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Health, Behaviour and Individual Differences of Capuchin Monkeys (*Sapajus* sp.) in Rehabilitation for Reintroduction

Guillermina Hernández Cruz

A dissertation submitted to the University of Bristol in accordance with the requirements for award of the degree of PhD in the Faculty of Health Sciences, Bristol Veterinary School, January 2022.

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We’re not the only
beings on the planet
with personalities,
minds, and emotions.

Dr Jane Goodall

Abstract

Thousands of trafficked Neotropical primates enter Latin American wildlife rehabilitation centres every year. There is a lack of research related to their health and behaviour and how these relate to personality. Moreover, the ethical issues on their reintroduction have not been discussed. The first aim of this dissertation was to investigate the health of capuchin monkeys in two Brazilian rescue centres and obtain their haematological and physiological values and parasitological status. Results showed that they had similar physiological and haematological values to previously published data. Five individuals were positive to *Ancylostoma* spp. The second aim was to analyse the behaviour of the capuchins to provide an initial assessment of their rehabilitation. This was performed by using changes in behaviours (activity budgets) and a behavioural diversity index. When comparing the baseline and final observational phases, behavioural diversity, affiliative behaviours and inactivity increased whilst human interaction decreased. The third aim was to assess the personality structure of the capuchins by utilising behavioural observations, tests and observer trait ratings. Four dimensions were obtained from trait ratings (Openness, Neuroticism, Assertiveness and Sociability) and five from tests (creative, aggressive, stereotypic, sociable to humans and risk-averse). The fourth aim was to investigate the association between personality and health (body condition and neutrophil-to-lymphocyte ratios) of the individuals. There was a significant negative correlation between Sociability and neutrophil-to-lymphocyte ratios and between stress-related behaviours and body condition. The fifth aim was to investigate the association of personality with behavioural changes during rehabilitation. Results suggested stereotypy, aggression and Assertiveness influenced vigilance behaviours and Neuroticism influenced space and substrate use. The sixth aim was to analyse the ethical issues related to the reintroduction of trafficked Neotropical primates. I developed an Ethical Matrix and suggested a set of recommendations as guidelines that may be used when confronted with these issues.

Covid-19 statement

Originally, this PhD dissertation included two data collection phases, planned in several government wildlife rescue centres in Northeast Brazil. The first occurred as planned, between the 28th of February 2019 and the 23rd of July 2019. The second, which was planned to take place between 30th of March 2020 and 30th of August 2020, could not occur due to the COVID-19 pandemic and the consequent travel restrictions imposed by the University of Bristol and the UK government. Unfortunately, these restrictions were still in place during the last year of my PhD (2021). Hence, the second data collection could not take place at any time of my PhD research project.

We (i.e., the author and my PhD supervisors) made several modifications to the PhD research project to try to ensure it could be performed safely and in accordance with the University and government regulations without sacrificing its scientific quality. As the main goals of my second data collection phase were (1) to enlarge the sample size of the study and (2) to perform an environmental enrichment experiment, I had instead to address my research questions using the data I collected from the first data collection phase alone. I also replaced the environmental enrichment experiment (planned to be Chapter 7) with an ethical review of the rehabilitation and reintroduction of Neotropical primates rescued or confiscated from the illegal pet trade in Latin America. This meant that the sample sizes of the final studies were smaller than originally expected, and this inevitably impacts on the robustness of my findings – the originally planned larger sample size would have given me a better sample of personality data from which to draw conclusions. Nevertheless, I was able to perform statistical analyses as planned, bearing this clear and unavoidable proviso in mind.

Dedication and Acknowledgements

I would like to acknowledge the support and funding given by CONACyT and the University of Bristol to perform this PhD research project. To my supervisors and collaborators Professor Michael Mendl, Dr Nicola Rooney, Dr Renata Ferreira and Dr Siobhan Mullan, thank you for all your patience, feedback and support; as well as to my examiners Dr Suzanne Held and Dr Blake Morton for their very helpful suggestions and insight. Finally, to all members from the Animal Welfare and Behaviour group at the University of Bristol and everyone from Operação Sapajus and CETAS–RN and CETAS–CE IBAMA, who supported the development of this research project.

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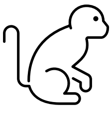

Author’s declaration

I declare that the work in this dissertation was carried out in accordance with the requirements of the University's Regulations and Code of Practice for Research Degree Programmes and that it has not been submitted for any other academic award. Except where indicated by specific reference in the text, the work is the candidate's own work. Work done in collaboration with, or with the assistance of, others, is indicated as such. Any views expressed in the dissertation are those of the author.

SIGNED: .... DATE: ....

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Chapter 1: General Introduction and Aims

Chapter 1: General Introduction and Aims

The main focus of this dissertation was the study of the association of individual differences in personality with the health and behaviour during rehabilitation of trafficked (i.e., born in the wild and raised in captivity as pets) bearded capuchin monkeys (*Sapajus libidinosus*). Capuchin monkeys, as well as many other wildlife species, are frequently found in wildlife rescue centres in Latin America. The rehabilitation and reintroduction of these animals are challenging from a practical and ethical perspective as there are few examples in the scientific literature regarding the pre-release training and assessment of Neotropical primates and previous studies have yielded low to medium post-release survival rates. Furthermore, few studies have been made on their health and behaviour during rehabilitation as well as on the relationship between individual differences in personality and these variables.

This Chapter is organised into several subsections: 1.1. Primate Rehabilitation and Reintroduction, 1.2. Personality, 1.3. Personality and Reintroduction Biology and 1.4. the Aims of the Study.

1.1. Primate Rehabilitation and Reintroduction

1.1.1. The illegal primate trade: conservation and welfare-related issues

Approximately 3,500 non-human primates (hereafter, primates) are exported annually from countries where primates naturally occur (known as primate-range countries). USA, Japan, and China are the main importers of primates, whilst China and Mauritius are the main exporters (Nijman et al., 2011). Trafficked primates may be sold to biomedical laboratories, pharmaceutical industries or as bushmeat. However, many of them are sold to private collectors or as family pets (Nijman et al., 2011). It is unknown how many primates are kept as pets globally; nonetheless, it is believed the number has increased every year, particularly in the Americas, Africa and Asia (Soulsbury et al., 2009).

The illegal wildlife trade is one of the major illegal activities worldwide, with its annual value being estimated by the United Nations as between seven and 23 billion US dollars (TRAFFIC, 2021). Furthermore, the illegal trade represents a major threat to both primate conservation and wellbeing. In fact, approximately 65% of primate species are currently considered as endangered and 75% have declining populations (Estrada et al., 2020); this is mainly due to human activities, such as habitat destruction and hunting for illegal trafficking (Guy et al., 2014; Estrada et al., 2017; Estrada et al., 2020).

Extracting primates from their natural habitats and keeping them as pets may negatively impact their individual health and welfare (Duarte-Quiroga and Estrada, 2003; Soulsbury et al., 2009). The concept of the ‘Five Freedoms’, originally developed to provide a framework for animal welfare (i.e., of domestic animals), may also be used as a framework for considering the welfare of wild animals kept as pets, including primates (Schuppli and Fraser, 2000; Soulsbury et al, 2009). According to this framework, all animals in captivity should have freedom from hunger, thirst, and malnutrition; freedom from disease and injury, freedom from physical forms of thermal discomfort, freedom from fear, distress, and negative psychological states, and freedom to express their natural behavioural repertoire (Schuppli and Fraser, 2000; Soulsbury et al., 2009).

Nonetheless, trafficked primates are not kept (as pets) in conditions where their needs associated to the Five Freedoms are met. They usually experience maternal and social deprivation and, in many cases, inadequate housing conditions that can lead to health and behavioural abnormalities, such as nutritional and infectious diseases and the exhibition of motor stereotypies and/or self-injurious behaviours (Duarte-Quiroga and Estrada, 2003; Soulsbury et al., 2009). Injuries are also common, with burns, electrocution and strangulation within the home environment being the most frequent (Duarte-Quiroga and Estrada, 2003).

Frequently, the response of the government in primate-range countries is to confiscate the individuals that are kept illegally. However, what should be done with these animals next? Primate rehabilitation and reintroduction programmes have arisen as a response to this question.

1.1.2. Primate rehabilitation and reintroduction

Beck et al. (2007) define primate rehabilitation as ‘*the process by which captive primates are treated for medical and physical disabilities until they regain health, are helped to acquire natural social and ecological skills, and are weaned from human contact and dependence, such that they can survive independently (or with greater independence) in the wild’* (Beck et al., 2007; p. 5). Similarly, the International Wildlife Rehabilitation Council (IWRC) defines wildlife rehabilitation as ‘*the treatment and temporary care of injured, diseased and displaced indigenous animals, and the subsequent release of* *healthy animals to appropriate habitats in the wild*' (Miller et al., 2012, p. ix). Thus, wildlife/primate rehabilitation programmes' main objective is to counteract the effects of the illegal trade on the health and welfare of confiscated wild animals to reintroduce them back into their natural habitats (Guy et al., 2014).

However, the rehabilitation and reintroduction of ex-captive primates has been –and still is– highly controversial, and raises many questions among primate conservationists (Shanee, 2007). Historically, the post-release mortality rates of primate reintroduction programmes have been considerably high. For example, a Bornean gibbon (*Hylobates muelleri*) rehabilitation and reintroduction programme studied in the 1990s resulted in less than 10% of the individuals surviving after release in a period of three months to 12 years (Bennett et al., 1992). More recent studies have had slightly better outcomes. Two studies performed with rehabilitated vervet monkeys (*Chlorocebus aethiops*) in South Africa in 2008 and 2009 recorded 31% and 56% survivorship after 10 and 6 months (respectively) post-release (Wimberger et al., 2010; Guy et al., 2012). The high mortality rates of many reintroduction programmes contribute to the primate rehabilitation and reintroduction controversy. Nonetheless, some relatively recent studies, such as de Palomino et al. (2013) and Bennett et al. (2013), in which a group of nine spider monkeys (*Ateles chamek*) and a group of 11 woolly monkeys (*Lagothrix lagothrica*) were released, reported that 100% of the monkeys survived after 12 and 6 months, respectively (de Palomino et al., 2013; Bennett et al., 2013). Interestingly, these reintroduction projects had extensive post-release monitoring and food provisioning as well as community support.

1.1.3. Methods for primate pre-release training and assessment

Studies describing rehabilitation techniques and methods of pre-release behavioural assessment in primate reintroduction projects are scarce in the scientific literature (Ongman et al., 2013). Here, I will make a summary of the studies I have found, focusing on primate rehabilitation and including some examples of the rehabilitation of other wildlife species where appropriate (for example, when there is not enough information on primate rehabilitation). I conducted this scientific literature search using online scientific and open search engines, such as Web of Science, Science Direct, Google Scholar and Google. I used the words 'primate' + 'rehabilitation', 'primate' + 'reintroduction' and 'primate' + 'translocation'. This was performed in English and

Spanish. A similar process was used for other wildlife studies. I also reviewed the International Union for the Conservation of Nature (IUCN) available information on reintroduction such as the IUCN Global Reintroduction Perspective Series edited by Soorae (2002, 2008, among others), the IUCN Guidelines for Non-human Primate Re-introductions (Baker, 2002), etc. For primate reintroduction, the review entitled 'A History of Primate Reintroduction' by Beck (2017) was particularly useful.

1.1.3.1. Behavioural assessment and pre-release training

The behaviour and socioecology of the species of interest must be considered as part of any primate rehabilitation and reintroduction programme. Before releasing primates back into the wild, it must be clear that they are behaviourally fit to survive (Baker, 2002). Nonetheless, few studies have assessed the effectiveness of rehabilitation techniques and/or established behavioural indicators of rehabilitation success in primates (Ongman et al., 2013; Schwartz et al., 2016).

Primates that have been kept as pets often develop behavioural abnormalities and lose their natural behavioural repertoire, which may compromise their welfare and post-release survival (Soulsbury et al., 2009; Cheyne, 2006). Hence, most primate reintroduction programmes include some form of behavioural modification (Shanee, 2007), also known as pre-release training. Behavioural modification may aid reintroduction programmes by helping the animals develop the necessary survival skills (Shier, 2006).

It has been proposed that reintroduction programmes should be conducted as scientific experiments, by transforming reintroduction from an animal management practice into a scientific discipline (e.g., by establishing clear research questions and hypotheses and publishing the results) (Seddon et al., 2007; Reading et al., 2013; Shier, 2016). In this sense, behavioural assessment and training must be performed with clear objectives and measured variables.

The behavioural skills that primates should possess to survive in the wild differ between species; overall, they should be able to forage, socialise with conspecifics, recognise and avoid predators, and move in their natural environment adequately (Reading et al., 2013, Melfi and Marples, 2000). Many reintroduction programmes fail partly due to the reintroduced individuals being unable to recognise predators, food, or water resources at the release site (Cheyne, 2005). Therefore, all programmes should assess and train (if needed) animals in these basic skills or provide them with opportunities to develop these skills. I will briefly discuss each f these skills, dividing them into (1) social and (2) ecological skills. As stated by Ongman et al. (2013), both domains are of equal importance but require the development of different abilities (Ongman et al., 2013).

1.1.3.1.1. Social skills

Behavioural problems due to poor socialisation are common in trafficked primates as they are often kept in social isolation (Soulsbury et al., 2009). They often exhibit increased aggression, motor stereotypies and/or self-injurious behaviours, which may be detrimental for their wellbeing (Swett, 1993; Soulsbury et al., 2009).

Suárez et al. (2001) and Cheyne et al. (2012) suggested introducing all individuals that will be part of a release group to a new enclosure simultaneously to reduce the likelihood of fighting, provided they do not have severe behavioural abnormalities that may pose a risk to conspecifics, such as extreme aggression or fearfulness (Suárez et al., 2001; Cheyne et al., 2012). In these cases, individuals could be gradually exposed to conspecifics. Auditory contact may be established first, followed by visual and tactile contact (Cheyne et al., 2012).

Moreover, the social structure of the species of concern must be considered when attempting to form social groups that will be released together (Baker, 2002). Gibbons (*Hylobates* spp.), for example, should be released in pairs (male and female adults) that have already developed a strong social bond, due to their monogamous social system (Cheyne, 2009; Cheyne et al., 2008; Cheyne et al., 2012). Cheyne et al. (2012) recommended gradually exposing fearful or aggressive individuals to conspecifics to avoid fights. Furthermore, selecting a method that mimics the natural formation of social groups may improve release outcomes in reintroduction projects (Gusset et al., 2006).

Social isolation may result not only in poor socialisation with conspecifics, but also in a strong attachment to human beings (Soulsbury et al., 2009). This may be a risk worth considering for individuals that are going to be released in the future, as approaching humans may result in the individuals' re-capture. Centro de Primatología Araguatos (2004), for example, reported that rescued capuchin monkeys (*Cebus* sp.) that were released back into the wild, kept trying to establish contact with humans during the first weeks after release (Centro de Primatología Araguatos, 2004). To avoid this, Suárez et al. (2001) and Cheyne et al. (2012) suggested keeping contact with humans to the minimum during the rehabilitation period, which could have the added benefit of preventing zoonoses and injuries to the staff working with the animals (Suárez et al., 2001; Cheyne et al., 2012). Nonetheless, I have not found any study that has demonstrated experimentally that avoiding contact with humans during rehabilitation improves the survival prospects of reintroduced individuals and/or helps to avoid human-wildlife conflict. This could be a potential gap to fill regarding primate rehabilitation and reintroduction studies.

1.1.3.1.2. Ecological skills

The ecological skills that primates need to learn before being reintroduced mainly include those related to foraging and locomotion. Primates spend up to 60% of their waking time foraging (Santra, 2008); thus, the development of species-specific foraging skills is vital for reintroduced primates to survive. Similarly, locomotion is another key requirement to achieve reintroduction success (Ongman et al., 2013). Factors affecting the development of these skills mainly include husbandry practices, but may also include pre-release training.

Captive-raised primates are often provided with diets that are inappropriate for their species or even given substances which their consumption may be harmful, such as tobacco and illegal drugs (Duarte-Quiroga and Estrada, 2003; Soulsbury et al., 2009). In rehabilitation, it is recommended to change their diets gradually until they resemble those found in the wild. In several primate rehabilitation studies, the individuals were fed with fruits, vegetables, and insects (Suárez et al., 2001; Vogel et al., 2002; Araguatos, 2004; Arango-Guerra, 2013; Schwartz et al., 2016; Guy et al., 2012; Guy, 2013). Commercial food and cereals may be given as well, provided they are gradually reduced before release to avoid future crop-raiding behaviours (Vogel et al., 2002; Araguatos, 2004; Guy et al., 2012; Guy, 2013). The use of wild fruits and leaves is also recommended so that the animals can identify them upon release (Cheyne et al., 2012; Arango-Guerra, 2013; Schwartz et al., 2016). Moreover, it has been recommended to feed the animals in a way that prevents them from developing an association between food and humans, such as using poles and/or elevated feeders which may also promote locomotion and simulate a ‘natural’ environment (Suárez et al., 2001; Araguatos, 2004; Arango-Guerra, 2013; Vogel et al., 2002).

Even though most wildlife species do not require special training in locomotion, arboreal species, including many primates, need opportunities to develop the necessary skills to move efficiently in complex three-dimensional environments (Reading et al., 2013). Thus, the design and enrichment of enclosures is an important factor to consider. Cheyne et al. (2012), for example, recommended enclosures of rehabilitant gibbons to be pyramidal with approximately five meters high, three meters wide and three meters long; and ideally constructed with materials that are inexpensive and available locally (Cheyne et al., 2012). Moreover, enriching the enclosures with tree branches and other structures may improve aerial locomotion (Suárez et al., 2001; Araguatos, 2004; Arango-Guerra, 2013). As with other aspects of primate rehabilitation, these are mainly recommendations made by primate behaviour experts based on years of experience working in primate rehabilitation, rather than experimental studies with control groups.

1.1.3.1.3. Predator avoidance

Mortality by predation is one of the most common reasons for failure in wildlife reintroduction projects (Griffin et al., 2000). Hence, the ability to recognise, detect and respond to potential predators is a key aspect of behavioural competency that must be assessed and taught to achieve reintroduction success (Griffin et al., 2000; Shier, 2016).

The lack of anti-predator skills in wild animals may be related to one of two causes (Griffin et al., 2000). Firstly, it may be the case that the individuals have been isolated from predators only throughout their development, known as *ontogenetic isolation*. This happens, for example, when individuals from free-ranging groups have been captured as infants and raised in captivity (i.e., ‘captive-raised’). Conversely, there are individuals that have been isolated from predators over evolutionary time (*evolutionary isolation*), meaning several generations have been either in captivity or in a place where there are no predators (Griffin et al., 2000). This is the case of individuals that are part of reintroduction projects that include captive-born animals raised in zoos or other captive facilities. Griffin et al. (2000) have suggested that individuals that have been isolated ontogenetically from predators will be more easily trained in anti-predator skills than individuals that have been evolutionary isolated (Griffin et al., 2000). However, this is difficult to test in a reintroduction programme, as most programmes include either captive-raised or captive-born animals, but not both.

Generally speaking, anti-predator training will include some type of associative learning (Griffin et al., 2000; Shier, 2016). This is commonly performed by classical conditioning: the animals to be reintroduced are submitted, whilst in captivity, to the predators' presence, by presenting them with models or cues of a natural predator species (the conditioned stimulus or CS) and pairing this with an aversive stimulus (the unconditioned stimulus or UCS) (Griffin et al., 2000; Shier, 2016). Depending on its nature, the UCS may cause discomfort, pain and/or fear in the individuals (e.g., a splash of water may cause discomfort, an electric discharge may cause pain, an alarm call from conspecifics may cause fear). It has been proposed that for UCSs to be useful, they should elicit the same motivational state that natural predatory events do (Griffin et al., 2000). In this sense, UCSs that elicit a fear-like state would be more useful than stimuli that cause pain or discomfort (Griffin et al., 2000). However, anti-predator training may be a stressful experience for animals (Teixeira et al., 2007). Because of this, it may be seen as detrimental for their short-term welfare (in captivity). Nonetheless, this may be a case in which the long-term benefits may outweigh the costs, as animals equipped with anti-predator skills will have better survival prospects when released, thus improving their long-term welfare (Shier, 2016).

The IUCN Guidelines for Non-human Primate Re-introductions (2002) recommend assessing the individuals' ability to cope with predation before release (Baker, 2002). Guy et al. (2012) reported that the rehabilitated vervet monkeys (*Chlorocebus aethiops*) released in their study responded to predators such as birds of prey and small felids by performing alarm calls and seeking shelter, and this was used as a release criterion (Guy et al., 2012). Similarly, in a study by Suárez et al. (2001), they reported providing audio-visual stimuli to eight brown capuchin monkeys (*Sapajus apella*) as part of their rehabilitation and reintroduction programme. This included artificial models of predators, as well as bird and other primate species' vocalisations (Suárez et al., 2001). However, it is not reported how the animals responded to the anti-predator training during captivity nor after release; thus, the results and potential benefits of the training are unclear. Furthermore, there was no control group in this study. This would have been useful to show that their methods did indeed have a significant and desired effect on the behaviour of the animals.

In a study by Centro de Primatología Araguatos (2004), 16 white-fronted capuchin monkeys (*Cebus albifrons*) in rehabilitation were presented with artificial models of several species of predators, such as eagles and wild felines, and a live dog, a live snake and a person as anti-predator training. This training yielded mixed results. While in captivity, several monkeys responded adequately (e.g., seeking shelter) to the presence of the live predators (the snake and the dog) as well as the person (which acted aggressively towards them, so the individuals would learn to identify humans as potential predators) by showing defensive behaviours and climbing to the upper part of the enclosure. The eagle models did not elicit the appropriate response in the monkeys; however, it was reported they reacted adequately to the presence of the feline predator model (Centro de Primatología Araguatos, 2004).

