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**PRE-TRANSLOCATION HEALTH ASSESSMENT AND CONSERVATION
PRIORITIES FOR *CONOLOPHUS* IGUANAS IN THE GALAPAGOS ISLANDS,
ECUADOR**

A thesis presented in partial fulfilment of the requirements for the degree of

Master of Science
in
Conservation Biology

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Figure 1. . Members of the genus *Conolophus*. From top: *C. pallidus*; *C. marthae* and *C. subcristatus*. Photos: L. Ortiz-Catedral

DECLARATION

I declare that this thesis is an accurate and original account of my research and that the contents have not previously been submitted for a degree at Massey University, or any other tertiary institution. Except where acknowledged within, the material contained in this thesis has not been written or published by any other individual and to the extent of my knowledge, does not infringe upon copyright restrictions. The research presented here is part of a broader research program on *Conolophus* iguanas developed since 2012 by my supervisor Dr. Luis Ortiz-Catedral in collaboration with staff from the Directorate of the Galapagos National Park, Island Conservation, Ministry of Environment and Ministry of Agriculture, Ecuador. Dr. Ortiz-Catedral allowed me to complete the research presented here following approved activities by the Ministry of Environment, Ecuador via the Directorate of the Galapagos National Park under permit PC-96-18 (Appendix I). Due to the ongoing COVID-19 pandemic, I was unable to complete fieldwork for this thesis. Instead, I worked on analyses of datasets collected by staff of Island Conservation and the Directorate of the Galapagos National Park. Financial support for field activities was provided by the Galapagos Conservation Trust, Mohamed bin Zayed Species Conservation Fund and Island Conservation. Dr. Ortiz-Catedral provided all the supervision necessary for the development of this project. I assume all responsibility for omissions or mistakes present in this document.

Colin Heng

Auckland, New Zealand, February 2021

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Figure 2. Park ranger Johannes Ramirez examines a male Pink Iguana (*Conolophus marthae*) during the February 2019 expedition to Wolf volcano. Photo: L. Ortiz-Catedral.

GENERAL ABSTRACT

The conservation of biological diversity worldwide requires an understanding of the biology of species and ecosystems to protect, as well as information sharing between scientists and managers. However, it is common to encounter delays between the production of research results and the refinement of conservation plans based on these findings. The Galapagos terrestrial iguanas (*Conolophus* spp) are a group of three endemic species in the Galapagos Archipelago, Ecuador. In the last 90 years there have been numerous actions with the goal to preserve these species, significantly the reintroduction of Galapagos land iguanas (*Conolophus subcristatus*) to various islands in the archipelago. However, important aspects of the biology of Galapagos terrestrial iguanas remain undocumented. In this thesis I present the first analysis of pre-translocation effects of ectoparasites on the body condition of Galapagos land iguanas on the island of Seymour Norte. This analysis is however incomplete due to significant logistical challenges imposed by the global 2019 COVID-19 pandemic. I also present information on the body condition and approximate density of the elusive Pink iguana (*Conolophus marthae*) on Wolf Volcano, and a preliminary description of the status of Galapagos land iguanas reintroduced to Santiago Island. Once again, the major logistical difficulties imposed by the ongoing COVID-19 global pandemic mean that the results presented in this thesis are only preliminary as there no possibility to complete data collection in the field at the time of submission of this thesis. Nevertheless, I hope that some of the information contained in this thesis can assist the Directorate of the Galapagos National Park in its mission to preserve all the known populations and species of Galapagos terrestrial iguanas.

RESUMEN

La conservación de la diversidad biológica en todo el mundo requiere una comprensión de la biología de las especies y los ecosistemas a proteger, así como el intercambio de información entre científicos y conservacionistas. Sin embargo, es común encontrar retrasos entre la producción de los resultados de la investigación y el refinamiento de los planes de conservación basados en estos hallazgos. Las iguanas terrestres de Galápagos (*Conolophus* spp) son un grupo de tres especies endémicas del Archipiélago de Galápagos, Ecuador. En los últimos 90 años se han realizado numerosas acciones con el objetivo de preservar estas especies, destacando la reintroducción de iguanas terrestres de Galápagos (*Conolophus subcristatus*) a varias islas del archipiélago. Sin embargo, aspectos importantes de la biología de las iguanas terrestres de Galápagos siguen sin documentarse. En esta tesis presento el primer análisis de los efectos de pre-translocación de ectoparásitos en la condición corporal de las iguanas terrestres de Galápagos en la isla Seymour Norte. Sin embargo, este análisis está incompleto debido a los importantes desafíos logísticos impuestos por la pandemia mundial de COVID-19. También presento información sobre la condición corporal y densidad aproximada de la esquiua iguana rosada (*Conolophus marthae*) en el Volcán Wolf, y un análisis preliminar de las iguanas terrestres de Galápagos reintroducidas en la Isla Santiago. Una vez más, las grandes dificultades logísticas impuestas por la pandemia global de COVID-19 en curso hacen que los resultados presentados en esta tesis sean solo preliminares ya que no existe la posibilidad de completar la recolección de datos en el campo al momento de la presentación de esta tesis. Sin embargo, espero que parte de la información contenida en esta tesis pueda ayudar a la Dirección del Parque Nacional Galápagos en su misión de preservar todas las poblaciones y especies conocidas de iguanas terrestres de Galápagos.



Figure 3. Release of land iguanas (*Conolophus subcristatus*) on Santiago Island. Photo: L. Ortiz-Catedral, 2019.

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CHAPTER 1: GENERAL INTRODUCTION



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ABSTRACT

Biodiversity loss is a global issue. There are many drivers of biodiversity loss but some of the most important ones include: habitat loss and invasive species. More importantly, species found on islands are more susceptible to decline from said drivers due to the water body surrounding them. Despite this, conservationists tend to utilize oceanic islands for translocations to prevent population decline and extinction. Many species of iguana (Squamata: Iguanidae) inhabit oceanic islands and some of them represent the most threatened reptiles in the world. The Galapagos land iguana (*Conolophus subcristatus*) is an excellent example to showcase the effects invasive species can have on iguana populations. Furthermore, the translocation of the land iguanas within the Galapagos archipelago has been ongoing since the 1930s. Moreover, in the recent years, iguanid species have been studied and observed to act as seed dispersers in their local habitat ranges and the Galapagos land iguana is one of them. In this chapter, I outline the threats to iguanas, the use of translocation as a conservation tool to aid in the recovery of iguanid species globally and present a general description of this thesis.

Biodiversity loss

Biodiversity loss, from the genetic level (i.e. the loss of haplotypes, reduction in heterozygosity etc.) to the loss of ecosystem functions is a widespread global phenomenon that requires urgent action to prevent the extinction of species, the disruption of ecological interactions and the collapse of ecosystem services to humanity. Perhaps the most well-known example of biodiversity loss is the extinction of species. Although extinction is a natural process (Olson, 1989), in the last few hundred years, human activities have accelerated the rate of extinction across taxonomic groups to unprecedented rates. Ceballos et al. (2015) conducted an analysis that revealed the number of species gone extinct in the past century, among various vertebrate taxa, would have taken at least 800 to 10,000 years to disappear without human interference. Instead, extinctions for birds, reptiles, and mammals since the 1800s have occurred at a faster rate. Global biodiversity is constantly affected by human interference, interspecific interactions (such as predation, parasitism and competition), environmental changes and chance events (Kotze & O'hara, 2003). It is important to note that anthropogenic threats work synergistically with one another; for example, habitat loss can cause decline in species directly and it can also affect other species indirectly, with eliminating prey and improving hunter access (Brook et al., 2008). Among vertebrates, 25% of all mammal species and 13% of all bird species are at risk of extinction, representing nearly 3000 species (Tilman et al., 2017). Habitat fragmentation and the introduction of invasive species represent two of the most severe examples of threatening factors to biodiversity, predominantly for island ecosystems. For instance, due to the severe modifications in native habitats, 10% of historically known native plants found in Hawaii have gone extinct, with another 40% threatened (Paulay, 1994). Furthermore, native species of flightless birds found in New Zealand declined significantly following the introduction of invasive mammal species due to the arrival of Polynesian and European settlements in the 1800s (Bellingham et al., 2010).

The economic prosperity and well-being of human societies is tightly linked to biodiversity: medicines, food, timber etc. are examples of biodiversity resources used by humans (Díaz et al., 2006). Ecologists have indicated that the decrease in biodiversity will lead to reductions in ecosystem functioning; consequently, leading to the decline in provisioning of the services and resources indicated above (Dobson et al., 2006). In addition, a study conducted by Worm et al. (2006), suggested that marine biodiversity loss is increasingly impairing the oceans abilities to provide food and maintain water quality to human societies. Hence, the need to consider a species role in ecosystem functions during conservation management is crucial. With faster rates of species extinctions and a growing number of endangered species, conservationists are required to act fast and efficiently in the race to preserve biodiversity. However, historically there have been substantial gaps between the results of conservation scientists and the practical application of their findings. Bridging the gap between conservation scientists and wildlife managers, conservation biologists and government agencies remains a major challenge in the field (Habel, et al., 2013).

Drivers of biodiversity loss

To date, the diversity of life on Earth has been severely impacted by humans. For example, within half a century, the range of tropical forests globally has shrunk by approximately nine million km² (Pimm et al., 2001) as a result of the rapid expansion of agricultural practices, construction of residential housings and timber usage and many more (Derouin, 2019; Pimm et al., 2001). Freshwater and marine animal species are also threatened in areas such as Mesa Central (Mexico) and central Asia due to accessible rivers being dammed and channeled. In addition, water for irrigation poses a threat to the ecosystem as salt accumulation rapidly destroys soil fertility (Mabbutt, 1984). Although extinctions across

taxonomic groups are occurring worldwide on terrestrial and marine environments (Díaz et al., 2019), the drivers and scale of biodiversity loss varies between regions. Consequently, local solutions to biodiversity loss require an understanding of the scale and tempo of threatening processes.

Habitat loss

A major contributor to global species decline is the loss of habitat. According to the International Union for Conservation of Nature (IUCN) red list, habitat loss affects 85% of all species classified as ‘Threatened’ and ‘Endangered’ (IUCN, 2015). The increase in human developments, has led to the increase in deforestations. A product of habitat lost is habitat fragmentation. For some species, this results in isolated ageing populations, where potential mates are limited, resulting in population decline (Conner & Rudolph, 1991). According to Todd et al. (2010), the decline in reptilian abundance is evident after the clearing or conversion of forests as habitat loss indirectly affects reptiles by limiting their ability to meet ecological needs for survival and reproduction. Similarly, amphibian species richness in the wetlands of southwestern Minnesota showed a greater decline as a function greater wetland isolation and road densities (Lehtinen et al., 1999).

Invasive species

The introduction of invasive species is currently recognized as one of the major threats to global diversity (Park, 2004). Apart from habitat loss, recent expansion of human

populations and the increase in global transportation and trade, have resulted in the introduction and establishment of many species outside of their native geographical range (Todd et al., 2010). Many of which have grown and established rapidly, causing severe ecological and economic damages (Pimentel et al., 2000).

The effects of invasive species have been widely documented on archipelagoes around the world. For example, prior to the arrival of humans to New Zealand in the 13th century AD (Wilmschurstl & Higham, 2004), numerous species of flightless birds existed (Clout & Lowe, 2000). The introduction of mammals following Polynesian and European settlements had cause severe declines in native bird species. Combined with habitat loss and hunting, the effects of Polynesian rats or “kioore” (*Rattus exulans*), black rats (*Rattus rattus*), cats (*Felis catus*) and stoats (*Mustela erminea*) as predators of endemic birds are considered the main drivers in the decline and extinction of up to 27 species of endemic New Zealand birds (Bellingham et al., 2010). Other introduced mammals such as goats (*Capra hircus*), brush-tailed possums (*Trichosorus vulpecula*) and red deer (*Cervus elaphus*) have caused severe damage to native plant communities through selective grazing (Clout & Lowe, 2000).

Another example of the effects of introduced species on islands, can be found in the Galapagos archipelago. For instance, large populations of the native giant tortoises (*Chelonoidis nigra*) and the Galapagos land iguana (*Conolophus subcristatus*) were historically found on Santiago island (Cruz et al., 2005). However, the introduction of mammalian species in the 17th century severely disrupted native animal communities; goats (*Capra hircus*), pigs (*Sus scrofa*.) and donkeys (*Equus asinus*) grazed on native vegetations, resulting in habitat destruction and competition for resources., while rats (*Rattus rattus*), cats (*Felis catus*) and dogs (*Canis lupus familiaris*) predated on juvenile iguanas and tortoises (Carrion et al., 2007; Cruz et al., 2005; Fabiani et al., 2011).

Data deficiency

The use of the IUCN red list as a system to classify species based on their extinction risk is almost universal. However, when data availability is inadequate, species are classified as data deficient (Morais et al., 2013). Recent studies have reported a decline in data deficient species due to anthropogenic and natural threats, which stresses the fact that some of these species, in reality, might actually be threatened (Eterovick et al., 2005; Lips et al., 2005; Morais et al., 2013; Pounds et al., 2006; Verdade et al., 2012).

Species classified as critically endangered, endangered and vulnerable are often prioritized for conservation actions. However, data deficient species should also be part of this priority as they contribute to uncertainties in estimates for extinction (Bland & Böhm, 2016). To date, governmental and international agencies consider extinction risk of species an important parameter when allocating financial resources for conservation. For example, species classified as data deficient are often excluded from national planning and investment schemes, only two percent of awards from the Mohamed bin Zayed species conservation fund (MBZ) and the World Association Zoos and Aquaria exclusively focus on data deficient species (Bland & Böhm, 2016).

Thus, while the classification of a species as “data deficient” per se, is not a threatening factor, lack of information to properly assess species mean that a number of extinctions are occurring “under the radar” of wildlife managers. It is therefore important not only to gather basic biological information on data deficient species, but also to accelerate data sharing between relevant stakeholders, to properly assess the threat status of as many species as possible.

The biodiversity found on oceanic islands is characterized by unique biogeographic, phylogenetic and functional characteristics (Russell & Kueffer, 2019). Islands frequently have unusually high endemism as compared to mainland ranges due to low species immigration; making them hotspots for biodiversity (Goldman et al., 2008). An analysis conducted by Mittermeier et al. (2011) revealed 34 biodiversity hotspots around the world. These regions collectively contain more than 50% of vascular plants and 42% of terrestrial vertebrates as endemics. However, due to rapid habitat loss in those regions, they are concentrated to 2.3% (3.4 million km²) of the world's land area. Island ecosystems have long been model cases for the study of extinctions and have thus significantly contributed to the development of the science in conservation biology (Nogué et al., 2017). For example, the role of vicariance and dispersal as biogeographical avenues for speciation, the relationship between area and species richness, and the evolution of convergent morphological traits under similar ecological conditions (Maunder et al., 2008).

The biodiversity of islands is highly susceptible to extinction. Approximately two-thirds of recent extinctions consist of island species (Jones et al., 2016). The large amount of water surrounding islands, is an indisputable factor that contributes to their greater vulnerability. For example, if there were a natural disaster (i.e. tsunami), species on the island without the ability of flight would perish. Further, many island species have evolved in the absence of human presence and introduced species, which makes them vulnerable to human activities (Nogué et al., 2017). For example, the endemic avifauna of New Zealand have evolved to be flightless and lack the behavioral defense mechanisms against invasive species (Goldman et al., 2008; Medina et al., 2011; Wikelski et al., 2004). Bird species such as the Moa (*Dinornithiformes* spp) and Takahē (*Porphyrio hochstetteri*) are just two examples of the many endemic avifauna species that have suffered. To date, competition and predation by

mammalian species remain a major threat to New Zealand's flightless birds (Clout & Craig, 1995).

Two main threats to island biodiversity are deforestation and invasive species (Thaman, 2002). Feral cats have profound impacts on island ecosystems. In a global review, feral cats were considered responsible for at least 14% of global bird, mammal and reptile extinctions. In addition, this invasive species is considered a threat to approximately 8% of critically endangered birds, mammals and reptiles (Medina et al., 2011). More importantly, invasive pathogens and their diseases have often been overlooked by conservation biologists. For example, the Hawaiian Iiwi (*Vestiaria coccinea*), were found to be susceptible to avian malaria. Infected Iiwis showed significant declines in food consumption which resulted in weight loss increasing mortality risks (Atkinson et al., 1995). Other populations of endemic birds in Hawaii have been severely affected due to the effects of exotic diseases and pathogens (Wikelski et al., 2004). In another case study, there was an ongoing debate on the extinction of the Christmas island rat (*Rattus macleari*). Some studies hypothesized that a pathogenic trypanosome carried by fleas (*Siphonaptera* spp) were responsible for the demise of the Christmas island rat, while others suggested hybridization between black rats (*Rattus rattus*) (Pickering & Norris, 1996). However, Wyatt et al. (2008), confirmed that the cause of extinction was due to the pathogen as there was no evidence of hybridization.

Conservation of island biodiversity

Ecological restoration

The practice of ecological restoration and the science of restoration ecology have experienced a rapid recent development and application. Restoration ecology provides a theoretical framework to identify and implement actions that can restore native or indigenous

ecological interactions which can help address current conservation problems (Young, 2000). Furthermore, ecological restoration practices often require help from ecological history; as the systematic monitoring of ecosystems rarely last more than a few decades (Jackson & Hobbs, 2009). This situation leaves restoration ecologists to utilize indirect means, such as fossils and old documentations to obtain ecological history. For example, based on skeletal remains (Cruz et al., 2005) and old document records by Charles Darwin during his voyage (Darwin, 1959), the island of Santiago was once filled with a high abundance of Galapagos land iguana (*Conolophus subcristatus*) populations; however, due to anthropogenic threats and the introduction of invasive species, they were brought to extinction till the recent translocation conducted in 2019 (Kumar, 2019).

Ecological restoration has often been misconstrued as “repairing” what can be repaired, and thus plays a secondary role to conservation biology (Young, 2000). Nonetheless, as threats to endangered species accelerate, it is imperative that none should be the latter but instead applied synergistically.

Translocation

Translocation is a popular method adopted by conservation biologists to aid in the deceleration of species decline. It is known as the deliberate movement and release of wildlife (Miskelly & Powlesland, 2013). Previously, translocation was widely used as a species conservation tool to establish populations of non-native species, and to restore native species extirpated by hunting (Griffith et al., 1989). In addition, it was also used to remove problem species to allow regeneration in the environment (Linnell et al., 1997). For example, the translocation of black bears (*Ursus americanus*) in British Columbia was conducted in response to predation on livestock and nuisance to humans (Linnell et al., 1997). However, due

to the rapid increase species extinction rates, the translocation of rare species has now become an important conservation technique (Griffith et al., 1989).

Successful translocations of rare species are a staple of New Zealand conservation. Since 1960, 68 taxa (55 species) of New Zealand birds have been translocated in over 1,100 separate releases; with new populations of 50 taxa (41 species) successfully established (Miskelly & Powlesland, 2013). For example, the translocation of the North Island saddleback (*Philesturnus rufusater*) conducted in 2008, showed high survival rates with a minimum of 11 juvenile birds observed during the breeding season (Parker & Laurence., 2008). Additionally, results from the translocation of rare lizards in New Zealand also showed stable and self-sustaining populations (Towns & Ferreira, 2001). Despite many positive results, global translocation success is considered low (Dodd Jr & Seigel, 1991). The criteria accounting for translocation success varies depending on the author. Translocated species showing population growth, evidence of reproduction, stable and self-sustaining populations are all considered successes (Goudarzi et al., 2015; Robert et al., 2015; Towns & Ferreira, 2001). More importantly, post translocation monitoring is key to assessing translocation success (Dodd Jr & Seigel, 1991). Post translocation monitoring is often hindered by the amount of funds available for the project. Moreover, they generally range from one month to five years (Tarszisz et al., 2014).

The synergy of threats towards species decline is globally evident (Brook et al., 2008). Hence, conservation biologists should try encompassing methods that can fulfil two needs with one deed. For example, the translocation of endangered keystone species, such as the Galapagos land iguanas (*Conolophus subcristatus*) can not only preserve them but shape and alter ecosystem functions; as they act as seed dispersers (Traveset et al., 2016).

Island translocation

The translocation of endangered species to oceanic islands have been a popular method used in conservation biology to date (Komdeur, 1994; Ostendorf et al., 2016; Parker & Laurence, 2008) despite their high susceptibility to decline (Mills et al., 2004). They are used to remove vulnerable species from threatening processes or as a source of animal reintroduction (Ostendorf et al., 2016). The inaccessibility from invasive species and the often pristine nature from anthropogenic threats allow oceanic islands to be more ideal for translocation as compared to mainland ranges (Romijn & Hartley, 2016). In addition, the eradication of invasive species on oceanic islands tends to be more viable due to the water body surrounding them as this allows for easier maintenance, and that in some cases, some of them are uninhabited by humans (Moro, 2003).

New Zealand was amongst the first few countries to practice island translocations to conserve their mainland native bird species in a sanctuary setting; with translocated populations of 41 species successfully (Towns et al., 2016). Some successful examples include the North Island saddleback (*Philesturnus rufusater*), which showed positive long-term success with breeding of new juveniles on Moutihe Island (Parker & Laurence, 2008). Australia too presented successful results with island translocations of the southern hairy-nosed wombat (*Lasiorninus latifrons*), black-footed rock-wallaby (*Petrogale lateralis pearsonii*) and the brush-tailed bettong (*Bettongia penicillate*) on Wedge Island. All three species have been observed to show substantial population increase (Ostendorf et al., 2016). To date, hundreds of invasive species eradications and endangered species translocation have been successfully completed globally (Couchamp et al., 2014).

Invasive species management

The introduction of invasive species plays a major role in global biodiversity decline (Bellingham et al., 2010; Todd et al., 2010; Park, 2004; Pimentel et al., 2000). Competition for resources, habitat destruction and depredation are just a few examples of their impacts. Hence, the removal of invasive species is paramount in the conservation of endemic and native populations.

The combination of the eradication of invasive species and island translocations of native fauna to islands, are powerful tools in conservation biology. New Zealand is the first country to have developed methods to manage invasive species that has been well established for over four decades (Howald et al., 2007). Methods used to control invasive species usually include trapping or using ground based or aerial based poisoning; such as sodium monofluoroacetate (1080), brodifacoum and cyanide. Many studies report an increase of native and endemic species populations after the eradication of invasive ones (Atkinson & Cameron, 1993; Carrion et al., 2007; Phillips et al., 2005). For example, the three-year program to eradicate feral cats on the island of Baltra using sodium monofluoroacetate (1080), followed by trapping and shooting, has allowed the recovery of the resident land iguanas (Phillips et al., 2005). Further, numerous captive-bred land iguanas have been released to Baltra to boost the local numbers (Kumar, 2019). Although increase in reproduction numbers could be due to iguanas reaching higher fecundity and sexual maturity, the decrease in feral cat populations does play a crucial factor, due to the lack of predation. Similarly, Turks and Caicos rock iguanas found in West India showed positive results with successful establishment and evident breeding populations after the eradication of feral cats (Mitchell et al., 2002).

Post translocation monitoring

The post-translocation monitoring of populations is a crucial process to implement as it not only allows the improvement of wildlife management and conservation actions but identifies the causes of both translocation successes and failures (Pinter-Wollman et al., 2009; Van Winkel, 2008). Unfortunately, in some studies, the process of post translocation monitoring is lacking or has been conducted inaccurately (Dodd Jr & Seigel, 1991). For example, south England has conducted many translocation projects; however, specific details on populations, such as founder populations health or evidence of breeding were absent in their literature. More specifically, the sand lizard (*Lacerta agilis*) population that was reintroduced in 1981 to their original location after a severe fire occurred in the nature reserve had no follow up samplings and information on population numbers.

Both short- and long-term strategies are equally viable options for conservation biologists to adopt. Long-term monitoring can ensure accurate evaluation of the species establishment. In addition, it can also ensure the maintenance of biodiversity integrity at release sites (Van Winkel, 2008). However, this requires a substantial amount of resources which may not be deemed feasible. The duration of long-term monitoring is highly influenced by the target species life history traits, such as sexual maturity, recruitment rate and their reproductive lifespan (Dodd Jr & Seigel, 1991). For example, a study conducted by Burke (1991) suggested translocation success on gopher tortoises (*Gopherus polyphemus*) after two years of monitoring or only 10% of the time it takes for tortoises to reach sexual maturity.; which, according to Dodd Jr and Seigel (1991), is not enough time to constitute translocation success.

Short-term monitoring, on the other hand, is much more feasible. Indicators such as founder recaptures and evidence of island born young have often been used to determine translocation success. For example, results from a study conducted by Knapp (2001), suggested

translocation success of the Allen Cay's iguana (*Cyclura cychlura inornata*) as 88% of the original population were recaptured during the post translocation monitoring phase.

