adapted to their natural environment and remained in the area of release. These findings confirm the short-term success of this reintroduction.

5.3. Management implications of research

The reintroduction of captive-bred juveniles has been shown to be a promising tool for ploughshare tortoise conservation. Based on the success of the released individuals in this study, future reintroductions should continue to use individuals within the same age and size range. Results also suggest that Beaboaly has been a good choice as a release site. The tortoises all remained well within its boundaries and were able to successfully exploit available resources. The compositional analysis carried out in this study confirms that bamboo scrub and forest habitat types are of ecological importance to ploughshare tortoises and suitably large areas of both of these habitat types are available at the release site. This site should continue to be used in future releases; however, the carrying capacity is unknown, and should be investigated. Also, the bamboo habitat types are concentrated in the north-west portion of the region (Fig.4.6) and whether or not releases carried out in different areas of Beaboaly would be as successful is unclear.

Based on the initial success of this reintroduction, it is critical that a commitment be made to the ongoing monitoring of this project and that assessments are frequently updated and made available to the public, whether they be positive or negative. The reintroduction of ploughshare tortoises to the wild should be seen as a dynamic process; one that is able to respond and adapt to feedback generated through detailed studies carried out as part of an efficient monitoring strategy over both the short and long-term. The ploughshare tortoise has a generation time of around 30 years (Pedrono et al 2004) and, as a result, monitoring should be carried out for at least another 40-50 years. It will only be after this amount of time that the long-term viability of the released population can begin to be seen. In this time, the first generation of released individuals will have hopefully reproduced and allow for recruitment to be assessed. Of course, an intensive programme of continuous monitoring for this amount of time would be impractical and extremely costly and, therefore, a sensible time-frame for monitoring should be developed. Given the survival and settlement observed in this study, it would seem unnecessary to continue carrying out yearly investigation over the long-term. Currently, future ploughshare tortoise releases will consist of the yearly release of 20 individuals (Richard Young, Pers. Comm. 22 August 2009). However, due to the small number of individuals so far released and their extremely high value, it is preferable that all tortoises continue to be tagged and monitored for at least the next release. As the released population gets larger after future reintroductions, it will be possible to reduce the number of tagged individuals and still obtain a representative sample. The number of individuals needed to obtain such a sample in future studies may be calculated by using bootstrapping analysis techniques. Lowering the number of tagged individuals will reduce expenditure and use of resources, a critical consideration in terms of the future viability of the project.

As the reintroduction programme progresses, a number of different response variables will need to be monitored. When the current group of released tortoises reach maturity, it will be important to monitor intra-species interactions, specifically those related to mating. Nest establishment, egg production and fertility, hatching success and hatchling survival are all important in terms of monitoring and assessing success. Monitoring for disease should also be ongoing and, regular counts of individuals should be carried out to check if individuals have been lost or stolen. By carrying out monitoring to include the above, it will be possible to make assessments of medium-term success and will allow for it to be seen if any additional management or intervention is necessary.

5.4. Future research

In the completion of this study, a number of interesting questions have arisen that should be addressed in the future. Firstly, a study into the carrying capacity of Beaboaly would be extremely useful in terms of determining the optimum number of individuals that should be released into this area. Although Beaboaly has a high proportion of bamboo habitat types compared with other regions in Baly Bay National Park, they only occur within a relatively small area. As a result, it may be necessary to identify other alternative areas for future releases, depending on results. A more detailed study of habitat use would also be useful, especially if tortoises are released into area of the release site where availability of bamboo habitat is low (i.e. in the south of Beaboaly, Fig. 3.1). Compositional analysis could again be used but a finer resolution habitat map would be beneficial. Population viability analyses (PVA) should also be carried out as part of future studies and would be highly useful in determining the number of individuals that should be released in order to obtain a stable population. PVA will aid prediction of long-term outcomes and can help guide evolution of management and release strategy.

It would also be interesting to investigate the effects of age and sex on post release movement, home range establishment and habitat use. In this study, age and sex effects were unclear and the heavily female-biased sex ratio rendered any such investigation in this study moot. The influence of sex and age is likely, however, to become more of an issue worthy of investigation as the released tortoises reach sexual maturity and behaviour and habitat requirements may alter. Investigating the factors that drive some individuals to make rare short-duration, long-distance forays would also be of interest. What triggers the foray? How long do they last? Is movement in a straight line and continuous or is it non-direct and periodical? What is their behaviour during these forays? Do they seek out and utilise specific resources? Are certain individuals, sexes, and age groups more likely to carry out forays than others? These are all interesting questions that could provide vitally important information regarding this extremely rare and scientifically under-studied species.