In another example, Campbell and Snowdon (2009) wanted to induce mobbing (i.e., '*the gathering of members of a group around a potentially dangerous individual*', Davis and Arkin, 2012; p.276) responses in captive-born cotton-top tamarins (*Saguinus oedipus*) by adapting a method previously used for birds. To achieve this, they introduced a Plexiglas box containing a snake (i.e., the CS) to the enclosure of 15 pairs or small groups whilst alarm/mobbing calls were being played (i.e., the UCS), previously recorded from conspecifics. This was repeated several times. The tamarins responded to the sound of the mobbing calls with vocalisations and piloerection; however, when they were presented with the snake without the calls, they failed to produce any mobbing or alarm call. This could mean that they did not recognise the snake as a predator even after hearing the mobbing calls from conspecifics; in other words, the conditioning was not successful. As stated by the authors, auditory playback may not be enough to produce mobbing responses in captive-born primates as it is in other species due to the specific learning strategies of primates (Campbell and Snowdon, 2009).

Even though some of these results are encouraging, without the use of control groups and post-release monitoring, there is no way of knowing if the anti-predator training really had a positive effect on the post-release survival of the individuals or even in eliciting the appropriate behaviours in the monkeys whilst in captivity. There is a need of more research in anti-predator training in trafficked primates, with adequate control groups and clear objectives.

1.1.3.2. Health assessment

From a veterinary and public health perspective, health assessment is one of the most important parts of rehabilitation in reintroduction programmes. In fact, the IUCN states that primate reintroduction projects must have a veterinary programme that includes quarantine and disease screening (Baker, 2002). Health assessments also help to identify potential issues that may prevent individuals from being suitable for release, such as diseases or injuries acquired during captivity. Slow lorises' (*Nycticebus* spp.) teeth, for example, are often cut or removed in illegal markets, which may compromise their survival after release (Moore et al., 2014). Injuries acquired during captivity may also prevent confiscated gibbons (*Hylobates* spp.) or spider monkeys (*Ateles geoffroyi*) from being suitable candidates for reintroduction (Cheyne, 2005; personal observation). Therefore, animals with permanent injuries that may negatively affect their post-release survival should not be released in any case.

According to the IUCN, the ideal quarantine period for primate reintroduction programmes is 90 days. If this is not possible, 31 days should be considered as the minimum, but 60 days is preferred (Baker, 2002). Many of the rehabilitation and reintroduction programmes reported in the literature include quarantine periods as part of their programmes, in which the animals are isolated from other animals for 42 to 56 days (Suárez et al., 2001; Arango-Guerra, 2013; Kenyon et al., 2014). Moreover, individuals must be screened before reintroduction to avoid carrying infectious agents that may pose a risk to conspecifics, other wildlife, or human beings. Some infectious agents naturally occur in certain species; thus, the specific tests to perform depend on the taxon and geographical area of interest (Baker, 2002). Tests performed before release commonly include full-body radiographs, haematology, clinical chemistry, screening for TB, toxoplasmosis and HVB; electrocardiograms and echocardiograms; parasitological tests and bacterial and mycotic cultures from faecal samples (Suárez et al., 2001; Araguatos, 2004; Arango-Guerra, 2013; Cardenio et al., 2020).

It must be considered that any procedure that requires handling a primate will be potentially stressful. Even seemingly simple procedures such as sexing or weighing can be a source of stress for wild animals during veterinary examinations (Teixeira et al., 2007). Hence, invasive procedures are generally performed using chemical restraint (i.e., anaesthetics). Small primates may be physically restrained for some procedures, whilst larger species require chemical restraint for most veterinary examinations. Because of this, the use of non-invasive health indicators may be preferred for frequent (routinary) health examinations.

Body condition, which can be measured non-invasively, has been used as a health indicator in primate reintroduction projects. In a study by Strum (2005), in which several troops of baboons (*Papio* spp.) were translocated, body condition was used to assess the health of the individuals and as a measure of reintroduction success, along with other indicators including birth and death rates, mortality and survivorship patterns, and internal parasites loads. It was found that body condition was inversely correlated to internal parasite load, meaning that individuals with higher parasite loads had lower body condition measurements (Strum, 2015). Similarly, body condition has been utilised to assess health and reproductive status in free-ranging primates. In a study with ring-tailed lemurs (*Lemur catta*), it was found that coat and body condition varied with seasos and resource availability; as food availability decreased during the dry season so did body condition of the individuals. Furthermore, body condition seemed to be related to reproductive status: females with offspring had lower body condition measurements when compared to males and females without offspring (Millete et al., 2015). The findings of these studies have important implications for rehabilitation/reintroduction projects, as some health indicators such as coat quality and body condition can be assessed remotely, and can help guide decisions on whether to nutritionally support, manage, or even remove released individuals for further treatment. Moreover, these indicators could be used to assess reintroduction success or even predict rehabilitation outcomes whilst the animals are still in captivity.

1.1.4. Measures of rehabilitation and reintroduction success

In the above sections, I mentioned several skills –social and ecological– that have been proposed as necessary for the post-release survival of reintroduced primates as well as, some studies that explored ways to assess and train animals in these skills. However, this does not necessarily mean that such efforts lead to successful outcomes after release.

In primate reintroduction projects, reintroduction success is commonly measured by initial post-release survival of the releasees as well as their reproduction (King et al., 2014). More recently, population viability analysis has been used to assess reintroduction success among some long-lived primate species, such as Western lowland gorillas

(*Gorilla gorilla*) (King et al., 2014). However, rehabilitation success –which could be considered as a requirement for reintroduction success– seems to be overlooked. There are few scientific studies which specifically propose how to decide when rehabilitant primates are ready for release; in other words, how to measure rehabilitation effectiveness (Ongman et al., 2013).

Cheyne et al. (2012), proposed several behavioural indicators as release criteria in gibbon reintroduction programmes: these criteria stated that (1) the activity budgets of the rehabilitant animals should approximate those of free-ranging conspecifics, (2) future releasees should exhibit adequate locomotion and spatial use: they should mainly move through brachiation and spend more than 40% of the time in the higher tiers of their enclosures, less than 5% on the ground and should not sleep on the ground at all and (3) they should spend less than 3% of the time performing severe abnormal behaviours, such as motor stereotypies and self-injurious behaviours (e.g., self-biting, hair-pulling) (Cheyne et al., 2012).

Release criteria like these are useful because they include species-specific behaviours and are based on the available scientific literature on the behaviour of free-ranging individuals of the same species. However, I have not found published release criteria or indicators of rehabilitation effectiveness for New World monkey species. In the study by Suárez et al. (2001), for example, in which a group of confiscated capuchin monkeys was released back into the wild, the individuals' progress was monitored and assessed, but it is not mentioned how the decision to release the animals was finally made, or which indicators were taken as release criteria (Suárez et al., 2001). Similarly, in a study by Arango-Guerra et al. (2013), five cotton-top tamarins were rehabilitated and reintroduced. In the subsequent report, is the authors mention how the progress of the animals was monitored (e.g., the behavioural data collection method) (Arango-Guerra et al., 2013), but not specifically which behaviours were considered as release criteria.

This issue could be related to the fact that in some primate reintroduction programmes, the decision to finally release the animals is influenced not only by the animals' suitability for release but also by other economic, political and social factors. The pressure to release animals due to space constraints in government rescue centres may be an example of these factors (Osterberg et al., 2015; personal observation). Nonetheless, more research is needed on potential indicators of primate rehabilitation effectiveness and release criteria, particularly among New World monkey species. Having this knowledge could help to fulfil the goals of primate reintroduction projects, such as ensuring the releasees' welfare after release and establishing long-term viable populations.

Primate rehabilitation and reintroduction programmes are necessary to counteract the illegal pet trade. As proposed by Reading et al. (2013), reintroduction programmes should be based on scientific knowledge and should be conducted as scientific experiments. This involves addressing specific research questions and controlling for as much variables as possible. For example, the suitability of a certain behaviour to be used as an indicator of behavioural proficiency in a key ecological skill such as locomotion or foraging. This could be done by analysing the results of an enrichment programme targeting the development of those specific behavioural skills and measuring the outcomes e.g., if the behaviour significantly increased, decreased or changed as needed for the rehabilitation and post-release survival of the individuals (Reading et al., 2013) and following the appropriate guidelines. The outcomes of these programmes –positive or negative– should always be assessed and shared with the scientific and conservation communities to learn from previous experiences and improve the outcomes of these programmes.

1.1.5. Ethical concerns on primate rehabilitation and reintroduction

As seen throughout this review, there may be several concerns related to the rehabilitation and reintroduction of primates. These could be grouped into two main categories: those related to conservation (e.g., of the taxon or biodiversity) and those related to animal welfare.

Primate reintroductions often yield low to medium survival rates (e.g., 10%, Bennett et al., 1992; 31%, Wimberger et al., 2010; 56%, Guy et al., 2012) which may be a concern for the long-term welfare of reintroduced individuals. Moreover, the reintroduction of trafficked individuals from non-native primate species could threaten the conservation of other primate or wildlife species. This is the case of the buffy-tufted-ear marmoset (*Callithrix aurita*) and the buffy-headed marmoset (*Callithrix flaviceps*). These marmoset species are native to the Atlantic rainforest in southeast Brazil; however, they are currently endangered and listed in the IUCN Red List. This is partly due to the introduction of black-tufted marmosets (*Callithrix penicillata*) and common marmosets (*Callithrix jacchus*) which are native to the northeast of Brazil. These species were severely trafficked during the 1980s and 1990s leading to abandonment in the Atlantic rainforest, reproduction and genetic mixing with *C. aurita* and *C. flaviceps* (Zanon, 2020).

Even though genetic testing is one of the requirements listed in the IUCN Guidelines for Non-human Primate Re-introductions as well as disease screening and post-release monitoring for at least 12 months after release (Baker 2002), this is seldom performed in Latin America (Mitman et al., 2021). Interestingly, some relatively recent studies, such as de Palomino et al. (2013) and Bennett et al. (2013) reported 100% of survivorship of the monkeys after 6-12 months (de Palomino et al., 2013; Bennett et al., 2013). These studies reported extensive post-release monitoring and food provisioning as well as community support. Thus, one could argue that it is the characteristics of the reintroduction project what defines the post-release survival and implications on animal welfare and conservation, rather than reintroducing trafficked primates *per se*.

There is the need for the ethical analysis of the issues surrounding primate rehabilitation and reintroduction. However, I have only found one study in which a similar approach was taken. Palmer (2018) examined the reasons behind the ongoing rehabilitation and reintroduction programmes of trafficked orangutans (*Pongo* spp.), despite the known low post-release survival of primate reintroduction projects and the little value they seem to have for conservation purposes (Palmer, 2018). They argued that even though people involved in orangutan rehabilitation and reintroduction programmes may be aware of these frequent negative outcomes, these programmes continue because they are viewed as the only solution for trafficked orangutans, preferable to life in captivity or death (i.e., euthanasia) (Palmer, 2018). Euthanasia, for example, is illegal in Indonesia, one of the two countries where orangutans are found (the other being Malaysia), and, if it was performed, it could lead to a severe (social) backlash in these and other countries (Usher, 2016 in Palmer, 2018). Furthermore, it was argued that there are valid ethical reasons to rehabilitate and reintroduce trafficked orangutans, as it is not their fault they have ended up in the illegal pet trade (Cheyne, 2015 in Palmer, 2018) and it is only fair to take responsibility for them, as humans put them in this situation (Galdikas, 2016 in Palmer, 2018). Palmer (2018) concludes that albeit many conservationists seem to be aware that rehabilitation and reintroduction of trafficked orangutans does not really benefit the conservation of their species, it will continue because of the reasons exposed above (Palmer, 2018). Nonetheless, a systematic analysis (i.e., utilising an ethical framework) of the ethical issues on trafficked New World monkeys' rehabilitation and reintroduction has never been performed. This could be a valuable contribution, since the rehabilitation and reintroduction of these animals is frequently performed in Latin American primate-range countries (Mitman et al., 2021).

1.2. Personality

1.2.1. What is personality? History and definition

The study of animal personality started with Pavlov's studies of canine behaviour and the distinctions he described between individual dogs' nervous systems (Pavlov, 1928 in Gosling et al., 2008). Over the last few decades, research on personality has broadened across many scientific disciplines, including behavioural ecology, psychobiology, veterinary science and reintroduction biology; moreover, this (research) has demonstrated the existence of personality in many animal species, ranging from invertebrates and reptiles to birds and mammals (Gosling et al., 2008).

There are several definitions used for the concept of 'personality'. In simple terms, Wilson et al., (2019) define it as '*individual differences in behaviour and emotion'* (Wilson et al., 2019, p. 1). Réale et al. (2007) define animal personality as stable inter-individual differences in behavioural patterns (Réale et al., 2007). Similarly, Uher (2011) defines personality as behavioural patterns expressed by an individual (Uher, 2011). From these definitions, two important characteristics of personality can be identified: (1) behavioural patterns specific to a certain individual vary consistently across time and (2) behavioural patterns will vary among individuals (Réale et al., 2007; Uher, 2011).

Animal personality is sometimes referred to as 'temperament', 'behavioural types' or 'behavioural syndromes', particularly in behavioural ecology, to avoid anthropomorphic associations with the term 'personality' (Mehta and Gosling, 2008; Gosling et al., 2008). However, the use of this term (i.e., 'personality') provides certain advantages, such as avoiding confusion and allowing for comparison between studies (Gosling et al., 2008).

Furthermore, as proposed by Gosling (2008), the use of the term 'personality' may help to bridge the gap between human and animal personality studies (Gosling, 2008). This is useful and of value because of several reasons, some beneficial to human studies, and some beneficial to animal studies. In terms of benefitting human personality studies, one important reason is that certain research questions are much easier to address in animal experiments than in human experiments as, in general, researchers have more control over the experimental variables in animals subjects than in human subjects, and many animals have considerably shorter lifespans when compared to humans (Gosling, 2008). Moreover, studies performed with animals may allow for genetic or other types of biological manipulation and physiological assessment that are forbidden or more difficult to perform in human studies (Gosling, 2008).

In terms of the benefits of using a 'human personality' approach for the study of animal personality, this may have the advantage of focusing on a broader set of potentially interesting and relevant traits, rather than focusing only on the personality traits that are frequently found in behavioural ecology studies: 'boldness', 'exploration', 'activity', 'aggression' and 'sociability', what Koski (2014) calls 'going beyond the five measured traits' (Koski, 2014). Some of these overlooked traits may be important for the survival of the animal species in question; such as individual differences in 'maternal styles', cooperation, and problem-solving skills (Koski, 2014). Using an approach based on human psychology may allow for easier appraisal of these traits (Koski, 2014), which could be supported by the use of the same term ('personality'). Indeed, Weiss (2017) stated that the 'human personality model' has been successful in setting up a framework for the study of animal personality because of its simplicity and scientific robustness (Weiss, 2017).

1.2.2. Human and non-human primate personality: The Five Factor Model

In the 1960s, the Five Factor Model of personality (hereafter, FFM) was first used as a way to describe human personality. The early version of this model, composed by the factors *Surgency*, *Agreeableness*, *Dependability*, *Emotional Stability* and *Culture* (Tupes and Christal, 1961 in Digman, 1990), later evolved into five stable factors named *Assertiveness*, *Likeability*, *Emotionality*, *Intelligence* and *Responsibility* (Borgatta, 1964; Digman, 1990). Personality theorists agree that there are four levels of abstraction when addressing personality traits: *behaviours* are specific responses to specific situations, and these responses aggregate into habits, act frequencies, dispositions or *items*, which in turn aggregate into characteristics. These (characteristics) may belong to one or more *traits*, also referred to as *factors* or *dimensions* (Digman, 1990). Goldberg (1980) suggested that the FFM could encompass many previous studies performed in relation to personality, providing a framework for this field and its conceptual organisation (Goldberg, 1980 in Digman, 1990). These studies were based on *ratings* which could have been performed by several persons that were acquainted with the individual being rated (i.e., raters), or self-ratings. Moreover, Goldberg (non-published data) provided a set of 50 self-rating scales, with ten of these related to each of the Five Factors (Digman, 1990). The scores obtained by using these scales was highly correlated with the Neuroticism, Extraversion, Openness Personality Inventory (NEO-PI) from Costa and McCrae (1985), another notable questionnaire for personality assessment in line with the FFM (Costa and McCrae, 1985; Digman, 1990).

Overall, the first factor in the FFM is agreed to be ‘Extraversion’, also called ‘Extraversion/Introversion’. Individuals with higher scores on this factor (i.e., extroverts) tend to direct their attention and energy to the outer world, whilst individuals with lower scores (introverts) tend to focus on their subjective experience and inner world (Digman, 1990; APA, 2020). The second dimension, ‘Agreeableness’ (also called ‘Friendliness’), refers to the tendency to act in a cooperative and selfless way (Digman, 1990; APA, 2020). There is general agreement on the name of the third dimension: ‘Conscientiousness’, which relates to the tendency to act in an organised, responsible and hardworking way (APA, 2020). The fourth dimension, which represents negative affect, is commonly referred to as ‘Neuroticism’ but has also been called ‘Neuroticism vs. Emotional Stability’ or ‘Negative Emotionality’. More neurotic individuals tend to have higher levels of psychological stress and less emotional stability than less neurotic individuals (Digman, 1990; APA, 2020). Finally, the fifth dimension, which has been called ‘Intelligence’ in some studies, has been related to flexibility and being ‘open’ about new ideas, feelings and fantasies; hence, it has been commonly referred to as ‘Openness’ (Digman, 1990). After Tupes and Christal (1961), the FFM and the dimensions that compose it (i.e., Extraversion, Agreeableness, Conscientiousness, Neuroticism and Openness) have been widely studied and have guided many areas of psychology, such as personality, cross-cultural, organisational and clinical psychology (McCrae and Costa, 2008).

Primatology –the branch of zoology that studies primates– has also benefited from the establishment of the FFM. King and Figueredo (1997) used a questionnaire based on the FFM to assess chimpanzee (*Pan troglodytes*) personality and obtained the five (human) factors plus another factor which was named ‘Dominance’ (King and Figueredo, 1997; Weiss et al., 2011). Similarly, Weiss et al. (2006) found the factors Agreeableness, Neuroticism and Extraversion in orangutans (*Pongo* spp.) plus another factor, ‘Intellect’, which comprised traits from Openness and Conscientiousness using an expanded version of King and Figueredo’s (1997) questionnaire (Weiss et al., 2006; Weiss et al., 2011). This questionnaire, known as the Hominoid Personality Questionnaire (hereafter, HPQ) has been subsequently used to assess the personality structure (based on the FFM) of other primate species, such as gorillas (Eckardt et al., 2015), rhesus macaques (Weiss et al., 2011), brown capuchin monkeys (*Sapajus apella*) (Morton et al., 2013) and yellow-breasted capuchin monkeys (*Sapajus xanthosternos*) (Fernández-Bolaños et al., 2020).

Hence, various personality dimensions are shared by several primate species: Extraversion, Conscientiousness and Agreeableness, are found in humans and chimpanzees; Openness and Neuroticism, are found in humans, chimpanzees, brown capuchin monkeys and yellow-breasted capuchin monkeys; Sociability, found in brown capuchin monkeys, yellow-breasted capuchin monkeys and gorillas (*Gorilla beringei beringei*) and Dominance, Intellect, Confidence and Attentiveness, uniquely distinctive to chimpanzees, orangutans, rhesus macaques and brown capuchin monkeys, respectively (King and Figueredo, 2007; Weiss et al., 2006, Weiss et al., 2011; Morton et al., 2013, Eckardt et al., 2015; Fernández-Bolaños et al., 2020). Studies on primate personality based on the FFM have focused traditionally on great apes, and to a lesser extent on Old and New World monkeys, with New World monkeys included in fewer studies compared to Old World monkey species (Fernández-Bolaños et al., 2020).

1.2.3. Personality assessment methods

Overall, there are two methodological approaches used to assess animal personality: (1) behavioural profiles obtained from behavioural observations, also called *behavioural coding,* and (2) *observer trait ratings*, which are ratings performed by two or more observers (also known as raters) on the behavioural patterns expressed by a specific individual (Freeman et al., 2011). According to Gosling (2001), almost all primate personality research is based on these methods, with approximately 90% of studies using behavioural coding and 40% using observer ratings (Gosling, 2001; Freeman et al., 2011).

Several instruments have been used to assess personality traits in non-human primates over the years. According to Freeman and Gosling (2010), these mainly include the Hominoid Personality Questionnaire (HPQ; Weiss et al., 2009), originally used for chimpanzees (*Pan paniscus*); the Emotions Profile Index (EPI; Buirski et al., 1973), originally developed for olive baboons (*Papio anubis*); and the Maddingley Questionnaire (Stevenson-Hinde and Zunz, 1978), originally developed for rhesus macaques (*Macaca mulatta*) (Freeman and Gosling, 2010). Other instruments include, for example, questionnaires developed by Capitanio et al. (2004) for assessing personality in rhesus macaques and those developed by Uher and Asendorpf (2008) for great apes (Capitanio et al., 2004; Uher and Asendorpf, 2008; Freeman and Gosling, 2010).

Questionnaires or instruments to rate animal personality are developed using one of two methods, called ‘etic’ and ‘emic’ approaches (Freeman et al., 2011). An etic approach involves using a questionnaire developed for a particular species as a base to develop a questionnaire for another species e.g., using a questionnaire for chimpanzees to develop a questionnaire for macaques. On the other hand, an emic approach involves using systematic behavioural observations, performed by expert observers, to develop the definitions of species-specific personality traits (Freeman et al., 2011; Uher and Visalberghi, 2016). Because of the ways they are developed, the etic approach allows for easier comparisons between species (than the emic approach), whereas the emic approach decreases the possibility of species-specific, ecologically relevant behaviours being overlooked (Freeman et al., 2011).

For personality measures to be of value in scientific research, they need to be **reliable** and **valid** (Freeman and Gosling, 2010). Reliability means that there must be a high level of agreement between observers or raters and is also called inter-observer reliability. Validity means that the instrument is indeed measuring what it is supposed to be measuring (Freeman and Gosling, 2010). In other words, there should be a significant correlation between the personality measure (for example, a personality trait as defined in a questionnaire) and a measure obtained objectively and directly; in this case, an observable behaviour (the ‘gold standard’). Here, I will briefly discuss the reliability and validity of both methodological approaches (observer trait ratings and behavioural coding), as well as their advantages and disadvantages.