Reptilian diversity and threats

Reptiles are one of the most remarkable groups of living organisms. They have successfully colonized most of the planet and inhabits all major land masses and some marine waters; except Antarctica (Gibbons & Luhring, 2009; Pincheira-Donoso et al., 2013). Reptiles have been around for approximately 320 million years, since the late Carboniferous period, and they have evolved ever since (Laurin & Reisz, 1995). There are currently six orders under the class Reptiles: *Amphisbaenia* (worm lizards), *Crocodylia* (Crocodilians), *Rhynchocephalia* (Tuatara), *Sauria* (Lizards), *Serpentes* (Snakes) and *Testudines* (Turtles). The class Reptiles can further be divided into two clades; non-squamate and squamate. Non-squamate reptiles (turtles, crocodilians and tuataras), consists of approximately 350 species globally while Squamata (lizards, snakes, worm lizards) consists of more than 9,100 species (Pincheira-Donoso et al., 2013). The figure below shows the conservation status of reptiles found globally and also the number of species within it.

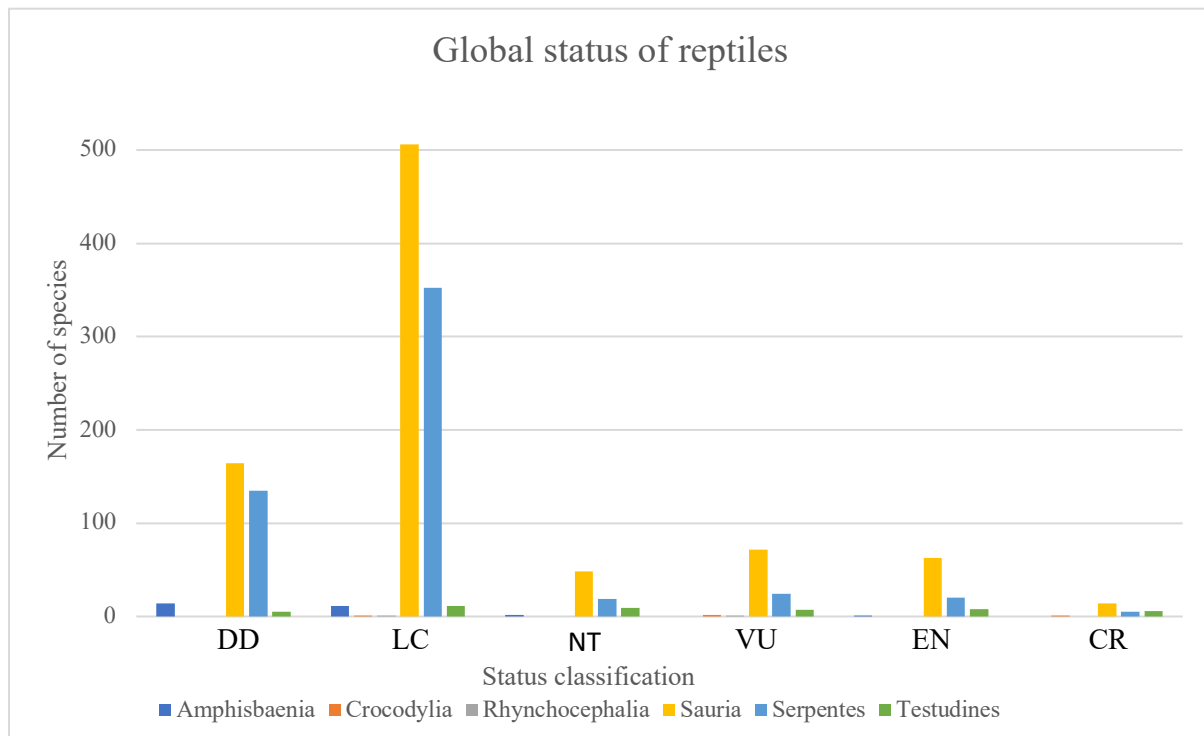


Figure 5. Global status of reptiles taken from Böhm et al. (2013), IUCN Threat Status Key: (DD = Data Deficient; LC = Least Concern; NT = Not Threatened; VU = Vulnerable; EN = Endangered; CR = Critically Endangered).

Anthropogenic threats are recognized as key accelerators for the decline in reptiles globally (Gibbons et al., 2000; Todd et al., 2010). Habitat degradation and the introduction of invasive species are two of many infamous factors (Gibbons et al., 2000). For example, the loss of 97% of southeastern longleaf pine habitats caused the decline in gopher tortoises (*Gopherus polyphemus*), eastern indigo snakes (*Drymarchon corais*) and the eastern diamondback rattlesnakes (*Crotalus adamanteus*) populations in the southern USA (Guyer & Bailey, 1993; Ware et al., 1993). Next, the introduction of invasive species disrupting indigenous reptile communities is evident (Gibbons et al., 2000). The Tuatara (*Sphenodon punctatus*), an endemic species of lizard found in New Zealand became extinct on the mainland and 10 offshore islands due to the introduction of rats. In addition, feral pigs introduced to the

Galapagos islands were just one of the many species responsible for the near extinction of the Galapagos tortoises (*Geochelone elephantopus*).

As mentioned before, data deficiency itself is not a threatening factor. However, the lack of information to properly assess reptilian species could mean that extinctions are happening without conservationists' knowledge. Reptiles are considered the second most species rich group of amniotes after birds (Pincheira-Donoso et al., 2013), however, conservation assessments relating to them are few (Todd et al., 2010). Most conservation assessments conducted relate to birds, mammals and amphibians. One plausible reason for this could be due to detectivity difficulties as most reptiles are characterized by cryptic coloration, seasonal or daily timings and environmental conditions (Todd et al., 2010). 52% of reptiles are classified as data deficient with only a few species known from specimen types and old records. Most of these data deficient reptiles lack information on population status and trends (Bland & Böhm, 2016).

The global decline in reptile populations, whether unnoticed or widely documented is troubling. Over the past few years, reptiles have been recognized to play an important role in natural ecosystems and as indicators of environmental quality (Böhm et al., 2013; Gibbons et al., 2000). For example, lizards and snakes have been shown to detect environmental pollution, particularly, organochlorines (Manolis et al., 2002). A study in the Canary Islands where agrochemicals were used showed low levels of lead and pesticides in the liver and fats of Gopher snakes (*Pituophis catenifer*) and Prairie rattlesnakes (*Crotalus viridis*), which suggested low levels of agrochemical use in the study area (Bauerle et al., 1975). In addition, radiocesium levels found in snakes species inhabiting contaminated sites were much higher than those from uncontaminated ones (Brisbin Jr et al., 1974). Reptiles play an important role in the ecosystem. Iwamoto (1986), suggests that reptiles, such as geckos, skinks and snakes play the role of a higher consumer in the ecosystem. Furthermore, studies have suggested

iguanas to act as efficient seed dispersers in their habitats (Benítez-Malvido et al., 2003; Lasso & Barrientos, 2015; Laurel et al., 2000). A study conducted by Lasso and Barrientos (2015), suggested faster seed germination attached to the snout of Green iguanas (*Iguana iguana*) as compared to those taken from feces and the fruit directly.

Iguanas

Iguana (*Iguanidae*) is a family that falls under the class Squamata. There are currently eight extant genera with approximately 44 species globally (Buckley et al., 2016). The genera consist of: *Cyclura*, *Ctenosaura*, *Amblyrhynchus*, *Conolophus*, *Sauromalus*, *Iguana*, *Brachylophus* and *Dipsosaurus* (Etheridge & de Queiroz, 1988); with *Ctenosaura* being the most diverse with 18 species and *Amblyrhynchus* the least, with only one species (Buckley et al., 2016). Iguanas are large herbivorous lizards that inhabit a range of habitats, from deserts to island coasts (Norris, 1953; Wikelski & Thom, 2000). *Iguana* is a common species known to inhabit a large geographical range, from southern Brazil to North Mexico. There are currently two extant species within its genus. *Iguana* species are arboreal and can often be found in wet forests. Furthermore, they have often been used as test subjects in the laboratory for physiological and behavioral experiments (Alberts et al., 2004). *Dipsosaurus*, or more commonly known as the desert iguana, inhabit desert areas in southwestern regions of the United States and North Mexico. There are four extant species found within this genus. *Sauromalus*, more commonly known as Chuckwallas, are often found in the arid regions of southwestern United States and northern Mexico (Alberts et al., 2004). They currently have five extant species in its genus. Both *Dipsosaurus* and *Sauromalus* are known as desert iguanas and can also be found on some islands of the Southwestern United States and Mexico (Lamb et al., 1992). *Ctenosaura* iguanas commonly known as the spiny-tailed iguana are the most

diverse, with 18 species in its genera. They are terrestrial and are native to Central America and Mexico. They can generally be found in dry forests (Alberts et al., 2004). *Conolophus* and *Amblyrhynchus*, are restricted to the Galapagos archipelago and are sister taxa. They are more commonly known as the Galapagos land iguana and marine iguana respectively. There are currently three recognized species within the genus of *Conolophus*; *Conolophus subscristatus*, *C. pallidus* and *C. Marthae* (Gentile et al., 2009; Rassmann et al., 2004). *Amblyrhynchus* on the other hand is considered a monotypic genus (meaning having only one species). They have a wide distribution within the Galapagos archipelago and can often be found in large groups on the coasts (Rassmann et al., 1997). *Cyclura*, more commonly known as rock iguanas have 11 species, with one listed as extinct, *Cyclura onchiopsis*. They are often found in subtropical regions of west India (Knapp & Hudson, 2004). This particular species has a high level of endemism, and are restricted to an individual island (Blair, 1991; Malone & Davis, 2004). Lastly, *Brachylophus*, or more commonly known as crested iguanas are found on the islands of Fiji and Tonga and are considered the only true iguanas to be found outside of the Americas (Gibbons, 1984). There are only four extant species found in this genus.

The majority of iguana species are found to inhabit islands; hence making them highly susceptible to decline and extinctions. 75% of recorded animal extinctions since the 1600s were island dwelling species; this includes 28 known reptile species (Alberts et al., 2004). Invasive species have been recognized to play a major role in the population decline of iguanas. Some excellent examples include the extinction of the Turks and Caicos iguana (*Cyclura carinata*) on Pine Cay island, where 15,000 individuals were extirpated due to the introduction of feral cats and dogs (*Canis lupus familiaris*) (Iverson, 1978); and the local extinction of Galapagos land iguanas on Santiago island in the 17th century, where introduced mammalian species such as cats, dogs and pigs severely predated and altered the environment (Cruz et al., 2005). Iguanas generally prefer large undisturbed areas unlike small lizard species such as geckos

(Gekkonidae), which thrives in smaller and altered habitats such as urban environments (Alberts et al., 2004). Based on the IUCN red list, there are currently eight species of iguanas that are listed as Critically Endangered, 12 listed as Endangered, 11 listed as Vulnerable, three listed as Not Threatened, 11 listed as Least Concern, two listed as Data Deficient and one listed extinct (IUCN, n.d.). The figure below shows the conservation status of the number of iguana species in each genus, found globally.

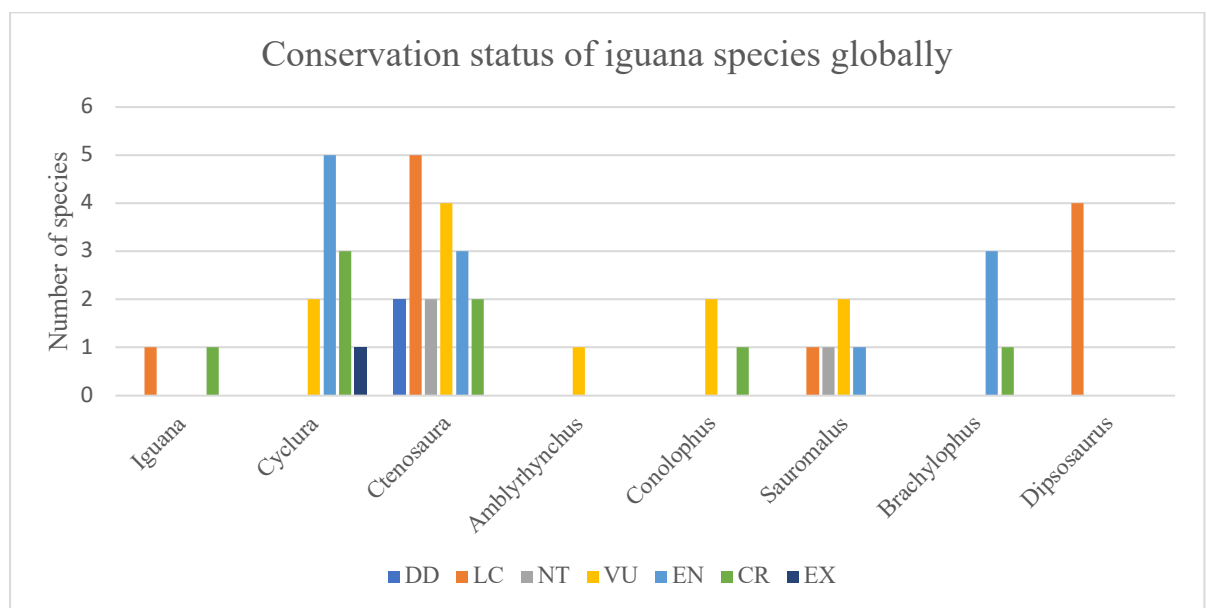


Figure 6. Figure showing the conservation status of iguana species found globally.

Iguanas play an important role in the ecosystem, primarily seed dispersal (Traveset et al., 2016; Vásquez-Contreras & Ariano-Sánchez, 2016). Endozoochory (the study of seed dispersal via ingestion) among reptiles is not commonly studied and few taxa such as iguanids have been reported to show this type of behavior (Vásquez-Contreras & Ariano-Sánchez, 2016). The Galapagos land iguana is one example that have been reported to play an important role in seed dispersal for native plants throughout the Galapagos archipelago (Traveset et al., 2016). Other studies have supported this notion showcasing similar results with other species

of iguanas, with one showing higher germination rates from ingested seeds (Benítez-Malvido et al., 2003; Lasso & Barrientos, 2015; Laurel et al., 2000).

Threats to Iguana populations and species

According to the IUCN, iguanas are amongst the world's most endangered animals due to habitat degradation and invasive species (IUCN, n.d.-c). The effects of invasive species, primarily feral cats, poses a threat to both iguanas and the ecosystem. The depredation of iguanas by feral cat populations is globally evident (Iverson, 1978; Mitchell et al., 2002). For example, fecal pellets collected by feral cats found on Pine Cay, revealed evidence of depredation on juvenile West Indian rock iguanas (*Cyclura carinata*) (Iverson, 1978). Foraging strategies by feral cats include excavating shallow burrows and patiently waiting outside burrows. Their exceptional sense of smell allows them to track iguana burrows which exhibits a distinct odor (Iverson, 1978). More importantly, the depredation of iguanas by feral cats can alter the ecosystem indirectly. Iguanas play the role of seed dispersers in their local habitat (Benítez-Malvido et al., 2003; Lasso & Barrientos, 2015; Laurel et al., 2000). Hence, as resident populations of iguanas decline, so does local vegetation.

The removal of feral cats from islands to protect endangered species of iguanas have been a popular method adopted by conservation biologists (Mitchell et al., 2002; Pérez-Buitrago et al., 2008; Phillips et al., 2005). Popular cat eradicating methods include poisons such as sodium monofluoroacetate (1080), hunting and trapping or disease introduction (mainly virus) (Muller et al., 2000; Nogales et al., 2004; Veitch, 2001). Many studies have suggested positive results on local iguana populations after the removal of feral cats (Álvarez et al., 2007; Mitchell et al., 2002; Phillips et al., 2005). For example, the translocation of Turks and Caicos rock iguanas (*Cyclura carinata*), after the eradication of feral cats, to Long Cay and Caicos

Bank had suggested successful establishment with hatchlings recorded in January 2001 (Mitchell et al., 2002).

THE PRESENT STUDY

The high proportion of threatened iguanas in the world, in particular island-dwelling species (see section above) requires the implementation of conservation management practices not only to assist in the recovery of iguana populations, but also to restore the ecological role of iguanas in their native ranges. For example, the implementation of translocation on iguanid species have been ongoing since the late 1980s to today, with many of them proven successful (see table below), The Galapagos Islands are home to four iguana species, three of which are exclusively terrestrial, plus one coastal species that forages in open sea waters (see section below). There have been a number of studies providing information on the terrestrial iguanas of the Galapagos Islands but there is still a significant gap in information about their current status, and basic biological data. In this thesis I present new information on species and populations of iguanas that will hopefully assist the Directorate of the Galapagos National Park in its mission to conserve these reptiles. Specifically, I present new information on a recently reintroduced population of Galapagos land iguanas to Santiago Island, and results from a recent survey of the critically endangered Pink Iguana (*Conolophus marthae*) on Wolf Volcano.

Table 1. Table of iguana translocations from 1980s to 2000s. Symbols: ? = unknown

Common Name	Genus	Species	Source Population	Translocated Population	Year	Number	Success	Failed	References
Cuban iguana	<i>Cyclura</i>	<i>Nubile</i>	Isla de la Juventude	Isla Magueyes (Puerto Rico)	1960	?	1	0	(Christian et al., 1986)
Acklins iguana	<i>Cylcura</i>	<i>Rileyi nuchalis</i>	Fish Cay (Acklin islands)	Bush Hill Cay (Northern Exuma islands)	1973	5	1	0	Iverson et al. (2016)
Rock iguana	<i>Iguana</i>	<i>Pinguis</i>	Anegada island	Guana island	1984 - 1987	8	1	0	Goodyear and Lazell (1994)
Allen Cay iguana	<i>Cyclura</i>	<i>Cychlura inornata</i>	Leaf Cay	Alligator Cay (Exuma islands, Bahamas)	1988 - 1990	8	1	0	Knapp (2001)

Galapagos land iguana	<i>Conolophus</i>	<i>subcristatus</i>	North Seymour	Baltra	1991	35	1	0	(Cayot & Menoscal, 1994)
Turks and Caicos rock iguana	<i>Cyclura</i>	<i>Carinata</i>	Big Ambergris Cay	Long Cay (Caicos bank)	1999	25	1	0	Mitchell et al. (2002)
Green iguana	<i>Iguana</i>	<i>iguana</i>	Sixaola River	Caribbean coast of Limon (Costa Rica)	2001	11	1	0	Escobar et al. (2010)
Mona island iguana	<i>Cyclura</i>	<i>Cornuta stejnegeri</i>	Mona island	Mona island	2002	10	1	0	Álvarez et al. (2007)
Sandy cay rock iguana	<i>Cyclura</i>	<i>Rileyi cristata</i>	Illegal poaching	Crown land cay	2014	9	?	?	Hayes et al. (2016)

COVID-19 Declaration

The 2020 COVID-19 pandemic severely affected my thesis plans and ability to conduct field research and data collection, as well as my writing. Initially, I planned to conduct research on the endangered Mariana Fruit Bat (*Pteropus mariannus*) on the island of Rota, in the Commonwealth of the Northern Marianas. I devoted substantial efforts to develop ideas, discuss these with staff from the US Fish and Wildlife Department in Saipan, and in preparing grant applications. In 2019, I prepared a research proposal and research grants for this project and presented these to the Auckland Zoo Conservation Fund and the Mohamed bin Zayed Species Conservation Fund (Appendix II, III). However, my applications were unsuccessful. The original intention was to re-apply in early 2020 based on feedback obtained from these grant applications, however after discussions with my supervisor, Dr. Luis Ortiz-Catedral, we agreed on developing a field project in the Galapagos Islands monitoring a reintroduced population of Galapagos land iguanas on Santiago Island. Specifically, I would be investigating the health status of Galapagos land iguanas after their release into the wild on Santiago Island, in particular the effects of ectoparasite removal on body condition and growth between age classes (see Chapter 2). This latest project was set to start in March 2020; however, this was not possible as New Zealand went into lockdown on March 25th 2020. Considering the COVID-19 pandemic and the travel restrictions in place both in New Zealand and the Galapagos Islands, my supervisor and I decided that a more productive course of action would be to develop a thesis without a field component as at the time it was uncertain how long the COVID-19 restrictions would be in place. The high degree of uncertainty about the extent of the pandemic and the duration of the lockdown in New Zealand and the rest of the world, as well as extended periods of isolation made it very difficult to make progress on my thesis. These circumstances also made it difficult to start an alternative project in New Zealand as

travel restrictions within the country relaxed. As a result, following the recommendations of my supervisor, I developed a thesis without a field component but covering aspects that had not previously been analysed for populations of Galapagos terrestrial iguanas. The thesis presented here is based on unpublished datasets collected in 2017-2019 by park rangers of the Galapagos National Park, members of the NGO Island Conservation and my supervisor. The data collection followed the approved activities outlined in the permit PC-96-18 by the Ministry of Environment, Ecuador (Appendix I).

The Directorate of the Galapagos Islands conducts semi-annual visits to key iguana localities on various islands to obtain basic information about the general status of populations. The approximate density of iguanas, their size and weight as well as sightings of juveniles are of interest because that information is used to prioritise management interventions. The present study focuses on data collected on populations of Galapagos terrestrial iguanas, and pink iguanas. This study also provides some recommendations for future research based on published and unpublished information.

THESIS AIMS

This thesis addresses a number of significant gaps in knowledge for the management and conservation of Galapagos terrestrial iguanas. The overall aim is to provide an update on the status of populations and species of Galapagos terrestrial iguanas and to present ecological information that can assist the Directorate of the Galapagos National Park on its mission to preserve the biodiversity of the Galapagos Islands.

Objectives

1. To determine the pre-translocation effects of *Amblyomma* ectoparasites on the body condition of Galapagos land iguanas (*Conolophus subcristatus*) from Seymour Norte (Chapter 2).
2. To quantify changes in body mass and body condition of Galapagos land iguanas (*Conolophus subcristatus*) reintroduced to Santiago Island in January 2019 (Chapter 3).
3. To present preliminary information on the dispersal and behavior of reintroduced Galapagos land iguanas to Santiago Island (Chapter 3).
4. To describe the current status of the world's only population of Pink iguanas (*Conolophus marthae*) on Wolf volcano (Chapter 3).
5. Provide recommendations on conservation management for three endemic species of iguanas (*Conolophus subcristatus*, *Conolophus marthae*, *Conolophus pallidus*) found in the Galapagos (Chapter 4).

STUDY SPECIES

Galapagos land iguana

The Galapagos land iguana (*Conolophus subcristatus*) (Fig. 8) is one of three *Conolophus*, a genus endemic to the Galapagos Islands. The Galapagos land iguanas form a sister taxon to the Galapagos marine iguanas (*Amblyrhynchus cristatus*), from which they diverged 10 to 20 million years ago based on osteological, immunological and mitochondrial DNA data (Tzika et al., 2008). At present, *C. subcristatus* inhabits six islands, Plaza Sur, Baltra, Santa Cruz, Isabela, Fernandina (Tzika et al., 2008) and more recently, Santiago island where the species was reintroduced in January 2019 (Kumar, 2019). According to the IUCN red list, Galapagos land iguanas are classified as ‘Vulnerable’, with its last assessment conducted in February 2020. In addition, they are protected under Appendix II of Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) (IUCN, n.d.-a). The need to update key information for the Galapagos land iguanas on the IUCN red list website is imperative, as the information from the red list is widely used for identifying species and sites for conservation assessment (Rondinini et al., 2014). The conservation of the Galapagos land iguana focuses on reducing the threat posed by introduced species, and reintroducing individuals via a head-start program or translocation. Starting in the 1930s, Galapagos land iguanas were translocated to Seymour Norte from Baltra Island by the Hancock expedition (Perkins, 1930). This action, conducted before the establishment of the Galapagos National Park, ensured the survival of the Baltra Island population. In the 1970s, a head-start program with headquarters at the Charles Darwin Research Station begun and oversaw the production of hundreds of Galapagos land iguana hatchlings, which were released on Santa Cruz, Baltra and Isabela Islands between 1970s and 2005. The head-starting program concluded in 2005 with the release of the remaining hatchlings on Baltra Island (A. Llerena, pers. comm.). Published articles that focus on the conservation of the Galapagos land iguanas

mostly discuss the eradication of invasive mammals (Fabiani et al., 2011; Phillips et al., 2005). There are only two published papers evaluating local population sizes and threats of the land iguanas and they are by Marquez et al. (2019) and Kumar (2019).