In terms of wider studies involving the investigation of tortoise home range, an in-depth evaluation of home range estimation methods would be greatly beneficial. No such review for tortoises has yet been carried out and gathering inferences from broad reviews and studies involving wider-ranging animals is difficult and of questionable value. MCP and kernel methods, whilst by far the most popular, are by no means the only options available and a thorough investigation of the suitability of the various methods available would be an invaluable contribution to the fields of tortoise ecology and conservation management.

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Appendix

Tortoise ID	Release Group	Pre-Release BM (kg)	BM (kg) at End of 2006 Study Peiod	Mean % Successive Change in BM	% Change in BM, Pre-Release to End of 2006 Study
T3020	2006	3.50	3.50	0.24	0.00
T3095	2006	6.75	6.25	-1.30	-7.41
T3096	2006	5.75	5.40	-0.82	-6.09
T3097	1998	7.00	7.00	0.51	0.00
T3098	1998	6.75	8.50	5.21	25.93
T3101	1998	7.00	7.50	2.15	7.14
T3102	2006	5.50	4.50	-3.62	-18.18
T3104	1998	7.00	6.80	-0.19	-2.86
T3106	2006	4.25	3.80	-1.56	-10.59
T3112	2006	4.75	5.40	3.00	13.68
T3117	2006	5.00	4.30	-2.88	-14.00
T3119	2006	4.50	5.00	2.21	11.11
T3123	2006	3.75	4.00	1.62	6.67
T3124	2006	4.50	4.10	-1.57	-8.89
T3125	2006	4.50	4.00	-1.74	-11.11
T3128	2006	4.50	3.75	-3.05	-16.67
T3130	2006	4.50	3.10	-5.68	-31.11
T3134	2006	4.00	4.00	0.01	0.00
T3146	2006	4.50	4.25	-0.16	-5.56
T3160	2006	3.50	3.80	1.98	8.57
T3196	2006	2.50	2.60	1.56	4.00
T3207	2006	2.25	2.40	1.42	6.67
T3332	2006	4.00	4.00	0.14	0.00
T3409	2006	4.00	4.00	0.37	0.00

Table A.1. Body mass of 2006 release tortoises during pre-release captivity and at end of 2006 sampling period. Percentage change in body mass is also shown. The same data is also listed for trial release tortoises over the same period.

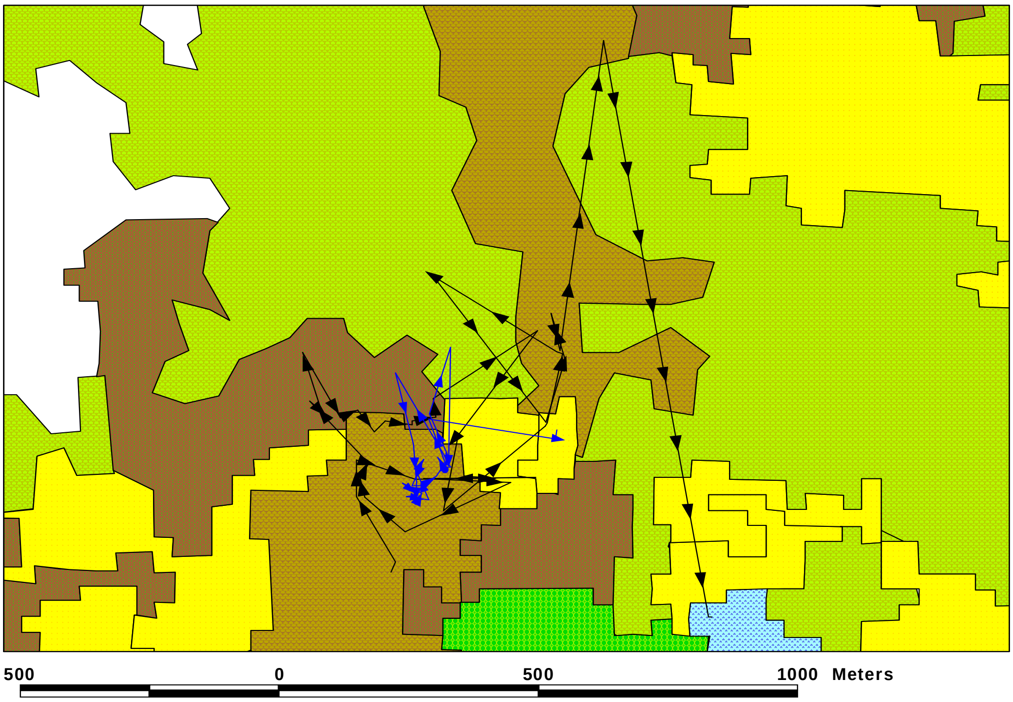

Figure A.1. 2006 movement polylines for ploughshare tortoises T3134 (Blue) and T3146 (Black) released in 2006. The polyline shows straightline distances between observations from release on 22 January 2006 until end of sampling on 23/24 May 2006. T3134 remained nearest to release site of all 2006 release tortoises, whilst T3146 moved the farthest from release site.