In terms of reliability, most primate personality studies performed with questionnaires (i.e., observer trait ratings) have measured inter-observer reliability (Weiss, 2017) and have found acceptable (i.e., significant) levels, often in the form of intraclass correlation (ICC 3,1 and ICC 3,k) or Pearson correlation (Freeman and Gosling, 2010; e.g., Morton et al., 2013; Manson and Perry, 2013; Robinson et al., 2016; Nunes, 2017). Moreover, most primate personality studies have found that certain personality traits, as measured in questionnaires, are strongly associated with the behaviour of the individuals (e.g., ‘aggressive’ is strongly associated with the exhibition of agonistic behaviours; Pederson et al., 2005 in Freeman and Gosling, 2010; Sociability is strongly correlated with affiliative behaviours, Manson and Perry, 2013); thus, this approach has been deemed as valid in many studies (Freeman and Gosling, 2010; Weiss, 2017).

Behavioural coding involves the direct observation of the behaviour of the subject, in which data may be collected in terms of duration (behavioural states) or frequency (behavioural events) (Freeman et al., 2011). Inter-observer reliability may or may not be assessed, depending on the number of observers in the specific study. Moreover, external validity cannot be assessed, as the ‘gold standard’ –the behaviour– is being measured directly. Nonetheless, it is considered a valid method because it is measuring individual behavioural patterns directly and should always be included when using observer trait ratings, if time and resources allow for its inclusion (Gosling, 2008).

Even though both methods –observer trait ratings and behavioural coding– provide information regarding personality, they do this in different ways. This means they have different characteristics that may be seen as advantages or disadvantages depending on the specific study and its research objective, but they may also complement each other. The advantages and disadvantages of both methods can be seen in Table 1.1 below.

Table 1.1

Advantages and disadvantages of personality assessment methods (based on Freeman et al., 2011)

	Behavioural coding	**Observer trait ratings**
Advantages	Direct observation of behaviour Less subjective Easier to compare objectively between subjects	Easier to perform Data may be collected in an efficient, quick way More control over inter-individual behavioural variability May focus on more than one behaviour or situation simultaneously Use of complex descriptions to define traits Non-systematic
Disadvantages	More difficult to carry out Time-consuming Systematic Less control over inter-individual behavioural variability Focus on one behaviour/situation at a time Certain personality traits may be difficult to define or describe in an ethogram (e.g., unpredictable, thoughtless)	Indirect – based on questionnaires answered by one or more ‘raters’ More subjective More difficult to compare objectively between subjects

As mentioned above, albeit some characteristics of each methodological approach are listed as disadvantages in Table 1.1, some may not be disadvantages necessarily. This depends on the specific research question being addressed in the study. Performing behavioural coding, for example, may require a more systematic method and may be more time-consuming than performing observer trait ratings, as it requires the use of an ethogram (i.e., the *code*) and familiarisation with the individuals. However, this may allow for more objective comparison between individuals (Freeman et al., 2011). Hence, if the aim of the study is to compare between individuals, it may be necessary to include behavioural coding.

Furthermore, observer trait ratings and behavioural coding can be applied in three different contexts. These include (a) *naturalistic observation*, where the animals are observed for a specific time frame in their ‘natural’ or familiar environment (i.e., not necessarily in the wild, but in the environment they are used to, which may be in the wild or in captivity); (b) *cumulative observation*, which accounts for all the cumulative experience the observer (rater) has with the individuals who are being rated; and (c) *behavioural tests*, in which the animals are expected to engage voluntarily in a test or experiment designed to promote the expression of specific personality traits, such as ‘boldness’ or ‘aggression’ (Freeman et al., 2011).

Similarly to what happens with the methodological approaches, each context has certain characteristics, that may be viewed as advantages or disadvantages, depending on the specific aim, research question and research hypotheses of the study. Overall, naturalistic observations are less difficult to perform in captivity than in the wild and (arguably) less prone to the effect of observer familiarity (Freeman et al., 2011). This means that a high level of familiarity between the observer and the target animal may not be necessary, as long as the observer is proficient in performing behavioural observations on individuals from the species in question. In other words, the observer may not need to be highly acquainted with the individual s/he is observing (as opposed to what happens in observer trait ratings, for example). Moreover, naturalistic observations are based on the behaviours exhibited in only one context (e.g., the individual’s ordinary enclosure) (Freeman et al., 2011).

Conversely, cumulative observation, often used to obtain observer trait ratings, depends completely on the observer’s experiences with a specific individual. Hence, the observer/rater needs to be highly acquainted with the target animal. This may include observations or experiences performed across several contexts and may provide a broader insight into the individual's personality (than naturalistic observations) as it is not restricted to only one context or situation. In general, observer trait ratings are considered as the fastest and easiest to perform of all methods, provided there are enough raters that are sufficiently acquainted with the individuals being rated (Freeman et al., 2011). Finally, performing behavioural tests has the advantage of greater control over variables that may influence the expression of certain personality patterns (Uher, 2011; Freeman et al., 2011). Nonetheless, in most cases it requires handling the animals, which may not always be feasible or beneficial for them (Freeman et al., 2011). This may be the case of individuals aimed for reintroduction (personal observation).

Several studies have been performed to examine the correlation of personality traits obtained using observer trait ratings and direct observations of behaviour (Freeman et al., 2011). Capitanio (1999), for example, compared several traits (obtained with questionnaires) such as 'sociability' with direct observations of social behaviour of Rhesus macaques, and found a strong association (≥ 0.60) between them (Capitanio, 1999). In a more recent example, Morton et al. (2013), Manson and Perry (2013), Uher and Visalberghi (2016), Nunes (2017) and Fernández-Bolaños et al. (2020) have found similar associations between specific behaviours obtained by direct observation and observer trait ratings in capuchin monkeys (*Cebu*s spp. and *Sapajus* spp.) (Morton et al., 2013; Manson and Perry, 2013; Fernández-Bolaños et al., 2020; Uher and Visalberghi, 2016; Nunes, 2017). Indeed, several studies use a combination of the two methodological approaches in different contexts to assess personality structure. This is performed, presumably, to overcome the limitations of each context and/or methodological approach and complement the data collected.

An example of this is the study by Uher and Visalberghi (2016). In this study, the authors assessed the personality of 150 brown capuchin monkeys (*Sapajus apella*) using behavioural coding and observer trait ratings in different contexts. These included: (1) behavioural observations in the animals' ordinary enclosures in two situations (i.e., 'pre-feeding' and 'social'), (2) a set of behavioural tests, and (3) a questionnaire developed using an emic approach (Uher and Visalberghi, 2016). Inter-rater reliability of the questionnaire items was significantly high in this study (≥ 0.89). Moreover, validity of the personality constructs was significant as well, meaning there was a strong correlation between the direct behavioural observations and the observer trait ratings and/or the behavioural tests (Uher and Visalberghi, 2016). The advantage of using the two methodological approaches in different contexts or situations relies on the distinct information that each of them can provide. For example, the direct behavioural observations could provide the ability to compare objectively between individuals and a mean to validate other forms of personality assessment (e.g., the questionnaire), whereas the use of the observer trait ratings could provide the advantage of accounting for the individuals' behaviour in other situations, different to their ordinary enclosures. Finally, the behavioural tests could elicit specific behaviours related to certain personality traits (e.g., 'boldness') that may not be observed in other contexts. Hence, together, these methods provide a stronger base for the construction of personality structure than if they were used separately, and a way of objectively comparing between individuals (e.g., by using direct behavioural observations) and taxa or subspecies (e.g., by using standardised questionnaires for a taxonomic order i.e., such as the HPQ, used for non-human primates, or questionnaires developed by Uher and Visalberghi (2016) for capuchin monkeys or by Capitanio et al. (2004) for rhesus macaques, for example.

In conclusion, the use of behavioural coding (in behavioural tests and direct observations) and observer traits ratings can complement each other by broadening the available data on the individual behavioural patterns of animals across different situations and contexts. Even though observer trait ratings may be seen as the easiest and quickest way to assess animal personality, they can also be subjective (Freeman et al., 2011). Thus, personality traits in questionnaires need to be correlated with observable behaviours, and described as accurately and objectively as possible (Uher and Visalberghi, 2016). Because of this, including direct behavioural observations may be advantageous in all animal personality studies. Firstly, to provide a systematic, objective way to assess individual behavioural patterns (i.e., personality traits); and secondly, to provide a way to validate other personality measures obtained, either in the form of questionnaires or behavioural tests.

1.2.4. Personality and health

The study of the association between individual differences and health started around the 1950s and has identified, over the years, several important personality factors that influence health outcomes positively such as intelligence, self-control, higher social class and social inclusion; and negatively, such as stress reactivity, pessimism, negative affect, social isolation and Neuroticism (Gosling, 2008; Kupper et al., 2013; Strickhouser et al., 2017; Jandackova et al., 2017). Nowadays, personality traits are considered an integral part of human health psychology (Gosling, 2008; Ferguson, 2013) and neurotic traits, in particular, have been used as predictors of clinical disease in many studies (Friedman et al., 2010).

Similarly, health and physiological parameters have been associated with individual differences in animals (Cavigelli, 2005). The study of animal personality and its relationship with health may complement human studies, as animal models have several advantages over human models such as greater control over experimental variables (Cavigelli, 2005; Gosling, 2008; Uher, 2011). Moreover, the study of the relationship between animal personality and health may aid improvement of animal welfare, as physical health is strongly associated with overall wellbeing (Finkemeier et al., 2018).

In non-human primates, Sociability and social position (i.e., dominance rank or social rank), emotionality (i.e., reactivity) and behavioural inhibition seem to have a strong influence on health outcomes and immune function, particularly in highly social and/or despotic species (Gosling, 2008; Capitanio, 2011; Howell et al., 2012; Shively and Day, 2015). In rhesus macaques (*Macaca mulatta*), for example, social rank plays an important role in aspects of their health such as reproduction, certain physiological parameters (e.g., cardiovascular) and glucocorticoid levels (Shively and Day, 2015). Moreover, Sociability can be used as predictor for antibodies levels after inoculation with simian immunodeficiency virus (SIV) in this species (Gosling, 2008).

The link between certain personality traits (e.g., Neuroticism, Sociability) and health outcomes is not entirely clear. However, it has been hypothesised that personality traits influence the way in which individuals react to the environment; therefore, the way in which the immunological and other physiological systems respond (Gosling, 2008). Thus, personality influences the ability of the individual to appraise and cope with a situation (Capitanio, 2011). If a situation is threatening, for example, a more neurotic individual may become more stressed than a less neurotic individual. Here, I will provide a very brief summary of the classical stress response systems. As mentioned above, personality traits may influence the strength of these responses with potential effects on health.

In the body, there are two major stress response systems: (1) the sympathetic-adrenal-medullary (SAM) system and (2) the hypothalamic-pituitary-adrenal (HPA) system. The SAM system works by releasing the neurotransmitter norepinephrine (noradrenaline) and stimulating the adrenal medulla, which in turn segregates epinephrine (adrenaline)

(Capitanio, 2011). The perception of a stressor activates the HPA axis resulting in corticotrophin releasing hormone (CRH) release. The pituitary gland releases, in turn, adrenocorticotrophic hormone (ACTH) into the blood stream. In response, the adrenal cortex releases glucocorticoids (e.g., cortisol) (Capitanio, 2011). The major action of glucocorticoids is to mobilise energy and prepare the animal for action via the effects on a range of target organs. Therefore, glucocorticoids have effects in many types of cells, including those of the immune system, in which they reduce activity (i.e., they have immunosuppressant effects) (Capitanio, 2011; Yasir, 2020). When an individual is more prone to perceive environmental changes or situations as challenging, s/he may become stressed more easily. This will lead to activation of the SAM and HPA systems (Capitanio, 2011). If the release of stress hormones exceeds the capacity of the immune system to adapt (e.g., when the individual is chronically stressed), it may lead to disease (Bae et al., 2019). Thus, personality traits may either aggravate (e.g., Neuroticism) or help to protect from (e.g., Sociability) the effects of chronic glucocorticoid segregation on the immune system of the individuals by mediating the way animals evaluate and cope with their environment. As explained by Capitanio (2011): *'...personality affects an individual's appraisals of its environment (e.g., what in the environment is considered a challenge, as well as the ability of the individual to cope with what has been appraised as challenging'* (Capitanio, 2011; p. 239). In other words, if an individual tends to view situations as more challenging, s/he will get stressed more easily, and, in turn, will segregate more stress hormones than animals that view situations as less challenging. This may lead to a weakened immune system, making the animal more prone to disease (Bae et al., 2019). There are several examples of this in the scientific literature on animal personality. Overall, animals considered as more 'risk-averse' produce more glucocorticoids, are more prone to cardiac and gastrointestinal diseases, and have weaker immune systems (de Azevedo and Young, 2021) than animals that are less 'risk-averse' (or more 'risk-taking').

1.3. Personality and Reintroduction Biology

In reintroduction biology, it has been proposed that personality traits may influence reintroduction outcomes, as certain individuals may survive for longer than others, for example, when released back into the wild (Bremner-Harrison et al., 2004; de Azevedo and Young, 2021). As rehabilitation is a pre-requisite for the reintroduction of trafficked primates, it would be valuable to know which personality traits (if any) influence the rehabilitation of rescued wild-born individuals. However, I have found few examples in the scientific literature on the association between personality traits and rehabilitation and/or reintroduction outcomes of trafficked primates.

Sita et al. (2016) used environmental enrichment to enhance the rehabilitation and promote the welfare whilst in captivity of ten trafficked black capuchin monkeys (*Sapajus nigritus*). Several methods of environmental enrichment were used: (1) physical, in which various novel objects and food items as well as structures such as tree branches and trunks were introduced to the enclosure of the individuals, and (2) social, in which conspecifics that the individuals had not met before were introduced to the enclosure. The behaviour of the individuals was recorded during the environmental enrichment. Some differences were found among individuals with different behavioural profiles: more neophilic individuals showed a significantly higher increase in foraging behaviours, whilst less neophilic individuals exhibited an increase in stereotypies and other abnormal behaviours during the physical enrichment of the enclosure.

Moreover, more active individuals exhibited a decrease in locomotion, and individuals with higher scores in exploration showed a decrease in investigation and an increase in locomotion and abnormal behaviours. During the social enrichment phase, individuals with higher scores on sociability increased exploratory behaviours (Sita et al., 2016). Overall, this study showed that environmental enrichment is capable of eliciting different behavioural changes amongst capuchin monkeys in rehabilitation. However, as mentioned by the authors, it is not clear whether environmental enrichment was really supporting the pre-release training of the individuals or if the environmental enrichment was only giving them the opportunity to express their natural behavioural repertoire whilst in captivity (Sita et al., 2016).

Personality assessment may aid reintroduction programmes by helping select the individuals with the highest chances of survival after release. Individuals with inadequate *boldness* (i.e., fearlessness) levels may be less fit to survive in the wild than individuals with more appropriate levels of this trait (Bremner-Harrison et al., 2004). To explore this, Bremner-Harrison et al. (2004) performed a study in which captive-born swift foxes (*Vulpes velox*) were released as part of a reintroduction programme (n = 49, 15 adults and 34 juveniles). Before release, the individuals’ levels of boldness were assessed with several tests based on presenting them with novel stimuli to elicit behaviours that help to differentiate ‘shy’ and ‘bold’ individuals. The novel stimuli selected were: a beach ball with different colours, a grey plastic box, a grey sack with paper inside, and an unknown person. Each session lasted for 50 minutes and was performed on a different day. The sessions were video-recorded from outside the enclosure and repeated six to eight weeks later. The behaviour of the foxes was recorded in each session, using an ethogram. Behaviours related to the different personality traits (i.e., ‘bold’ and ‘shy/cautious’) were recorded and given a score, one (1) for behaviours related to ‘shyness/cautiousness’, such as fleeing or approaching hesitantly; and two (2) for behaviours related to ‘boldness’, such as investigating or approaching boldly. Thus, each fox ended up with an ‘overall boldness score’, which consisted of the sum of the behaviours it had performed during the sessions with the novel stimuli (i.e., bolder foxes had higher scores, as bold behaviours had a higher value than cautious behaviours). Furthermore, the time of first appearance after each stimulus was presented (i.e., latency) and the closest distance to the novel stimulus was recorded for every fox.

Before release, 16 juveniles were given radio-collars to monitor them for six months after release (Bremner-Harrison et al., 2004). High concordance levels were found between different tests. This means that individuals with high boldness scores with one stimulus also had high scores with the other ones. There were no differences found between females and males, but adults had higher boldness scores than juveniles. Moreover, juveniles had higher boldness scores in the second trials than in the first trials. Regarding survival, the individuals that were found dead or presumed dead (n = 5; 4 dead and 1 presumably dead) after release had higher scores in the tests; furthermore, they had approached the stimuli closer and had lower latencies to approach the stimuli. In other words, they were ‘bolder’ than those that survived after release. As stated by the authors, bolder individuals may be more prone to approach predators or other situations that may pose a risk for them after release, as they may be less fearful of novel stimuli. In this sense, selecting ‘shy’ individuals or individuals with low scores on boldness may be an advantage in reintroduction programmes (Bremner-Harrison et al., 2004).

Similarly, in a study by Lopes et al. (2017), personality tests were performed to 15 blue-fronted Amazon parrots that were then released back into the wild. To assess the birds’ personality, boldness scores were calculated following the methodology of Bremner-Harrison et al. (2004, see above paragraph). To achieve this, two novel objects were introduced into the birds’ enclosure, and their ‘shy/cautious’ and ‘bold’ behaviours were recorded. After this, individual boldness scores were calculated for each bird. After release, there were no differences found between the survival rates of bold and shy individuals, as opposed to the study of Bremner-Harrison et al. (2004) in which shy swift foxes had better survival rates than bold ones (Bremner-Harrison et al., 2004). However, bold parrots interacted less with members of their release group, interacted less positively with wild parrots, and emitted human vocalisations more frequently (Lopes et al., 2017). The emission of human vocalisations, in particular, is an undesirable behaviour for parrots after release (Lopes et al., 2017).

As mentioned above and stated by Bremner-Harrison et al. (2004), it appears that bold individuals are more prone to exhibit behaviours that may be considered as risky for their post-release survival (Bremner-Harrison et al., 2004). Therefore, it may seem obvious that selecting individuals with high boldness scores may be a disadvantage in reintroduction programmes. Nevertheless, some authors, such as Watters and Meehan (2007), have suggested that social groups composed by individuals with different personality traits (i.e., ‘shy’ and ‘bold’) may be more stable and fare better against environmental variations than release groups composed by individuals with similar personality traits (Watters and Meehan, 2007). An explanation for this, is that individuals with different personality traits not only respond differently in terms of behaviour to distinct scenarios, but also possess different physiological responses to pathogens and environmental stressors (Watters and Meehan, 2007). Thus, it is necessary to construct groups with individuals with different personality traits to promote resilience to environmental change in populations aimed for reintroduction (Watters et al., 2003; Watters and Meehan, 2007). To achieve this, we need methods to assess personality traits whilst in captivity, as well as tools (e.g., environmental enrichment) to promote their expression (Watters and Meehan, 2007).

An example of this is a study by Sita et al. (2016). In this study, the behavioural profiles and short-term post-release survival of 69 capuchin monkeys (*Sapajus* spp.) and nine black capuchin monkeys (*Sapajus nigritus*) were assessed as part of a reintroduction programme in Brazil. These monkeys were confiscated from the illegal pet trade and were not born in captivity. Their personality was assessed utilising a method based on (1) the five proposed dimensions (i.e., boldness, exploration, sociability, aggressiveness, and activity) by Reále et al. (2007), (2) the five factor components based on the Hominoid Personality Questionnaire (HPQ) from Morton et al. (2013), and (3) the 20 working constructs developed specifically for capuchin monkeys by Uher et al. (2013). Eight personality dimensions were defined: boldness, neophilia (i.e., food neophilia), sociability, aggressiveness, exploratory, activity, vigilance (i.e., fearfulness), and food-orientation (Sita et al., 2016). The tests used to assess these traits were performed in the social groups whilst in captivity; overall, they include presenting the animals with novel objects and foods, as well as mirrors and new conspecifics (i.e., conspecifics that the animals had not met before) and recording their behaviour (see Sita et al., 2016 for a complete description of the tests).

Confirmed overall post-release survival after three months for the 78 individuals was of 34.7%. In the group of the 69 capuchin monkeys, females had a significantly longer post-release survival when compared to males. Individuals that were more affiliative and neophilic (whilst in captivity) survived longer after release than less affiliative and less neophilic individuals; moreover, more active individuals survived for less time (Sita et al., 2016). This study has been the first to correlate personality and post-release survival of rehabilitated primates (Sita et al., 2016). As mentioned above, more neophilic, affiliative and active individuals survived longer after release than individuals that were rated as being less neophilic, affiliative or active (Sita et al., 2016). More research is needed to understand the relationship between individual differences in personality and rehabilitation and reintroduction outcomes of non-human primate reintroduction projects, as it has been studied in other wildlife species.

1.4. Aims of the Study

The review of the scientific literature I performed for this dissertation revealed there are several clear gaps of knowledge related to the rehabilitation and reintroduction of trafficked Neotropical primates. Most studies have focused on reintroduction outcomes and were conducted with great apes and Old World monkeys. Conversely, few studies have focused on the health and behaviour during rehabilitation of trafficked New World monkeys, and even less have focused on their association with personality traits. In the review of primate personality research performed by Freeman and Gosling (2010), for example, trafficked primates (i.e., born in the wild and raised in captivity by humans) are not even mentioned among the different rearing conditions of individuals in primate personality studies, with the closest being individuals raised by peers or in nurseries (Freeman and Gosling, 2010). In the literature review performed for this dissertation, I found only a single, unpublished study which aimed to investigate the relationship between personality traits and rehabilitation and reintroduction of trafficked capuchin monkeys (Sita, 2016), and this study mainly focused on post-release survival of the individuals.

Personality assessment is an important part of reintroduction biology and may provide practical, real-world advantages for the conservation of primates and other wildlife species. These may include but are not restricted to (1) allowing conservationists and wildlife rehabilitators to identify potential issues and/or predict reintroduction outcomes based on physiological measures, such as indicators of stress, for example, by identifying which individuals are at more risk of predation or disease by analysing the correlation between these issues, physiological stress and personality traits; (2) selecting the best candidates for a desired reintroduction outcome, such as reinforcement of a population, by assessing behavioural patterns *before* release, based on behaviour during rehabilitation and, in some cases, post-release survival (e.g., Mendoza-Nakano, 2016; Sita, 2016); (3) selecting the best candidates for a desired reintroduction outcome, by assessing personality traits *after* release, based on post-release survival (e.g., Bremner-Harrison et al., 2004) (de Azevedo and Young, 2021). Even though approximately 65% of non-human primate species are currently considered as endangered and 75% have declining populations (Estrada et al., 2020), in most primate-range countries (i.e., countries where primates naturally occur such as Mexico, Brazil and Kenya) social inequalities, low educational levels and an excessive use of their natural resources make financial resources very difficult to access for primate conservation-related activities (Estrada et al., 2020). Thus, the use of these resources must be carefully planned. The study of primate personality in rehabilitation and reintroduction projects may help to ensure that these resources are used in a thoughtful, effective way, by helping to predict and improve reintroduction outcomes.