Galapagos land iguanas are primarily herbivorous and mainly feed on the endemic Galapagos Prickly Pear cactus (*Opuntia galapageia*) and native Palo Santo tree (*Bursera graveolens*) (Traveset, 1990). However, they occasionally also ingest animal matter such insects, centipedes or carrion (Charles Darwin Foundation, 2006). Sexual dimorphism is evident with males being significantly larger than females. In addition, males are often characterized with possessing more distinct head scales and larger crests (Kumar, 2019; Snell et al., 1984) (Fig. 7). Land iguana adults have the capacity to grow to more than a meter in length and can weigh up to 13 kilograms. They have life span of approximately 60 years with maturity age ranging between eight to 15 years old (Charles Darwin Foundation, 2006). The Galapagos race snakes (*Pseudalsophis* spp) and Galapagos hawks (*Buteo galapagoensis*) are known natural predators that commonly prey on juvenile iguanas that are smaller in size. However, introduced species such as goats, rats, dogs, pigs and cats represent a threat to the iguana population via habitat destruction, competition for resources and predation (Kumar, 2019). Some studies have suggested that the Galapagos land iguana play an important role in their local ecosystem by acting as seed dispersers. Results showed, 10% of seeds ingested by the iguanas germinated. This is especially useful for the restoration of habitats (Hendrix & Smith, 1986; Traveset et al., 2016).



Figure 7. Galapagos land iguana. Male (left) and female (right), not the larger and more robust dorsal scales on the male and the more gracile look of the female. Photos: L. Ortiz-Catedral.



Figure 8. Land iguana (*Conolophus subcristatus*) in its habitat on Seymour Norte. Photo: L. Ortiz-Catedral.

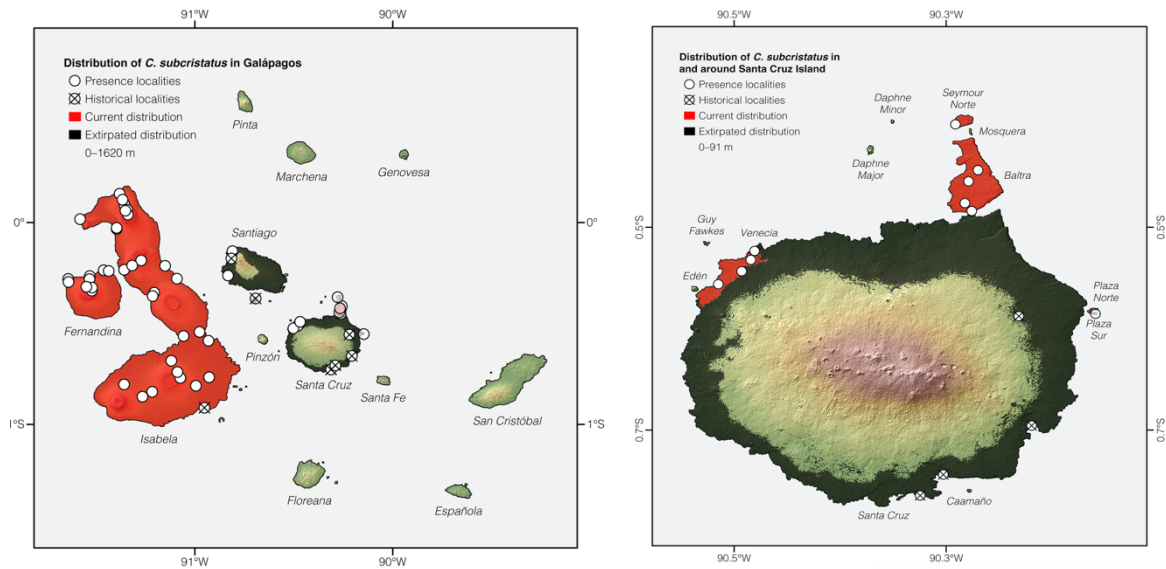


Figure 9. Distribution of the Galapagos land iguana (*Conolophus subcristatus*) in the Galapagos Islands. Left: distribution across the archipelago; right: distribution on islands and islets off the coast of Santa Cruz Island. Maps: Arteaga et al., 2019.

Galapagos pink land iguana

The Galapagos pink land iguana (*Conolophus marthae*) or “Iguana rosada” is a recently discovered new species endemic to the Galapagos archipelago. They are distinguished by having a pinkish head and black body and legs (Fig. 11). Typically, they have a black-striped pattern on the mid to posterior dorsal body (Gentile & Snell, 2009). Their distinctive pattern of head bobbing to establish territory and attract mates are also unique and is found to be much more similar to the marine iguanas than any of the other Galapagos iguanas. Based on stool samples collected in the lab, the Galapagos pink land iguana is believed to be strictly vegetarian (Pierson & Durham). They are currently classified as ‘Critically Endangered’ under the IUCN red list. In addition, information regarding their ecology and population trend is limited due to the nature of their habitat. The estimate population size is 192 and they are known to be found only on Volcán Wolf, on Isabela island (Gentile et al., 2016; IUCN, n.d.-b).

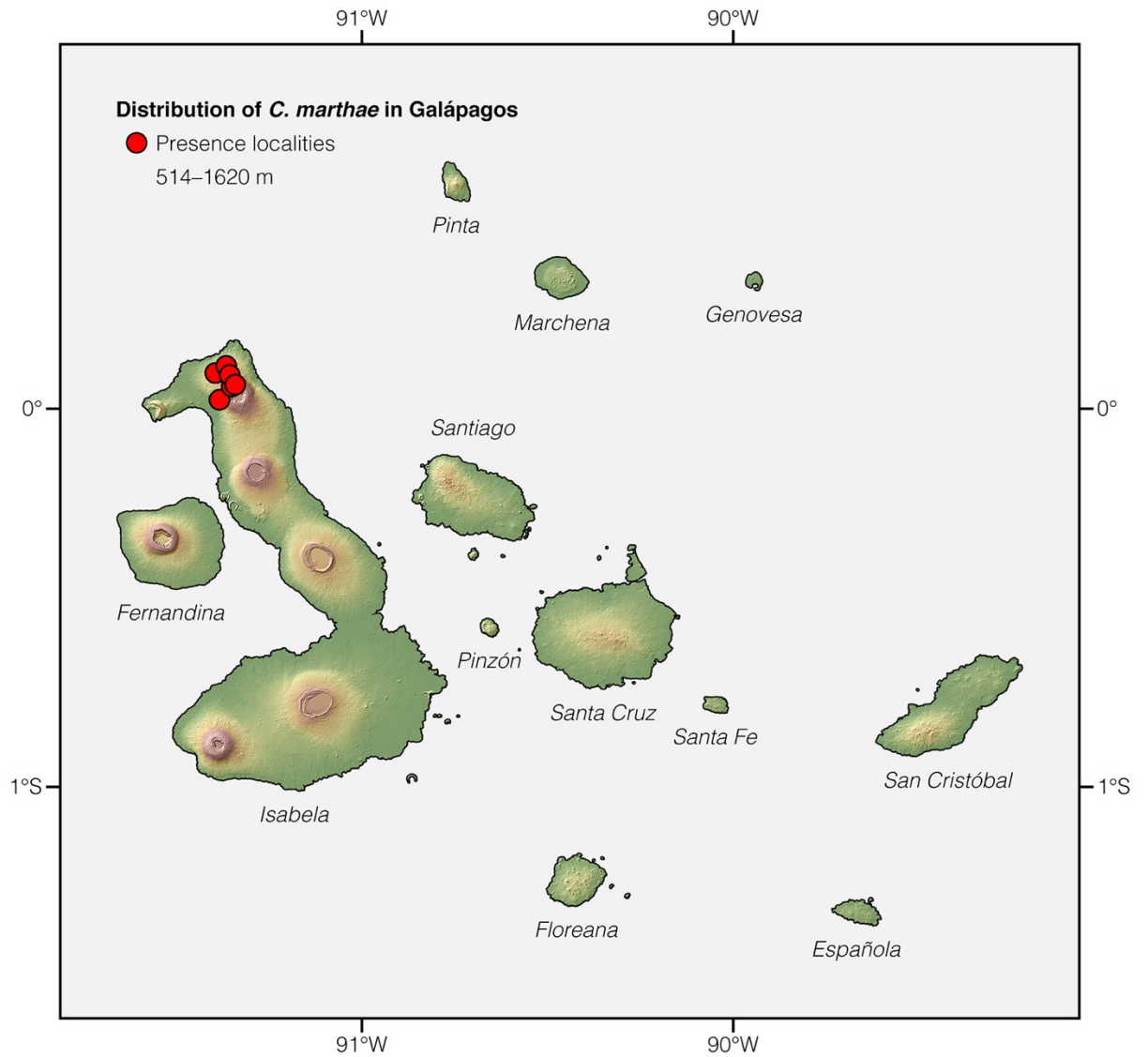


Figure 10. Distribution of the Pink iguana (*Conolophus marthae*). Map: Arteaga et al., 2019.



Figure 11. Pink iguana (*Conolophus marthae*) on Wolf Volcano. Photo: L. Ortiz-Catedral

Santa Fe Land iguana

The Santa Fe iguana (*Conolophus pallidus*) (Fig. 13) also known as the Barrington land iguana is a species of land-dwelling iguanids found only on the island of Santa Fe. According to the IUCN red list, the Santa Fe iguanas are classified as ‘Vulnerable’ (IUCN, n.d.). The most recent survey conducted in 2005 estimated their population to be approximately 3,500 to 4,000 mature individuals with their population potentially being stable (Gentile & Grant, 2020). However, whether the island has reached its maximum carrying capacity is currently still unknown. The Santa Fe iguana is morphologically similar to the Galapagos land iguana; apart from the fact they possess paler yellow scales and have a longer, more tapered snout with more pronounced dorsal spines. They are able to grow to approximately 0.91 m, including tail length and a body weight of approximately 11 kg.

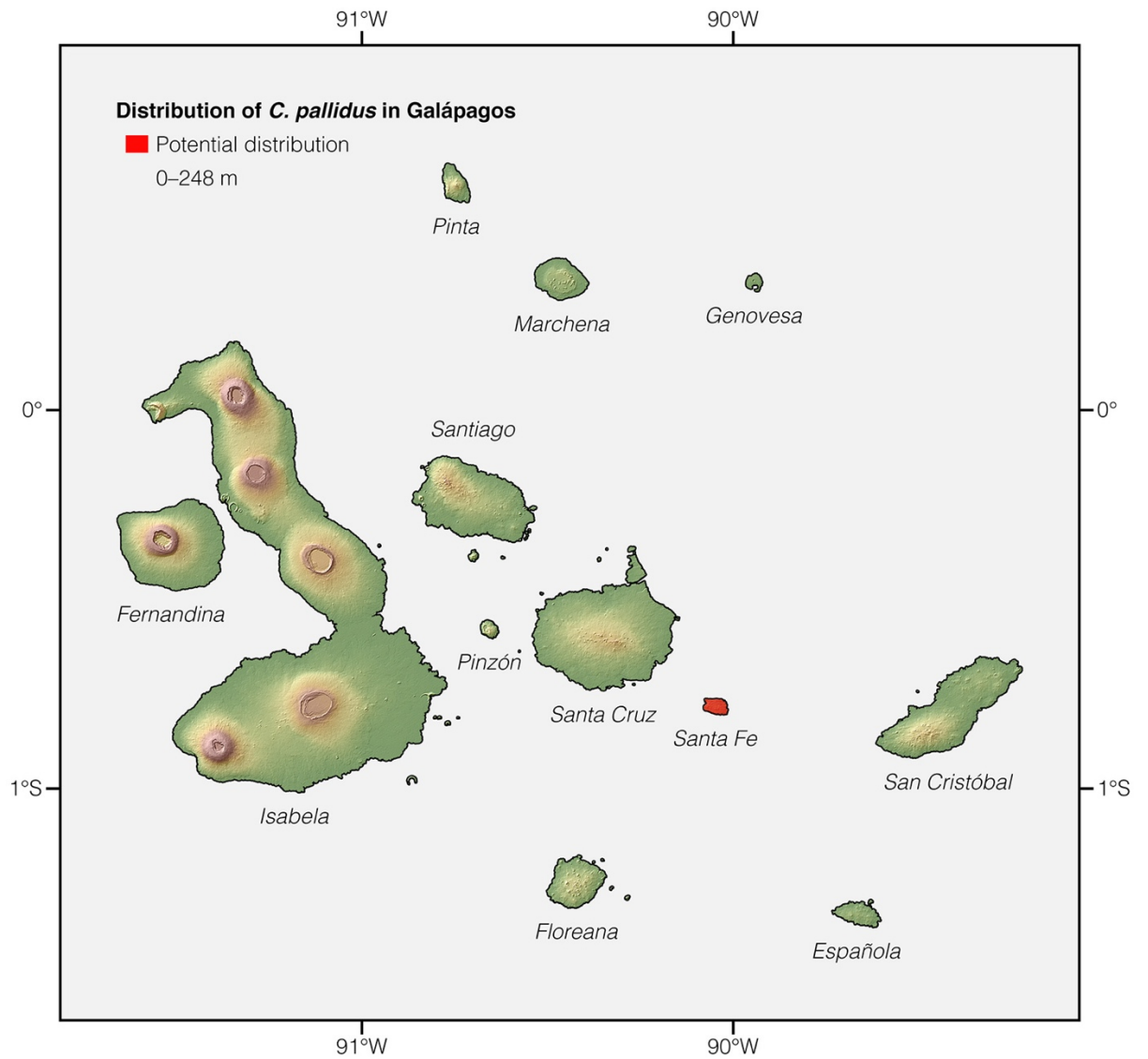


Figure 12. Distribution of Santa Fe iguana (*Conolophus pallidus*). Map: Arteaga et al., 2019.



Figure 13. Santa Fe land iguana (*Conolophus pallidus*) on Santa Fe island. Photo: L. Ortiz-Catedral.

STUDY SITES

Study sites from my data collection includes Fernandina island, Santiago island, Baltra, Isabela and Seymour Norte island in the Galapagos archipelago. The Galapagos archipelago is part of the Republic of Ecuador and is made up of several volcanic islands. It is found in the Pacific Ocean approximately 1,000 kilometers from the mainland of Ecuador, South America (Claudino-Sales, 2019). The Galapagos archipelago sits relatively close to the equatorial line, however, three main currents; Humboldt, Panama and Equatorial allows the island to maintain

average temperatures around 22°C (Ader, 2000). The climate of the Galapagos archipelago consists of two season, wet and dry. Wet season, also known as the hot season, usually occurs from June to November, while the dry season is characterized by cool temperatures with fogs that envelops the highlands (Claudino-Sales, 2019). More details of individual site descriptions are provided below.



Figure 14. Map of the Galapagos islands.

Fernandina island (0°22'0"S, 91°31'20"W) (July 2018)

Area size: 64248 ha (Snell et al., 1996). Fernandina island is the westernmost island found within the Galapagos archipelago and is known to be the youngest of the islands. It is one of the few islands in the Galapagos archipelago that has no introduced mammals. Fernandina island is approximately less than one million years old and is famous for its continuing series of volcanic eruptions. (Galapagos Conservancy, n.d.; Schatz, 1998). Approximately 80% of the island consists of barren lava fields that has limited vegetation with crevices formed by old lava flow on slopes.

Isabela island (0°25'30"S, 91°7'W) (February 2019)

Area size: 458812 ha (Snell et al., 1996). Isabela island is found on the western edge of the Galapagos archipelago and is approximately one million years old. It is also the largest island within the Galapagos archipelago. It consists of six shield volcanos, one of which, is the Wolf volcano; home to the rare, critically endangered, Galapagos pink land iguana (*Conolophus marthae*). In addition, Isabela island houses many native fauna and flora which are threatened by several different introduced mammals and plants (Eckhardt, 1972).

Baltra island (0°25'30"S, 90°16'30"W) (December 2017)

Area size: 2620 ha (Snell et al., 1996). Baltra island is a small, low and flat island situated near the northern coast of Santa Cruz island. Unlike the other islands, Baltra houses the Galapagos airport and is managed by the Ecuadorian Air Force as a military base. Vegetation on the island is sparse due to the large surfaces of weathered boulders which

prevents shrubs and grasses to establish. However, they do support arid-adapted shrubs and cactus (Phillips et al., 2005).

Santa Fe island (0°49'0"S, 90°3'30"W)

Area size: 2413 ha (Snell et al., 1996). Santa Fe island is one of the oldest islands in the Galapagos archipelago, with volcanic rocks found to be approximately four million years old (Galapagos Conservancy, n.d.). The island is low and arid and has a relatively open desert vegetation. Most vegetation is dominated by *Bursera graveolens* and cactus (*Opuntia echios*). The island houses the only population of the Santa Fe iguanas (*Conolophus pallidus*) and is also home to the Barrington leaf-toed gecko (*Phyllodactylus barringtonensis*). There are currently no introduced mammals on Santa Fe. No field data were collected on Santa Fe, but it is included here to outline the characteristics of this island.

Seymour Norte (0°23'30"S, 90°17'0"W) (December 2017)

Area size: 184 ha (Snell et al., 1996). Seymour Norte or North Seymour island is located just north of Santa Cruz island and is separated by a narrow channel less than one kilometer wide. It has volcanic rock surfaces that graduals upwards from a sandy beach in the west to a 60 meters cliff (Alpert, 1946). Vegetation on Seymour Norte is heavily dependent on exposure and elevation and hence can be seen in patches along the shores of the island in certain places.

Santiago island (0°15'30"S, 90°43'30"W) (February 2019, August 2019)

Area size: 58465 ha (Snell et al., 1996). The island of Santiago consists of two overlapping volcanoes. Historically, the island was inhabited by an abundant number of native species such

as the giant tortoises (*Chelonoidis nigra*) and the Galapagos land iguana (*Conolophus subcristatus*). According to a scientist, Charles Darwin, he documented a thriving population of Galapagos land iguanas on the island with and quoted he had no place to pitch a single tent due to their burrows (Darwin, 1959). Unfortunately, due to the introduction of mammalian species in the 17th century and anthropogenic threats, the ecology of the island was torn asunder, and the Galapagos land iguana populations were brought to extinction. Goats (*Capra aegagrus hircus*), pigs (*Sus scrofa*) and donkeys (*Equus asinus*) destroyed plants through grazing while rats (*Rattus rattus*) and cats (*Felis catus*) depredated on juvenile iguanas and tortoises (Carrion et al., 2007; Cruz et al., 2005). In addition, the endemic rice rat (*Nesoryzomys swarthi*) was considered extinct since 1996 due to depredation by black rats but was then later rediscovered in 1997 (Dowler et al., 2000).

To date, the island of Santiago has undergone tremendous changes. It has been declared free of all large introduced mammals with the exception of rats (Galapagos Conservancy, n.d.). More importantly, the translocation of the Galapagos land iguana back to their historical site was conducted in 2019. Furthermore, with the island undergoing exceptional recovery in vegetation regrowth, wildlife such as the Galapagos rails were being sighted once again.

Santiago island is one of the many islands found in the Galapagos archipelago. It is a perfect example of a large-scale restoration site. Historically, the island was inhabited by many native species such as the giant tortoises (*Chelonoidis nigra*) and the Galapagos land iguana (*Conolophus subcristatus*). According to a scientist, Charles Darwin, his report stated a thriving population of Galapagos land iguanas on the island with ‘no place to pitch a single tent due to their burrows’ (Darwin, 1959). Unfortunately, due to the introduction of mammalian species in the 17th century, the ecology of the island has been torn asunder. Goats, pigs, donkeys, rats, dogs and cats are highly considered to be the cause of the destruction of Santiago

island. (Carrion et al., 2007; Cruz et al., 2005; Fabiani et al., 2011). These introduced mammals destroyed habitats and competed with native species for resources.

In addition, according to Cruz et al. (2005), an endemic species of rice rat (*Nesoryzomys swarthi*) was considered extinct since 1996 but was later rediscovered in 1997. Although all pigs on Santiago island were eradicated in 2005 other introduced mammals such as goats, donkeys, rats, house mice and smooth billed anis were still present. Following the completion of project Isabela in 2006, Santiago island has been declared free of all large, introduced mammals and only rats (*Rattus rattus*) remain on the island. With the island undergoing exceptional recovery in vegetation regrowth and most mammalian species removed, reintroduction of the Galapagos land iguanas on Santiago island was conducted in 2019 to help with seed dispersal and maintaining open areas free from vegetation.

GENERAL FIELD METHODS

The data presented in this thesis was collected in the course of five field trips to five islands in the Galapagos archipelago from December 2017 to August 2019. As stated above, I did not participate in the data collection. Here I describe the general methods used to capture and measure iguanas on each of the fieldtrips. Galapagos terrestrial iguanas are diurnal and active from early in the morning to sunset (Ortiz-Catedral, pers. comm.). Capture of iguanas is by hand and only done by skilled park rangers of the Galapagos National Park. Iguanas are firmly grabbed by the base of the tail and the nape and turned on their back (Fig. 15). In this position, they remain relatively motionless and they can be processed within a few minutes. Each iguana is weighed, and measured by one person, while the other restrains the animal. To distinguish between individuals each iguana is marked with a PIT-tag. Due to their longevity many iguanas exhibit old brands in their chest, applied before PIT technology was available.

Following the measurements of snout-vent length (SVL), tail length (TL) and overall examination of body condition, iguanas are released at the site of capture.



Figure 15. Luis Ortiz-Catedral (Massey University) and Marcelo Gavilanes (Galapagos National Park) processing a Galapagos Land iguana (*Conolophus subcristatus*) on Baltra Island, December 2017. From top left: Marcelo Gavilanes restraining an iguana on its back to facilitate measuring; top right: iguana weighed using a Pesola® scale and a piece of rope around the forelimbs; bottom left: measuring of SVL; bottom right: inserting a unique subcutaneous PIT tag in the right leg to facilitate ID on recapture. Photos: H. Sollis ©.



Figure 16. Wilson Cabrera, Johannes Ramirez (Galapagos National Park) and Luis Ortiz-Catedral (Massey University) processing a Pink Iguana (*Conolophus marthae*) on Wolf Volcano, February 2019. From Top left: Wilson Cabrera examining an iguana; top right: Wilson Cabrera measuring the SVL of an iguana; bottom left: Luis Ortiz-Catedral inserting a unique subcutaneous PIT tag on the left leg of the iguana; bottom right: dorsal pattern of the individual photographed before release.

Photos: Luis Ortiz-Catedral and Johannes Ramirez ©.

Every iguana was handled by experienced park rangers, and all captures, measurements and photographs were obtained according to approved methods by the Directorate of the Galapagos National Park (Appendix I). Iguanas from Baltra Island and Fernandina Island (both discussed in Chapter 3, were also individually photographed, Appendix IV).

THESIS OUTLINE

The present thesis consists of four chapters, the following is a brief description of each chapter and their connection to the thesis aims and objectives outlined above.

CHAPTER 1

This is a general introduction to the global biodiversity extinction crisis and the status and diversity of iguanas around the world. I include here a description of the study sites and general field methods used in this thesis.

CHAPTER 2

In this chapter I present a preliminary analysis of the effect of ectoparasites on the pre-translocation body condition and health of Galapagos land iguanas (*Conolophus subcristatus*) prior to their reintroduction to Santiago Island in January 2019.

CHAPTER 3

In this chapter I summarize information of an expedition to the remote Wolf volcano in search of the critically endangered Pink Iguana (*Conolophus marthae*). This chapter includes information on the morphology and general observations of one of the least studied species of iguana in the world. I also present preliminary information of the biology of Galapagos land iguanas reintroduced to Santiago Island one month after their release. Finally, I provide an overview of the current status of populations of *Conolophus* iguanas in the archipelago.

CHAPTER 4

In this chapter I present and discuss significant gaps in knowledge on the terrestrial iguanas of the Galapagos Islands, and propose a number of priorities for future research that can help protect these endemic reptiles, and that can assist the Directorate of the Galapagos National Park in fulfilling its mission to preserve the biodiversity of the Galapagos archipelago.

**CHAPTER 2: EVALUATING THE PRE-TRANSLOCATION EFFECTS OF
AMBLYOMMA ECTOPARASITES ON (*CONOLOPHUS SUBCRISTATUS*) FROM
SEYMOUR NORTE.**



Figure 17. Park ranger Jean-Pierre Cadena holding a juvenile land iguana translocated to Santiago Island. Photo: L. Ortiz-Catedral

ABSTRACT

Conservation translocation is an important tool used to prevent a species from population decline and extinction. However, there are important factors to consider before the implementation of translocation, specifically physiological considerations such as ectoparasites and body condition. The study on the effects of ectoparasites on iguanas are few, and many of which are inconclusive. In this study, I compared the average mass, SVL and body condition of 2,120 pre-translocated Galapagos land iguanas (*Conolophus subcristatus*) to Santiago island with or without ectoparasites. In addition, I compared the average mass, SVL and body condition index between sex classes. The results yielded showed no significance between average mass, SVL and body condition index and sex classes, but previous studies on land iguanas have evidently shown sexual dimorphism; with males being significantly larger than females. Similarly, there were no significant correlation observed when comparing average mass, SVL and body condition index with the presence of ectoparasitism. However, I have insufficient evidence to conclude that ectoparasitism does not have an effect on mass, SVL and body condition index. I suggest that the mass, SVL and body condition index of the Galapagos land iguanas be investigated for post-translocation monitoring on Santiago island to ensure physiological growth of individuals. Further, the effects of ectoparasitism on land iguanas should be explored for long-term effects such as reproductive success and annual survivorship.

INTRODUCTION

Translocation is a broad term to describe the human-mediated movement of living organisms from one area to another. It comprises of moving living organisms from the wild or from captive origins (Reinert, 1991). Wildlife translocations or conservation translocations, on the other hand, is defined as the intentional movement and release of a living organism where its primary purpose is a conservation benefit (Seddon et al., 2007). The goals of conservation translocation can be divided into two groups: population restoration and conservation introduction. Population restoration is any conservation translocation within indigenous range, and it comprises of two activities: reinforcement and reintroduction (AWMS, 2019). Reinforcement (the intentional movement and release of an organism into an existing population of conspecifics) aims to enhance the viability of a population by increasing population size or genetic diversity (IUCN/SSC, 2013). For example, otter (*Lutra lutra*) populations found in the UK and mainland Europe have undergone various translocations to increase population abundance due to them being heavily persecuted from fishery protection and sport from the late 18th century to the early 20th century (White et al., 2003). Next, the aim of reintroduction (the intentional movement and release of an organism inside its indigenous range from which it has disappeared) is to re-establish a viable population of the focal species within its indigenous range (Armstrong & Seddon, 2008). A good example is the recently translocated Galapagos land iguanas (*Conolophus subcristatus*) from Seymour Norte to Santiago island (Kumar, 2019) which was once inhabited by the land iguanas according to Charles Darwin's notes (Darwin, 1959) and skeletal remains (Cruz et al., 2005).

Conservation introduction is the intentional release of an organism outside of its indigenous range and can be further divided into two activities: assisted colonization and ecological replacement (IUCN/SSC, 2013). Assisted colonization is the intentional movement

and release of an organism outside of its indigenous range to avoid extinction (Ricciardi & Simberloff, 2009) and it has been well established. Many endemic birds, reptiles and invertebrates threatened by introduced mammalian predators in New Zealand have been translocated to predator-free offshore islands that may not be part of the species range (Saunders & Norton, 2001). Next, ecological replacement (the intentional movement and release of an organism outside of its indigenous range to perform a specific ecological function), is used to re-establish an ecological function through the loss of extinction (Seddon & Soorae, 1999). A stark example is a case study on using extant non-indigenous tortoises as a restoration tool on Round Island found on the Mauritian offshore. Following the extinction of native giant tortoises, the plant communities found on Round Island were altered as tortoises are known to alter the competitive balance between plant species (Cayot, 1989; Grubb, 1971). The results of this study suggest that the introduction of surrogates to Round island was successful in promoting ecological restoration; non-indigenous tortoises fed on exotic vegetation and avoided native plants initially; however, when released from their enclosures, they were seen consuming and dispersing seeds of an endemic palm (*Latania loddigesii*) (Griffiths et al., 2010).

Wildlife translocation is an important conservation tool utilized by conservationists to date. In the year 2018, a total of 49 species of animals (six invertebrate species, four fish species, three amphibian species, five reptile species, six bird species and 25 mammal species) and 12 species of plants were translocated globally (Soorae, 2018). For example, a total of 31 Chinese giant salamander (*Andrias davidianus*) was reintroduced back to Heihe and Donghe river in the Qinling mountains, due to severe decline from habitat destruction and water pollution, where they were historically abundant according to local villagers; the results from this study was deemed partially successful due to the lack of resources to conduct further post translocation monitoring. More importantly, critical information, such as knowledge on

survivability were obtained (Zhang et al., 2018). In another example, the reintroduction of the Telfair's skink (*Leiopisma telfairii*) to Gunner's Quoin was deemed highly successful with results showing a good understanding of the species ecology and behavior and that the threats the species once faced were no longer present (Cole et al., 2018).

Conservation translocation have been occurring at increasing rates with improvements over the years (Seddon et al., 2007); however, there have still been mixed outcomes due to poorly planned programs and the lack of post-translocation monitoring (Armstrong & Seddon, 2008; Fischer & Lindenmayer, 2000; Scott & Carpenter, 1987; Scott et al., 2010; Seddon, 1999; Dodd Jr & Seigel, 1991). In order to address these difficulties, the IUCN's Species Survival Commissions Reintroduction Specialist Group (IUCN RSG) was formed in 1988 and released the first set of guidelines for reintroductions (IUCN, 1998). Post-translocation monitoring is important as it adds to our knowledge base and ability to carry out more effective releases (Ewen et al., 2012). For example, Burton and Rivera-Milán (2014), provided a technical evaluation of monitoring the Grand Cayman blue iguana (*Cyclura lewisi*); their results suggested that less intensive and invasive survey methods could provide accurate and precise counts of animals. Further, a study on jewelled geckos (*Naultinus gemmeus*) suggested that a period of penning significantly reduced dispersal and provided released animals opportunities to interact, breed and contribute effectively to population establishment (Knox & Monks, 2014).

There are many ways to define the criteria for success of translocation, but these criteria should be compatible with the biology of the focal species and the socio-economic context in which the translocation is developed (Germano et al., 2014). For example, investigating juvenile recruitment is a way to quantify translocation success. For example, a post-translocation study of Northern Bobwhites (*Colinus virginianus*); this study investigated the growth and survival rates of Bobwhite offspring and the results suggested translocated

Bobwhites had similar reproduction and offspring survival as their resident counterparts (Lunsford et al., 2019). Another method to quantify translocation success is physiological growth in the translocated species (i.e. mass and body length). Physiological evaluation of an individual before and after translocation could improve translocation success by identifying potential problems that may not be apparent through ad hoc observations. Physiological considerations can be classified into four categories: condition (i.e., distress and body condition), nutrition (i.e., wild food and supplementary feeding), health (i.e. parasite management) and ‘traditional’ (i.e. thermoregulation, immunoecology, micronutrients and stress). Based on a literature review conducted by (Tarszisz et al., 2014), out of the 72 studies that reported the condition of the translocated animals, 86% of them were rated successful. Furthermore, out of the 26 studies that noted distress in the animals, 81% of them demonstrated success. A post-translocation study conducted on Woylies (*Bettongia penicillate ogilbyi*) utilized body mass as a proxy for an individual’s success in securing resources in the new habitat and fitness; the results suggested that body mass on translocated Woylies increased post translocation due to the absence of predators in the new habitat (Page et al., 2019). More importantly, the authors concluded that the strongest predictors of body mass gain were sex, heart rate lability and escape behavior when released.

Before implementing translocation, there are many factors to consider. Knowledge of habitat quality, location of release area within species range, number of animals released, program length and reproductive traits are some examples (Griffith et al., 1989). Another important factor to consider is the presence and absence of ectoparasites. Ectoparasites have a large diversity which relates to the independent origin of many taxa: ectoparasitism is considered to have evolved at least seven times in insects (Waage, 1979). Ectoparasites rely on their host to survive but more so than often they inflict detrimental effects to their host. Weight loss, reduced production of milk, eggs, meat, hide and wool, fetal abortions and death are some

effects that ectoparasites can have on its host (Marshall, 1981; Nelson et al., 1977). For example, tick (*Ixodida* spp) infested cattle were shown to portray anorexia due to tick toxins; however, results have shown that even intense irritation could lead to reduced feeding (Meleney, 1985). In other cases, ectoparasites relating to poor insulation and resulting in mortality can be seen. Results from Clayton (1991), suggested that louse infected doves (*Columbidae* spp) had damaged feathers leading to poor insulation, which resulted in mortality. Furthermore, tick infested moose (*Alces alces*) showed more than 80% hair loss during winter, which is suggested to have influenced winter survivability (Glines & Samuel, 1989). Most ectoparasites pay a minimal price for killing their hosts (Lehmann, 1993). Results from Ewald (1987), suggested that ectoparasites that move between hosts have extremely low costs associated with extensive use of host resources; they are able to increase their fitness by faster conversion of host tissues and hence minimizing their dependance on their individual host by shortening the duration of host.

The role of ectoparasites, or their removal prior to translocation has received growing attention (Ewen, Acevedo-Whitehouse, et al., 2012; Moir et al., 2012; Sainsbury & Vaughan-Higgins, 2012). Some argue that ectoparasites too may face co-extinction and that their extinction rates are even higher than their hosts (Colwell et al., 2012; Dunn et al., 2009; Stork & Lyal, 1993). Further, some believe ectoparasites should be able to be listed as endangered or threatened based on their host's status (Durden & Keirans, 1996; Mihalca et al., 2011). Currently, only the pygmy hog sucking louse (*Haematopinus oliveri*) is listed on the IUCN red (Jørgensen, 2015; IUCN, n.d.). Despite the detrimental effects that ectoparasites bring to most host species, they have been given the benefit of the doubt and have been considered for co-reintroduction. The IUCN Species Survival Commission has also added their considerations for parasite co-reintroduction: If an extinct host had parasites that also became extinct, then it is desirable from a restoration perspective to re-establish those parasites with the translocated

host; but, this should subject to especially rigorous assessment of risks (IUCN/SSC, 2013). One example of host-specific ectoparasites is the beaver beetle (*Platypsyllus castoris*) that was unknowingly reintroduced from beavers (*Castor* spp). *P. castoris* was first observed in 1869 when European beavers (*Castor fiber*) were reduced to approximately 1,200 individuals in scattered areas (Halley & Rosell, 2003; Ritsema, 1869). However, ever since the abundant reintroductions of beavers in most Europe nations, *P. castoris* can now be seen in many areas such as Scotland and Poland, where they are believed to have existed before (Buchholz et al., 2008; Duff et al., 2013; Jørgensen, 2015). The Galapagos land iguana (*Conolophus subcristatus*) is a widespread species in the Galapagos archipelago and has been translocated to various localities, such as Seymour Norte in 1930s and Venecia in 1977 (Kumar et al., 2020). However, the potential effect of ectoparasites on translocation planning has not been studied in detail. It is known that tick infesting reptiles have the possibility to transmit viruses, bacteria and hemoparasites (Kho et al., 2015; Labuda & Nuttall, 2004). Lewbart et al. (2019), performed a health assessment on species of the Galapagos land iguana and Santa Fe iguana (*Conolophus pallidus*), including investigating the effects of blood-feeding ectoparasites. Their results suggested no differences in packed cell volume (PCV), body condition, refractometric plasma total solids and hemoglobin between the ectoparasitized and non-parasitized. There was, however, a slight negative correlation observed among ectoparasitized *C. subcristatus* and PCV, hemoglobin and body condition. In addition, Onorati and Gentile (2014) observed that iguanas with *Hepatozoon* infections had higher heterophils/lymphocytes (H/L) ratio than non-infected iguanas, which indicates stress (Davis et al., 2008). However, the question on whether ectoparasite and hemoparasite affects fitness of these iguanas are still unknown (Gentile et al., 2016). Further, a study on the Galapagos marine iguanas (*Amblyrhynchus cristatus*), suggested that ectoparasite infestation affects the annual energy budget by 5.4% due to tissue removal

and that individuals with lower ectoparasite infestations had a higher mating success (Hayes et al., 2004; Wikelski, 1999).

The Galapagos terrestrial iguanas (*Conolophus* spp) are an endemic group of three species with wide distribution in the Galapagos archipelago occurring across a range of habitats from sea level to 1700 meters. One species, the Galapagos land iguana (*Conolophus subcristatus*) occurs on at least nine populations on seven islands (Kumar, 2019). The conservation of the Galapagos land iguana has involved translocation of adults and juveniles to predator-free or predator-controlled areas since the 1930s ((Kumar et al., 2020). In general, these translocations are considered successful with local recruitment registered in all translocated or supplemented populations (A. Llerena & D. Rueda, pers. comm.). The genetics of Galapagos terrestrial iguanas has been extensively studied from looking at their phylogeny to intrapopulation genetic diversity (Snell et al., 1984; Tzika et al., 2008). Several aspects of their ecology have also been documented, for instance preferred diets (Christian et al., 1984), behavioral and morphological adaptations (Snell & Tracy, 1985) and population abundance (Kumar, 2019). Despite the numerous translocations of Galapagos land iguanas, some aspects of this practice have not been studied, for example the potential role of ectoparasites. Galapagos terrestrial iguanas are hosts to various species of ticks in the genus *Amblyomma* spp (Voltzit, 2007). This is a generalist genus of ticks that also parasitizes other species in the Galapagos Islands including the Galapagos giant tortoises (*Geochelone elephantopus*) and Galapagos marine iguanas (Voltzit, 2007).

A translocation of the Galapagos land iguana from Seymour Norte to Santiago was conducted in January 2019. A total of 2,120 iguanas were reintroduced back to their historical habitat, after 150 years since their extinction on Santiago Island. The extinction of the Galapagos land iguana on Santiago Island has puzzled scientists for many years. The available information, and knowledge about the susceptibility of island species to introduced predators

suggests that feral pigs (*Sus scrofa*) and feral dogs (*Canis familiaris*) introduced to Santiago Island in the late 1800s were responsible for the extinction of iguanas (Snell et al., 1984). Following decades of extensive management, Santiago Island is now free of feral pigs and dogs (Coblentz & Baber, 1987; Cruz et al., 2005). In addition, the introduced browsers, goats (*Capra hircus*) and donkeys (*Equus asinus*) have also been eradicated (Carrion et al., 2007; Cruz et al., 2009). Thus, the reintroduction of Galapagos land iguanas contributes to the ecological restoration of Santiago Island.

Prior to this translocation, Kumar (2019) analysed the effect of cat predation on the Galapagos land iguana's body condition index and juvenile recruitment across the range of the species, as part of the pre-translocation assessment of Galapagos land iguana populations. Her results suggested that land iguana populations found on cat present islands exhibited a positive correlation with body condition indices but had a negative correlation regarding juvenile recruitment; this could potentially increase the risk of local extinction from cat present populations (Marquez et al., 2019; Kumar, 2019). Kumar's (2019) research provided information to assist with the planning of Galapagos land iguana translocation, but other aspects not included in her research, for instance the effects of ectoparasites on iguanas, were identified as priorities for investigation. In 2018-2019, Island Conservation, the Directorate of the Galapagos National Park and Massey University, produced a translocation plan for Seymour Norte Iguanas to Santiago (Castaño & Ortiz-Catedral, 2019). One of the sections of this plan recommended investigating the effects of ectoparasite removal on the establishment of iguanas on Santiago Island, an element I had originally plan to examine in this thesis. Unfortunately, the ongoing COVID-19 pandemic prevented further field research. Therefore, I present only preliminary information in this chapter. Here I examine the effect of *Amblyomma* spp ectoparasites on Galapagos land iguanas on Seymour Norte.

Objective

1. To determine whether the presence/absence of ectoparasites has an influence on mass, SVL and body condition across sex, age and size classes of Galapagos land iguanas.

METHODS

Data collection

Data collection was conducted from 3rd to 15th of December 2018 a total of 2,120 Galapagos land iguanas were captured by hand on Seymour Norte, by a team of 20-30 park-rangers. Iguanas were captured following the general field methods described in Chapter 1. These land iguanas were originally translocated from Seymour Norte as a reintroduction back to Santiago island. Upon capture, morphometric measurements were taken for each iguana. These include; Snout Vent Length (SVL), the measurement from the tip of the chin to the cloaca of the animal; tail length, the measurement from the base of the tail to the tip of the tail; and the weight. In addition, for those iguanas that had no Passive Integrated Transponder (PIT), tags were inserted in the hind thigh of the iguana. The presence or absence of ectoparasites on the body of each iguana was determined by two trained veterinarians on site (Paula Castaño, Island Conservation and Andrea Loyola, Galapagos National Park). Due to the volume of iguanas to process, only two qualitative categories were used “presence” or absence” of ticks, and no estimation of number of ticks per individual iguana (Paula Castaño, Island Conservation and Andrea Loyola, Galapagos National Park). After examination each iguana was sprayed with permethrin (0.5%) on their body and placed in a burlap sack and transferred to a boat (Fig. 18). Permethrin is used routinely in the Galapagos Islands on tortoises (*Chelonoides* spp) as well as range of Galapagos finches (*Geospiza* spp, *Chamarhynchus* spp), to control the

introduced chick parasite *Philornis downsi* (Bulgarella et al., 2020; Flanagan, 2021). No negative effects of permethrin on the physiology of these species have been reported. Iguanas were transferred by boat to Santa Cruz Island, and kept in captivity on purpose-made corrals at the offices of the Directorate of the Galapagos National Park in Puerto Ayora. The captivity of Galapagos land iguanas lasted for 30 days in purpose-built corrals at the Directorate of the Galapagos National Park in Puerto Ayora, Santa Cruz Islands, according to recommendations outlined in the Galapagos land iguana translocation plan (Castaño & Ortiz-Catedral, 2019). The diet and other husbandry information can be found in Castaño and Ortiz-Catedral (2019).



Figure 18. Capture and transfer of Galapagos land iguanas from Seymour Norte. Left: Galapagos land iguanas captured on Seymour Norte inside burlap sacs; right: individual iguanas transferred to a speedboat for transfer to holding corrals in the Directorate of the Galapagos National Park. Photos. J. Ramirez.

Data analysis

I used R Studio version 1.3.1093 to conduct analyses. Data from a total of 2,068 Galapagos land iguanas were used for this analysis. It is important to note that I only included iguanas that were unambiguously classified as either male or female. I excluded the iguanas that were sex undetermined from this analysis. The total number of land iguanas with and without the presence of ectoparasites were different, 910 and 1,158 respectively. This prompted me to conduct a Shapiro-wilk test to test for normality in the data, the results showed that the data sets were non-normally distributed ($P < 0.05$). Despite this, I opted to use a paired T test to analyze the relationship of SVL, mass and body condition index of iguanas with and without ectoparasites. This is because the paired T-test is known to be quite robust and since the data

sets had relatively the same distribution and I did not have a ranked variable, I proceeded to use the more powerful paired T test instead of a Mann-Whitney U-test (McDonald, 2009). Finally, the average SVL, mass and body condition index of males and females were computed and compared with iguanas sampled on Fernandina, Isabela and Baltra island. The data of the land iguanas from these three islands were taken from the analysis conducted by Kumar (2019).

RESULTS

Results obtained from the analysis showed that there was an overlap between SVL and mass between sexes. There was no clear segregation between the females being smaller and lighter than males as known from iguana sexual dimorphism. The figure below shows the sexes of the sampled land iguanas in relation to their mass and SVL.

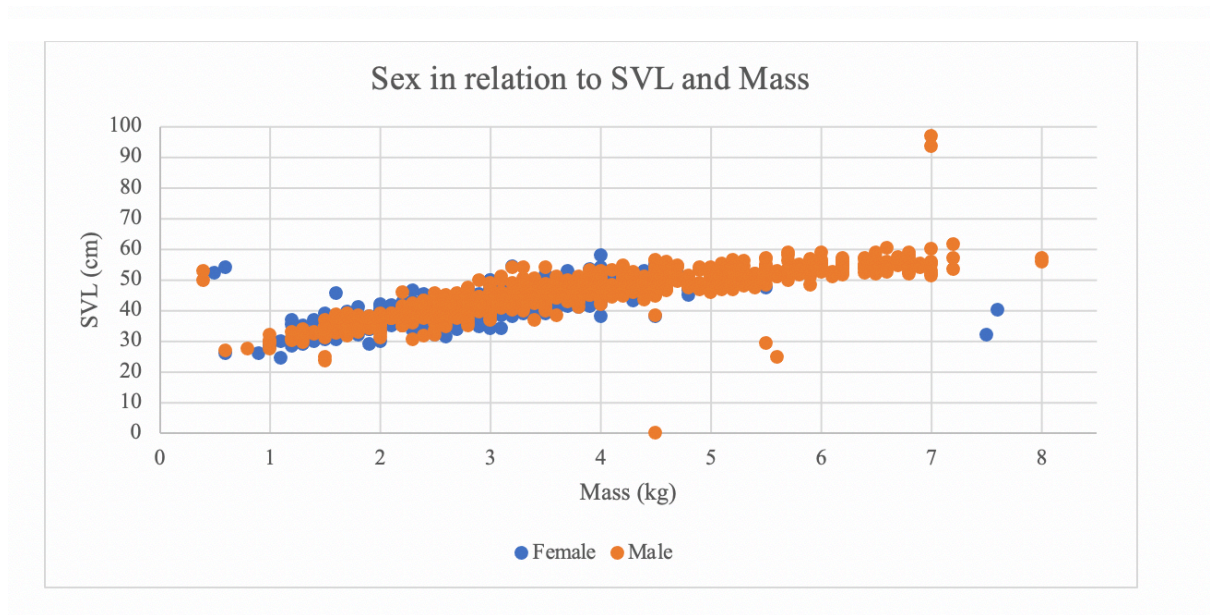


Figure 19. Relationship between SVL and mass for Galapagos land iguanas.

Results for average SVL, mass and body condition index of land iguanas found on Fernandina, Isabela and Baltra island were taken from Kumar (2019). The results show that the land iguanas that were translocated to Santiago island had the lowest average SVL, mass and body condition index as compared to those found on the other three islands. Female average SVL, however, was relatively similar to those sampled on Baltra island, 41.75 cm and 41.83 cm respectively.

Table 2. Table showing the average SVL, mass and body condition index between sex across Santiago, Fernandina, Isabela and Baltra island.

	Santiago	Fernandina	Isabela	Baltra
n (males)	852	140	151	72
n (females)	1216	230	71	49
Total	2068	370	222	121
Average SVL (cm) (males)	45.1	49.5	51.26	47.45
Average SVL (cm) (females)	41.75	43.33	45.03	41.83
Average Mass (kg) (males)	3.68	5.35	6.75	5.29
Average Mass (kg) (females)	2.85	3.42	4.5	3.52
Average Body Condition Index (males)	16.91	43.99	50.22	48.42
Average Body Condition Index (females)	12.74	41.94	49.82	47.76

***values Fernandina, Isabela and Baltra was taken from Kumar (2019).**

Results from the analysis showed that there was no obvious relationship between SVL, and mass of land with presence of *Amblyomma* ticks. Figure 20 shows and overlap in mass and SVL between iguanas that had ectoparasites. However, averages from the table showed iguanas infected with ectoparasites had lower average mass, SVL and body condition index. Upon further analysis, my paired T-test showed no correlation between the presence of ectoparasites

and SVL, mass and body condition index of the land iguanas sampled ($P > 0.05$). The figure below shows the presence and absence of ectoparasitic iguanas in relation to SVL and mass.

Table 3. Table showing the average mass, SVL and body condition index between infected and uninfected iguanas with ectoparasites.

	Ticks	No Ticks	P-value
n	927	1193	
Average mass (in kg)	3.11	3.21	0.696
Average SVL (in cm)	42.8	43.06	0.548
Average body condition index	14.03	14.56	0.72

*indicates significance

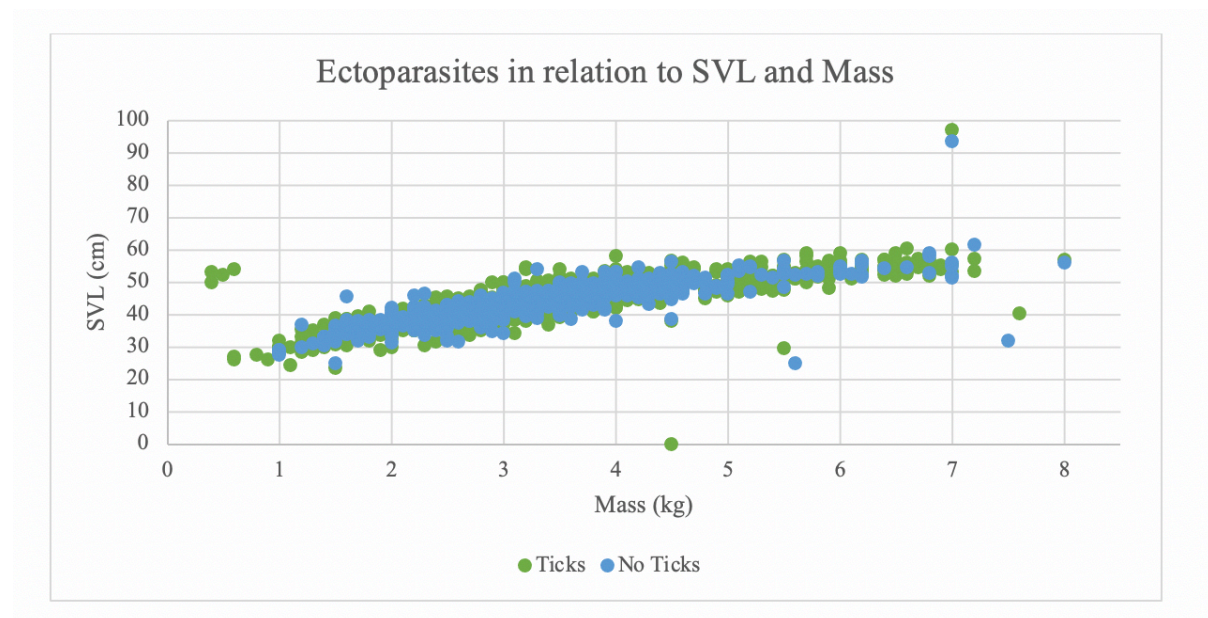


Figure 20. Figure showing the SVL and mass of iguanas with and without the presence of ectoparasites within the group of Galapagos land iguanas destined for translocation to Santiago Island.