Furthermore, in the literature review performed for this dissertation, I did not find any study which purpose was to specifically address the ethical challenges related to the rehabilitation and reintroduction of trafficked Neotropical primates. This is another potential valuable contribution, as this is a controversial subject among primate conservationists (Shanee, 2007; personal observation).

Thus, the overall aim (i.e., research objective) of this dissertation was to study the relationship between individual differences in personality and health and behaviour during rehabilitation of trafficked bearded capuchin monkeys that were part of a government reintroduction programme in Northeast Brazil. There were six specific aims or research objectives in this dissertation: the first research objective (Chapter 3) was to investigate the health of rehabilitant bearded capuchin monkeys (*Sapajus libidinosus*) maintained in Northeast Brazilian rescue centres by (1) developing and validating a set of non-invasive health indicators for this species and (2) performing full physical examinations and blood collections to obtain physiological and haematological values as well as morphometry and parasitological status of these individuals and compare them with previously published data. The second objective (Chapter 4) was to analyse the behaviour of rehabilitant bearded capuchin monkeys to provide an initial assessment of the efficacy of the rehabilitation programme and propose potential improvements. This was performed by using changes in behaviour across time (i.e., activity budgets, space and substrate use and behavioural diversity) and comparing with previously published data, both in captivity and in the wild. The third objective (Chapter 5) was to broaden the current scientific knowledge on bearded capuchin personality structure by utilising two methodological approaches to measure personality: behavioural coding in naturalistic and testing contexts and observer trait ratings. The fourth objective (Chapter 6) was to investigate the association between personality traits (obtained from Chapter 5) and health parameters (obtained from Chapter 3) of rehabilitant bearded capuchin monkeys. The fifth objective (Chapter 7) was to investigate the association between personality traits (obtained from Chapter 5) and behaviour in rehabilitation or ‘rehabilitation outcomes’ (obtained from Chapter 4) of bearded capuchin monkeys. Finally, the sixth research objective (Chapter 8) was to investigate the ethical issues that may arise from the reintroduction of rescued/confiscated Neotropical primates back into the wild by utilising a revised version of the Ethical Matrix for conservation-related issues.

As mentioned throughout this Chapter, there are only a handful of studies on the rehabilitation of trafficked capuchin monkeys. Because of this lack of available scientific data, I considered this dissertation as exploratory, descriptive research, understanding ‘exploratory’ as preliminary or early stage (Babbie, 2020) and ‘descriptive’ as having more than one research question but not being necessarily driven by highly specific research hypotheses (Sue and Ritter, 2012). As such, I used broad, *working hypotheses* i.e., ‘*hypotheses that are subject to change, are provisional and the possibility of finding contradictory evidence is real’* (Casula et al., 2021, p. 1709), rather than highly specific predictions, typical of confirmatory or hypothesis-testing research (Sue and Ritter, 2012; Casula et al., 2021).

As explained by Swedberg (2020), the objectives of performing exploratory studies include (a) to provide a preliminary analysis of the issue in question and/or (b) to explore a topic to obtain new insight and hypotheses, even if they are not verifiable in said study (Swedberg, 2020). Thus, I focused on the general aim or research objective: performing a preliminary analysis on the relationship of individual differences in personality and health and rehabilitation of trafficked bearded capuchin monkeys (*Sapajus libidinosus*) and exploring this topic to provide a new insight and a more solid base, scientifically speaking, for future studies on capuchin monkey reintroduction biology.

Note that the aim of this dissertation was **not** to provide release criteria for rehabilitated trafficked capuchin monkeys, as (1) these criteria would not have been verifiable because post-release monitoring was not performed and (2) release criteria, as well as other indicators of the animals' suitability for release, must be based on peer-reviewed scientific data (Seddon et al., 2007), which, to the best of my knowledge, does not exist to this day for trafficked capuchin monkeys in Brazil or other Latin American countries.

Chapter 2: General Methodology

Chapter 2: General Methodology

2.1. Introduction

This chapter contains an overview of the biology of bearded capuchin monkeys (*Sapajus libidinosus*), the species studied in this dissertation, as well as a description of the study sites, subjects and the ethical statement for the study. For the methodology of data collection and analysis, please refer to the 'Methods' section of each individual chapter (Chapters 3 to 8).

2.2. Bearded Capuchin Monkeys

2.2.1. Natural area of occurrence

As other Neotropical primates (also known as Platyrrhines or New World monkeys), capuchin monkeys occupy a wide range of tropical environments in Central and South America (Lynch-Alfaro et al., 2011; Püschel et al., 2017; NPC, 2021). Bearded capuchin monkeys are native to the Brazilian states of Bahia, Piauí, Rio Grande do Norte, Mato Grosso, Minas Gerais, São Paulo, Mato Grosso do Sul, Pernambuco, Alagoas, Pará, Ceará, Maranhão, Goiás, Paraíba and Tocantins (Martins et al., 2019). *Sapajus libidinosus* was previously classified as a subspecies of *Sapajus apella* (Groves, 2005; Bacalhão et al., 2016).

2.2.2. Conservation status

Bearded capuchin monkeys are considered as Near Threatened by the International Union for the Conservation of Nature (IUCN), with habitat fragmentation and hunting for the illegal pet trade being two of the major drivers for their declining populations (Martins et al., 2019). In fact, some studies have predicted that approximately 42% of forest area will remain in 30 years in certain parts of Northeast Brazil (e.g., Morro do Boi) whereas in other parts (e.g., Fazenda Boa Vista) most of the available land which is suitable for agriculture is already being used for this (Presotto et al., 2020).

As mentioned above, the illegal pet trade is a major threat for the conservation of bearded capuchins (Martins et al., 2019); hence, they are frequently found in wildlife rescue centres in these countries (Levacov et al., 2011). After reception, these centres may keep the monkeys in temporary captivity or send them to permanent captivity in other institutions (e.g., zoos). However, some individuals are rehabilitated and reintroduced back into the natural area of occurrence of the species (Levacov et al., 2011; Mitman et al., 2021), posing a valuable opportunity to study the association between personality and rehabilitation and/or reintroduction outcomes as well as other suitable research questions related to reintroduction biology, animal health or behavioural research.

2.2.3. Morphology and physiology

Bearded capuchins are medium-sized monkeys. Their body size ranges between 34 – 44 cm and their body weight between 1.3 and 4.8 kg (Souvignet et al., 2019). Robust capuchin monkeys (*Sapajus* spp.), as their name indicates, have moderately proportioned, strong bodies (Fragaszy et al., 2004). Wild, free-ranging bearded capuchin monkeys have been reported to weigh approximately 2.1 kg for adult females and 3.5 kg for adult males (Fragaszy et al., 2016). Bearded capuchins start exhibiting sexual dimorphic characteristics at approximately four years of age, with adult males having on average a 1.9 larger body mass than adult females (Fragaszy et al., 2016). All robust capuchin monkey species (*Sapajus* spp.) exhibit a tuft of fur on their heads, whilst gracile capuchin monkeys (*Cebus* spp.) do not (Fedigan et al., 2016). Bearded capuchins' body coat (fur) may be beige or yellow with darker hands, feet and face/head (Bacalhão et al., 2016). As well as other capuchin monkey species (*Cebus* spp. and *Sapajus* spp.), bearded capuchins possess semi-prehensile tails, which are fully furred and help the animals to maintain balance and support diverse activities such as feeding and foraging, locomotion, social interaction and resting (Bezanson, 2018).

Bearded capuchins have a gestational period of around 154 to 162 days (Fragaszy et al., 2004). They are considered as infants after they are born and before they reach 12 months old, and then as juveniles and subadults until they reach sexual maturity at approximately eight to 10 years of age (Souvignet et al., 2019). Even though they reach sexual maturity until eight years old (females) and 10 years old (males), male capuchins start displaying interest in females around their third year of age and females (on males) around their fourth year of age (Fragaszy et al., 2004). Female bearded capuchins born in captivity have oestrus cycles of approximately 21.07 (±1.07) days (Lima et al., 2012). In captivity, capuchin monkeys may become pregnant and give birth throughout all year. Conversely, free-ranging capuchin births occur mostly when there is high food availability (Souvignet et al., 2019). Anecdotally, captive brown capuchin monkeys (*Sapajus apella*) can live up to 46 years of age (HAGR, 2021), but the life expectancy of bearded capuchin monkeys in the wild is unknown.

Figure 2.1. Bearded capuchin monkeys (Sapajus libidinosus): adult female (right) and juvenile male (left). Note the beige/yellow colouration of their coat (fur) and the darker coloration of their hands, face, head and tuft (adult female).

2.2.4. Behaviour and socioecology

Generally, bearded capuchin monkeys live in social groups composed of 10 to 17 individuals (Souvignet et al., 2019), although this can vary greatly. Free-ranging bearded capuchins have been reported to live in small groups of up to eight individuals, or larger groups with almost 20 individuals (e.g., Verderane et al., 2013). This species has a polygynous mating system; moreover, females tend to stay in the group they were born (i.e., are philopatric) whilst males disperse to other groups (Izar et al., 2012). Females establish stable, linear hierarchical relationships which include coalitions maintained via behaviours such as grooming (Izar et al., 2012). Interestingly, forming and keeping social groups is often a challenge for the maintenance of capuchin monkeys in captivity, as conflicts between individuals may occur in an attempt to gain a better social position within the group (Souvignet et al., 2019; personal observation).

Bearded capuchins live in various types of habitats, including rain forests and deciduous tropical forests, secondary forest fragments, and semi-arid areas with different levels of human presence (Sabbatini et al., 2008; Verderane et al., 2013). The population density of bearded capuchin monkeys varies among studies. In a study performed by Verderane et al. (2013) in Fazenda Boa Vista, an open woodland area located in Piauí, Northeast Brazil, a population density of 2.3 monkeys per km^2 was reported (Verderane et al., 2013).

Overall, capuchin monkeys are considered as frugivores-insectivores (Souvignet et al., 2019) or omnivores (Sabbatini et al., 2008). Their diet varies according to the environment in which they live, or are housed, in the case of captive individuals. Capuchin monkeys in captivity may prefer sugary fruits such as fruits for human consumption (e.g., papaya, banana, etc.), food items that are high in fat, or novel food items rather than foods with an adequate nutritional and/or caloric content (Souvignet et al., 2019). Thus, providing a well-balanced diet is important to prevent obesity in captive capuchin monkeys.

In semi-arid areas with high predation risk, such as the Cerrado–Caatinga biome of Fazenda Boa Vista (Piauí, Brazil) free-ranging bearded capuchin monkeys feed mainly on high quality, clumped food items such as fruits, obtained from palms and trees, and less on other food items obtained by manipulating them (e.g., nuts), often with the use of tools such as stone hammers and anvils (Izar et al., 2012; Verderane et al., 2013; Presotto et al., 2020). In areas with human presence, such as the Parque Nacional de Brasília in Brazil, the diet of bearded capuchins exhibits seasonal variation; in fact, capuchins rely more on foods provided by humans during the dry season (Sabbatini et al., 2008). In this area, bearded capuchins feed mainly on fruits, small animals (e.g., lizards) and insects (e.g., butterflies) (Sabbatini et al., 2008).

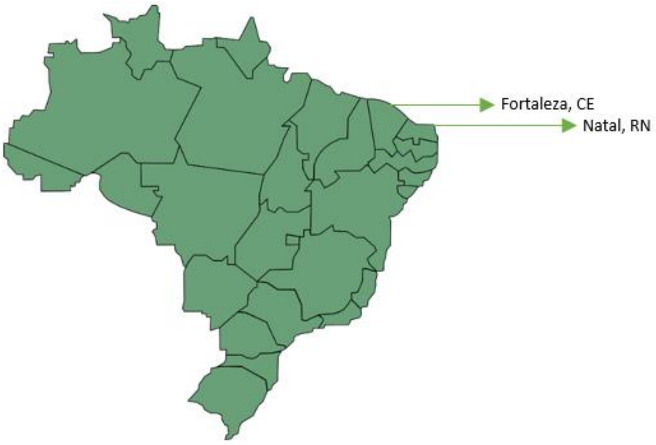
There are several sex-related and age-related differences in foraging behaviour in capuchin monkeys. Adult females tend to spend more time foraging than adult males; however, adult males spend more time hunting and eating small animals or insects than adult females (Fragaszy et al., 2004). In addition to this, adult females often spend more time foraging in the canopy whereas adult males spend more time foraging on the ground. These differences are already observable before juveniles reach sexual maturity. Nonetheless, capuchin monkeys become more proficient in foraging-related behaviours as they age, and many differences on foraging behaviours are actually related to individual differences (Fragaszy et al., 2004). Infant capuchins (< 12 months of age) rely mostly on their mother for feeding, but older juveniles mostly feed themselves (Fragaszy et al., 2004). Unsurprisingly, bearded capuchin monkeys tend to spend more time manipulating food items that are tougher (e.g., foods that have a hard shell) and stiffer than food items that are softer and less stiff (Laird et al., 2020). Capuchin monkeys engage more time in ingesting foods, followed by mastication and manipulation of food items (Laird et al., 2020).

2.3. Study Sites

2.3.1. Rescue centres

I performed the practical work for this dissertation between March and July 2019. Two Brazilian government wildlife rescue centres were part of the study. Known as *Centros de Triagem de Animais Salvagens* (CETAS), both centres were located in the Northeast region of Brazil, in Fortaleza, Ceará (CETAS–CE) and in Natal, Rio Grande do Norte (CETAS–RN). CETAS–CE rescue centre was included only in Chapter 2 of this dissertation. CETAS–RN, where I was based, was included in Chapters 3, 4, 5, 6 and 7. Unfortunately, the distance between the cities where the rescue centres were (~ 435 km) made it impossible to work at both of them simultaneously. Figure 2 shows a map of Brazil with the cities of Fortaleza, Ceará and Natal, Rio Grande do Norte pointed out.

Figure 2.2. Map of Brazil pointing out the cities of Fortaleza, Ceará (CE) and Natal, Rio Grande do Norte (RN).



2.3.2. Rehabilitation and reintroduction programme

The rehabilitation and reintroduction programme run by CETAS–RN and CETAS–CE had an expected duration of approximately four months, with releases (i.e., reintroductions) being performed twice per year (in February/March and July/August). Hence, capuchin monkeys received during the first two months of every year were rehabilitated to be released in July or August of that same year, and capuchins received after February were kept until the next year. After the monkeys arrived, they were placed either in an individual cage, or directly in an enclosure with other monkeys. In most cases, there was no quarantine period (i.e., the monkey was placed in a social group in the first few days after his or her arrival) or physical examination under anaesthesia when a new individual arrived. Nonetheless, a quick, non-invasive health assessment was performed and injured and/or ill monkeys were medically treated accordingly.

2.3.3. General husbandry

In both rescue centres, the monkeys were fed once per day during the morning or early afternoon (8:00-12:30) with fresh fruits and vegetables (mango, chayote, cucumber, beetroot, banana, oranges, etc.) given daily, and potatoes and eggs, given several times per week. Both CETAS had one resident veterinarian, several caregivers, and several bachelor and postgraduate students performing behavioural research (mostly CETAS–RN). Hence, the animals housed in CETAS–RN were placed in individual cages (1 m^2 cages) and transferred to a room without the presence of other monkeys several times per month, for up to 30 minutes per session. People working with the animals in both rescue centres were instructed to avoid interaction with them at all times to help in dishabituation from human beings.

2.3.3.1. CETAS–CE housing

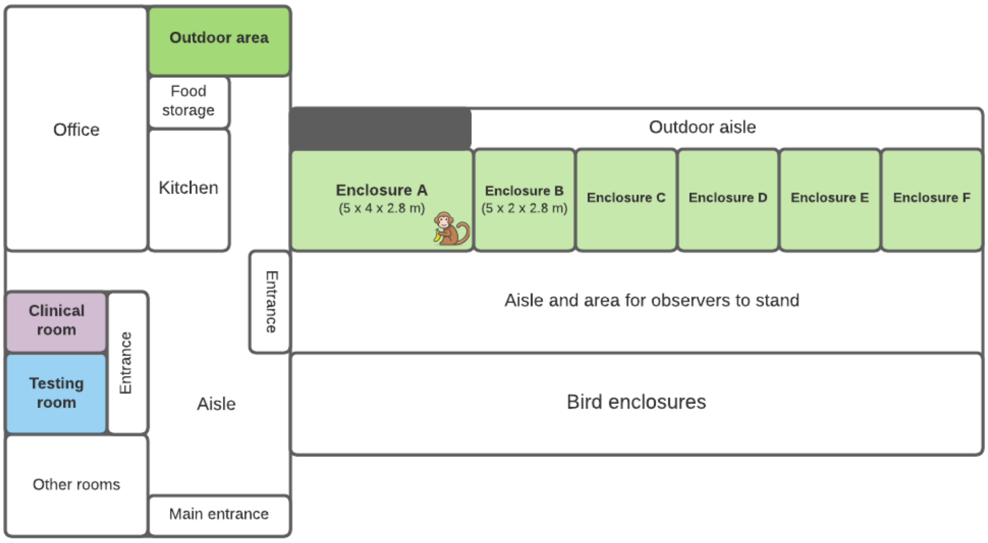
CETAS–CE had six enclosures to house monkeys, all of them located in one building. The enclosures had ceramic walls and mesh doors and windows. The enclosures measured approximately 5.0 x 2.0 x 2.8 metres (m) and contained a few items of environmental enrichment such as ropes and branches.

2.3.3.2. CETAS–RN housing

CETAS–RN had seven enclosures used to house capuchin monkeys, which I called A, B, C, D, E, F and G. Enclosures A to F ,were located in the same building (I), whilst enclosure G was located in the building next to it (II). Enclosures B to G measured 5.0 x 2.0 x 2.8 m, whilst enclosure A was approximately two times larger, measuring 5.0 x 4.0 x 2.8 m. Overall, all enclosures were structured mostly with hard substrates, such as concrete walls and floors. Most of the front wall (approximately ¾) of every enclosure, where the door was located, as well as the ceiling and most (¾) of the back wall were made of metallic mesh; except in enclosure A and B, where the back wall was made completely of concrete. There was an inner concrete ‘cage’ (i.e., a small enclosure with a door) in each enclosure where the animals were placed whilst the caregivers cleaned, once per day between 8:00 and 12:30. However, the monkeys had access to these inner cages at all times; thus, these spaces provided a place where they could hide from viewers or other monkeys if desired. There were several elements of environmental enrichment in all the enclosures, mainly ropes, and fixed and mobile branches and trunks. Enclosure C had a mobile bed since the start of the study, whereas enclosures A, D and E were enriched with mobile beds throughout the study.

Figure 2.3. Front (large) and back (small) views of enclosures C, D, E and F. (1) Metallic mesh ceiling, (2) concrete side wall, (3) back wall made of metallic mesh and concrete, (4) concrete inner cage (entrance seen in the small picture) and (5) concrete floor. Environmental enrichment structures can also be seen.

Figure 2.4. Floor plan of building I in CETAS–RN rescue centre.



2.4. Subjects

2.4.1. Origin of the individuals

Most of the monkeys considered in the present study (94.44% or 34/36) were born in the wild and captured, trafficked and kept as pets before being rescued or confiscated by the Brazilian environmental authority (*Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis*, IBAMA). Some of these monkeys may have been surrendered voluntarily by their 'owners' to the rescue centres (CETAS). The remaining two individuals (5.56% or 2/36) were male juveniles born in captivity to rescued females. Little information is often provided by previous 'owners' of trafficked primates. Even when information is given by the 'owners', it must be considered cautiously, as in some cases it is unlikely to be true. For example, 'owners' may report finding the monkeys in a park or garden outside the natural area of occurrence of the taxon (personal observation). Hence, in most cases, little is known about the experiences of the monkeys before entering the rescue centre. Thus, their psychological and physical state can only be known after a few days or weeks in the rescue centre.

2.4.2. Demographics

The study included 35 bearded capuchin monkeys: 15 adult females, 12 adult males, three juvenile females and five juvenile males. The exact age of every animal was unknown in most cases. Therefore, morphological characteristics were used to calculate the approximate age of the individuals. Table 2.2 shows the name, rescue centre, group (only in the case of individuals from CETAS-RN that were included and behavioural and personality assessments), age category, sex class and origin of the individuals included in this study.