DISCUSSION

Differences in mass, SVL and body condition index among sex classes

Based on the results acquired, there was no clear differentiation between average mass and SVL among sex classes, despite sexual dimorphism being known. However, when comparing the averages of mass, SVL and body condition, the results showed that land iguanas that were selected for translocation had the lowest mass, SVL and body condition when compared to those found on Fernandina, Baltra and Isabela island. One potential explanation for this could be due to the fact the land iguanas that were translocated to Santiago were originally taken from Seymour Norte. Kumar (2019), had obtained results showing low average mass of male and female land iguanas, 5.62 kg and 3.42 kg respectively. It was mentioned and speculated that the island of Seymore Norte has reached carrying capacity and an increase in competition for food resources, as there was observations made of several land iguanas grazing on the same *Opuntia* cacti (*Opuntia echios zacana*). More importantly, there were high juvenile counts recorded during the field visit which could provide an explanation to the low average mass, SVL and body condition index (Kumar, 2019). I propose that long-term post translocation monitoring be continued to ensure growth in these translocated iguanas. In addition, the average mass, SVL and body condition indices should be measured post-release to provide a post translocation report on the land iguanas.

Effects of ectoparasites on mass, SVL and body condition index

Next, the results from my analysis showed that there was no correlation between the presence or absence of ectoparasites in relation to the land iguanas' SVL, mass and body condition index ($P > 0.05$). This is surprising as most studies have shown that ectoparasites do affect the host's in one way or another. Studies conducted on the relationship between birds and ectoparasites are abundant (Cantarero et al., 2013; Christe et al., 1996; Johnson & Albrecht, 1993). In a study conducted on Great tit (*Parus major*) and Hen fleas (*Ceratophyllus gallinae*), the results suggested that nestlings of parasitized broods not only doubled their begging rate but also showed increased frequencies of feeding trips of 50% from parents and food competition among siblings also increased (Christe et al., 1996). In another example, a study conducted by (Møller et al., 1994) suggested that ectoparasites found on House Martins (*Delichon urbica*), showed a correlation between average metabolic rate and the intensity of ectoparasite infestations. They found that as intensity of infestation increased, so did the average daily metabolic rate. In relation to iguanas, results from a study conducted by Wikelski (1999) on the Galapagos marine iguanas (*Amblyrhynchus cristatus*), suggested that the minimum cost of ectoparasite infestation would have 5.4% lower annual energy budget due to tissue removal, excluding potential internal infections. In addition, results from another study conducted on the Galapagos marine iguanas by Hayes et al. (2004), suggested that marine iguanas with lower parasite loads, especially ticks, had a higher mating success rate. This result supports the theory from Hamilton and Zuk (1982), proposing that individuals that mate more successfully possess the genes for a resistance to parasites. Furthermore, individuals that are pathogen resistance are able to signal their quality by the development of extravagant ornamentation or brighter colouration. Unfortunately, this theory has had some debates on the relationship between ectoparasite loads, male phenotypic traits and female choice in some species (Poulin & Hamilton, 1997). For example, a study conducted on Sand lizards (*Lacerta*

agilis), suggested that females do not reject males based on their parasite loads (Olsson & Madsen, 1995).

A possible explanation for the non-significant correlation between ectoparasites, mass, SVL and body condition index could be that the infestation of ectoparasites on the iguanas sampled were not severe enough. The observations recorded during the field were limited to a qualitative estimation of the presence or absence of *Amblyomma* ticks, rather than an estimation of ectoparasite load. This represents a major limitation of the analysis presented here. Coupled with my inability to access field sites for a BACI (Smith, 2014) comparison between the control and translocated population of iguanas (see COVID-19 statement). Hence, the results achieved does not provide me sufficient evidence to conclude that there are no effects of ectoparasites on the iguanas' mass, SVL and body condition index. However, during a health assessment study by Lewbart et al. (2019), the results suggested that ectoparasites had an effect on PCV, hemoglobin levels and body condition.

I propose that the relationship between the severity of ectoparasite infestation on the Galapagos land iguanas, mass SVL and body condition be investigated; especially the long-term effects of ectoparasites. Studies have shown that ectoparasites have an impact on long-term survivorship on their hosts. For example, cliff swallows (*Hirundo pyrrhonota*) infected by ectoparasites (cimicid bugs, fleas and chewing lice) were observed to have at least 12% lower annual survivorship as compared to non-infected birds (Brown et al., 1995). Some studies have also shown that ectoparasites have an effect on reproduction. Oppliger et al. (1994) conducted study on great tits (*Parus major*) and hen fleas (*Ceratophyllus gallinae*) and suggested that while no significant effects were observed in clutch sizes; hatchling success and brood size at hatching were significantly smaller in infested nests. Furthermore, nest desertion after egg-laying and hatching were significantly higher in infected nests.

More importantly, the type of ectoparasites should be taken into consideration. Interestingly enough, a study conducted on the Galapagos male marine iguanas showed an increase in tick (*Ixodida* spp) infestation when body mass of the iguanas was low and a reduction in mite (*Acari* spp) infestations (Hayes et al., 2004). *Amblyomma* species are also known to alter the cells of its host. For example, the saliva of *Amblyomma cajennense* are known to have the capability to modulate host immune cells, including dendritic cells (Carvalho-Costa et al., 2015). It is important to obtain more information on this as if this is the case, it is not ideal to translocate individuals with low body condition indices. A way to conduct this would be to count the number of ectoparasites on the sampled individual, focusing on areas such as the tail vein and pre-dorsal spine vein where *Ornithodoros* spp are usually found; furthermore, the ventral part of the body and skin folds should be checked for *Amblyomma* spp where they are most likely to be found (Wikelski, 1999). More specifically, to determine blood extraction from ectoparasites; ectoparasites should be removed from their hosts when they appear full of blood and submerged in water to measure their full body volume.

The information presented here is preliminary due to logistical limitations. However, this exercise has been useful to identify ways to investigate the effects of *Amblyomma* ectoparasites on health parameters of Galapagos land iguanas. One recommendation for future studies is including a seasonal sampling on a control population (i.e., Seymour Norte) and a parallel sampling on the reintroduced population on Santiago Island. This approach could allow an analyses of the temporal changes in the health of iguanas according to ectoparasite load. Another recommendation consists on estimating ectoparasite load per individual. The current available data (i.e. presence/absence) is insufficient potential subtle differences in weight or other parameters. These and other recommendations (see Chapter 4) can assist the Directorate of the Galapagos National Park in fulfilling its mission to safeguard the diversity of the Galapagos Islands.

**CHAPTER 3: AN UPDATE ON THE POPULATION STATUS AND STATE OF
KNOWLEDGE OF *CONOLOPHUS* IGUANAS**



Figure 21. The elusive Pink iguana (*Conolophus marthae*) in its habitat, Wolf Volcano, Isabela. Photo: L. Ortiz-Catedral.

ABSTRACT

Obtaining information on population status is paramount in the conservation of species. Information such as adult sex ratios and body condition can provide conservationists information on the demographics of a population. The Galapagos land iguana (*Conolophus subcristatus*) inhabits several different islands in the Galapagos archipelago, and the demographics of some of these populations have not been studied in recent years. More importantly, the information regarding the elusive Galapagos pink land iguana (*Conolophus marthae*) is few. I compare the adult sex ratios of the Galapagos land iguanas found on Fernandina, Baltra and Seymour Norte and also the elusive Galapagos pink land iguana found on Isabela island. I further compare the average mass, SVL and body condition index between males and females of iguanas sampled on each individual island. I also present some information on Galapagos land iguanas recently reintroduced to Santiago Island. The results show a significantly higher proportion of females than males on Fernandina island. I further analysed the average mass, SVL and body condition index between male and female land iguanas found on all four islands. The results achieved showed that the land iguanas on Seymour Norte had the lowest average mass, SVL and body condition index, which could potentially mean the island of Seymour Norte have reached its carrying capacity. Finally, since the mass and body condition index of the Galapagos pink land iguana could not be obtained, I used average SVL to infer potential population growth. The results suggest that the pink iguanas sampled have all reached sexual maturity. The adult sex ratios found on Fernandina island reflect seasonal migration of females to reach nesting sites near the summit rim, but more sampling needs to be conducted again to ensure accurate sampling of adult sex ratios. Lastly, more research should be conducted on the elusive pink land iguana, important information such as life history traits should be investigated.

INTRODUCTION

The implementation of conservation actions to protect species threatened with extinction requires a substantial amount of information, one of which is population abundance estimates (Reed et al., 2003). Population abundance is the number of individual animals globally and it is a fundamental ecological parameter that is crucial when making management and conservation decisions (Caughley & Gunn, 1996; Gaston, 1994; Lancia et al., 1994). An analysis conducted on 35 species of turtles and squamates found within the islands of the Mediterranean Sea suggested population abundance and habitat specialization significantly affected the variation seen in the studied species extinction rates (Foufopoulos & Ives, 1999). Population abundance of a species and the geographical range they inhabit are not independent from one another. Previous analyses on plants and birds indicate that species occurring over large geographical ranges tend to have greater local abundances than those that are geographically restricted (Gaston & Lawton, 1990); although, it is important to note that there usually some outlier species that are widely spread and also rare (Lawton, 1993). The typical correlation between high population abundance and large geographical ranges serves as an important conservation tool as it allows conservation biologists to understand that geographically restricted taxa usually have smaller local populations, making them highly susceptible to decline.

Population density estimates (i.e., the number of individuals within a geographical area), on the other hand, can provide information on demographic stochasticity, environmental stochasticity and inbreeding depression (Lancia et al., 1994; Ryan & Siegfried, 1994). For example, 11 lions (*Panthera leo*) found in Madikwe Game Reserve showed results of inbreeding just after five years of reintroduction despite management plans to prevent this. This included translocating individuals, trophy hunting and culling of sub adult lions. Furthermore, population growth within the reserve decreased significantly when females were reported to

delay reproduction and decrease litter sizes when the reserve had reached carrying capacity of 61 lions (Trinkel et al., 2010). Counting every single individual from a particular species is nearly impossible except for a few extremely rare species. One example is the Po'ouli (*Melamprosops phaeosoma*), an endemic species from Hawaii. The Po'ouli was reduced to only three individuals in the rainforests of Maui and every individual was accounted for during intensive studies (Groombridge et al., 2004). Genetic sexing of these three individuals have produced conflicting results but available information has indicated that the sex ratio of the remaining population consists of one male and two females. In addition, during the six-year observation study conducted, they have not been observed together, indicating the unlikely possibility of breeding (VanderWerf et al., 2003). It is important to note that this scenario is extremely rare and conservation biologists today require the estimates of larger populations of species found in different geographical locations with a larger number of individuals.

Extinction risk in species is inversely associated with population growth rates and one way to ensure population growth is a species reproductive capability. From a management perspective, understanding the number or proportion of males and females in a population, or adult sex ratio, and age to first reproduction are valuable parameters to assess the viability of populations of conservation interest. For instance, historically in the critically endangered Kakapo (*Strigops habroptila*) females made up only a small proportion of the adult population, creating a dilemma whereby the management at the time successfully produced more kakapo chicks, but the majority of survivors to reproductive age were males (Clout et al., 2002). Since the early 2000s emphasis on adjusting the supplementary feeding of reproductive females to fine-tune the proportions of males and females has resulted in more females being produced (Robertson et al., 2006).

Obtaining knowledge on a species population's sex ratios is important because it provides information on whether conservation intervention is required to aid in the

survivability of the species. For example, species affected by high sex biased mortality rates will affect their population growth in obvious ways. If a species has higher male counts as compared to females, this directly affects fecundity as it reduces juvenile recruitment. Sex ratio biasness can be altered in many ways, some include sex differences in behavior, size, morphology or simply by hunter's preferences (Bunnefeld et al., 2009; Marealle et al., 2010; Tryjanowski et al., 2009). Furthermore, sex ratios can be altered by environmental changes. For example, two close related species of salamanders, *Pleurodeles poireti* and *Pleurodeles waltl* showed opposite results when larvae were kept at high temperatures ranging from 30 °C to 32 °C; genotypic males became phenotypic females and genotypic females became phenotypic males respectively (Dournon et al., 2003). Many species of reptiles are known to show sensitive sexual differentiation when eggs are incubated at certain temperature, at high temperatures, lizards and crocodilians produces males but chelonians produce females (Deeming & Ferguson, 1988). Maturity age is known to show a primary correlation in extinction risk Hutchings et al. (2012). For example, Devil rays (*Mobula* spp) studied in Mexican fisheries were reported to potentially reach local extinction at low fishing mortality; this is due to the fact that Devil rays exhibits low somatic growth resulting in low reproductive output, causing lower population growth (Pardo et al., 2016). The need to take into account a species reproductive capability is imperative today due to the high anthropogenic threats most species face globally.

The Galapagos terrestrial iguanas (*Conolophus* spp) are a group of three species with marked differences in abundance and distribution. The Galapagos land iguana (*Conolophus subcristatus*) is the most widespread and numerous of the three species, occurring on at least nine populations on seven islands on an area of 5000 km² (Kumar, 2019; Arteaga et al., 2019). The two largest islands where Galapagos land iguanas occur; Santa Cruz and Isabela, are inhabited and a range of introduced predators overlap in distribution with iguanas, representing

a threat to local recruitment (Kumar, 2019). The Santa Fe land Iguana (*Conolophus pallidus*) is restricted to the island of Santa Fe, occupying an area of approximately 24 km². Although restricted to a smaller area, the Santa Fe Iguana inhabits an area free of introduced predators and the population is considered viable, with approximately 4500-5800 individuals (Marquez et al., 2019). The Pink iguana (*Conolophus marthae*) is the most range-restricted species of iguana in the Galapagos archipelago, occurring in an area of approximately 11 km² on Wolf Volcano, on the northern part of Isabela Island (Arteaga et al., 2019). However very little is known about this species in the field due to the significant logistical effort required to reach the area where Pink iguanas are found.

The management of the Galapagos terrestrial iguanas by the Directorate of the Galapagos National Park has largely benefited from research studies from visiting scientists (list some papers here). In 2017 – 2019 there was substantial field activity on various iguana populations thanks to fieldtrips to Baltra and Seymour Norte to assess the body condition and adult sex ratio of Galapagos land iguanas, the historical reintroduction of Galapagos land iguanas to Santiago Island, and an expedition to Wolf volcano in search for the elusive Pink iguana. All of these trips responded to the necessity by the Directorate of the Galapagos National Park to generate a baseline of population status of *Conolophus* iguanas. In this Chapter, I summarize the findings of these field visits and provide novel information on the population parameters of the populations visited. This information can help in the conservation of these reptiles. Specifically, I describe morphological parameters of iguana populations of conservation interest, and also compare changes in body mass of reintroduced Galapagos land iguanas to Santiago Island.

METHODS

Data collection

Data collection was conducted in December 2017 on Baltra island, in January and July 2018 on Seymour Norte and Fernandina respectively and on Santiago Island in February and August 2019. Also, in late February 2019, an expedition to Isabela island (Wolf Volcano) took place. The locations for this study are of conservation interest due to the history of the populations of iguanas: The Galapagos land iguanas of Baltra Island had decline to local extinction by 1940s, however an artificial population introduced to nearby Seymour Norte in the 1930s acted as insurance (Hofkin et al., 2003; Kumar, 2019). Repatriation of captive-bred Galapagos land iguanas in the 1990s provided an initial boost to their local recovery (Cayot et al., 1992) greatly aided by the eradication of feral cats from Baltra Island in 2001-2003 (Phillips et al., 2005). The Baltra population of iguanas has recovered but the Directorate of the Galapagos National Park commissioned a visit in December 2017 to assess the body condition of iguanas and their gross density (iguanas per hectare). The sampling of iguanas in the field is conducted to capture every iguana sighted along pre-determine tracks that criss-cross the habitat. To avoid re-capturing iguanas already processed every iguana was painted with water soluble paint on the tip of the tail as described by Kumar (2019).

Field trips lasted an average of 5.5 days (range 3 to 11) with a team of three to five participants. Iguanas were captured and handled according to the general field methods described in Chapter 2 and following the approved protocols by the Directorate of the Galapagos National Park (Appendix I). On Baltra and Fernandina iguanas were also individually photographed (Appendix IV, V). The iguanas captured on Seymour Norte, represent a different group than the one presented in Chapter 2, as the sampling at that locality occurred 11 months before data collection for iguanas destined for translocation to Santiago

Island. Galapagos land iguanas on Baltra, Seymour Norte and Fernandina Islands inhabit Palo santo (*Bursera graveolens*) scrub, arid scrublands and Opuntia/Palo santo scrub (Fig. 22). Preliminary surveys indicate they occur more or less evenly across habitat patches, or vegetated spots on these islands, outside of the breeding season which extends from April to June (A. Llerena, pers. comm). The localities visited, have been the focus of semi-regular surveys since the year 2002 (Marquez et al., 2019; A. Llerena, pers. comm.). The most recent survey on Baltra and Seymour Norte Islands took place in 2012 (see Kumar, 2019), and on Fernandina in 2017.

Lastly, the data for Santiago Iguanas was collected in a similar manner but due to the scattered distribution of the then recently reintroduced iguanas, the sample size is very limited. In spite of this limitation, the data obtained is useful to compare changes in body mass of iguanas in a new environment. I also report here field observations on the behaviour of reintroduced Galapagos land iguanas. The sampling of Pink iguanas (*Conolophus marthae*) on Wolf Volcano was conducted as part of an expedition led by the Directorate of the Galapagos National Park. This expedition was intended to obtain a ‘snapshot’ of the status of the Pink iguana population. Specifically, there was an interest to determine age and size classes, and to determine whether reproductive pairs were present in the population.

Due to the high value of Galapagos terrestrial iguanas in the black market (W. Tapia, pers. comm, see also Gentile et al., 2013) and the high threat of extinction on the species, no geo-spatial data is included in this thesis following recommendations by the Directorate of the Galapagos National Park. A visit to assess the status of the only population of Santa Fe iguana (*Conolophus pallidus*) on Santa Fe island was discussed for 2020, however due to the ongoing COVID-19 pandemic this component was not pursued further. Although no data on Santa Fe iguanas were collected, there are some recommendations for the future conservation of the species, discussed in Chapter 4.

Data analysis

There were morphological differences, such as weight, sample size, and SVL, between each iguana sampled from each island. A total of 204 Galapagos land iguanas (*Conolophus subcristatus*) and 22 Galapagos pink land iguanas (*Conolophus marthae*) are included in this analysis. Averages of mass, SVL and body condition index were taken for the total population of iguanas on each individual island. Unfortunately, the mass of the Galapagos pink land iguana could not be accurately taken because the portable pesola scale was lost in during the hike to the summit of Wolf volcano. Body condition was calculated using Laurie and Brown (1990) index given for marine iguanas (*Amblyrhynchus cristatus*):

$$\text{Body condition index} = \text{Body mass} / \text{SVL}^3$$

The body index formula is proportional measurement that is body mass relative to SVL, quantifying the “fatness” of an individual. Individuals with large or intermediate SVL lengths with lower masses will show lower body indices indicating a thinner individual on average and vice versa for those with higher masses. The indices have no relation to the age of the individual, thus is purely just a value to indicate the level of fatness of the iguana at the time.

For all of the analysis conducted in this chapter, I used RStudio version 1.3.1093. In order to further compare the population sex ratios of iguanas on each island, I conducted a two-sided Binomial test. Since there were differences in sample sizes and temporal sampling, I conducted a Shapiro-Wilk test to test for normality in the data. The results obtained varied; showing non-normal data for the iguanas sampled on Fernandina and Seymour Norte ($P < 0.05$). However, the iguanas sampled on Baltra and Isabela showed normal data ($P > 0.05$). With this in mind, I was unable to perform any statistical analysis as the results would prove

to be inaccurate. To compare the weight, SVL and body condition of the iguana populations, I used averages that were calculated using Microsoft Excel version 16.42.



Figure 22. Examples of habitat types inhabited by *Conolophus* iguanas. Top left, Baltra Island Palo Santo scrub; Top right: Fernandina island arid scrub; bottom left: Seymour Norte *Opuntia*/Palo Santo scrub; bottom right: Wolf Volcano *Scalesia*/grassland shrub. Photos: L. Ortiz-Catedral.

RESULTS

Adult Sex ratio

The results collected in section is separated into population sex ratios of iguanas found on Fernandina, Baltra, Seymour Norte and Isabela islands. The iguanas found on Isabela island were Galapagos pink land iguanas (*Conolophus marthae*), while on the other three islands, iguanas sampled were Galapagos land iguanas (*Conolophus subcristatus*). The column labelled ‘Undetermined’ shows the sexes of the iguanas that were inconclusive. Results from the analysis shows significantly higher proportions of females as compared to males on Fernandina island ($P < 0.05$). However, the results from Baltra, Seymour Norte and Isabela islands showed no significance in population sex ratios. All islands consisted of iguanas that were sex undetermined, but Seymour Norte had the highest value of 24.

Table 3. Table showing population sex ratios of Galapagos land iguanas (*Conolophus subcristatus*) found on Fernandina, Baltra and Seymour Norte; and Galapagos pink land iguana (*Conolophus marthae*) on Isabela island.

	Males	Females	Undetermined	Total	P value
Fernandina	30 (30%)	65 (65%)	5 (5%)	100 (100%)	0.0004*
Baltra	20 (57%)	14 (40%)	1 (3%)	35 (100%)	0.3915
Seymour Norte	28 (41%)	17 (24%)	24 (35%)	69 (100%)	0.1352
Isabela	14 (61%)	8 (35%)	1 (4%)	23 (100%)	0.2863
*indicates significance					
*iguanas sampled on Isabela are Galapagos pink land iguanas (<i>C. Marthae</i>)					

Differences in length, mass and body condition

Results from my analysis shows that the Galapagos land iguanas (*Conolophus subcristatus*) sampled on Fernandina island had the highest average in mass (4.92 kg) and the iguanas sampled on Seymour Norte had the lowest (3.5 kg). However, iguanas sampled on Baltra showed the highest averages in SVL (49.27 cm) and Seymour Norte iguanas were found to be the lowest (12.45 cm). Body condition index of iguanas sampled on Fernandina and Baltra island were found to be relatively similar while Seymour Norte was the lowest.

Table 4. Table showing averages in mass, SVL and body condition index of all iguanas sampled on Fernandina, Baltra, Seymour Norte and Isabela island.

	Fernandina	Baltra	Seymour Norte	Isabela
N	100	35	69	23
Average mass (kg)	4.92	4.81	3.5	NA
Average SVL (cm)	47.12	49.27	12.45	46.94
Average body condition index	22.39	22.57	16.2	NA
*iguanas sampled on Isabela are Galapagos pink land iguanas (<i>C.Marthae</i>)				

The averages in SVL among males from Fernandina and Baltra island are relatively similar, 53.14 cm and 53.80 cm respectively. Average SVL of males sampled from Seymour Norte and Isabela were found to be the lower than the other two islands, 51.84 cm and 51.19 cm respectively. Average female SVL showed varied results, with Isabela iguanas having the highest average SVL, 45.36 cm, followed by Fernandina, 44.83 cm. The average SVL of females from Baltra and Seymour Norte were found to be relatively similar, 43.34 cm and

43.82 cm respectively. Averages in masses varied slightly in both sexes. Average mass of males sampled on Fernandina island was found to be 6.64 kg, while Seymour Norte was recorded to be the lowest, 5.15 kg. Average mass in females across Fernandina, Baltra and Seymour Norte was similar to males, with land iguanas on Fernandina being the highest, 4.24 kg and Seymour Norte being the lowest, 3.05 kg. Finally, average body condition of males and females was also the highest in land iguanas found on Fernandina, 31.25 and 18.82 respectively; Seymour Norte was also recorded to be the lowest in male and female body condition, 23.22 and 13.75 respectively.

Table 5. Table showing the averages of lengths, mass and body condition index between sexes among Fernandina, Baltra, Seymour Norte and Isabela island.

	Fernandina	Baltra	Seymour Norte	Isabela
n (males)	30	20	28	14
n (females)	65	14	17	8
Total (N)	95	34	45	22
Average male SVL (cm)	53.14	53.80	51.84	51.19
Average females SVL (cm)	44.83	43.36	43.82	45.36
Average male mass (kg)	6.64	5.89	5.15	NA
Average female mass (kg)	4.24	3.39	3.05	NA
Average male body condition index	31.25	28	23.22	NA
Average female body condition index	18.82	15.38	13.75	NA
*iguanas sampled on Isabela are Galapagos pink land iguanas (<i>C. Marthae</i>)				

***Note: weight of the *C. Marthae* could not be taken**

Current status of populations

Baltra Island

A total of 34 individuals (20 males; 14 females) were captured, in an area of 93 ha, over a period of 18 field hours, for three days. The gross density of iguanas in the area sampled is 0.36 iguanas per hectare. No juveniles were encountered during this sampling period, however only two of the captured iguanas (6%) were recaptures (i.e., individuals marked previously).

Seymour Norte Island

On Seymour Norte, a total of 69 iguanas were captured in an area of 28 ha, over a period of 24 hours, for three days. The gross density of iguanas in the area sampled is 2.4 iguanas per hectare. Of the total of iguanas captured, 45 (65%) were adults, and the other 24 (35%) were juveniles. Five juveniles were captured (5% of total) and the rest were adults. Only one iguana (0.01%) was a recaptured individual.

Fernandina Island

A total of 100 iguanas were captured in an area of 57 ha, over a period of 45 field hours, for five days. The gross density of iguanas in the area sampled is 1.75 iguanas per hectare. Of the total of iguanas captured, only seven (0.07%) were recaptures.

Santiago Island

A total of 15 iguanas were captured in an area of 82 ha, over a period of 19 field hours for three days. Of these, seven were males and four females, with another four classified as juveniles but their sex could not be determined. Another 12 iguanas were sighted but could not be captured. The gross density of iguanas including captured and sighted iguanas is 0.32 iguanas per hectare. Evidence of iguana presence on Santiago Island was indicated by track marks and chew marks on vegetation (Fig. 23). Of interest was the recapture of a juvenile (PIT tag: 982126055989106) which weighed 2 kg. A change of 0.8 kg relative to the weight in December during capture on Seymour Norte 262 days prior (Fig. 24).

Pink Iguana

Wolf Volcano

A total of 22 iguanas were captured, eight females and 14 males, over a period of 25 field hours over 11 days. The area sampled is equivalent to 438 ha. With the exception of three pink iguanas, all iguanas encountered were captured. The gross density of pink iguanas is 0.06. No juveniles were encountered. Of the total of pink iguanas captured, three were new individuals, or individuals without PIT tags. Two of these were females and one male. In addition, the skeleton of a dead pink iguana was located (PIT tag ID: 010623086).



Figure 23. Indirect evidence of presence of reintroduced *Conolophus subcristatus* on Santiago Island. Top left: fresh dropping; top right: distinctive iguana bite marks on cactus (*Opuntia galapageia*) stem; bottom left: feet and tail tracks on loose substrate, bottom right: iguana tracks amongst vegetation on Santiago Island. Photos: L. Ortiz-Catedral.



Figure 24. Individual 982126055989106 (PIT tag reference) recaptured on Santiago Island.

Photo. Jean Pierre Cadena.

DISCUSSION

Inter-population comparison of adult sex ratios

The proportion of mature females on Fernandina island, relative to mature males, appears to be higher (Binomial test, $P < 0.05$) than during previous sampling events at the same site. However, there were no significance between the proportions of mature males and females sampled on Baltra, Seymour Norte and Isabela island. Sex ratios in animal populations are affected by the quality and stability of the immediate habitat, life history traits, competition and dispersal, and environmental effects (Sapir et al., 2008). For example, the adult sex ratio

of the endangered Allens Cay rock iguana (*Cyclura cyclura inornate*) found in the Bahamas was changed to nearly 1:1 in 2006; results suggested that this was due to the recovery from intense harvesting of females over 100 years and also the removal of large males by poachers and tourists (Smith & Iverson, 2006). Furthermore, environmental factors such as temperature plays a major role in the reproduction of reptiles, as it can be a determinant for the sex produced in a hatchling (Deeming & Ferguson, 1988). Results from a study conducted on the Red-eared slider (*Trachemys scripta elegans*) suggested that their embryonic growth is highly influenced by the incubation conditions inside the nest. Nests with cool and wetter substrates produced 100 % of males while warm and dryer nests produced only 42% males (Sifuentes-Romero et al., 2018).

Obtaining sex biased results can sometimes relate to the time of sampling. For example, a study conducted by Hayes et al. (2016), suggested that male Acklins ground iguanas (*Cyclura rileyi nuchalis*) found on North Cay were significantly more likely to be seen as compared to females. However, the San Salvador rock iguanas (*Cylcura rileyi rileyi*) found on Green Cay suggested equal encounter occurrence from both sexes. The results from both of these studies suggests that the time of sampling does affect the occurrence of iguanas; with further analysis suggesting that there was no correlation between the occurrence of a sex and the time of day. In addition to time of sampling, location of sampling could also produce sex biased ratios results. According to Rauch (1985), female marine iguanas (*Amblyrhynchus cristatus*) prefer locations that have a higher number of shaded areas and low temperatures of rocks at noon while males were noted to follow their individual site preferences rather than female density in their choice of territory. Hence, if sampling was conducted in a particular area with high density of females, it is wrong to assume that sex ratios of a species is skewed.

A potential explanation for a significantly higher proportion of females sampled on Fernandina island is due to the time and location of population sampling. The sampling of the

Galapagos land iguana on Fernandina island was conducted near the crater rim in July. Adult female iguanas can be easily differentiated from males based on their morphological features (see Chapter 1). According to historical research conducted on land iguanas found on Fernandina island, females in the western part of the island leave mating areas to migrate to the crater rim prior to July as the first two weeks of July marks the peak laying season (Burghardt & Rand, 1982; Werner, 1983). Furthermore, previous analysis conducted by Kumar (2019) suggested similar female biased results when sampling was conducted during the dry season (June to November). More interestingly, sampled iguanas at the same location during wet seasons (December to May) were male biased indicating a segregation in habitats based on sex. Additionally, the analysis from Marquez et al. (2019) on the land iguanas sampled near the crater rim in July 2006 suggested high female biasness (25.40% males). These results strongly support my reasoning as to why I believe the adult sex ratio obtained from my analysis is not an accurate estimation of the Galapagos land iguana population found on Fernandina island. The argument for the global change in temperatures from climate change could be made to explain the high female biased result. For example, an analysis on the relationship between local climatic variation and the offspring sex ratio on Painted turtles (*Chrysemys picta*) showed that a small increase in mean temperature ($< 2^{\circ}\text{C}$) could drastically skew sex ratios (Janzen, 1994). However, this does not provide enough evidence to relate to the results I have obtained, as the population sex ratios of land iguana populations from Baltra and Seymour Norte showed no significance (Binomial test, $P > 0.05$).

Furthermore, islands found in the Galapagos archipelago all have relatively similar temperatures all year round with wet seasons (December to May) range from 27°C to 32°C and dry seasons (June to November) range from 21°C to 27°C . I strongly suggest that the adult sex ratios for the Galapagos land iguanas found on Fernandina island be conducted again to ensure the population sex ratio was accurate as it influences population growth and suggests if

conservation interference is required. Comparing adult sex ratios from different seasons should be conducted for significance to provide a more accurate estimation of adult sex ratio of Galapagos land iguanas found on Fernandina island.

Comparison of mass, SVL and body condition index

The average mass, SVL and body condition index of the land iguana populations from Fernandina and Baltra island were found to be relatively similar. On the other hand, average mass, SVL and body condition index from the land iguana population sampled on Seymour Norte was the lowest compared to the other two islands. There was a high number of land iguanas sampled on Seymour Norte with undetermined sexes (35%). The assumption I have made was that they were juveniles as it is difficult to identify a juvenile without a cloacal probe. Furthermore, there were no photographs available from the field visit that could allow me to identify them based on their coloration or pronounced head crests. Additionally, results from a recent analysis conducted by Kumar (2019), showed that there were high proportions of juveniles recorded (28.65%). Since averaging the mass, SVL and body condition index of the total sample size from each island was an inaccurate comparison, I decided to exclude the iguanas that were sex undetermined. The results obtained showed that all three populations of iguana (Fernandina, Baltra and Seymour Norte) showed relatively similar SVL between males and females. However, average mass and body condition index from the land iguanas on all three populations of land iguanas slightly differed. Body condition index have often been used in ecological and morphological studies to measure an individual's fitness. It is the proportionate measurement that is body mass relative to SVL. This quantifies the individual's 'fatness'. Individuals with larger SVL lengths and lower masses would produce lower body condition indices and vice versa. These indices obtained have no relation to the age of the

individual but primarily exhibits the level of fatness of the iguana at the point of capture. According to Costantini et al. (2005), the body condition index of individuals < 25 are considered as starving or dying individuals. Only the male iguanas sampled on Seymour Norte exhibited body condition index < 25 , while females sampled on all three islands exhibited < 25 body condition index.

A possible explanation for this could be the growing population on Seymour Norte and Baltra island. A study conducted by Dudek et al. (2015) suggested that juvenile lizards have a proportionally longer body and a smaller body mass; this suggests that the priority development in a lizard's body is non-linear and that investment in growth is contributed to their body length first rather than mass. This result corroborates with the results achieved by Kumar (2019) with average mass of male and female land iguanas on Seymour Norte being relatively low (5.62 kg and 3.42 kg respectively) and the high juvenile counts obtained during her field visit (28.65%). Another potential explanation could be due to Baltra island and Seymour Norte reaching carrying capacity. Both Baltra and Seymour Norte have significantly smaller land masses (21 km² and 1.9 km²) as compared to Fernandina island (642 km²). A high density of land iguanas on a small area size would result in increased competition for resources. The population on Baltra According to Kumar (2019), her analysis showed an estimation of approximately 7,979 iguanas inhabiting Seymour Norte. Furthermore, during her field visit, she observed several land iguanas grazing on the same *Opuntia* cacti (*Opuntia echios zacana*). Additionally, she observed degraded conditions, with rotting or black bases and minimal fresh green growth on many of the *Opuntia* cactus found on the island. Having a high density of land iguanas inhabiting a small area size such as Seymour Norte could potentially affect population growth by reducing juvenile recruitment. For example, a study conducted on the relationship between population density and reproduction in white rhinos (*Ceratotherium simun*) found in Zimbabwe, showed decreased rates of population growth and calf recruitment; the results

suggested that as density within the fenced area increased, and females were noted to delay reproduction (Rachlow & Berger, 1998). It is expected that if the no conservation interference is implemented, the body condition and the reproductive output of the current population of land iguanas on Seymour Norte will continue to decrease and finally resulting in local extinction. More importantly, the local species of *Opuntia* cacti will be threatened with extinction due to overgrazing and this in turn will affect other endemic species that feed on it such as Galapagos finches (*Geospiza*), doves (*Zenaida*) and Mockingbirds (*Nesomimus*) (Grant, 1981).

With regard to the lower body condition index on the females sampled on Fernandina island, a possible explanation could be the rigorous hike they would have to accomplish to reach the crater caldera. According to (Werner, 1983), female land iguanas found on Fernandina do not use suitable nesting areas used by other *iguanine* and land iguana populations, but instead migrate distances exceeding 10 km to a 1,495-meter summit and finally into a 900-meter-deep caldera. It is hypothesized that the fumaroles from volcanic activity aids in the incubation of the eggs (Werner, 1982, 1983). This requires a huge amount of energy expenditure which could explain the low weights seen in the females from my sample. Werner (1983) estimates that migration costs constitute half of the reproductive effort in land iguanas found on Fernandina island. A stark example to support migration weight loss is in migratory freshwater fishes. The American Shad (*Alosa sapidissima*) found in the Connecticut river experienced a mean somatic weight loss ranging from 41% to 51% during freshwater migration. Furthermore, the analysis suggested that individuals with larger body sizes experience a greater weight loss when compared to individuals with smaller body sizes (Leggett, 1972). Similarly, migratory birds are known to lose weight during their long flights as seen in Blackpoll Warblers (*Setophaga striata*).

The small group of iguanas captured on Santiago Island is insufficient to meaningfully compare body sizes and weights, however, of interest was the location of a juvenile 262 days after its initial capture on Seymour Norte (Fig. 24). This individual had gained 0.8 kg since its original capture and appeared in good body condition and overall health. It is unclear when the next available visit to Santiago Island will be possible, but priority should be given to re-capture iguanas to track changes in mass and size as well dispersal from release site. The information from Kumar (2019) and the present thesis are useful to understand inter-island variation in body mass and condition of iguanas and the variability in iguana density. Subject to a more structured sampling once COVID-19 restrictions are lifted, these parameters can be used to model population growth and viability.

Inference on the Galapagos pink land iguana

It is unfortunate that the weights were unable to be obtained for the Galapagos pink land iguana due to human error. Furthermore, the sample data obtained showed no evidence of juveniles as the lengths of all of the pink land iguanas sampled were > 30 cm (Kumar, 2019). However, the average lengths of the pink land iguanas caught were calculated to be 100.67 cm. Comparing these to the average lengths of the land iguanas sampled on Fernandina and Baltra, this estimate is highly similar. More specifically, average length of male pink land iguanas sampled were highly similar to average male land iguanas sampled on Fernandina, Baltra and Seymour Norte. In addition, average female pink land iguanas had lengths that were longer than all three populations of land iguanas sampled. According to Iverson et al. (2004), Allen Cay iguanas (*Cyclura cychulra inornata*) reach sexual maturity at SVL lengths of approximately 26 cm to 27 cm. It is likely that Galapagos pink land iguanas sampled on Isabela island are all sexually matured which provides reassurance that there could be potential population growth. More interestingly, during the field visit to Isabela island, Galapagos pink

land iguana sampled were found in pairs (one male and one female). There is limited information on the life history traits of the Galapagos pink land iguana due to their habitat location and it is imperative to obtain this information as this critically endangered species is already threatened with extinction.

CHAPTER 4: CONSERVATION PRIORITIES AND FUTURE RESEARCH NEEDS



Figure 25. Santa Fe Iguana (*Conolophus pallidus*) on Santa Fe Island. Photo: L. Ortiz-Catedral

ABSTRACT

This thesis has provided information on key demographics of the Galapagos land iguana (*Conolophus subcristatus*) populations found throughout the Galapagos archipelago. In addition, key information and inference have been provided on the critically endangered pink iguana (*Conolophus marthae*). In this chapter, I discuss the conservation recommendations for the three endemic terrestrial iguanas (*C. subcristatus*, *C. marthae*, *C. pallidus*) found within the Galapagos archipelago. Recommendations for conservation is highly important in every study as it not only provides future conservationists the opportunity to learn from past mistakes in execution, but also identifies practices and opportunities for the conservation of species.

INTRODUCTION

The conservation of species is a dynamic process that requires short, medium and long-term goals and specific spatial and temporal components (Pressey et al., 2007; Grantham et al., 2010). For endangered species, conservation management often operates with different levels of uncertainty given that several aspects of the system are unknown or poorly researched. For instance, the negative effects of commercial gillnet fisheries are well documented for Hector dolphins (*Cephalorhynchus hectori*), and a range of conservation decisions have been implemented due to the rarity of the species and high probability of extinction, even though several aspects of its biology remain poorly known (Slooten, 2007). In another example, large-scale control of introduced feral cats (*Felis catus*) and rats (*Rattus* spp) has been sustained for nearly eight years on Norfolk Island in order to preserve the world's only population of Tasman parakeets (*Cyanoramphus cookii*) despite significant gaps in our understanding of the biology of the species at the onset of the predator control program (Ortiz-Catedral et al., 2018; Skirrow, 2018). As conservation programs advance, they could benefit from the results of research that can help refine conservation goals and identify opportunities that could enhance the recovery prospects of target species. Further, research and review of conservation programs can provide future conservation biologists an overview of the gaps in knowledge in current research that are happening globally.

Recommendations taken from previous studies also provides the opportunity for conservation biologists to learn from mistakes in the execution of methods and improve them to obtain more effective results. For example, a popular cited paper published by Dodd Jr and Seigel (1991), showcased the many faults in relocation, repatriation and translocation (RRT) projects conducted in the past. Some common mistakes made by previous studies included lack of post RRT information on breeding evidence and also the lack of long-term post translocation monitoring. Many studies have taken precaution and consideration the proposed

recommendations provided by Dodd Jr and Seigel (1991). For example, a study conducted by Burke (1989), monitored relocated Gopher tortoises (*Gopherus polyphemus*) for only two years, which amounts to only 10% of the time it takes for these tortoises to mature, which is hardly enough time to quantify long-term relocation success. Following which, Tuberville et al. (2008), conducted a 12-year mark and recapture long-term post translocation monitoring to investigate apparent annual survival in Gopher tortoises. In another example, Tuberville et al. (2005) conducted a study on Gopher tortoises using penning experiments to investigate site fidelity and area size activity. Although, the methods from this study were adopted from previously conducted studies (Burke, 1989; Doonan, 1986; Lohoefener & Lohmeier, 1986), they were improved to provide more effective translocation results as compared to those previously executed. Tuberville et al. (2005) adopted longer penning durations, which were more suited for long living species, translocated intact populations of tortoises that included all size classes and provided the tortoises opportunities for social interactions.

Conservation recommendations allows conservation biologists to identify opportunities and practices for the conservation of species. Interestingly, this does not necessarily have to apply to the conserving the species the recommendations were intended for. For example, the practice of translocation was previously used as a species conservation tool to establish populations of non-native species; to restore native species extirpated by hunting and to remove problem-causing species to allow regeneration in the environment (Griffith et al., 1989; Linnell et al., 1997) (i.e. translocating black bears (*Ursus americanus*) in British Columbia to remove depredation of livestock and other nuisance behaviors). Conservation biologists in New Zealand have since adopted the method of translocation to aid in the recovery of many endemic avian fauna species by translocating them to uninhabited, invasive species free offshore islands (Miskelly & Powlesland, 2013).

Based on previous studies on the Galapagos iguanas and the results presented in this thesis, I provide a few recommendations that could aid in future conservation and management of three endemic species of iguanas: *Conolophus subcristatus*, *Conolophus pallidus* and *Conolophus marthae*. The following recommendations requires more research and aim to allow greater conservation success of these species across the Galapagos archipelago.

Galapagos land iguana

Long-term post translocation monitoring

One of the aims of this thesis was to conduct an evaluation of the status of reintroduced Galapagos land iguanas (*Conolophus subcristatus*) on Santiago island. However, as outlined in Chapter 1, the ongoing COVID-19 pandemic prevented me from completing this component. There is some preliminary information on the Santiago population (see Chapter 3) however, the monitoring of this population should not stop there. Research in other systems shows that some adults are usually lost from the founder population as a result of dispersal (Burke, 1989; Epperson & Heise, 2005). Thus, it is important to determine whether this has occurred in the Santiago Island Galapagos land iguana population.

Long-term post translocation monitoring provides an accurate evaluation of the species establishment and the biodiversity integrity at release sites (Van Winkel, 2008). For example, a 12-year mark and recapture study on Gopher tortoises (*Gopherus polyphemus*) showed high apparent annual survival which did not correlate to the tortoises being confined to the study area. In addition, a short-term release effect on apparent survivability of both immature and adult tortoises was observed; with newly released adult and immature tortoises having significantly lower apparent annual survivability than previously established tortoises (Tuberville et al., 2008). Obtaining results from a long-term post monitoring study is often rare

due to the fact that they are incredibly time consuming and expensive (Dodd Jr & Seigel, 1991). However, the information that is obtained from this process is crucial; as if large numbers of released animals are lost through mortality or dispersal, or if even small losses is sustained for many years, the method of translocation could become a catastrophic event rather than a beneficial one (Tuberville et al., 2008).

The recommendation for long-term post translocation monitoring should be a minimum of 12 years (20%), as the average life span of the Galapagos land iguana is approximately 60 years (Charles Darwin Foundation, 2006). During the monitoring phase, repeated census counts should be conducted as it can not only provide information on the survivability of the founder population but also provide information on breeding evidence, which is a crucial criterion for evaluating translocation success. Studies on post release behaviors should be conducted to analyze any behavioral differences that may surface in comparison to those from Fernandina, Seymour Norte and Baltra islands even though it is highly likely that there would not be any differences as results from head-started iguana programs showed no behavioral differences to those raised in the wild (Pérez-Buitrago et al., 2008). However, behaviors regarding habitat use might show slight differentiation due to the islands being different than Santiago (i.e. terrain and food sources).

Invasive species management on Santiago island

Throughout this thesis, there have been many examples illustrating the impact invasive species populations have on indigenous flora and fauna, especially feral cat populations. Feral cat populations play a major role in global biodiversity decline (Medina et al., 2011). More importantly, the detrimental effects they have on iguanas are globally evident (Iverson, 1978; Mitchell et al., 2002). Hence, even though Santiago island has been declared free of large invasive mammalian species, except rats (*Rattus rattus*), continuous monitoring should be

conducted to ensure the island remains a safe haven for the Galapagos land iguanas. Surveys for feral cat populations should be conducted every three months to ensure they do not return; if they do return, they should be eradicated immediately. In addition, another way to prevent the introduction of invasive species is to strengthen biosecurity at Santiago island's entrances.

One leading example is the country of New Zealand. Islands like Tiritiri Matangi has been declared invasive mammal free and there are strict laws to ensure none returns. Visitors are required to complete a biosecurity checklist in which to hand in to stationed rangers at the island's entrances. Furthermore, stationed rangers usually re-check gears brought in by visitors (Department of Conservation, n.d.). The prohibition of importing exotic plant and animal species should be implemented in the Galapagos archipelago to reduce the risks of introducing more invasive species to the islands. In addition, ships importing goods and supplies should be thoroughly checked by stationed rangers at island entrances before allowing them to dock.

The eradication of black rats from Santiago island requires more effort due to the existing Galapagos rice rat (*Nesoryzomys swarthi*) population. Studies have shown that the two species of rats have been co-existing for approximately 100 years (Trillmich, 1986). The cause of extinction of the Galapagos rice rats on Santa Cruz was believed to be caused by a disease or parasite that came with the introduced black rats, but the decline on Santiago island was due to competition for resources (Brosset, 1963). Further analysis by Harris and Macdonald (2007) indicated that the Galapagos rice rats increased in mass with supplementary food and there was evident interference by aggressive encounters between the two species of rats. Hence, the need to eradicate black rats from Santiago island in order to protect the endemic rice rats is necessary. The use of poison bait trappings such as diphacinone and brodifacoum is not ideal due to fatal effects that these poisons can have on the Galapagos rice rats. One plausible option is to use live trapping of the endemic rice rats and translocate them to an offshore island that is free of invasive mammalian species.

Endozoochory research

Seed dispersal plays an important role in the ecosystem and is paramount to plant reproduction, population genetics and ecology as it determines the movement of plant genes in space and time. Moreover, seed dispersal prevents high seedling mortality under and near parent trees as they prevent resource competition (Wenny & Levey, 1998). More importantly, seed dispersal allows the plants to reach specific habitats that are favorable for survival. For example, seed survivability by the three-wattled bellbirds (*Procnias tricarunculata*) was found to be higher than those dispersed by other birds found in the Monteverde Cloud Forest Preserve in Costa Rica due to a higher light condition and evading fungal pathogens (Wenny & Levey, 1998). Furthermore, seeds dispersed by ants, though only dispersed in short distances, are buried underground which provides the seeds shelter from unfavorable environmental conditions such as droughts and allows the seeds to reach nutrient-rich microsites (Lengyel et al., 2010).