Table 2.1

Individuals that were part of the present study (n = 35)

	Name/ID	Rescue centre	Group (at the start and end of the study)	Age category	Sex class	Origin
1	Acara	CETAS-CE	Unknown	Adult	Female	Wild-born
2	Aclara	CETAS-CE	Unknown	Adult	Female	Wild-born
3	Angélica	CETAS-CE	Unknown	Adult	Female	Wild-born
4	Fénix	CETAS-CE	Unknown	Adult	Female	Wild-born
5	Florentina	CETAS-CE	Unknown	Adult	Female	Wild-born
6	Silvana	CETAS-CE	Unknown	Adult	Female	Wild-born
7	Melequinha	CETAS-CE	Unknown	Adult	Male	Wild-born
8	Peruco	CETAS-CE	Unknown	Adult	Male	Wild-born
9	Walber	CETAS-CE	Unknown	Adult	Male	Wild-born
10	Amadeu	CETAS-CE	Unknown	Adult	Male	Wild-born

Table 2.1 (continued)

Individuals that were part of the present study (n = 35)

11	Magrela	CETAS-CE	Unknown	Juvenile	Female	Wild-born
12	Piolho	CETAS-CE	Unknown	Juvenile	Male	Wild-born
13	Dana	CETAS-RN	1, 1	Adult	Female	Wild-born
14	V	CETAS-RN	2, 2	Adult	Female	Wild-born
15	Fúria	CETAS-RN	2, 2	Adult	Female	Wild-born
16	Joana	CETAS-RN	3, 2	Adult	Female	Wild-born
17	Tapa	CETAS-RN	3, 3	Adult	Female	Wild-born
18	Capuccina	CETAS-RN	4, 4	Adult	Female	Wild-born
19	Preta	CETAS-RN	Other	Adult	Female	Wild-born
20	Cotoco	CETAS-RN	Other	Adult	Female	Wild-born
21	Caramelo	CETAS-RN	Other	Adult	Female	Wild-born
22	Lombinho	CETAS-RN	1, 1	Adult	Male	Wild-born
23	Tino	CETAS-RN	1, 1	Adult	Male	Wild-born
24	Café	CETAS-RN	2, 2	Adult	Male	Wild-born
25	Galápagos	CETAS-RN	2, 2	Adult	Male	Wild-born
26	Arrebite	CETAS-RN	3, 3	Adult	Male	Wild-born
27	Linguinha	CETAS-RN	4, 4	Adult	Male	Wild-born
28	Parambú	CETAS-RN	Other	Adult	Male	Wild-born
29	Padrinho	CETAS-RN	Other	Adult	Male	Wild-born
30	Li	CETAS-RN	1, 1	Juvenile	Female	Wild-born
31	Rabinha	CETAS-RN	4, 2	Juvenile	Female	Wild-born
32	Garrincha	CETAS-RN	1, 1	Juvenile	Male	Wild-born
33	Vitinho	CETAS-RN	4, 2	Juvenile	Male	Wild-born
34	Mãozinha	CETAS-RN	3, 3	Juvenile	Male	Captive-born
35	Tiquinho	CETAS-RN	3, 3	Juvenile	Male	Captive-born

2.4.3. Social groups

The social groups were formed of two to seven monkeys, with two to four adults (females and males) and two or more juveniles of both sexes. However, some groups were composed only by adult monkeys. The centre avoided groups composed only of juveniles, as agonistic interactions were deemed as more frequent in those cases. In CETAS–RN, an individual was removed from a group if s/he was being constantly threatened, rejected, and/or injured by other member(s) of the social group and/or if s/he was spending most of the time in isolation or performing stress-related behaviours, such as motor stereotypies or self-injurious behaviours. I do not know if this was also performed in CETAS–CE rescue centre.

Since the composition of the social groups I observed for this study changed several times i.e., there was a lack of group stability as members were changed from one group to another frequently (see Table 2.1 above for the composition of groups at the start and end of the study) and this was out of my control, I decided not to consider 'social group' as a variable for analysis in the experimental chapters (Chapters 3 to 7).

2.5. Ethical Statement

The project received ethical approval by the Animal Welfare and Ethical Review Body of the University of Bristol in January 2019 (reference number UB/18/087) and complied with the NC3Rs Guidelines for Primate Accommodation, Care and Use (NC3Rs, 2017).

Chapter 3: Health of Capuchin Monkeys (*Sapajus* sp.) in Northeast Brazilian Wildlife Rescue Centres

Chapter 3: Health of Capuchin Monkeys (*Sapajus* sp.) in Northeast Brazilian Wildlife Rescue Centres

3.1. Introduction

The IUCN in its Guidelines for Nonhuman Primate Re-introductions recommends performing health assessments on all individuals before being released back into the wild (Baker, 2002). Pre-release health assessments help to ensure that primates are healthy before and after release by:

(a) identifying potential issues such as diseases or injuries acquired during captivity. Slow lorises' (*Nycticebus* spp.) teeth, for example, are often cut or removed in illegal markets, which may compromise their survival after release (Moore et al., 2014). Injuries acquired during captivity may also prevent confiscated gibbons (*Hylobates* spp.) or spider monkeys (*Ateles geoffroyi*) from being suitable candidates for reintroduction (Cheyne, 2005; personal observation)

(b) identifying treatable injuries and/or diseases to ensure adequate health, so that the animals are fit to endure the rehabilitation, pre-release, transport and release process (Baker, 2002)

(c) finally, ensuring that the individuals are not carriers of pathogens that may affect the health and viability of wild primates, other wild animals, and/or human populations (Baker, 2002). Some infectious agents naturally occur in certain species; thus, the specific tests to perform depend on the taxon and geographical area of interest (Baker, 2002). In several primate reintroduction projects, tests performed before release commonly include full-body radiographs, screening for TB, toxoplasmosis and HVB; electrocardiograms and echocardiograms; parasitological tests and bacterial and/or mycotic cultures from faecal samples (Suárez et al., 2001; Centro de Primatología Araguatos, 2004; Arango-Guerra, 2013).

However, diagnostic tools, such as haematology screening, clinical biochemistry and ultrasonography are seldom used in the health assessment of rescued primates (Cardenio et al., 2020). These tools may provide valuable information regarding animal health and help clinicians and wildlife rehabilitators make decisions in primate reintroduction projects. Furthermore, laboratory diagnostic tests may help to ensure that the individuals have a good health and welfare level while in captivity (Ferreira et al., 2018).

Haematological parameters vary with geographical, dietary and environmental factors (Ferreira et al., 2018; Cardenio et al., 2020). Hence, it is recommended that data are compared with studies made in the same geographical area when assessing animal health. It is also important to know the age and sex class of the individuals, as certain parameters may be influenced by these factors. Erythrocyte counts and haemoglobin concentrations, for example, tend to be higher in males when compared to adult females, probably due to blood loss in menstruation and the effect of oestrogen on the formation of red cells (i.e., erythropoiesis) in mature females (Wirz et al., 2008; Ferreira et al., 2018). Similarly, certain parameters tend to differ between adults and juveniles, such as leucocytes, lymphocytes, band neutrophils and eosinophils being higher in juveniles when compared to adult bearded capuchin monkeys (Ferreira et al., 2018). Flaiban et al. (2008) found similar differences in *Sapajus cay* and *Sapajus nigritus*, with adult monkeys having lower leucocyte and lymphocyte counts than juveniles (Flaiban et al., 2008).

Only a few studies have assessed haematology of bearded capuchin monkeys in Brazil, such as those by Ribeiro et al. (2015), Ferreira et al. (2018) and Abreu Sousa et al. (2020). Moreover, I have not found any study in which the relationship between hematologic values and parasitological status has been assessed in rescued bearded capuchin monkeys. Certain parasites are capable of infecting multiple animal species, including domestic animals and non-human primates, and some of them can act as zoonotic agents, passing from animals to humans (Ngui et al., 2012). Sampling captive monkeys for gastrointestinal parasites is easier than sampling free-ranging individuals and provides useful information about the potential risk for humans (Solórzano-García and Pérez-Ponce de León, 2018). Furthermore, infections with worms such as hookworms (e.g., *Necator* spp., *Ancylostoma* spp.) and whipworms (*Trichura* spp.) frequently lead to acute and chronic issues such as intestinal obstruction, malnutrition and iron-deficiency anaemia in humans and animals (Else et al., 2020) and may lead to the disruption of haematological values.

Performing health assessments on wildlife is not always an easy task. Most primate species require anaesthesia to be examined, as even simple procedures such as sexing or weighing can be a source of stress for wild animals during veterinary examinations (Teixeira et al., 2007). Hence, procedures that require handling are generally performed using chemical restraint (i.e. under general anaesthesia). Because of this, non-invasive indicators can be used to minimise the stress of handling required to monitor certain aspects of primate health, such as coat and body condition. Millette et al. (2015), for example, developed a coat condition score and a body condition score for wild (i.e., free-ranging) ring-tailed lemurs (*Lemur catta*). They found that the individuals’ coat condition scores were higher during the wet season (as opposed to the dry season) and that females had lower coat condition scores soon after giving birth and during lactation in comparison to males (Millette et al., 2015). Moreover, body condition did not vary between the dry and wet season nor between females and males (Millette et al., 2015). Unfortunately, this study did not include if inter-rater reliability analyses were performed.

In a previous set of studies, Clingerman and Summers (2005, 2012) aimed to develop and validate a body condition score for non-human primates, using laboratory Rhesus macaques (*Macaca mulatta*) as models (Clingerman and Summers, 2005; Clingerman and Summers, 2012). Notably, this score did require handling the animal to score it, and was meant to be performed as part of a full examination under anaesthesia. The body condition score of 616 macaques was assessed by four veterinarians. The agreement between raters on the body condition score of 15 randomly selected individuals was approximately 83% (Clingerman and Summers, 2012).

In a study by Strum (2005), body condition was used as an indicator of success in a baboon (*Papio* sp.) translocation project. Several troops of wild, free-ranging baboons were translocated to different areas, and body condition varied depending on the local circumstances of where each group was moved to (Strum, 2005). Hence, it was not clear how body condition was related to survivorship or ‘success’ of the project. Nonetheless, even if coat or body condition cannot act as a predictors of ‘reintroduction success’, assessing them is still important in primate reintroduction projects, as it may help to provide valuable information such as general health status, nutrition and performance, reproductive status, resource use and the effects of different seasons on the releasees (Clingerman and Summers, 2005; Millette et al., 2015). Moreover, capuchin monkeys, as many other primates, have a tendency to become overweight or obese in captivity, due to preference and/or excessive consumption of sugary fruits and/or fruits for human consumption (Souvignet et al., 2019; personal observation). Hence, a body condition score could help primate rehabilitators and veterinarians to monitor the individuals to prevent obesity.

Furthermore, collecting and publishing the health data obtained from the use of non-invasive indicators and other health assessments could be useful, as there are few primate veterinarians when compared to other veterinary specialists (personal observation) and the scientific and veterinary literature regarding primate health during rehabilitation and reintroduction is scarce, particularly regarding Neotropical primates. Thus, the development of non-invasive health assessment protocols designed specifically for capuchin monkeys (*Sapajus* spp.) is a relevant contribution for the present study and for future rehabilitation and reintroduction projects.

The aims of this chapter were: (1) to develop and validate a coat condition score and a body condition score for bearded capuchin monkeys (*Sapajus libidinosus*), to be used in a reintroduction project (this study) and future studies or in different institutions such as wildlife rescue centres, zoological institutions and other places where capuchin monkeys are kept in captivity; (2) to investigate the physiological parameters of the individuals in this study and compare them with previous studies made with individuals from the same species; (3) to investigate the haematological values and parasitological status of the individuals in this study; finally, (4) to broaden the current veterinary scientific knowledge on the haematological profiles of capuchin monkeys in Brazil, particularly in the Northeast region.

My hypotheses were:

1. The coat condition score and body condition score would have a high level of inter-rater reliability (≥0.60; Cardenio et al., 2020).
2. The coat condition score and body condition score would be valid, i.e., there would be a strong correlation between the ‘gold standard’ and the non-invasive scorings obtained (≥0.60; Cardenio et al., 2020).

To consider the individuals as clinically healthy, I hypothesised that:

3. The physiological values obtained from the individuals would be similar to previously published values from clinically healthy individuals of the same species, and within the published reference intervals for this species.
4. The haematological values obtained from the individuals would be similar to those found in previously published values from clinically healthy individuals of the same species, and within the published reference intervals for this species.
5. The individuals would be free of intestinal parasites.

3.2. Methodology

3.2.1. Ethical approval

The project received ethical approval by the Animal Welfare and Ethical Review Body of the University of Bristol in January 2019 (reference number UB/18/087) and complied with the NC3Rs Guidelines for Primate Accommodation, Care and Use (NC3Rs, 2006).

3.2.2. Study sites and subjects

The study was performed in two government wildlife rescue centres located in Rio Grande do Norte (CETAS–RN) and in Ceará (CETAS–CE), Brazil. These rescue centres receive primates and other wildlife that have been rescued/confiscated from the pet trade in Northeast Brazil. Thirty-five capuchin monkeys (*Sapajus sp.*) were part of the study (see below). Most of these animals were born in the wild, captured and sold as pets illegally. Refer to Chapter 2: General Methodology for a full description origin of the individuals as well as the rehabilitation procedures carried out in the rescue centre. A summary of the individuals considered in this Chapter as well as the data available for analysis can be seen in Table 3.1.

Table 3.1

Individuals that were part of the study (n = 35)

	Name	**Rescue centre**	**Age category**	**Sex class**	**Origin**	**Non-invasive health assessment**	**Physical examination/ biological sampling**
1	Acara	CETAS-CE	Adult	Female	Wild-born	Yes	Yes
2	Aclara	CETAS-CE	Adult	Female	Wild-born	Yes	Yes
3	Angélica	CETAS-CE	Adult	Female	Wild-born	Yes	Yes
4	Fénix	CETAS-CE	Adult	Female	Wild-born	Yes	Yes
5	Florentina	CETAS-CE	Adult	Female	Wild-born	Yes	Yes
6	Silvana	CETAS-CE	Adult	Female	Wild-born	No	Yes
7	Melequinha	CETAS-CE	Adult	Male	Wild-born	Yes	Yes
8	Peruco	CETAS-CE	Adult	Male	Wild-born	Yes	Yes
9	Walber	CETAS-CE	Adult	Male	Wild-born	No	Yes
10	Magrela	CETAS-CE	Juvenile	Female	Wild-born	Yes	Yes
11	Piolho	CETAS-CE	Juvenile	Male	Wild-born	Yes	Yes
12	Amadeu	CETAS-CE	Adult	Male	Wild-born	Yes	Yes
13	Dana	CETAS-RN	Adult	Female	Wild-born	Yes	Yes
14	V	CETAS-RN	Adult	Female	Wild-born	Yes	Yes
15	Fúria	CETAS-RN	Adult	Female	Wild-born	Yes	Yes
16	Joana	CETAS-RN	Adult	Female	Wild-born	Yes	Yes
17	Tapa	CETAS-RN	Adult	Female	Wild-born	Yes	Yes
18	Capuccina	CETAS-RN	Adult	Female	Wild-born	Yes	No
19	Preta	CETAS-RN	Adult	Female	Wild-born	Yes	No
20	Cotoco	CETAS-RN	Adult	Female	Wild-born	Yes	No
21	Caramelo	CETAS-RN	Adult	Female	Wild-born	Yes	No
22	Lombinho	CETAS-RN	Adult	Male	Wild-born	Yes	Yes
23	Café	CETAS-RN	Adult	Male	Wild-born	Yes	Yes
24	Galápagos	CETAS-RN	Adult	Male	Wild-born	Yes	Yes
25	Arrebite	CETAS-RN	Adult	Male	Wild-born	Yes	Yes
26	Linguinha	CETAS-RN	Adult	Male	Wild-born	Yes	No
27	Parambú	CETAS-RN	Adult	Male	Wild-born	Yes	No
28	Padrinho	CETAS-RN	Adult	Male	Wild-born	Yes	No
29	Li	CETAS-RN	Juvenile	Female	Wild-born	Yes	Yes
30	Rabinha	CETAS-RN	Juvenile	Female	Wild-born	Yes	Yes
31	Garrincha	CETAS-RN	Juvenile	Male	Wild-born	Yes	Yes
32	Vitinho	CETAS-RN	Juvenile	Male	Wild-born	Yes	Yes
33	Mãozinha	CETAS-RN	Juvenile	Male	Captive-born	Yes	Yes
34	Tiquinho	CETAS-RN	Juvenile	Male	Captive-born	Yes	Yes
35	Tino	CETAS-RN	Adult	Male	Wild-born	Yes	Yes

3.2.3. Health assessments

I assessed the health of the individuals using two different methods: (1) a series of non-invasive assessments performed by several raters, using indicators that could be measured from a distance (i.e., remotely); and (2) a physical examination and biological sample collection/analysis, performed under general anaesthesia (this was performed by a veterinary team that included me). Moreover, blood sampling for serological tests for detection of arboviruses was performed during this physical examination. An *arbovirus*, short for arthropod-borne virus, is a virus that is transmitted to humans beings and other vertebrate animals by arthropod species that feed on blood, such as mosquitoes and ticks (Yuill, 2018). Several arboviral diseases, such as yellow fever disease, are important for public (human) and wildlife health, as they can be transmitted between humans (i.e. urban cycle, from human to human) and between non-human primates (i.e. sylvatic cycle, from monkey to monkey) by mosquitoes (CDC, 2019). Hence, it was important to perform these tests to be certain that the CETAS individuals would not pose a threat to free-ranging individuals from the same or other primate species living in the release area.

The non-invasive general health indicators used were a body condition score and a coat condition score developed for this study and based on published scores for primates (Honess et al., 2005; Clingerman and Summers, 2005; Millette et al., 2015). A summary of the non-invasive health indicators can be seen in Table 3.2.

Table 3.2

Non-invasive health indicators utilised in this study and references

	Indicator	Scale	Reference
1	Coat condition	5-point scale	Honess et al., 2005 Millette et al., 2015
2	Body condition	5-point scale	Berman and Schwartz, 1988 Clingerman and Summers, 2005 Millette et al., 2015

3.2.3.1. Development of body and coat condition scores

I developed a body condition scoring system based on those proposed by Berman and Schwartz (1988), Clingerman and Summers (2005), and Millette et al. (2015). The first two were developed considering captive macaques (*Macaca* spp.) as models, whilst the last one was based on free-ranging ring-tailed lemurs (*Lemur catta*). The proposed score was based on an ordinal five-point scale, where the animals could be rated as (1) emaciated, (2) thin, (3) ideal, (4) overweight, and (5) obese. Each animal was rated for four aspects of body condition: (a) general appearance, (b) head/face, (c) body, and (d) palpable structures. The aspect *palpable structures* could be measured only during a physical examination as it required to handle the monkey; otherwise, the scale was the same for assessments performed from a distance (without handling) and when the animal was anaesthetised. I kept each score separate for analysis. The description of the complete score can be seen in Table 3.3.

Table 3.3

Body condition score (BCS) used non-invasively and during the physical examinations (based on Berman and Schwartz, 1988; Clingerman and Summers, 2005 and Millette et al., 2015)

Aspect	Score	Description
General appearance	1 (emaciated)	The monkey is extremely thin
	2 (thin)	The monkey appears thin, with low body mass and minimal fat reserves
	3 (ideal)	The monkey is neither thin nor fat
	4 (overweight)	The monkey seems very full or overstuffed, with a ‘light-bulb’ shape
	5 (obese)	The monkey is extremely obese
Head/face	1–2 (emaciated/thin)	The monkey has prominent facial bones; the face appears thin with eye orbits exaggerated
	3 (ideal)	The face seems full and the eye orbits do not protrude
	4 (overweight)	The head appears small in relation to the body
	5 (obese)	The head appears very small in relation to the body
Body	1 (emaciated)	Overall underlying skeletal structure is clearly visible, this includes spinous processes and hip bones
	2 (thin)	Spinous processes and hips are exposed with flanks depressed
	3 (ideal)	Hip bones and flanks may be slightly concave or not concave and there is no abdominal, axillary or inguinal excess fat apparent
	4 (overweight)	Fat deposits starting to accumulate in axillary, inguinal, or abdominal areas
	5 (obese)	Obvious, large fat deposits in the abdominal, inguinal and axillary regions that may alter posture/locomotion
Palpable structures (only during physical examination)	1 (emaciated)	The body is very angular, with no fat layer to smooth out bone structures
	2 (thin)	Hips, spinous processes, and ribs are easily palpable with only a small amount of muscle mass over hips and lumbar region
	3 (ideal)	Hip bones, ribs, and spinous processes are palpable with gentle pressure but generally not visible; well-developed muscle mass and subcutaneous fat layer
	4 (overweight)	Hip bones, spinous processes, and ribs may be difficult to palpate because of more abundant subcutaneous fat layer
	5 (obese)	Abdominal palpation is very difficult due to large amount of mesenteric fat. Hip bones, rib contours, and spinous processes only palpable with deep palpation

Following a similar methodology, I developed a coat condition score based on published scores for macaques (Honess et al., 2005) and ring-tailed lemurs (Millette et al., 2015). This score was also based on an ordinal five-point scale as described in Table 3.4.

Table 3.4

Coat condition score utilised remotely and during the physical examinations (based on Millette et al., 2015)

Score	Description
1 (good)	• The animal has excellent coat condition, with no imperfections or few of them • Little or no hair creasing present • Hair is unidirectional (cranial to caudal direction) • Coat is uniform (not matted) • 1 to 2 small size (<2 cm^2) holes permitted
2 (rough)	• General lack of grooming • Coat is slightly to significantly matted • Hair creasing and opaqueness frequently present • 1 or 2 small size holes permitted (<2 cm^2) or 1 large one (>2 cm^2)
3 (ragged)	• Several small (<2 cm^2) or large (>2 cm^2) patches of alopecia, that together account for less than 25% of the body • Hair is less than half normal length in affected areas
4 (sheared)	• Several small (<2 cm^2) or large (>2 cm) patches of alopecia, that together account for between 25% and 50% of the body • Hair is less than half normal length in affected areas
5 (bald)	• Generalised alopecia totalling more than 50% of the body • Hair is less than half normal length in affected areas • More skin visible than hair

I performed the non-invasive health assessments four times across a period of approximately three months (21 March 2019 to 25 June 2019). All raters, including me, were qualified veterinarians or veterinary/biology undergraduate students. A timeline of the health assessments performed can be seen in Table 3.5.

Table 3.5

General timeline of the health assessments performed in this study

	Activity	Date(s)	Rescue centre
1	Non-invasive health assessments	21/03/2019 10/04/2019 03/05/2019 17/06/2019 24/06/2019	CETAS–RN CETAS–RN CETAS–RN CETAS–RN CETAS–CE
2	Physical examinations and biological samples collection	18/06/2019 – 21/06/2019 24/06/2019 – 25/06/2019	CETAS–RN CETAS–CE

3.2.3.2. Physical examination and biological samples collection

These were performed on the same day (i.e., during the same anaesthetic procedure) in June 2019. We used two different methods of physical restraint depending on the size of the animal; adults and larger juveniles were transferred from their enclosure to an individual squeeze cage, which they entered voluntarily as they had been trained to do so previously. Small juveniles were restrained humanely by the caregivers using a net. Once restrained, a dose of ketamine (20 mg/kg) and xylazine (0.5 mg/kg) was administered intramuscularly by a member of the veterinary team of each rescue centre. After the animal was completely anesthetized, I or a member of the veterinary team performed a complete physical examination in which they assessed and recorded body weight, crown-rump length (CRL), heart rate, respiratory rate, and rectal temperature. CRL was measured from the external occipital protuberance to the first coccygeal vertebra (Andrade et al., 2004). Blood samples (2-5 mL) were collected from the femoral vein using disposable 3 or 5mL syringes. The collected blood was placed in a centrifuge tube and was sent to a commercial laboratory and analysed using an automated analyser (Mindray™ BC–2800Vet, Shenzhen Mindray Bio-Medical Electronics Co., Ltd. or Alere™ Bio 2900 VET™, Diagno Co., Ltd.). Body mass index was calculated using the formula BMI = (weight (kg)/crown-rump length (cm) x 1000 (Berman and Schwartz, 1988). Summarising, the following parameters were measured during the physical examinations (Table 3.6):

Table 3.6

Physiological parameters and measurements taken during the physical examinations

	Parameter	Unit/Score
1	Body weight	Kilograms (kg)
2	Body temperature	Celsius (°C)
3	Heart rate	Beats per minute (bpm)
4	Respiratory rate	Respirations per minute (rpm)
5	Crown-rump length (CRL)	Centimetres (cm)
6	Body condition	5-point scale[1]
7	Coat condition	5-point scale[1]

[1] The complete body and coat condition scores are described in the above sections. These measurements were taken by two veterinarians during the physical examinations in each of the rescue centres.