Endozoochory research in iguanas have just begun (Benítez-Malvido et al., 2003; Lasso & Barrientos, 2015; Laurel et al., 2000; Traveset et al., 2016). There have been some conflicting results with regards to seed germination in iguanas. For example, a study conducted in Puerto Rico showed lower percentage of seeds ingested by green iguanas (*Iguana iguana*) germinating, however, the time taken to germinate was reduced (Burgos-Rodríguez et al., 2016). Another study conducted on endangered rock iguanas (*Cyclura* spp) supported this notion showing similar results with ingested seeds germinating more rapidly than those that were not, however, their results did not show a reduction in germination success in ingested seeds (Laurel et al., 2000). If iguanas do in fact play an important role as seed dispersers, declines in iguanid populations could have a significant impact on plant communities found in habitats that rely on them as seed dispersers. The rapid germination of seeds facilitated by

iguanas could prove to be significantly advantageous to plants found especially in dry habitats throughout the Galapagos archipelago. Seeds that germinate early are less likely to desiccate and are able to take advantage of rainfalls and gain a developmental advantage to those that were not ingested. Hence, I propose conducting a more in-depth study of endozoochory in the Galapagos land iguanas. The study of endozoochory in the Galapagos land iguanas can provide key information on plant ecology and spatial distribution throughout the Galapagos archipelago. Results obtained from Traveset et al. (2016) showed that four species of native plants found in the Galapagos archipelago, *Jasminocereus thouarsii*, *Scalesia affinis*, *Stylosanthes sympodiales* and *Tephrosia cinerea* were dispersed only by the Galapagos land iguanas. Furthermore, endozoochoric studies can provide conservation biologists useful information on their foraging ecology, patterns and diet as a by-product. Studying seed deposition patterns throughout the Galapagos archipelago can provide spatial distribution information on the iguanas. It is important that the information obtained from endozoochoric studies be published and updated on the IUCN red list as it is universally used as a platform for conservation biologists to obtain key information on species and provides the opportunity for prioritizing the conservation of other endangered species of iguanid globally.

Galapagos pink land iguana

Head-starting programs

Head-starting programs is captive breeding of a species with the intention to release them back into the wild. This method has been widely used to protect endangered species by increasing survivorship (Pérez-Buitrago et al., 2008). A stark example is the conservation of the Grand Cayman blue iguana (*Cyclura lewisi*). This species of iguana once faced the threat of extinction due to anthropogenic threats and introduced invasive species. However, with the

combination of captive breeding programs and head-starting of wild hatchlings, plus zoological facilities and funding, the Grand Cayman blue iguana has managed to downgrade its IUCN red list classification from ‘Critically Endangered’ to ‘Endangered’ (Grant & Hudson, 2015). Head-starting programs have been ongoing in the Galapagos since the late 1900s and it has been used to recuperate population numbers for the Galapagos giant tortoises (*Chelonoidis nigra*) and the Galapagos land iguanas (*Conolophus subcristatus*). In 1975, the population of land iguanas on Santa Cruz was decimated due to feral dog populations. However, with the help of captive breeding coupled with eradication of invasive introduced species, the population has thrived, and the transfer of iguanas continues to date, approximately every three years (Cayot, 2008). Many studies have suggested that iguanas bred in captivity displayed similar behaviors to those reared in the wild, this suggest that iguanas as a species are highly adaptable (Álvarez et al., 2007; Escobar et al., 2010; Pérez-Buitrago et al., 2008). Growth rates on the other hand showed slightly different results. In a head start program conducted on the Mona island iguanas (*Cyclura cornuta stejnegeri*), growth rates were similar to those found in the wild during the first two years of captivity; however, a significant decline in growth rates at the beginning of the third year was noticed (Pérez-Buitrago et al., 2008). One plausible explanation could be due to the stress from overcrowding and aggression that comes with increasing age. In addition, growth rates prior to release decreased immediately. However, many studies suggested that the cause of growth rate decrease was due to the change in diet and the exponential increase in energy expenditure to forage and seek shelter (Lewis et al., 2008; Pérez-Buitrago et al., 2008).

I propose considering using head-starting programs to aid in the recovery of the Galapagos pink land iguana populations. Since the population numbers of the Galapagos pink land iguana is considered low, it is imperative that conservation actions be implemented before the numbers decrease further. Juveniles should be captured and reared in appropriate facilities

for a maximum of three years of age before releasing, as stress from overcrowding and increased aggression has proven to affect growth rates. The rearing facility should be able to mimic the natural environment they were found in; though it may be disputed that it would be difficult to mimic the volcanic environment the Galapagos pink land iguanas are found in, genetical analysis have shown that the Galapagos pink land iguana diverged roughly around 5.7 million years ago, which is before the existence of any of the archipelago's present islands (Pierson & Durham, 2009). This information allows us to suggest that their primary habitat was never restricted to Wolf volcano. More importantly, homing behavior, the ability to return to their usual home range which they are displaced from, should be taken into consideration with regards to the construction of this head-starting facility as lizard species have been reported to exhibit this, including the Mona island iguanas (Freake, 1998; Jenssen, 2002; Pérez-Buitrago et al., 2008). I propose that the facility be constructed on Santiago island as it was deemed large invasive mammals free in 2006; and is now considered a safe haven for the recently translocated Galapagos land iguana.

Translocation

Translocation is powerful tool used by conservation biologists to date. As mentioned in Chapter 1, there have been many successful translocations in not only iguanid but avian and mammal species. Studies have shown that the Galapagos pink land iguana is restricted to a small area of Wolf Volcano (Gentile et al., 2009; Gentile et al., 2016; Pierson & Durham, 2009). However, as aforementioned, they are highly speculated to be restricted to Wolf volcano due to anthropogenic threats and invasive species. One similar example is New Zealand's Takahe. Though it may seem these flightless birds have adapted to the alpine region, fossil records show they were widely distributed around the country in the past (Beauchamp & Worthy, 1988; Bunin & Jamieson, 1995; Mills et al., 1984). To date, 10 sanctuaries in New

Zealand houses an active breeding pair; although there were concerns for delayed breeding and lowered reproductive success, a study conducted Jamieson and Ryan (1999) suggested the effects from translocation were insignificant and translocated birds displayed similar hatching and fledging success to resident birds. Moreover, the results from Chapter 2 in this thesis, showed breeding success in the recently translocated Galapagos land iguana in 2019. This makes the proposition of translocation the Galapagos pink land iguana feasible.

There have been suggestions to translocate Galapagos pink land iguanas outside Isabela Island, for instance to the humid zone of Santiago island since 2013 (K. Campbell, pers. comm.). Since Santiago Island has been declared invasive mammal free in 2006 with the exception of rats (*Rattus rattus*), it could be an ideal sanctuary for these critically endangered iguanas. Translocating the Galapagos pink land iguanas would ensure the protection of the species. The Wolf volcano is found on Isabela island which is colonized by many invasive species such as dogs (*Canis familiaris*) and cats (Levy et al., 2008). Feral cat populations pose a major threat to the iguanas inhabiting Isabela island. Studies have shown, feral cats are able to travel large distances from their home range (Edwards et al., 2001; Jones & Coman, 1982; Morgan et al., 2009). Results taken from Edwards et al. (2001), showed several instances where feral cats moved distances of up to 34 km and that home range sizes are highly influenced by prey availability. In addition, the current habitat the Galapagos pink land iguana inhabit requires a long and tedious hike. Large amounts of water and food are required to ensure rangers and conservation biologists remain hydrated and satiated. Hence, translocating the Galapagos pink land iguanas could provide conservation biologists easier access to study these elusive creatures. To date, important information such as life history traits are currently missing and is urgently needed to help conserve the species.

Santa Fe iguanas

Invasive species prevention

As mentioned in Chapter I, biodiversity found on islands are highly susceptible to decline due to invasive species. The small area and water body surrounding the island makes it impossible for flightless native fauna to escape depredation from invasive species such as cats and rats. The island of Santa Fe was once invaded by invasive species, including goats (*Capra hircus*), but after several eradication programs, the island is now invasive species free (Phillips et al., 2012) and it has allowed native vegetation and the populations of the Santa Fe iguana to recover (Snell et al., 1984). One of the greatest challenges faced on Santa Fe island at the moment is ensuring that no invasive species establishes (Galapagos Conservancy, n.d.). I propose that biosecurity around the island's entrances should be enhanced to prevent invasive species, such as feral cat populations to establish. Similarly, to the above recommendations for continuous monitoring of invasive species on Santiago island, intensive checks should be conducted on ships that visit Santa Fe to ensure no black rats (*Rattus rattus*) are introduced to the island. Visitors to the island should be asked to fill in biosecurity forms and have their gears searched by stationed rangers before allowed to enter the island.

Population estimates monitoring






The current population of the Santa Fe iguanas on Santa Fe island is potentially stable. However, in order to keep track on their population estimates and to ensure the island has not reached its maximum capacity, monitoring of population estimates should still continue to be conducted. One similar example is measuring the carrying capacity of Takahē (*Porphyrio mantelli*) in four different offshore islands found in New Zealand. Mana island's (2.17 km²) carrying capacity was estimated to be approximately 22 – 53 pairs, Maud island (3.09 km²) was 7 – 34 pairs, Kapiti island (20.23 km²) was (5 – 33 pairs) and Tiritiri Matangi (2.2 km²)

was 25 pairs. From the information above, it is obvious that land size does not play a major role in the maximum number of breeding pairs, but instead the terrain and the area size of usable habitat for the Takahē (Ryan & Jamieson, 1998). More importantly, the high population of iguanas on Santa Fe island can result in detrimental effects rather than beneficial ones. High population numbers on a small land size would result in increased resource competition and increased competition for burrow space; and as iguanas are ectothermic, obtaining shelter in burrows is paramount to their survival. Furthermore, results from a study conducted by Cano Rodríguez (2018) suggested that the Galapagos giant tortoise (*Chelonoidis hoodensis*) fed on similar plant species as the Santa Fe iguanas. This increases the competition of food resource on the island and could potentially have a detrimental effect in time.

I propose that the monitoring of Santa Fe iguana populations be conducted every five years to ensure continual growth in their population numbers. Conducting population size monitoring could also provide information on breeding evidence and mortality rates on the island. In addition, the information obtained should be corresponded to previous years to analyze any patterns that may arise in their population fluctuations. Furthermore, studies on iguana carrying capacity of Santa Fe island should be conducted. The estimation of carrying capacity is complicated by habitat availability as regeneration on the island occurs (Ryan & Jamieson, 1998); in addition, seasonal variations should also be taken into consideration as studies have shown the Santa Fe iguanas exploited microclimates created by cliff faces during colder seasons (Christian et al., 1983). The home ranges of the Santa Fe iguanas are known to be relatively large and overlap extensively (Christian & Tracy, 1985), hence it is imperative that carrying capacity of Santa Fe island be reviewed to ensure the continued growth and recovery of the Santa Fe iguanas.

APPENDIX 1.

Research Permit from the Ministry of Environment / Directorate of the Galapagos National Park to conduct research on Galapagos Land Iguanas (*Conolophus subcristatus*).

    	
DIRECCIÓN DEL PARQUE NACIONAL GALÁPAGOS DIRECCIÓN DE GESTIÓN AMBIENTAL PERMISO DE INVESTIGACIÓN CIENTÍFICA: N° PC-96-18	
Título del Proyecto: Proyecto de translocación de la iguana terrestre de Galápagos (<i>Conolophus subcristatus</i>) a la isla Santiago como estrategia para contribuir a la restauración ecológica de la especie y el ecosistema en la isla.	
Nombre del Aplicante: Danny Rueda, Luis Ortiz- Catedral	Contraparte Institucional: N.A.
Dirección actual completa: Oficinas del Parque Nacional Galápagos, Pto Ayora Santa Cruz. Email: drueda@galapagos.gob.ec	
Otros participantes en el Proyecto: Christian Sevilla (DPNG), Wilson Cabrera (DPNG), Andrea Loyola (DPNG), Víctor Carrión, Karl Campbell (Island Conservation), Gabriele Gentile (U. Tor Vergata), Kirtana Kumar (Massey University, Nueva Zelanda).	
Clasificación del Proyecto: Conservación	Requiere Contrato Marco de Acceso a Recursos Genéticos: NO
Se requiere coleccionar muestras: SI	Factura: N.A.
Duración del Permiso de Investigación: Del 04 de diciembre del 2018 al 31 de diciembre del 2019	
Islas: Seymour Norte, Santa Cruz, Santiago.	Sitios: Seymour Norte: Centro de Crianza DPNG (Santa Cruz), Espumilla, Bucanero, Puerto Nuevo, Puerto Red, y Cerro Colorado arriba de la Bomba, Zona Árida, Zona de Transición (Santiago).
Instituciones auspiciantes: Dirección del Parque Nacional Galápagos, Island Conservation, Massey University, Universidad Tor Vergata, Galapagos Conservancy	
Condiciones de cumplimiento obligatorio: <ol style="list-style-type: none"> Los investigadores deberán cumplir estrictamente con todas las regulaciones establecidas en el Manual de Procedimientos para Científicos Visitantes y Protocolos para viajes de Campo y Campamentos en las Islas Galápagos. Únicamente el equipo de investigadores incluido en el presente permiso está autorizado para participar durante el desarrollo del proyecto, tanto en la fase de campo, análisis de muestras, tabulación de datos y desarrollo de publicaciones. En caso de requerir la participación de investigadores adicionales, los responsables del proyecto deberán solicitar la autorización correspondiente por escrito a esta Dirección, presentada las justificaciones necesarias. Previo a la salida de campo, el investigador principal deberá presentar a la DPNG el AVISO DE VIAJE DE CAMPO correspondiente, con mínimo 72 horas de anticipación y de acuerdo al formato establecido; y coordinar permanentemente con el Responsable del proceso de Conservación y restauración de Ecosistemas Insulares de la DPNG en Santa Cruz, a fin de sistematizar todas las actividades que se ejecuten dentro del proyecto. Para las salidas de campo del proyecto, los investigadores deberán contar con una copia legible del respectivo Permiso de Investigación y el Aviso de Viaje de Campo firmado por los técnicos de la DPNG. Los sitios e Islas autorizados serán exclusivamente: Seymour Norte, Centro de Crianza DPNG (Santa Cruz), Espumilla, Bucanero, Puerto Nuevo, Puerto Red, y Cerro Colorado arriba de la Bomba, Zona Árida, Zona de Transición (Santiago). Se autoriza la captura y manipulación de un máximo de 4000 Iguanas amarillas (<i>C. subcristatus</i>), de la isla Seymour Norte con la finalidad de ser puestas en cuarentena en instalaciones de la DPNG Isla Santa Cruz, y su posterior translocación a la isla Santiago. Se autoriza el monitoreo post-liberación (<i>flush transects</i> Captura-Recaptura y Radiotelemetría) de las Iguanas amarillas (<i>C. subcristatus</i>) translocadas a la isla Santiago con la finalidad de evaluar su estado luego de la translocación desde Seymour Norte. Se autoriza el marcaje mediante la implantación de microchip (Pit Tag) el cual será colocado subcutáneamente en el total de las iguanas amarillas (<i>C. subcristatus</i>) capturadas en la isla Seymour Norte con la finalidad de facilitar su identificación. Se autoriza la colocación de 30 transmisores (uno por individuo) VHF modelo AL-2 Holohii Systems, los cuales serán fijados a la cadera de los individuos de Iguana amarilla (<i>C. subcristatus</i>) mediante el uso de gel de Cianocriato. Se autoriza el uso de Permetrina como agente de control de parásitos externos como ácaros o garrapatas mediante su aplicación sobre el cuerpo de las iguanas amarillas (<i>C. subcristatus</i>) capturadas, este compuesto deberá ser en una dilución al 0.5%. 	

Santa Cruz, Pto. Ayora: (PBX) 593 51 2 528 189/190 • info@galapagos.gob.ec • Código Postal: 200102 • San Cristóbal, Pto. Baquerizo Moreno:
 Tel./Fax: 593 51 2 520 135/491/478 • Código Postal: 200101 • Isabela, Pto. Vilama: Tel: 593 51 2 528 178/268 • Código Postal: 200103
 Floreana, Pto. Velasco 189111. Tel: 593 51 2 533 008 • PUIG: 2090002010001

**DIRECCIÓN DEL PARQUE NACIONAL GALÁPAGOS
DIRECCIÓN DE GESTIÓN AMBIENTAL**

Versión: 01 Fecha: 09/01/17

PERMISO DE INVESTIGACIÓN CIENTÍFICA: N° PC-96-18

11. **Se autoriza** la colección de muestras de un máximo de 7500 muestras de sangre (1ml c/u) de iguanas amarillas (*C. subcristatus*), para determinar nivel de Hematocrito, además estas serán subdivididas de la siguiente forma: 500 muestras de plasma sanguíneo para análisis hormonales; 2 frotis sanguíneos de cada muestra (15000 placas) para conteo diferencial de células y presencia de hemoparásitos.
12. **Se autoriza** la colección de un máximo de 40 muestras de uñas de iguanas amarillas *C. subcristatus*, con la finalidad de determinar la dieta de las mismas mediante la aplicación de análisis de isótopos estables.
13. **Se autoriza** la colección del total de las heces producidas por las iguanas amarillas (*C. subcristatus*) durante su aislamiento y cuarentena en las instalaciones de la DPNG con la finalidad de evaluar la presencia de semillas restantes en el tracto digestivo de las iguanas.
14. **Se autoriza** la realización de necropsias a los cadáveres de iguana amarilla (*C. subcristatus*) que pudieran resultar durante el periodo de aislamiento y cuarentena en las instalaciones de la DPNG en la isla Santa Cruz.
15. Posterior al cumplimiento de los procedimientos establecidos por la DPNG, los investigadores podrán movilizar las muestras autorizadas para los análisis correspondientes en los laboratorios del Veterinary Diagnostic Laboratory - Iowa State University, Ames, IA, Estados Unidos el Edwards Lab, University of California, La Merced, CA, Estados Unidos, el Conservation Metrics Inc, Santa Cruz, CA, Estados Unidos y la Universidad Tor Vergata, Roma, Italia.
16. Las muestras colectadas dentro del marco legal del presente permiso de investigación NO podrán ser utilizadas en actividades de **BIOPROSPECCIÓN, NI ACCESO AL RECURSO GENÉTICO**; y estas únicamente podrán ser utilizadas bajo las líneas de estudio autorizadas por el Ministerio del Ambiente del Ecuador a través de Dirección del Parque Nacional Galápagos.
17. El desarrollo de actividades de investigación científica dentro de las Áreas Protegidas como el Parque Nacional y la Reserva Marina de Galápagos es un privilegio concedido por el Gobierno Ecuatoriano a través de la DPNG, por lo que los responsables del proyecto deberán citar el número de Permiso de Investigación Científica otorgada por la DPNG, e incluir los reconocimientos y/o agradecimientos en las publicaciones científicas, Tesis o informes técnicos científicos que se emita como producto generado en base al presente permiso de investigación.
18. El análisis de los datos y los avances de la investigación deberán estar disponibles permanentemente para los técnicos de la DPNG, existiendo el compromiso de usarlos únicamente para acciones de manejo y no publicarlos sin el consentimiento de los investigadores principales del proyecto.
19. Previa a publicaciones científicas como resultados del proyecto, se deberá remitir el documento final ya aceptado para publicación a la DPNG bajo la denominación de embargo, con la finalidad de conocer y preparar el boletín correspondiente con dicha información, existiendo el compromiso como DPNG de no realizar ninguna difusión hasta contar con la publicación científica oficial.
20. Una vez concluido el análisis de las muestras, estas, las bibliotecas de datos y/o cualquier material resultante deberán ser devueltos a la DPNG. Estas deben ser preservadas, curadas y depositadas correctamente en las colecciones de referencia en Galápagos, de lo contrario, se deberán sufragar los gastos que demanden la preparación del material para su ingreso a la colección correspondiente.
21. Previa a la renovación del permiso de investigación, el científico responsable del proyecto o su contraparte institucional de investigación deberán entregar a la DPNG lo siguiente:
 - Una copia digital (Tabla de Excel 97-2003) de todos los datos obtenidos durante la ejecución del proyecto.
 - Un INFORME TECNICO DE CAMPO por cada Aviso De Viaje De Campo presentado a la DPNG, para la ejecución del proyecto.
 - Dos conferencias para los guardaparques de la DPNG, guías naturalistas y otras personas interesadas sobre los avances del proyecto. Para establecer el cronograma se deberá coordinar con el Responsable de Investigación Aplicada de la DPNG.
 - Un INFORME DE AVANCES del proyecto con todos los detalles de la investigación y sus recomendaciones técnicas aplicables al manejo de las Áreas Protegidas.
 - Artículos y publicaciones resultantes de la siguiente manera: en el caso de publicaciones únicamente en formato digital, si son libros o tesis, 5 impresas y una digital, en el caso de la versión digital lo harán en dispositivos magnéticos adecuadamente identificados.

DIRECCIÓN DEL PARQUE NACIONAL GALÁPAGOS

DIRECCIÓN DE GESTIÓN AMBIENTAL

PERMISO DE INVESTIGACIÓN CIENTÍFICA: N° PC-96-18

Versión: 01

Fecha: 09/01/17

22. Los datos que se desprendan de esta investigación, no podrán ser utilizados para estudios posteriores sin la previa autorización del Ministerio del Ambiente, a través de la Dirección del Parque Nacional Galápagos.
23. Del incumplimiento de las obligaciones dispuestas anteriormente se responsabiliza a Danny Rueda (DPNG) y Paula Castaño (Island Conservation) Responsables del proyecto. Por lo tanto el incumplimiento de cualquiera de estas condiciones así como el uso indebido de este documento, serán sancionados conforme al Código Orgánico Ambiental y dependiendo de la infracción podría conllevar a suspensión inmediata de la investigación.

Valoración Técnica: Carlos Vera

Categoría: COLABORADOR DPNG 2018

Considerando que el Título II, numeral 2.2, literal V del Estatuto Orgánico de Gestión por Procesos de la Dirección del Parque Nacional Galápagos publicado en la Edición Especial N° 349 del Registro Oficial publicado el martes 16 de octubre del 2012, establece entre las atribuciones y responsabilidades del Director de Gestión Ambiental "Administrar y organizar las actividades de investigación que se desarrollen en las áreas protegidas de Galápagos, en coordinación con el proceso de investigación", además que mediante Resolución N° 71 del 14 de Diciembre del 2012, el Director del Parque Nacional Galápagos delegó al Director de Gestión Ambiental, para que en su nombre y representación tramite y suscriba los actos relacionados con el desarrollo de proyectos de investigación científica en las áreas protegidas de Galápagos; en uso de la facultad delegada y de conformidad a lo señalado en el Título Cuarto, capítulo I del Estatuto Administrativo de la Dirección del Parque Nacional Galápagos, otorga el presente PERMISO DE INVESTIGACIÓN.

Reporte de Avances o Final: 15 de junio 2019

Entrega de Propuesta para Renovación: 01 de diciembre 2019

FIRMA DEL INVESTIGADOR PRINCIPAL

Fecha de emisión:

28 de diciembre de 2018

Director de Gestión Ambiental
DIRECCIÓN DEL PARQUE NACIONAL GALÁPAGOS



APPENDIX 2

Mohamed bin Zayed Species Conservation fund application

29/10/2019	The Mohamed bin Zayed Species Conservation Fund Application Form (print version)
The Mohamed bin Zayed Species Conservation Fund Application Form (print version)	
This print version is for your reference only. All applications must be submitted online at: http://www.speciesconservation.org/grants/application/	
Project Title (brief sentence which explains the purpose of the project for which a grant is being sought): *	
20 word limit	
Population size, trends and distribution of the Mariana flying foxes (Pteropus mariannus) on Rota island	
To help our administrators process your application, please provide in the box below any information relating to a previous relationship or contact you may have had with the Fund. You may already have an active project with us and are seeking continuation funding, or you previously had a grant which is now completed, or you may have previously been rejected for a grant. If this is the case then please state ANY previous project numbers. If this is your first contact with the Fund, we would also be very interested to know how you heard of us.	
Please enter any previous relationship in the box below to help us process your application:	
50 word limit	
None	
Section 1 - Grant Amount	
1.1 - Size of grant application: *	
<input type="radio"/> Up to \$5000 <input checked="" type="radio"/> \$5000 - \$25000	
Section 2 - Your Details	
2.1 - Title (e.g. Mr / Mrs / Prof / Dr): * Mr	
2.2 - First Name: * Colin	
2.3 - Last Name: * Heng	
2.4 - Organization: Massey University	
2.5 - Website Address: www.islandsforever.com	
2.6 - Network: (if applicable - e.g. IUCN / SSC Specialist Group or NGO)	
Address	
2.7 - Street & Number: * 38 Totaravale drive	
2.8 - City / Town: * Totara Vale	
2.9 - State: * Auckland	
2.10 - Country: * New Zealand	
2.11 - Postal / ZIP Code: * 0629	
https://www.speciesconservation.org/grants/application/print.php	
1/7	

2.12 - Landline Phone Number: * +64 021 086 00328
 2.13 - Mobile Number: +64 021 086 00328
 2.14 - Fax Number:
 2.15 - Email Address: * C.t.heng@hotmail.com
 2.16 - Re-enter Email Address: * C.t.heng@hotmail.com
 2.17 - Secondary Email:

Section 3 - Agreement

3.1 - I agree that to the best of my knowledge, the information provided is true and correct. *
 ✓

Section 4 - Overview of Your Organization / Community / Individual Project

4.1 - For organizations, please provide the following information:

- Mission of the organization
- Objectives of the organization
- Date it was established
- Geographical area of the project
- Target species / habitats covered by the organization

500 word limit

Massey University (est. 1927) is the largest university in New Zealand and offers a range of programs for undergraduate and graduate students. Massey University has a successful Master of Science program, which attracts national and international students. In the Conservation Biology program we strive to provide solutions to the global biodiversity loss crisis and since 2010 have successfully completed six grants from MBZ. The present project will take place on Rota Island (Luta in Chamorro Language), one of the three Southern Islands in the CNMI. The target species is the "Fanihi" or Mariana Flying Foxes (*Pteropus mariannus*), a large fruit bat found in Guam, Japan, Federate States of Micronesia and CNMI. Although the species has a broad distribution, it is mostly on uninhabited islands. Rota holds the largest population of Mariana fruit bats, but their current population size and movements are little studied. This project will tackle two research priorities identified by the IUCN: 1. Estimate the local population size on Rota and 2. Contribute to a better understanding of its ecology.