In addition to the previous measures, a faecal sample was collected for 14 of the monkeys on the same day in which the physical examination and blood collection were performed. The samples were maintained in a refrigerator at approximately 4°C before being sent to commercial veterinary laboratories. The laboratories used the Willis-Mollay simple flotation technique to scan for parasites, as this is a useful technique for rapid detection of cestode and nematode eggs (RVC, 2021).

3.2.4. Statistical analysis

Statistics were performed using SPSS 27 (SPSS Inc. Released 2020. SPSS for Windows, Version 27.0. Chicago, SPSS Inc). I performed Schapiro-Wilk analyses on all dependent variables to test for normality deviations and applied non-parametric statistics to variables with non-normal distribution. Variables are shown as mean and standard deviation, with the minimum and maximum values (i.e., range) shown between parentheses to allow comparison with previously published data. A univariate GLM was used to assess the effects of age and sex and their interaction on physiological and haematological parameters. Any significant main or interaction effects were further tested using post-hoc Least Significant Difference confidence interval adjustments. I performed one sample t-tests to compare between the means obtained in the present study and the means published in previous studies (for physiological values: Cordeiro et al., 2015; Souvignet et al., 2019; de La Salles et al., 2020; for haematological values: Ribeiro et al., 2015; Ferreira et al., 2018; Abreu Sosa et al., 2020). Moreover, I performed independent t-tests to investigate the effects of parasitological status on the haematological profiles of the monkeys. Individuals infected with parasites were considered as 'positive to (parasite species)' whilst individuals that were not infected with parasites were considered as 'negative to (parasite species)'. We considered p-values ≤ 0.05 as significant.

I used intraclass correlations (ICCs) to assess the inter-rater reliability of the body and coat condition scores. I calculated the ICC(3,1) and ICC(3,*k*) with SPSS. The number of raters (e.g., *k*) varied among indicators and days of assessment as different raters were available on different days; overall, the number of raters ranged from two to four. ICC(3,1) is used to estimate the reliability of single ratings and ICC(3,*k*) is used to evaluate the reliability across mean ratings, based on *k* number of raters (Shrout and Fleiss, 1979; Robinson et al., 2016).

To assess the validity of these scores, I used Spearman or Pearson correlations. In this study, *validity* is understood as the correlation between a health indicator and a 'gold standard' measured whilst the animal is under anaesthesia. For example, to test the validity of the body condition score, I measured body weight and crown-rump length (CRL). Then, I calculated the body mass index (BMI) of each monkey and performed Spearman correlations between body weight, BMI, and the body condition score obtained for each animal. The measures taken under anaesthesia included rectal temperature, heart rate, respiratory rate, body weight, body condition and coat condition (assessed also non-invasively i.e., from a distance) and CRL. Blood samples were also taken under anaesthesia.

3.3. Results

3.3.1. Physiological parameters, body weight and morphometry

Bearded capuchin monkeys in this study had a mean body weight of 2.23 ± 0.75 kg (range: 1.15–4.32), a body mass index (BMI) of 23.30 ± 5.00 kg/m^2 (16.58–33.50), a crown-rump length (CRL) of 30.74 ± 3.28 cm (25.00–36.00), a rectal temperature of 38.02 ± 1.09 °C

(35.90–39.07), a heart rate of 101.08 ± 17.80 beats per minute (bpm) (64.00–128.00) and a respiratory rate of 52.08 ± 16.98 respirations per minute (rpm) (20.00–88.00). Table 3.7 shows a comparison of the present study and previous studies with the same species as well as published reference intervals.

Table 3.7

Physiological parameters and body weight of bearded capuchin monkeys in this study and previous studies

Parameters	**Unit**	**Present study (n = 26)**	**Cordeiro et al. (2015) (n = 10)**	**de La Salles et al. (2020) (n = 8)**	**Reference interval (Souvignet et al., 2019)**
Anaesthetic used	mg/kg	Ketamine/ xylazine (20.0 /0.5)	Tiletamine-zolazepam (5.0)	Tiletamine-zolazepam (5.0)	NA
Rectal temperature	°C	38.02 ± 1.09	37.40 ± 0.80**	36.63 ± 0.79***	36.7–38.4
Heart rate	Bpm	101.08 ± 17.80	185.00 ± 25.00***	182.75 ± 31.10***	165–230
Respiratory rate	Rpm	52.08 ± 16.98	63.00 ± 31.00**	48.00 ± 27.00	30–50
Body weight	Kg	2.23 ± 0.75	2.76 ± 0.75**	3.0 ± 0.63***	1.30– 4.80

***Significant differences observed between results from the present study and this study (p <0.001). **Significant differences observed between results from the present study and this study (p <0.01).

I found a significant interaction between age and sex class with both body weight and BMI, (body weight, F = 5.943, dF = 2, *p* = 0.023; BMI, F = 7.192, dF = 2, *p* = 0.014). Adult males (n = 8) had significantly higher body weights and BMIs than adult females (n = 11) (*p* <0.001 for both). Nonetheless, this difference was not observed between juvenile females (n = 3) and juvenile males (n = 4) (body weight, *p* = 0.580; BMI, *p* = 0.565) (Table 2). Moreover, there was a significant interaction between age and sex class in respiratory rate (F = 8.855, dF = 2, *p* = 0.007); overall, juvenile females (n = 3) had significantly higher respiratory rates than the other age–sex categories (i.e., adult males, n = 8; adult females, n = 11; juvenile males, n = 4). I did not find significant differences related to age and sex class in the parameters rectal temperature, heart rate and crown-rump length (*p* ≥ 0.156).

Table 3.8

Physiological parameters and morphometry by age and sex class category (mean ± SD)

Parameters	**Unit**	**Adult males (n = 8)**	**Adult females (n = 11)**	**Juvenile males (n = 4)**	**Juvenile females (n = 3)**
Body weight	kg	3.12 ± 0.56***	2.02 ± 0.36***	1.62 ± 0.27	1.44 ± 0.32
Body mass index	kg/m^2	29.14 ± 3.68***	20.18 ± 3.18***	22.13 ± 2.16	20.72 ± 2.15
Crown-rump length	cm	33.06 ± 2.80	31.64 ± 1.93	27.00 ± 1.08	26.25 ± 1.56
Rectal temperature	°C	38.33 ± 0.70	37.54 ± 1.09	38.58 ± 0.92	38.20 ± 1.87
Heart rate	bpm	102.00 ± 14.02	101.09 ± 22.63	100.00 ± 7.30	100.00 ± 24.98
Respiratory rate	rpm	50.00 ± 16.97	45.09 ± 10.29	52.50 ± 13.89**	82.67 ± 9.24**

***Significant differences observed between adult males and adult females (p <0.001). **Significant differences observed between juvenile males and juvenile females (p <0.01).

3.3.2. Haematological profiles

The overall haematological values obtained in this study are shown in Table 3.9.

Table 3.9

Haematological parameters of rescued bearded capuchin monkeys (n = 26)

Parameter	Unit	Mean	± SD	Median	Min-Max
Erythrocytes	x10^{12}/L	5.56	0.82	5.44	3.85 – 7.19
PCV	L/L	0.40	0.06	0.40	0.27 – 0.51
Haemoglobin	g/L	110.20	15.70	111.50	65.00 – 142.00
MCV	fL	72.15	3.64	72.05	64.00 – 78.30
MCHC	g/dL	27.64	3.46	25.65	24.00 – 33.30
Leucocytes	x10^{9}/L	13.93	4.45	13.20	7.30 – 21.66
Band neutrophils	x10^{9}/L	0.01	0.03	0.00	0.00 – 0.15
Segmented neutrophils	x10^{9}/L	7.81	3.66	6.95	2.69 – 16.32
Eosinophils	x10^{9}/L	0.60	0.54	0.47	0.00 – 2.55
Basophils	x10^{9}/L	0.01	0.07	0.00	0.00 – 0.37
Lymphocytes	x10^{9}/L	4.59	2.65	3.80	1.07 – 11.97
Monocytes	x10^{9}/L	0.86	0.54	0.69	0.00 – 2.17
Platelets	x10^{9}/L	345.12	132.68	312.00	176.00 – 831.00

PCV = packed cell volume, MCV = mean corpuscular volume, MCHC = mean corpuscular haemoglobin concentration.

I did not find any significant difference with age and/or sex class in any of the haematological parameters considered (*p* > 0.144) (Table 3.10). There was a marginally significant tendency for categories to differ in basophil counts (Kruskal Wallis H = 7.667, *p* = 0.053): juvenile females tended to have higher basophil counts than adult males, adult females and juvenile males. Table 3.10 shows the haematological values obtained for each age–sex class category separately.

Table 3.10

Haematological parameters of rescued bearded capuchin monkeys by age and sex class categories (n = 26)

Parameter	Unit	Adult males (n = 8) (Mean ± SD)	Adult females (n = 11) (Mean ± SD)	Juvenile males (n = 4) (Mean ± SD)	Juvenile females (n = 3) (Mean ± SD)
Erythrocytes	x10^{12}/L	5.86 ± 0.85	5.31 ± 0.84	5.62 ± 0.67	5.57 ± 0.91
PCV		0.43 ± 0.06	0.39 ± 0.06	0.39 ± 0.05	0.40 ± 0.03
Haemoglobin	g/L	119.38 ± 14.87	107.46 ± 18.38	103.11 ± 6.83	105.33 ± 4.93
MCV	fL	73.08 ± 2.32	72.50 ± 4.04	69.78 ± 1.69	71.60 ± 6.73
MCHC	%	28.15 ± 4.15	27.90 ± 3.31	26.48 ± 3.67	26.83 ± 3.08
Leucocytes	x10^{9}/L	13.07 ± 3.28	13.09 ± 4.77	13.18 ± 3.96	20.31 ± 1.80
Band neutrophils	x10^{9}/L	0.00 ± 0.00	0.00 ± 0.00	0.04 ± 0.07	0.00 ± 0.00
Segmented neutrophils	x10^{9}/L	7.48 ± 3.43	7.74 ± 4.32	7.11 ± 3.46	9.92 ± 2.71
Eosinophils	x10^{9}/L	0.65 ± 0.42	0.58 ± 0.73	0.75 ± 0.32	0.33 ± 0.11
Basophils	x10^{9}/L	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.13 ± 0.21
Lymphocytes	x10^{9}/L	4.10 ± 2.41	4.00 ± 2.02	4.48 ± 3.05	8.24 ± 3.25
Monocytes	x10^{9}/L	0.84 ± 0.39	0.68 ± 0.32	0.79 ± 0.88	1.70 ± 0.40
Platelets	x10^{9}/L	386.25 ± 192.72	299.55 ± 95.26	358.75 ± 102.83	384.33 ± 89.10

All of the mean haematological values obtained in this study were within the reference intervals for this species (Ferreira et al., 2018) (Table 3.11). However, most of the mean values obtained in the present study significantly differed from the mean published values reported for captive and free-ranging bearded capuchin monkeys in previous studies (Table 3.11).

Table 3.11

Haematological values of bearded capuchin monkeys in this study and comparisons with previous studies (t tests) and reference intervals

Parameter	**Unit**	**Present study (n = 26)**	**Abreu Sousa et al. (2020)[10] (n = 17)**	**Ferreira et al. (2018)[6] (n = 50)**	**Ribeiro et al. (2015)[9] (n = 15)**	**Reference intervals (Ferreira et al., 2018)[6]**
Erythrocytes	$x10^{12}/L$	5.56 ± 0.82	10.56 ± 0.37***	5.65 ± 0.97	3.91 ± 0.33***	3.66-7.64
PCV	L/L	0.40 ± 0.06	0.40 ± 0.32	0.39 ± 0.31	Not reported	0.32-0.45
Haemoglobin	g/L	110.20 ± 15.70	133.17 ± 10.74***	121.00 ± 13.00**	128.95 ± 9.60***	95.00-147.00
MCV	fL	72.15 ± 3.64	72.29 ± 4.38	69.70 ± 10.00**	101.04 ± 46.74 ***	49.30-90.10
MCHC	g/dL	27.64 ± 3.46	32.82 ± 1.01***	31.40 ± 2.20***	33.63 ± 15.69***	26.80-35.90
Leucocytes	$x10^9/L$	13.93 ± 4.45	10.56 ± 5.20**	9.70 ± 3.88***	14.87 ± 4.20	3.92-18.69
Band neutrophils	$x10^9/L$	0.01 ± 0.03	Not reported	0.08 ± 0.11***	Not reported	0.00-0.41
Segmented neutrophils	$x10^9/L$	7.81 ± 3.66	4.97 ± 5.57**	4.93 ± 3.24***	Not reported	1.11-14.31
Eosinophils	$x10^9/L$	0.60 ± 0.54	0.70 ± 0.24	0.21 ± 0.33**	Not reported	0.00-1.52
Basophils	$x10^9/L$	0.01 ± 0.07	Not reported	0.01 ± 0.07	Not reported	0.00-0.13
Lymphocytes	$x10^9/L$	4.59 ± 2.65	3.98 ± 1.98	4.08 ± 2.23	Not reported	0.33-9.26
Monocytes	$x10^9/L$	0.86 ± 0.54	0.53 ± 0.44**	0.46 ± 0.87**	Not reported	0.00-4.67
Platelets	$x10^9/L$	345.12 ± 132.68	Not reported	270.72 ± 52.12**	169.50 ± 72.31 ***	164.93-376.51

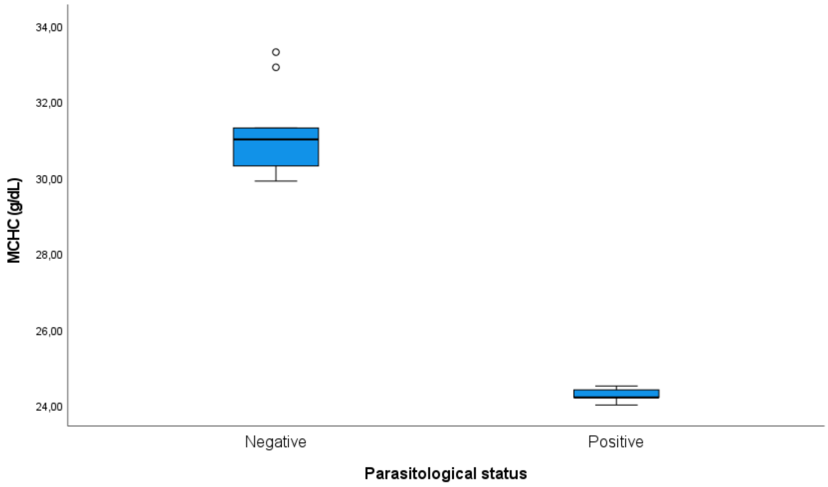
PCV = packed cell volume, MCV = mean corpuscular volume, MCHC = mean corpuscular haemoglobin concentration. ***Significant differences observed between results from the present study and this study ($p < 0.001$). **Significant differences observed between results from the present study and this study ($p < 0.01$).

3.3.3. Parasitological status: effects on physiological and haematological values

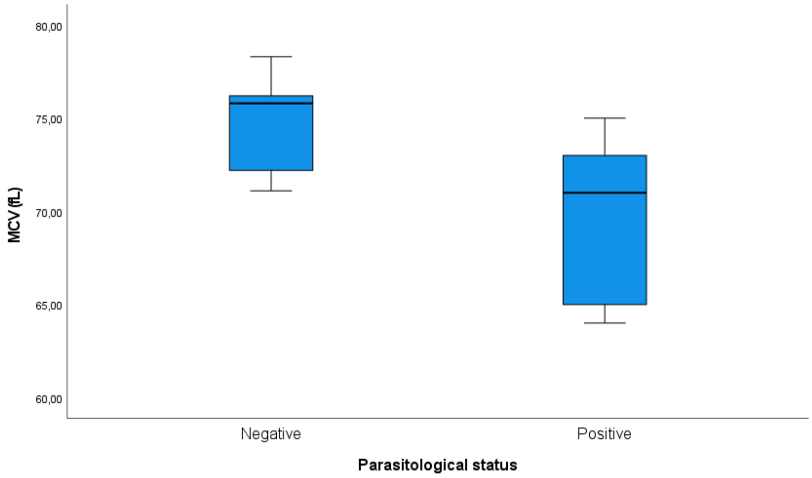
Faecal samples were collected from 14 monkeys. Five of these animals were positive to *Ancylostoma* spp. Positive animals (n = 5) had significantly lower haemoglobin concentrations (100.60 ± 11.31) than negative animals (n = 9) (120.56 ± 10.83) (t = 3.255, *p* = 0.007). I found similar results in the parameters MCV and MCHC, with positive animals having significantly lower MCV values (69.60 ± 4.88) and MCHC values (24.26 ± 0.20) than negative animals (MCV = 74.82 ± 2.43, MCHC = 31.18 ± 1.20) (MCV t = 2.719, *p* = 0.019; MCHC Mann-Whitney U = 0.000, Z = -3.003, *p* = 0.003). Positive animals also had significantly higher total erythrocyte counts (5.95 ± 0.56) when compared to negative animals (5.16 ± 0.44) (t = -2.926, *p* = 0.013). Moreover, there was a tendency towards a significant difference in monocyte counts (t = -2.034, *p* = 0.065) with positive animals having higher monocyte counts (1.07 ± 0.37) than negative animals (0.73 ± 0.26). There were no significant differences between positive and negative animals in the remaining haematological parameters (*p* > 0.132).

Figure 3.1. Boxplot graphs showing the significant differences between bearded capuchin monkeys that were positive and negative to Ancylostoma spp. The figure shows the differences observed in: (a) haemoglobin concentration, (b) mean corpuscular volume, (c) mean corpuscular haemoglobin concentration, and (d) total erythrocyte counts.

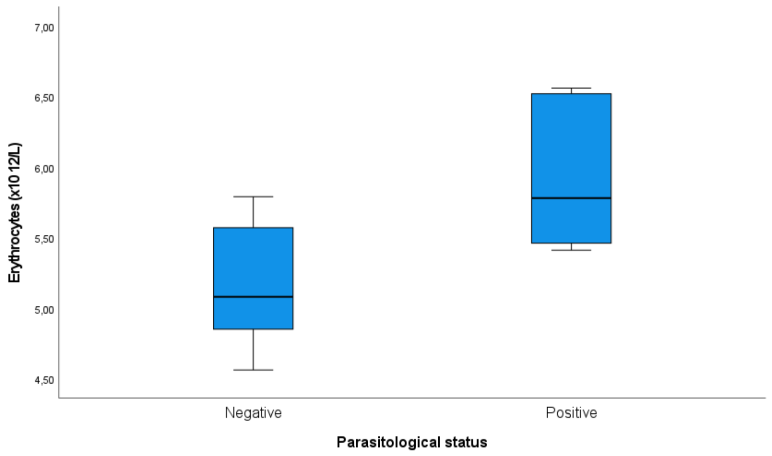
(a)



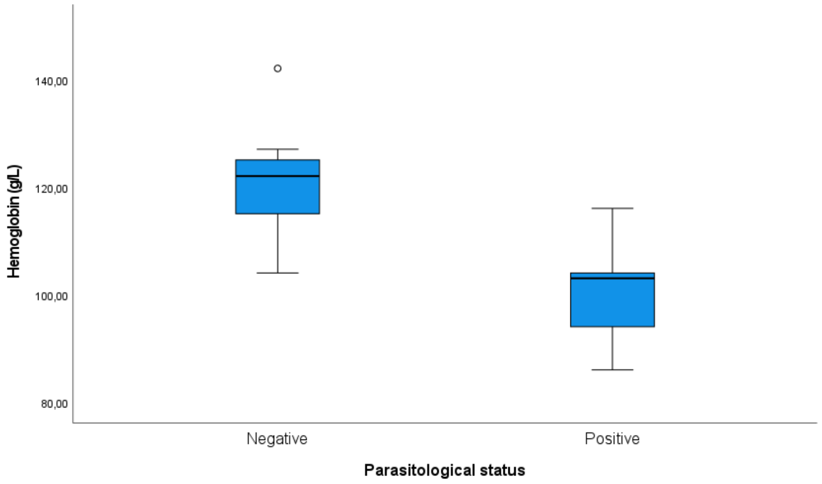
(b)



(c)



(d)



3.3.4. Serological tests

Serological arbovirus tests were performed to the 26 individuals from which blood samples were collected. They were all negative, meaning the animals did not possess antibodies for any arboviral disease.

3.3.5. Non-invasive health indicators

3.3.5.1. Body and coat condition scores

The inter-rater reliability of these scores can be seen in Tables 3.12 and 3.13 below. I considered as acceptable ICC(3,*k*) results with significant *p*-values.

Table 3.12

Inter-rater reliability of non-invasive coat and body condition scores (BCS)

	First assessment (n = 23)			Second assessment (n = 23)			Third assessment (n = 23)			Fourth assessment (n = 21)		
Indicator/item	ICC(3,1)	ICC(3,*k*)	*p*	ICC(3,1)	ICC(3,*k*)	*P*	ICC(3,1)	ICC(3,*k*)	*p*	ICC(3,1)	ICC(3,*k*)	*p*
Coat condition	**0.66**	**0.86**	**<0.001**	**0.84**	**0.94**	**<0.001**	**0.60**	**0.89**	**<0.001**	**0.60**	**0.72**	**<0.01**
BCS – general	0.27	0.53	0.017	0.18	0.39	0.079	**0.39**	**0.72**	**<0.001**	**0.74**	**0.84**	**<0.001**
BCS – head/face	**0.79**	**0.92**	**<0.001**	**0.60**	**0.82**	**<0.001**	**0.24**	**0.55**	**0.006**	-0.09	-0.22	0.669
BCS – body	0.30	0.56	0.010	0.07	0.18	0.284	**0.20**	**0.50**	**0.016**	**0.41**	**0.60**	**0.024**

k = first and second assessments, 3 raters; third assessment, 4 raters; fourth assessment, 2 raters.

Table 3.13

Inter-rater reliability of coat and body condition scores during the physical examinations (n = 26)

Indicator/item	ICC(3,1)	ICC(3,*k*)	*p*
Coat condition	**0.88**	**0.94**	**<0.001**
BCS – general	**0.84**	**0.91**	**<0.001**
BCS – head/face	**0.70**	**0.83**	**<0.001**
BCS – body	**0.69**	**0.82**	**<0.001**
BCS – palpable structures	**0.72**	**0.83**	**<0.001**

k = 2 raters.

To test the validity of the body condition score (BCS), I measured body weight and crown-rump length (CRL) and calculated the body mass index (BMI) of each monkey. Then, I performed Spearman correlations, in which I analysed the correlations between (1) the ratings obtained during the physical examinations and (2) the ratings obtained remotely with (a) the body weight and (b) the body mass index (BMI) of every individual, as performed by other authors to validate body condition scorings in primates (e.g., Berman and Schwartz, 1988). BMI was calculated utilising the formula BMI = (weight/crown-rump length) x 1000 (Berman and Schwartz, 1988). This index is used as a general measure of body fat in many primate species (including humans) and is highly correlated with tritiated water methods used to measure body fat (Berman and Schwartz, 1988). Because of this, I used BMI as the ‘gold standard’ for the body condition score.

When using body mass index (BMI), I performed the Spearman correlations considering all the animals in one group i.e., adults and juveniles of both sexes. However, when I used body weight, I performed the Spearman correlations for adult females, adult males, and juveniles of both sexes separately to account for differences in body mass due to sexual dimorphism. Adult brown capuchin monkeys (*Sapajus* spp.) exhibit sexual dimorphism, with adult males having on average a 1.9 larger body mass than adult females (Fragaszy et al., 2016). However, this does not necessarily mean adult males, in general, have a better body condition than adult females. For example, an adult male with a body condition score of 2 (thin) may still have a larger body weight than a female with a body condition score of 3 (ideal). Hence, it makes sense to compare using body weight only between individuals of the same sex after the age in which the differences in body mass start to appear. This happens at approximately four years of age (Fragaszy et al., 2016). The results of the Spearman correlations can be seen in Tables 3.14, 3.15, 3.16 and 3.17.

Table 3.14

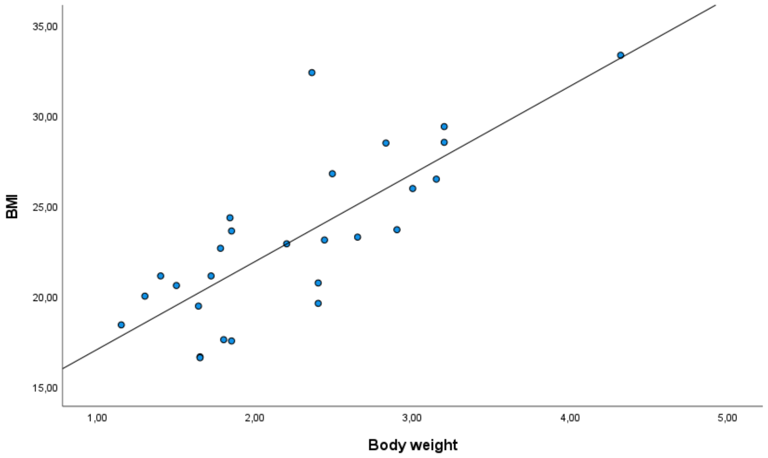
Spearman correlations between BCS ratings performed during physical examinations and non-invasively and body mass index (BMI) for all individuals (n = 15 to 26)

Physical examination scorings						
		Weight	BCS general	BCS head	BCS body	BCS palpable
BMI	Spearman Rho	**0.766**	**0.582**	**0.421**	**0.605**	**0.646**
	P	<0.001	0.001	0.029	0.001	<0.001
	p'	**<0.001**	**0.029**	0.318	**0.017**	**0.006**
Non-invasive scorings						
		Weight	BCS general	BCS head	BCS body	BCS palpable
BMI	Spearman Rho	**0.766**	**0.496**	**0.543**	**0.654**	NA
	P	<0.001	0.012	0.020	0.008	NA
	p'	**<0.001**	0.152	0.239	0.121	NA

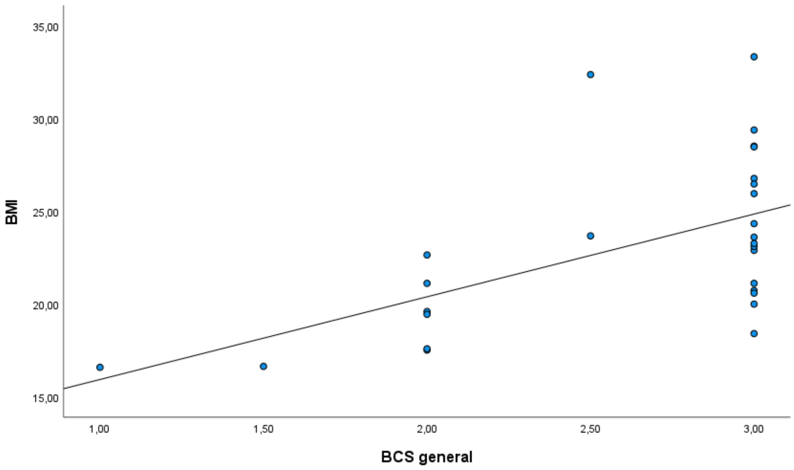
* n = 26, ** n = 18, *** n = 15. *p'* = *p*-value after Holm-Bonferroni sequential correction.

Figure 3.2. Scattergrams showing the correlations between body mass index (BMI) and (a) body weight (b) body condition general score, (c) body condition – head score, (d) body condition – body score and (e) body condition – palpable structures score.

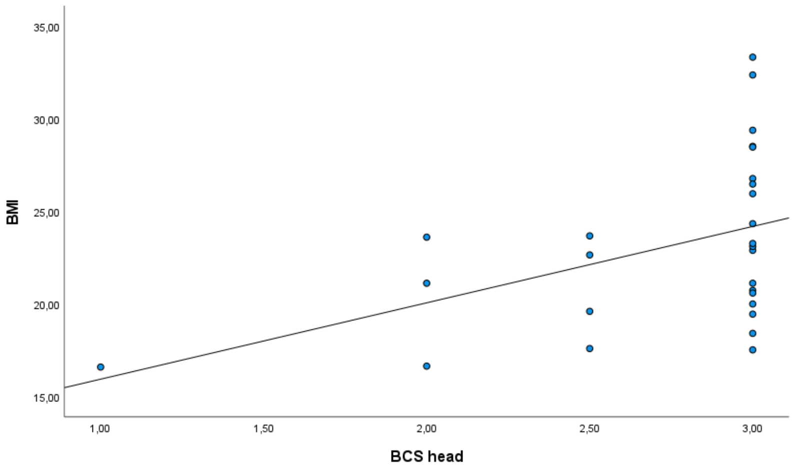
(a)



(b)

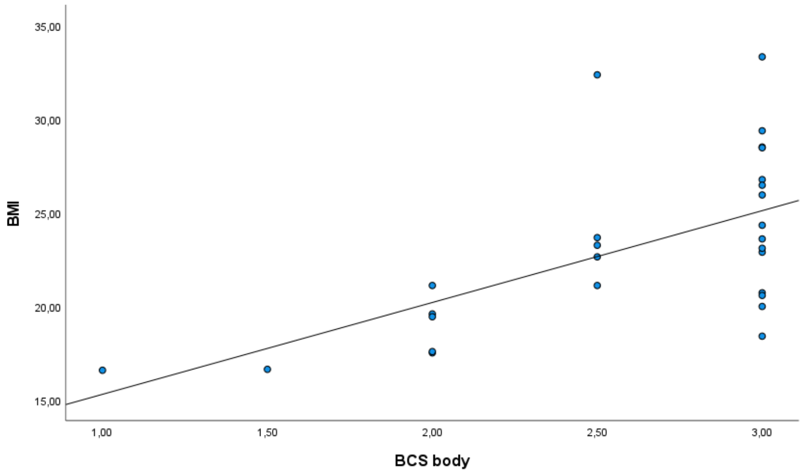


(c)



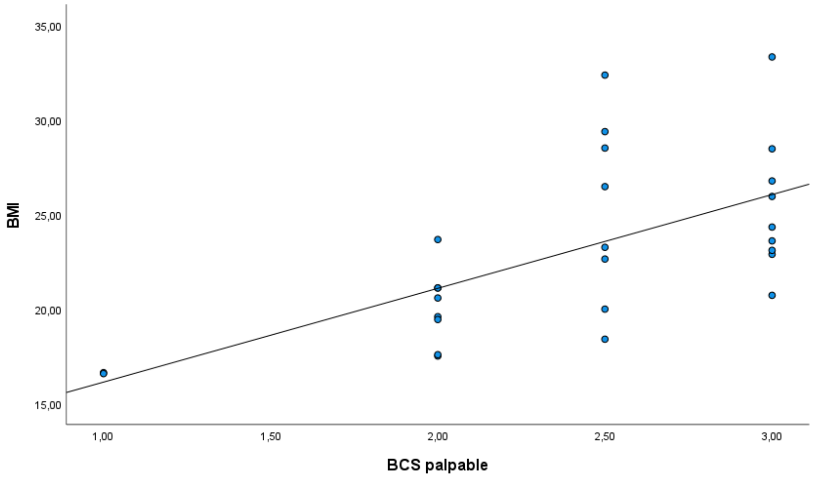
35,00
30,00
25,00
20,00
15,00
BMI
1,00
1,50
2,00
2,50
3,00
BCS head

(d)



35,00
30,00
25,00
20,00
15,00
BMI
1,00
1,50
2,00
2,50
3,00
BCS body

(e)



35,00
30,00
25,00
20,00
15,00
BMI
1,00
1,50
2,00
2,50
3,00
BCS palpable

Figure 3.3. Scattergrams showing the correlations between body mass index (BMI) and (a) body condition general score, (b) body condition – head score and (c) body condition – body score.

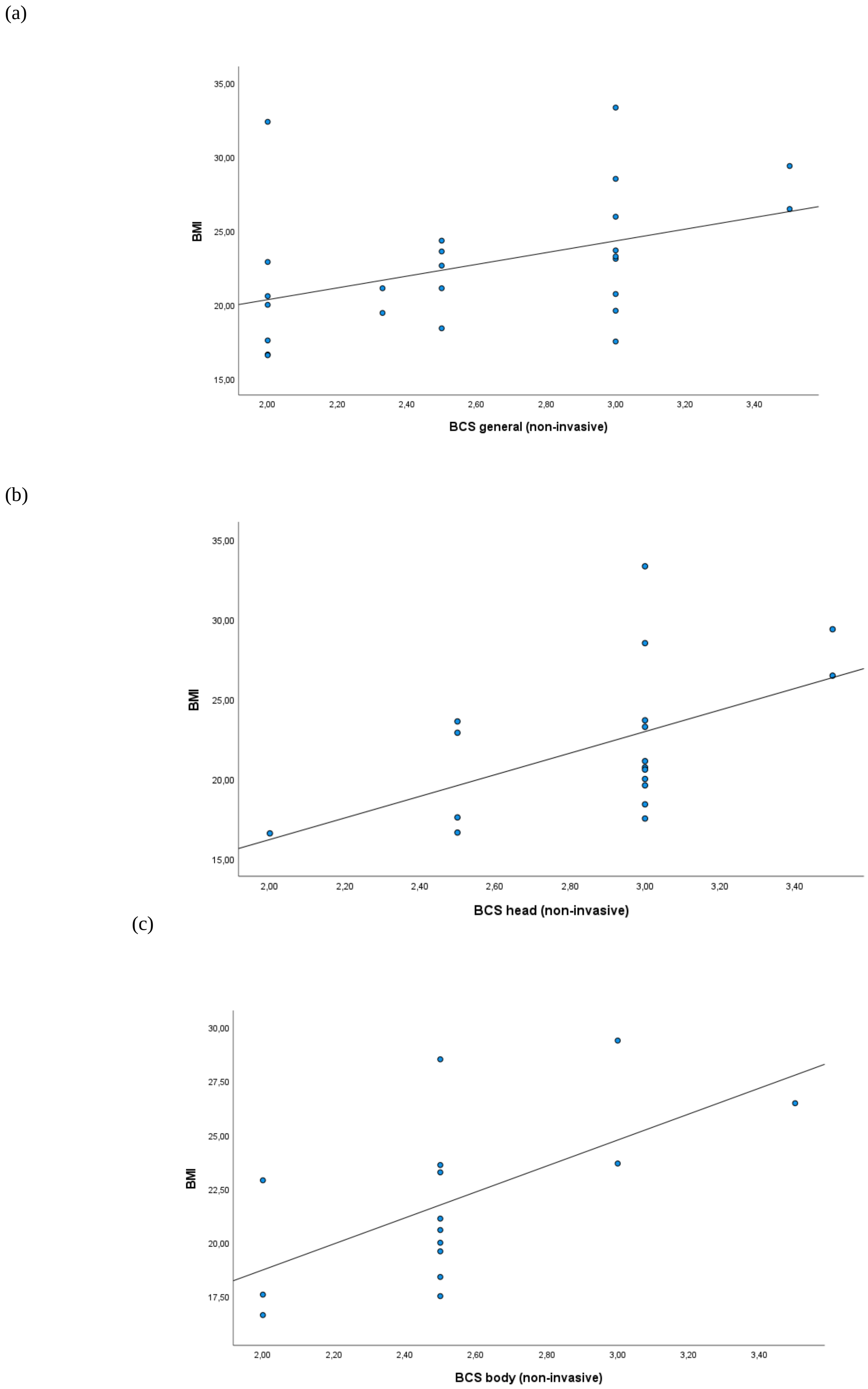


Table 3.15

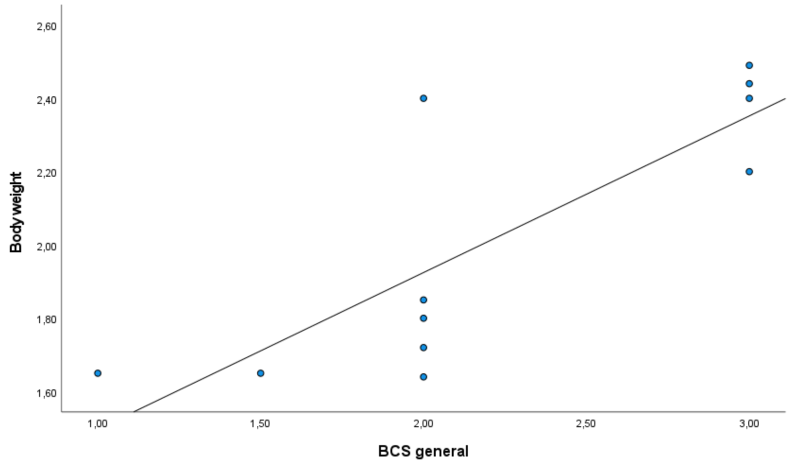
Spearman correlations for the BCS ratings and body weight obtained during the physical examinations for adult females (n = 11)

		BMI	BCS general	BCS head	BCS body	BCS palpable
Weight	Spearman Rho	**0.740**	**0.794**	0.533	**0.794**	**0.796**
	p	0.009	0.004	0.091	0.004	0.003
	p'	0.129	0.061	0.821	0.061	0.061

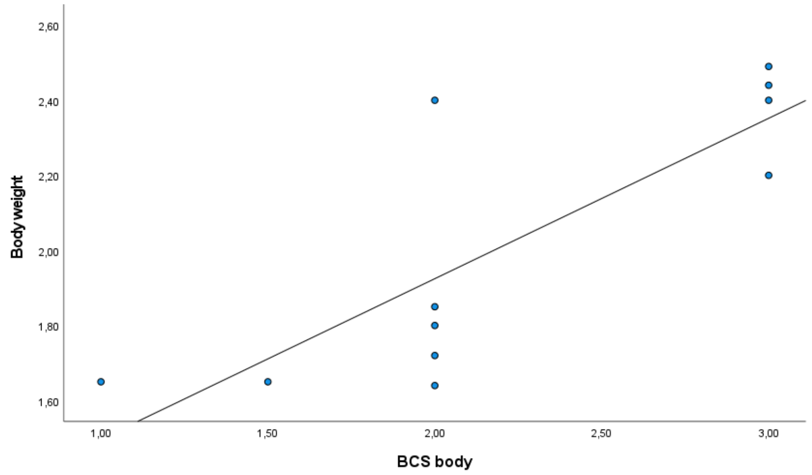
p' = *p*-value after Holm-Bonferroni sequential correction.

Figure 3.4. Scattergrams showing the correlations between body weight and (a) body condition – general score, (b) body condition – body score, and (d) body condition – palpable structures score. These scores were obtained during the physical examinations.

(a)



(b)



(c)

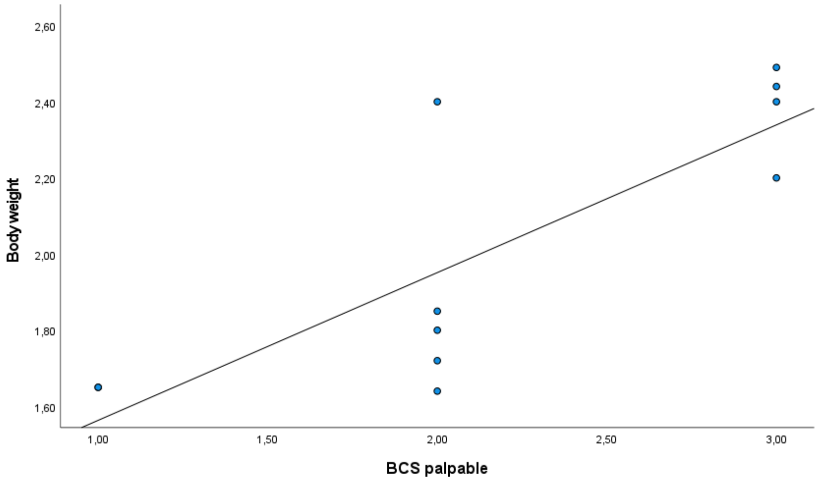


Table 3.16 **Spearman correlations for the BCS ratings and body weight obtained during the physical examinations for adult males (n = 7)**						
		BMI	BCS general	BCS head	BCS body	BCS palpable
Weight	Spearman Rho	**0.775**	0.412	0.412	0.412	0.039
	p	0.041	0.354	0.354	0.354	0.934
	p'	0.408	1.000	1.000	1.000	1.000

p' = *p*-value after Holm-Bonferroni sequential correction.

Table 3.17 **Spearman correlations for the BCS ratings and body weight obtained during the physical examinations for juveniles of both sexes (n = 9)**						
		BMI	BCS general	BCS head	BCS body	BCS palpable
Weight	Spearman Rho	**0.867**	-0.274	-0.228	-0.433	0.387
	P	0.002	0.476	0.555	0.244	0.303
	p'	**0.047**	1.000	1.000	1.000	1.000

p' = *p*-value after Holm-Bonferroni sequential correction.

As seen above, the correlation between weight and BMI was high for all age categories (r_s = 0.740, *p* = 0.009, *p'* = 0.129 for adult females; r_s = 0.78, p = 0.041, *p*' = 408 for adult males; and r_s = 0.87, p = 0.002, *p'* = 0.067 for juveniles of both sexes). Furthermore, the correlations between body mass index (BMI) and the body condition scores were significant for animals of all ages and both sexes. Nevertheless, correlations between body weight and the body conditions scores obtained during the physical examinations were not significant for any age/sex category after performing Holm-Bonferroni sequential correction; hence, I excluded body weight from further analysis.

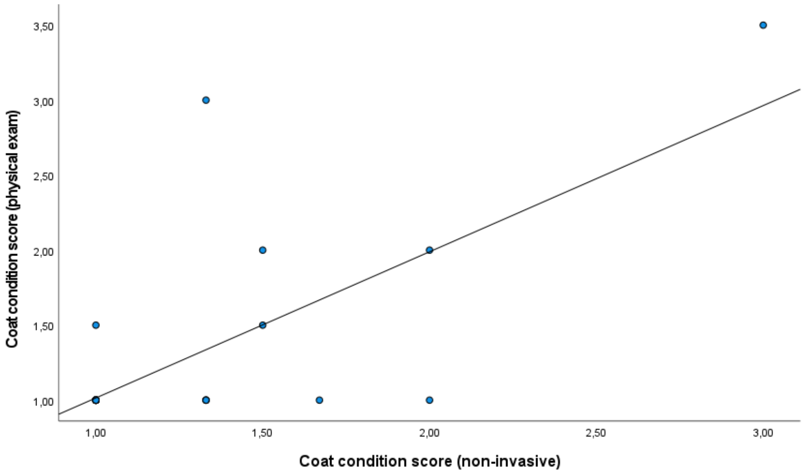
Regarding the coat condition score, I performed Pearson correlations between the scores obtained remotely for coat condition and the scores obtained during the physical examinations for all individuals to test the validity of the non-invasive scoring system. In this case, it was not necessary to analyse the scores separately for each age or sex category as there are no differences related to sex class or age in the coat condition of capuchin monkeys of different ages. The correlation between both scores was considered as moderate (r = 0.548, $p < 0.005$) and can be seen in Table 3.18 and Figure 3.5.

Table 3.18

Pearson correlation for the coat condition score ratings obtained during the physical examinations and non-invasively for all individuals (n = 25)

		Scores obtained through non-invasive health assessment
Scores obtained during physical examination	r	**0.548**
	p	**0.005**

Figure 3.5. Scattergrams showing the correlations between the coat condition score obtained during the physical examination and the coat condition non-invasively.