4.2 - Please list key personnel / titles / qualifications / email address:

Name	Title / Role	Qualifications	Email Address
<input checked="" type="checkbox"/> Colin Heng	Mr/Master's student	BSc	c.t.heng@hotmail.com
<input checked="" type="checkbox"/> Luis-Ortiz Catedral	Doctor/Master's supervisor	PhD, MSc, BSc	L.Ortiz-Catedral@massey.ac.nz
<input checked="" type="checkbox"/> Josh Guilbert	Doctor/Supervisor	PHD, Msc, BSc	guilbert.dfw@gmail.com

4.3 - Are there any key institutions the project is affiliated with, such as NGO and community partners, government agencies or corporate involvement?✓ Yes ☐ No

If yes, describe below. *

Organization	Description Of Affiliation	Contact Name	Email Address
<input checked="" type="checkbox"/> Massey University	Msc	Colin Heng	c.t.heng@hotmail.com
<input checked="" type="checkbox"/> Massey University	Supervisor	Luis-Ortiz Catedral	L.Ortiz-Catedral@massey.ac.nz
<input checked="" type="checkbox"/> CNMI Division of Fish and Wildlife	Supervisor	Joshua Guilbert	guilbert.dfw@gmail.com

Section 5 - Project description

Please note that any information provided on this application may be made available to the media if the application is successful.

5.1 - Please provide: *

- Scientific name(s) of the species
- Vernacular name(s) of the species
- Specify species type as one of the following: mammal, bird, reptile, amphibian, fish, invertebrate, plant or fungi

Scientific Name	Vernacular Name	Species Type
<input checked="" type="checkbox"/> Pteropus mariannus	Mariana Flying Foxes	Mammal

5.2 - Whilst your project may cover a vast area, please specify first the continent and then the country where your work is focused most: *

Continent: Oceania
Country: Northern Mariana Islands

5.3 - Please elaborate on this location and mention any additional countries or geographic areas (including oceans) where you aim to support the target species: *

200 word limit

The Mariana flying foxes are endemic to the CNMI and even though they have a broad distribution, the population sizes on most inhabited islands are scarce. Within the CNMI, Rota island is the only inhabited island containing a substantial population of these flying foxes.

5.4 - Please outline the conservation status of the target species and the importance of the associated habitat: *

200 word limit

The species is currently classified as endangered but population estimates have not been reviewed since 2008. Based on previous studies, we are certain current classification of the Mariana flying foxes does not reflect its true status. The target species is known to inhabit native tropical forests, coastal strands and mangroves with their preferred diet unknown. Roosting sites are commonly found where there are minimal wind and anthropogenic disturbances. On the island of Rota, apart from anthropogenic disturbances, Mariana flying foxes face

predation from the invasive brown tree snake. Hence, assessing population abundance of the target species on Rota island coupled with investigating foraging ecology are the main objectives of this research to not only update the status of these flying foxes but to understand the required conservation actions needed to be undertaken to ensure their survival.

5.5 - What is the conservation status of the primary target species, using the IUCN guidelines: *

Endangered

5.6 - Please outline in 20 words or less how this project will contribute to the conservation of your target species: *

20 word limit

We will estimate the current population and distribution of these bats in order to identify key areas for conservation.

5.7 - Please list the overall objectives of the project: *

200 word limit

1. Using observational studies at roosting sites coupled with radio-tracking tags or GPS loggers to determine population size and also the movement patterns of these bats 2. To obtain information on foraging behaviour 3. To investigate the effects of rehabilitation on these bats

Please also summarize these objectives in the table below:

Objective	Estimated time to spend on completing objective (in weeks and months)	Next steps
1. Determining population size and movement	12 months	Develop H&S plans, gear preparations, visit key roosting sites
2. Information on foraging behaviour	12 months	Develop H&S plans, gear preparations
3. Effects of rehabilitation	12 months	Visit rehabilitation centre located on Rota to discuss effects of rehabilitation on pups

5.8 Please outline briefly how this project relates to other work done previously with this species in this area (e.g. Species Action Plan, previous survey or monitoring work).

Please also include any scientific references: *

400 word limit

There has been a high fluctuations in Mariana flying fox populations on Rota island and there is a high priority for research and their migration patterns between islands as mentioned in the IUCN. Since the last published study was conducted in 2000 (Cruz et al., 2000), it is essential to provide a more updated assessment of these flying foxes.

5.9 - What are the specific conservation actions that the grant would support and how would each of these actions directly or indirectly contribute to conservation of the project's primary target species and its habitat? *

Activity	How Would It Contribute?
<input checked="" type="checkbox"/> Estimating population abundance of the Mariana flying foxes on Rota island	Providing a updated population assessment of the Mariana flying foxes
<input checked="" type="checkbox"/> Foraging habits	Provide information on the types of trees that requires conservation to ensure survivability
<input checked="" type="checkbox"/> Rehabilitation of pups	Provide information on the effects of rehab on pups compared to those being brought up in the wild

5.10 Approximately when will the activities that the grant would support take place?

Intended Start Date: (dd/mm/yyyy)

05/02/2020

Intended End Date: (dd/mm/yyyy)

05/02/2021

5.11 - What is the next step after the completion of this project for the conservation of the species? *

100 word limit

The population assessment for the Mariana flying fox resulting from this project, will help us better assess whether a reintroduction to Saipan and other islands is feasible.

5.12 - Why is this project important to your personal development as a conservation biologist and the work / objectives of your organization? *

100 word limit

I am currently pursuing my masters in conservation biology and this study will be for my thesis. I have always had a passion to work with mammals and more importantly conserve them. This project is important as it would allow me to complete my masters program, furthermore it will provide me with the experience required to kickstart my career as a biologist.

Section 6 - Media Support

Please note that any information provided on this application may be made available to the media if the application is successful.

6.1 Please also attach the following support material, to be used by the Fund for publicity:

A short biography / curriculum vitae of the project applicant and a list of his / her relevant scientific and popular publications *

✓ File uploaded.

A recent Annual Report or other description of your organization (e.g. strategic plan), if applicable

You have not uploaded a file.

Previous media coverage of your work to date, especially relating to the target species for this project. Successful applications will be able to provide more later

You have not uploaded a file.

6.2 Please use Google Maps to illustrate the project area: *

Whilst we understand your project may cover vast areas, we need to illustrate your work by placing it on a map. So please base this location on what you might consider to be the main

focus area (even if it is in the middle of an ocean). Simply click on the map below so that it updates the form fields below the map labelled as 'Latitude' and 'Longitude'. You can drag the map, or use the scroll and zoom tools on the left of the map to navigate to your location.

*** GOOGLE MAP TOOL APPEARS HERE IN ONLINE APPLICATION ***

Latitude: 14.146421740669552

Longitude: 145.21464396945998

6.3 Please provide one JPEG image of the project's target species. (Wild shots preferred). Successful applicants will be able to provide more later: *

Project's primary target species - *Pteropus mariannus* (Mariana Flying Foxes)

✓ File uploaded.

Section 7 - References

Please provide one to three references below:

Reference 1 *	
Name: *	Luis Ortiz-Catedral
Organization: *	Massey University
Phone (work): *	+64 21 073 3351
Mobile: *	+64 21 073 3351
Email: *	L.Ortiz-Catedral@massey.ac.nz

Reference 2	
Name:	Daniel Thomas
Organization:	Massey University
Phone (work):	+64 22 187 3670
Mobile:	+64 22 187 3670
Email:	D.B.Thomas@massey.ac.nz

Section 8 - Permits

8.1 Will this project require authorization, permits or licences, to complete the activity? *

✓ Yes ☐ No

If yes, describe below. *

Please note that the grantee is entirely responsible for obtaining all necessary authorization documents and adhering to the spirit and letter of all relevant international and national legal mechanisms (e.g. CITES and CBD).

Authorization, Permits Or
Licence Name

Description

Applied for?

<input checked="" type="checkbox"/>	Research permit from the Department of Fish and Wildlife CNMI	Document that approves the activities outlined here for visiting scientists	Yes
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Section 9 - Budget

9.1 Please provide a cost estimate for the specific activities that the grant would support:

*

Activity	Description	Budget Estimate (us \$)	Time It Will Take To Complete (Months / Weeks)	
<input checked="" type="checkbox"/>	Estimating the population size of the Mariana flying foxes on Rota island, CNMI	Visit roosting sites and conduct observational counts using a spotting scope at key roosting sites	3600	2 - 3 months
<input checked="" type="checkbox"/>	Analysing the effects of captive rehabilitation	Visit rehabilitation centre on Rota island, receive rabies vaccination prior to bat handling	2600	6 - 12 months
<input checked="" type="checkbox"/>	Investigating dietary preference	Visit roosting sites, analyse faeces for seeds	3200	2 - 3 months

Budget Total US: \$9400

9.2 - Please detail how you would spend the money you are requesting from the Species Fund: *

200 word limit

Funds will be used to cover the cost of accessing field sites by plane (overall US \$2000), and to purchase food for duration of stay on Rota island (US \$1700). Funds will also be used for vehicle transportation hire and petrol (US \$1150) to access key roosting sites for the duration of the research. In addition, funds will be used to obtain rabies vaccination prior to research (US \$130). The remaining funds will be used to cover the accommodation cost required during our stay on Rota island (US \$4420).

9.3 - Please specify if there is anything else you wish to highlight regarding this budget (e.g. This funding would be combined with funding from other organizations): *

200 word limit

Massey University and CNMI Division of Fish and Wildlife has offered in kind support with regard to equipment hire, and access to facilities. Equipment use trainings and bat handling techniques will be conducted by the staff of CNMI Division of Fish and Wildlife organisation.

Fields marked with an asterisk denote mandatory fields *

APPENDIX III. Auckland Zoo Conservation fund application



Auckland Zoo Conservation Fund Small Grants Application Form

- This form must be completed in English.

Section 1 – Contact Details

1.1	Date	3/9/2019
1.2	Principal applicant and title	Colin Heng Zhao Jie
1.3	Name of institution or organisation	Massey University
1.4	Contact telephone number	+64 21 086 00328
1.5	Contact address	23A Lewisham Street Highland park Auckland 2010
1.6	Email address	c.t.heng@hotmail.com
1.7	Website address	www.islandsforever.com
1.8	Project partners or other participants	Luis Ortiz-Catedral Joshua Guilbert
1.9	Individual applicants please include (as separate attachments) your résumé/ CV and two professional reference letters, with contact details for your referees, with your application.	

Section 2 – General Project Details

2.1	Project title	Population trends and distribution of the Mariana flying foxes (<i>Pteropus mariannus</i>) on Rota island
2.2	Project location and country	Rota island, CNMI
2.3	Focal species and/or habitat	Mariana flying foxes (<i>Pteropus mariannus</i>)
2.4	Focal species IUCN (and /or National) threat status	Endangered
2.5	Project start date	5/02/2020
2.6	Project completion date	5/02/2021
2.7	Project Summary (200 words or less)	
	<p>This project encompasses the population and ecological assessment of the unique Mariana flying foxes (<i>Pteropus mariannus</i>) on Rota island. Apart from being classified as endangered under the IUCN red list, Rota island is the only inhabited island in the CNMI to hold a population of these bats. They currently face many threats such as habitat modification, hunting and trapping, and introduced species causing their population numbers to decrease. The last population assessment conducted was in 2008 and there are limited and outdated publications on them. The importance of population assessment allows conservation biologists to decide on the necessary conservation actions required to ensure survivability of the species. Ecological assessment such as foraging habits and the effects of rehabilitation will too be analysed to provide more information on these bats.</p>	

Section 3 – General Grant Details

3.1	Amount of money requested from Auckland Zoo (in New Zealand dollars)	\$5,000
3.2	Total project budget (in New Zealand dollars)	\$20,000
3.3	Other <i>confirmed</i> financial sources of project support	NA.
3.4	Other <i>potential</i> sources of financial project support	Mohammed bin Zayed Species Conservation Fund
3.5	Details of any <i>in-kind</i> project support	<p>Equipment will be supplied by CNMI Division of Fish and Wildlife</p> <p>And the Animal Rehabilitation centre on Rota island</p> <p>Massey University contributes approximately \$10,000 in kind support with field equipment, and access to facilities.</p>
3.6	Previous awards from Auckland Zoo (give details)	None

Section 4 – Project Aims and Objectives

Conservation Issue or Problem	Project Aim or Objective
Outdated population assessment	To estimate the population size and distribution of the Mariana flying foxes on Rota island
Unknown foraging habits and dietary diversity	To determine key foraging resources and their location in the landscape
Effects of rehabilitation	Comparison of behaviour to the pups being raised in captivity to those being raised in the wild

Section 5 – Project Activities and Anticipated Achievements

Project Aim/Objective (from Section 4)	Project Activity or Method	Anticipated Result/Achievement
To estimate the population size of the Mariana flying foxes on Rota island	Observational studies by using a spotting scope at key roosting sites to estimate population abundance.	To be able to understand their population abundance and proceed with the necessary conservation actions
Foraging habits	Investigating food diet by analysing faeces.	Provides ecological information on their diet
Effects of rehabilitation	Discussing with the team on Rota island about the effects of rehabilitation on abandoned pups as compared to those being brought up in the wild.	To understand the effects of rehabilitation. Which could be used for other species in the future

Section 6 - Expertise, Experience and Knowledge

Project Activity or Method (from Section 5)	Relevant Expertise, Experience and Knowledge
Population assessment	Scientists from the CNMI Fish and Wildlife division have been studying these bats for years and have been using GPS trackers to track the movement of these bats.
Foraging habits	Liaising with scientists on the island and also speaking with the locals
Effects of rehabilitation	Using their rehabilitation centre coupled with results obtained from the researchers on the island to provide accurate findings

Section 7 – Project Evaluation

Anticipated Result/Achievement (from Section 5)	Measure of Success – give indicators
Population abundance and distribution	Acquiring a population estimate on the Mariana flying foxes found on Rota island
Foraging habits	To determine preferred fruit types consumed by the Mariana flying foxes
Effects of rehabilitation	Using results from past analysis and provide a comparison of different behaviours observed between captured and wild pups

Section 8 – Other Information

7.2	Details of permissions and/or permits and/or licenses required to carry out project	
Do you have permission to work at the site/s?		
Yes	<input checked="" type="checkbox"/>	If yes, give permit number if applicable:
No	<input type="checkbox"/>	
N/A	<input type="checkbox"/>	
7.3	Details of any animal capture, collection, handling, manipulation, sampling etc. required	
Will you be handling animals?		
Yes	<input checked="" type="checkbox"/>	
No	<input type="checkbox"/>	
N/A	<input type="checkbox"/>	
Occasionally, orphaned bat pups are encountered and brought into captivity for rehabilitation. I will receive training from qualified field biologists, in particular Dr. Joshua Guilbert. Handling will be limited to feeding, any required rehabilitation and also for regular weighing and measuring.		
If yes, please give details here:		
Do you have the necessary permits to capture / handle / take samples?		
Yes	<input checked="" type="checkbox"/>	If yes, give permit number if applicable:
No	<input type="checkbox"/>	
N/A	<input type="checkbox"/>	
7.4	Details of any potential human health or safety risks or issues and how they will be addressed	
Unforeseen injuries – health and safety plans will be implemented in the event of injuries according to regulations and recommendations as well as safety standards by Massey University.		
Potential rabies infection – Rabies vaccination will be taken before the start of the project.		
7.5	Details of any ethical considerations for the animals involved in your project	
As there will be handling of the bats, handling practices will be conducted by the CNMI Division of Fish and Wildlife organization to ensure the animals are not harmed during the process.		

Section 9 – Details of Project Budget

Description of Budget Item in Priority Order	Proposed Auckland Zoo contribution (please indicate in New Zealand dollars)	Proposed contribution from other source (please indicate in New Zealand dollars)	Total Cost of Item(s) (please indicate in New Zealand dollars)
Accommodation	\$0	\$5,000	\$5,000
Transport (including air fares)	\$3,000	\$1000	\$4,000
Rabies Vaccination	\$0	\$200	\$200
Food	\$700	\$300	\$3,700
Fuel for daily transport to study sites	\$1300	\$500	\$1800
Sub-total: Auckland Zoo contribution	\$5,000		
Sub-total: Contribution from other sources		\$7000	
Percentage	AZ%	Other %	
TOTAL PROJECT BUDGET			\$14,200

Section 10 – The Final Appeal

Describe, in 200 words or less, why this proposed project is particularly special and why it should be specifically funded by Auckland Zoo.

The Mariana flying foxes are an endemic species of bats found in the Mariana islands. They have a broad distribution and are mainly found on uninhabited islands. According to the IUCN, they are currently listed as endangered on the IUCN red list and has not been assessed since 2008. Populations of these flying foxes can be found in Guam, Japan, Federate States of Micronesia and CNMI but are decreasing. Rota island is the only island in the CNMI to have Mariana flying foxes cohabiting with humans.

Apart from having limited ecological information, the population abundance and distribution are currently out dated. The importance of reassessment provides conservation biologists the information needed to plan for future conservation actions. In addition, since New Zealand is considered the lead in island conservation, implementing similar conservation plans towards the Mariana flying fox could be useful in their conservation success.

**APPENDIX IV. PHOTOGRAPHS OF GALAPAGOS LAND IGUANAS
(*CONOLOPHUS SUBCRISTATUS*) ON BALTRA ISLAND.**

Photos: L. Ortiz-Catedral

Males



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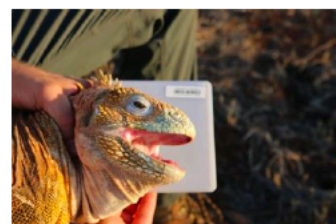
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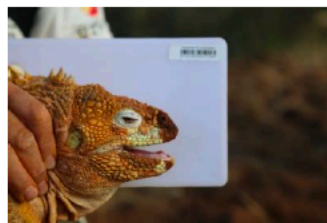
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Females



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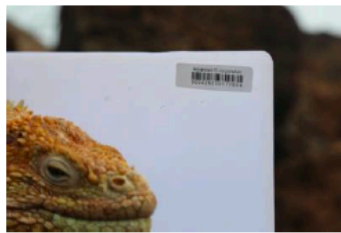
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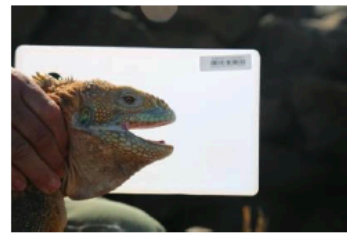
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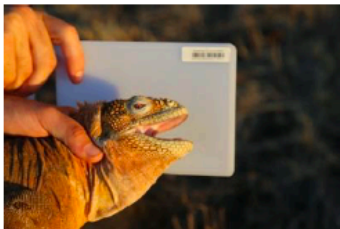
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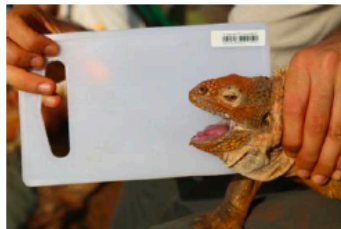
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**APPENDIX V. PHOTOGRAPHS OF GALAPAGOS LAND IGUANAS
(*CONOLOPHUS SUBCRISTATUS*) ON FERNANDINA ISLAND.**

Photos: L. Ortiz-Catedral

Males



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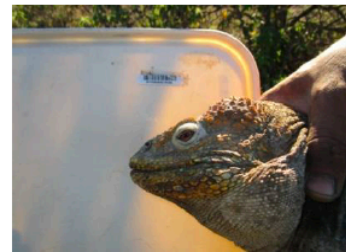
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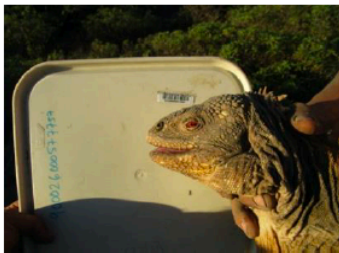
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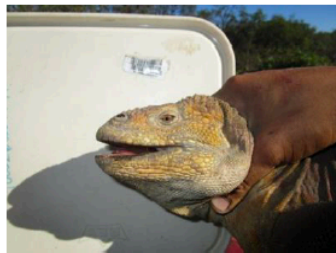
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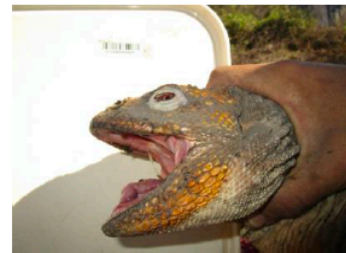
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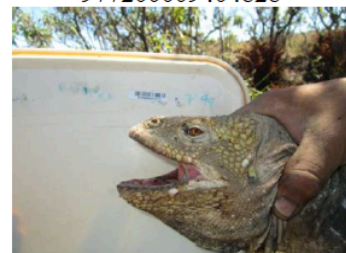
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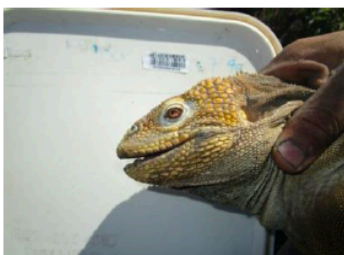
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Females



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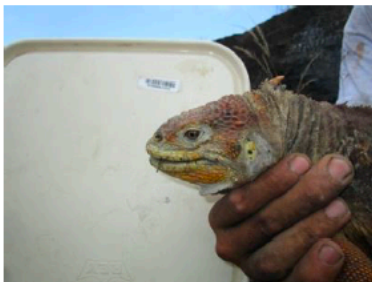
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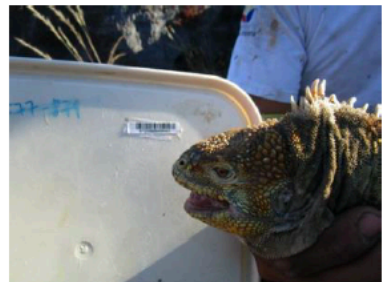
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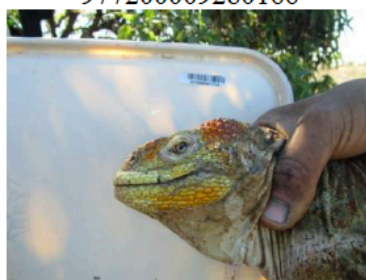
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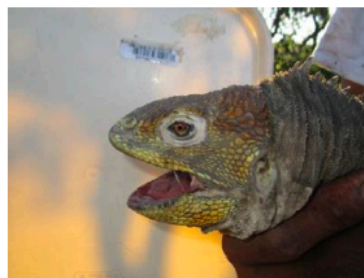
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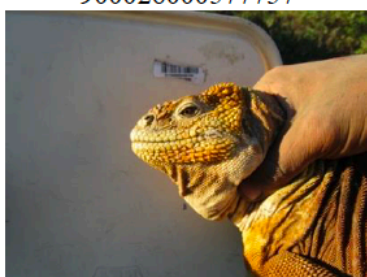
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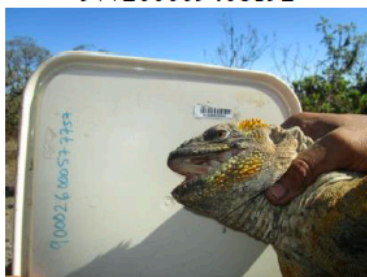
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Undetermined sex



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