3.4. Discussion

3.4.1. Physiological parameters

Overall, the physiological parameters obtained in this study were within the reference intervals proposed by Souvignet et al. (2019) in The Best Practice Guidelines for Capuchin Monkeys of the European Association for Zoos and Aquariums (EAZA) (Souvignet et al., 2019). Nonetheless, mean cardiac frequency (101.08 beats per minute) was significantly lower than in previous studies with captive *S. libidinosus* (Cordeiro et al., 2015; de La Salles et al., 2020) and lower than the reference value of this parameter for this species (165–230 beats per minute; Souvignet et al., 2019). I hypothesise this may be related to the drug and dosage used in the present study (xylazine 20mg/kg, ketamine 5 mg/kg) as certain anaesthetics can influence physiological parameters. Alpha2-agonists such as xylazine are known to promote bradycardia due to an increase in systemic cardiovascular resistance (Lester et al., 2012). In fact, a study with 24 black capuchin monkeys (*Sapajus nigritus*) reported a decrease in the mean cardiac frequencies of the individuals from approximately 130 beats per minute to 100-110 beats per minute after 40 minutes of induction with ketamine (7.5 mg/kg) and dexmedetomidine (30-50 µg/kg), another alpha2-agonist (Chagas et al., 2018). Furthermore, pronounced bradycardia (<70 bpm) was found in three individuals anesthetized with ketamine/dexmedetomidine (Chagas et al., 2018). In the present study, low cardiac frequencies and the pronounced bradycardia (<70 bpm, Chagas et al., 2018) observed in one adult female may be related to the administration of xylazine. Further studies need to be performed to understand the effect of alpha2-agonists and their dosages on the cardiovascular parameters of bearded capuchin monkeys.

3.4.2. Haematological profiles

The mean haematological values obtained in the present study were within the reference intervals proposed by Ferreira et al. (2018) for bearded capuchin monkeys (*Sapajus libidinosus*). The overall mean values obtained differed statistically from previous published studies (Ferreira et al., 2018; Abreu Sousa et al., 2020 and Ribeiro et al., 2015) for this species. Local geographical characteristics may influence haematological parameters (Ferreira et al., 2018); thus, it is recommended to compare with studies performed for the same species and in the same geographical area where available. In this sense, the present study and the Ferreira et al. (2018) study were performed with captive bearded capuchin monkeys from the same geographical area, Northeast Brazil, whilst the Abreu Sousa et al. (2020) and Ribeiro et al. (2015) studies were performed with free-ranging individuals within the area of natural occurrence of *S. libidinosus* (Bernardes et al., 2011; Martins et al., 2019), and hence I considered it appropriate to compare the data with these studies as well.

Interestingly, differences in white blood cell values have been reported when comparing between captive and free-ranging capuchin monkeys (*Sapajus* spp). Mean leucocyte counts tend to be higher in free-ranging capuchin monkeys when compared to captive conspecifics (Ribeiro et al., 2015). When comparing with previous studies on the same species, leucocyte counts in the present study were higher than those reported by Ferreira et al. (2018) in captive individuals and by Abreu Sousa et al. (2020) in free-ranging individuals, and similar to those reported by Ribeiro et al. (2015) in free-ranging bearded capuchins albeit the values observed in all studies were within the reference intervals proposed for this species (Ferreira et al., 2018). Leucocyte counts increase in response to acute stress and exposure to environmental pathogens; thus, animals may have lower values when maintained in captivity (Ribeiro et al., 2015; Abreu Sousa et al., 2020). In the present study, most of the animals (92%) were born in the wild and it was unknown how much time they had spent in captivity and in what conditions before being received by the rescue centres. Considering this, as well as the fact that the leucocyte values were within reference intervals (Ferreira et al., 2018), I do not consider this finding as clinically significant. However, further studies could be made to explore the relationship between stress, environmental conditions (including captivity) and immune function in bearded capuchin monkeys.

I did not find any significant difference related to age and/or sex class in the haematological values analysed in the present study, which could be related to the small sample size (n = 26). Several previous studies with captive, non-sterilized *S. libidinosus* and *S. apella* have found sex-related differences in total erythrocyte count and haemoglobin concentration, with males having higher values than females (Ferreira et al., 2018; Riviello and Wirz, 2011; Naves et al., 2006; Wirz et al., 2008). However, these differences have not been observed when comparing by sex class in studies with free-ranging *S. libidinosus*, *S. cay* or *S. nigritus* (Ribeiro et al., 2015; Abreu Sousa et al., 2020; Flaiban et al., 2008). Menstruation and oestrogen probably contribute to the sex-related differences observed in several studies regarding total erythrocyte counts, as females lose blood during menstruation and oestrogen may inhibit erythropoiesis (Wirz et al., 2008; Ferreira et al., 2018). Furthermore, lower haemoglobin values in females may be related to smaller muscle masses and hence less demand for haemoglobin, as this protein is responsible for carrying oxygen to tissues (Ihrig et al., 2001).

Previous attempts to analyse the effect of age on haematological parameters of capuchin monkeys have resulted in mixed findings. Ferreira et al. (2018) found that adult *S. libidinosus* had lower haemoglobin concentrations, PCV values, leucocytes, lymphocytes, band neutrophils and eosinophils when compared to juveniles (Ferreira et al., 2018). Moreover, Wirz et al. (2008) and Riviello and Wirz (2001) found age-related differences in neutrophils, lymphocytes and platelets of *S. apella*, with adults having higher neutrophil counts and lower platelet (Wirz et al., 2008) and lymphocyte counts (Riviello and Wirz, 2001) than juveniles. Finally, Flaiban et al. (2008) found that adult *S. cay* and *S. nigritus* had lower leucocyte and lymphocyte counts than juveniles (Flaiban et al., 2008). In this study, I did not find significant differences related to age in any of the haematological values considered.

3.4.3. Parasitological status

Faecal samples were collected from 14 monkeys, and five monkeys from CETAS-RN rescue centre were positive to *Ancylostoma* spp. *Ancylostoma* is a genus of helminth parasites commonly known as hookworms (Kaye, 2016) whose definitive hosts differ between species. Humans are the definitive hosts of *Ancylostoma duodenale*, while dogs are the definitive hosts of *Ancylostoma caninum* and *Ancylostoma braziliense* and cats of *Ancylostoma braziliense* and *Ancylostoma tubaeforme* (Fetouh, 2003; Lappin, 2013). Certain species, such as A. duodenale, possess the ability to cross-infect other animal species, different to their definitive hosts (Fetouh, 2003). Unfortunately, the non-invasive diagnostic test performed in the present study (i.e., simple flotation technique), does not allow for species identification or quantification of parasite loads. This is a limitation of several non-invasive parasite identification techniques, as they rely mainly on the morphological features of eggs, which may be very similar in related species (Solórzano-García and Pérez-Ponce de León, 2018). Nonetheless, simple flotation techniques are useful in the preliminary assessment of many parasite species (RVC, 2021).

When I compared between individuals infected with *Ancylostoma* spp. (positive) and non-infected (negative) individuals, I found significant differences in several haematological values. Infected animals had significantly lower haemoglobin, MCV and MCHC values than non-infected individuals. These findings are consistent with the pathogenic processes associated with infection by *Ancylostoma* spp., as these parasites promote blood loss and consequent anaemia by attaching to the intestinal wall (Lappin, 2013; CDC, 2019). When comparing the hematologic values of infected animals to the reference intervals (RI) proposed for *S. libidinosus* (Ferreira et al., 2018), only MCHM values were found to be below the RI. Chronic cases of hookworm infections, including ancylostomiasis, may lead to iron-deficiency anaemia; this has been observed in several species such as dogs (Lappin, 2013) and humans (Ngui et al., 2012). Hence, I hypothesise the changes in haemoglobin, MCV and MCHC observed in infected animals in the present study was related to infection with *Ancylostoma* spp. The mean lower values observed in infected animals when compared to non-infected individuals, albeit not lower than the RI proposed for this species (Ferreira et al., 2018), support this hypothesis. Furthermore, infected individuals had significantly higher erythrocyte counts than non-infected individuals. Even though erythrocytosis may be observed in severe cases of haemorrhagic gastroenteritis (Hall, 2013), the erythrocyte counts observed in infected animals in this study were only slightly higher than those of non-infected individuals and were within the RI proposed for this species (Ferreira et al., 2018); therefore, I do not consider this finding as clinically significant.

Unfortunately, it was not possible in the present study to identify the exact source of infection of *Ancylostoma* spp. nor quantify the parasite loads of the infected animals. I have not found any study reporting the presence of these parasites in free-ranging capuchin monkeys (*Sapajus* spp.). Nonetheless, *Ancylostoma* spp. has been identified previously in captive capuchin monkeys and may be related to anthropozoonotic transmission (Macedo, 2016; Solórzano-García and Pérez-Ponce de León, 2018). I hypothesise that the affected individuals in the present study could have become infected due to close contact with humans, dogs and/or cats when kept as pets, or even whilst in the rescue centre, where transmission may have occurred as all affected individuals were housed in the same rescue centre (CETAS-RN). Ancylostomiasis has been associated with several negative effects on the health and development of infected animals and humans, such as low growth rates, and relevant clinical signs such as abdominal pain, nausea, malabsorption, diarrhoea, weight loss, skin diseases and even pneumonia (Lappin, 2013; CDC, 2019). Unfortunately, irregularities in resource (financial) availability, and the continuous entry of animals prevented the rescue centre conducting routine tests for quantification of parasites loads. Sample collection to perform the Willis-Mollay flotation technique was conducted only before the animals were released, in order to ensure that the animals were fit for reintroduction and to avoid the introduction of infectious agents to naïve environments. In the present study, the animals were treated before being released back into the wild.

3.4.4. Body weight

Mean body weight (2.23 kg) of the individuals in the present study was significantly lower than those reported by Cordeiro et al. (2015) (2.76kg) and de La Salles et al. (2020) (3.00 kg) for captive individuals of this species. Nonetheless, our study included immature individuals, whilst Cordeiro et al. (2015) study only included adults and de La Salles et al. (2020) included only adult males. When comparing mean body weight of adults in the present study to Cordeiro et al. (2015) and mean body weight of adult males to de La Salles et al. (2020), our results were similar. Nevertheless, Fragaszy et al. (2016) reported higher body weights for free-ranging bearded capuchin monkeys, with adult males having a mean of 3.5 kg and adult females 2.1 kg (Fragaszy et al., 2016). In this sense, both females and males in this study had lower body weights, with males having the most noticeable difference, averaging 380 grams (10%) less than free-ranging bearded capuchin monkeys. I suggest further studies to understand the difference observed in body weight between captive and free-ranging bearded capuchin monkeys, which may be related to stress of captivity, a suboptimal diet and/or lack of physical activity.

In this study, unsurprisingly adult monkeys had significantly higher body weights, body mass indexes (BMI), and crown-rump length measures (CRL) than juveniles. Adult males were also significantly heavier and had higher BMIs than adult females. However, this difference was not observed between juvenile males and females. This is consistent with the current scientific literature, as bearded capuchin monkeys start exhibiting sexual dimorphic characteristics at approximately four years of age, with adult males having on average a 1.9 larger body mass than adult females (Fragaszy et al., 2016).

3.4.5. Body and coat conditions scores

The inter-rater reliability of the coat condition score was considered as high (i.e., ≥ 0.70) in all assessments, both remotely and during the physical examinations. Conversely, the body condition score was not considered consistently reliable during the non-invasive assessments. However, it was considered reliable in all four aspects (i.e., general appearance, head/face, body, and palpable structures) in the ratings obtained from the physical examinations. This suggests there may be an issue with the non-invasive application rather than with the score per se. I suggest further research to improve the body condition score, as well as application in a larger sample size.

Spearman correlations between body weight and BMI were higher than 0.70 for all age categories. This is consistent with previous findings in other primate species, such as macaques (Berman and Schwartz, 1988). Even though there are more accurate methods available to validate primate body condition scorings, such as dual-energy X-ray absorptiometry (Summers et al., 2012), it was unrealistic to perform this or other techniques (e.g., ultrasound) in the present study, due to unavailability of the necessary equipment. Nonetheless, Spearman correlations between body weight, BMI and the aspects of general appearance, body, and palpable structures of the body condition score obtained during the physical examinations were considered high (i.e., > 0.70) for adult females. The scorings of adult males and juveniles, however, were not highly correlated with body weight or BMI. A possible explanation for this is that the sample size was smaller (i.e., n = 7 for adult males and n = 9 for juveniles of both sexes) than for adult females (n = 11). Furthermore, there was more variability in the scores of adult females, where individuals with scores of 1 (emaciated), 2 (thin) and 3 (ideal) were observed. Conversely, most adult males and juveniles of both sexes were rated as 3 (ideal). Thus, enlarging the sample size could help to improve the validity of the body condition score in further studies.

3.5. Conclusion

The first aim for this chapter was to develop and validate a coat condition score and a body condition score for bearded capuchin monkeys (*Sapajus libidinosus*), that could be used in this study and other studies or institutions (e.g., wildlife rescue centres) in the future. I hypothesised that both non-invasive scores would yield high levels of inter-rater reliability (≥0.60) and that there would be strong correlations between the 'gold standard' and the scores obtained (≥0.60), representing their 'validity'. The coat condition score had high levels of inter-rater reliability in all assessments and a strong correlation was found between the scores obtained non-invasively in the last assessment and the scores obtained whilst the animals were anaesthetised (i.e., the 'gold standard'). Conversely, the body condition score did not have high levels of inter-rater reliability in most assessments, and I did not find a strong association between the scores obtained non-invasively in the last assessment and the body mass index (BMI) calculated using the body weight and crown-rump length (CRL) obtained whilst the animals were anaesthetised (i.e., the 'gold standard'). Based on these results, the coat condition score can be used to assess coat condition in capuchin monkeys (*Sapajus* spp.) non-invasively. However, the body condition score should not be used until its inter-rater reliability is improved and its validity can be ensured. In other words, until it is clear that there is a strong association between BMI and the score.

My second aim for this chapter was to investigate the physiological parameters of the individuals in this study and compare them with previous studies performed with bearded capuchin monkeys. I hypothesised that the physiological values obtained would be similar to previously published values from clinically healthy individuals of the same species and would be within published reference intervals for *Sapajus libidinosus*. The physiological values of the monkeys were indeed similar to the values from individuals in other studies, except for heart rate. However, as explained above, this was likely related to the use of the alpha-2agonist xylazine.

The third aim of this chapter was to investigate the haematological values and parasitological status of the capuchins. I hypothesised that the haematological values obtained would be similar to previously published values from clinically healthy bearded capuchins and within published reference intervals for *Sapajus libidinosus* and that the individuals would be free from intestinal parasites. I found that five of the capuchins in this study were infected with *Ancylostoma* spp. This was likely to be the reason why some of their haematological values (e.g., MCHC) were lower than the reference intervals proposed for this species. This could also account for some of the differences observed between the haematological values obtained in this study and previous studies. However, most of the haematological values were within the reference intervals for the species and the changes observed in the infected monkeys were only mild. Thus, based on the physiological and haematological values obtained, I considered the animals as clinically healthy.

Finally, the third aim of this chapter was to broaden the current veterinary scientific knowledge on the haematological profiles of capuchin monkeys in Brazil, particularly in the Northeast region. Some of these values were possibly affected by the ancylostomiasis observed in some monkeys. However, as the available data on haematology of bearded capuchins is limited and there is only one other study performed in the Northeast of Brazil, I consider that this study achieved the aim of broadening the current veterinary scientific knowledge on bearded capuchin haematology.

Chapter 4: Behaviour of Rehabilitant Capuchin Monkeys (*Sapajus* sp.)

Chapter 4: Behaviour of Rehabilitant Capuchin Monkeys (*Sapajus* sp.)

4.1. Introduction

Primates and other animals need to develop certain behavioural species-specific skills to survive. According to Melfi and Marples (2000), these mainly include those related to (1) locomotion and orientation, (2) foraging and feeding, (3) intra- and inter-specific social behaviour, including predator avoidance and/or hunting skills, and (4) finding suitable places to rest (Melfi and Marples, 2000). Without these skills, animals are less likely to survive in the wild; therefore, I will refer to these skills as *survival skills.*

Survival skills differ between animal species. This is particularly true in primates, widely known for their behavioural flexibility. Because of this, a thorough study of the behaviour and socioecology of the species in question is necessary when working with primates in any context; either in a zoo, a research laboratory, or any other captive or wild setting. However, when dealing specifically with primates in rehabilitation programmes, considering rehabilitation as '*the process by which captive primates are treated for medical and physical disabilities until they regain health, are helped to acquire natural and social and ecological skills, and are weaned from human contact and dependence, such that they can survive independently (or with greater independence) in the wild* (Beck et al., 2007, p. 5)', survival skills gain the utmost importance. Simply because without them, the future survival and welfare of the rehabilitant individuals and ultimately the success of the whole rehabilitation/reintroduction programme would be at stake. The IUCN addresses this issue in its Guidelines for Non-human Primate Re-introductions (2002), stating that the species' behaviour and socioecology must be studied before attempting any reintroduction (Baker, 2002). However, it does not give any further guidance in how to assess the efficacy of the rehabilitation programme nor in how to perform pre-release survival skills training. There are several questions that arise when considering this issue. For example, *which* specific behaviours or skills are necessary –for the species that is part of the rehabilitation programme– to survive after release, and *how* to assess if the rehabilitation programme is actually successful at helping the animals develop these skills.

Several approaches have been proposed to try to answer these questions. A study on captive-raised spider monkeys (*Ateles geoffroyi*), for example, assessed the outcome of their rehabilitation programme by focusing on changes across time (i.e., before and after rehabilitation) related to foraging and feeding, social, and locomotion skills as well as space use (Mendoza-Nakano, 2016). In this case, the author suggested space use as an indicator of suitability for release as spider monkeys are arboreal primates who spent most of the time on the canopy and descend to the floor only occasionally. Increases in genus normative behavioural patterns, such as foraging and social behaviours, were used to compare between phases and analyse the activity budgets and assess their similarity with free-ranging counterparts (Mendoza-Nakano, 2016).

Another example is a study performed with rehabilitant gibbons (*Hylobates* spp.) by Cheyne et al. (2012). Here, the authors proposed to assess the similarity between the *activity budget* of rehabilitant gibbons and that of wild individuals of the same species (Cheyne et al., 2012). In this sense, the behavioural patterns and activity budgets of rehabilitant individuals should be similar to those of wild, free-ranging individuals before considering the animals suitable for release (Cheyne et al., 2012). Thus, specific release criteria could include observing the appropriate type of locomotion (i.e., the type of locomotion that the primate species use in the wild; for example, brachiation), space use i.e., use of the highest level of the enclosure for at least 40% and no more than 5% on the ground (if the primate species is arboreal such as gibbons), an increase in foraging and social behaviour, and a decrease in stereotypic behaviour to less than 3% (Cheyne et al., 2012). It must be considered that *similarity* of activity budgets means that there are certain behaviours considered as 'key' for the species survival; hence, these behaviours need to be learned (if not known before) and exhibited during the future releasees' rehabilitation. For example, gibbons must learn to socialise and exhibit a certain level of pair cohesion (i.e., because they are monogamous) before being deemed as suitable for release, and spider monkeys need to learn to utilise the 'arboreal' space of their enclosures and use it as much as possible before being released (Cheyne, 2004; Mendoza-Nakano, 2016). This does not mean the activity budget of the future releasees needs to be exactly the same as observed in free-ranging counterparts, as this may depend heavily on the specific characteristics of their captive environment. Small enclosures, for example, may limit space use and promote the exhibition of social behaviour as a way of decreasing stress (Mendoza-Nakano, 2016).

Activity budget is a term taken from bioenergetics research. It refers to the way in which animals divide their time to perform different activities that are necessary for their survival and reproduction, such as foraging, eating, moving, and resting; and may be obtained from behavioural observations (Christiansen et al., 2013). Furthermore, calculating activity budgets may also be useful for behavioural profiling, which is a method for measuring animal personality traits (Pankhurst et al., 2009).

Several studies have focused on behavioural profiles and activity budgets of robust capuchin monkeys (*Sapajus* spp.), both in the wild and in captivity. According to Ferreira et al. (2016), genus normative behavioural patterns (GNB) of robust capuchin monkeys include eight broad behavioural categories: *locomotion*, *feeding and foraging*, *social positive*, *social negative*, *vigilance*, *inactivity* (resting), *environmental manipulation*, and *solitary play*, whilst *behaviours potentially indicative of stress* (BPIS) include mainly behavioural patterns related to self-directed and/or repetitive behaviours, such as motor stereotypies and excessive self-grooming (Ferreira et al., 2016).

Social behaviour is of particular interest when considering capuchin monkeys, as it is often challenging to maintain captive capuchin groups socially stable (Souvignet et al., 2019). Moreover, most Neotropical primate species dedicate little time to social behaviour, about 5% of their overall activity budget, according to Sussman et al. (2005). Agonistic behaviours, which are behaviours that arise from competition for resources and include submissive and aggressive behaviours, can range from ‘mild’ (e.g., displacements) to ‘severe’ (e.g., fighting), that can lead to injuries or even death (Sussman et al., 2005). Among Neotropical primates, rates of agonistic interactions are commonly found around 0.60 per minute. These are much less common than affiliative behaviours. Indeed, more than 80% of social interactions observed in New World monkeys were classified as ‘affiliative’ in several studies (Sussman et al., 2005).

Some genus normative behaviours, such as foraging and grooming, differ distinctly between juvenile and adult capuchin monkeys and/or between adult females and males (Fragaszy et al., 2004). In general, adult females spend more time foraging than adult males, and juveniles spend more time foraging than adults, but tend to be less efficient or successful. Similarly, juveniles spend more time engaged in environmental manipulation than adult monkeys, but are less likely to be successful. Regarding social behaviour, adult females spend more time grooming others than adult males, and adult males spend more time in social play than adult females. Juveniles also spend more time playing with others than adults. Furthermore, adult monkeys spend more time in vigilance behaviour than juveniles (Fragaszy et al., 2004).

Even though several studies have focused on captive-raised capuchin monkeys in wildlife rescue centres, most studies related to behavioural patterns and activity budgets have been performed with objectives different from analysing the efficacy of rehabilitation, such as assessing welfare (Ferreira et al., 2018) or measuring personality traits (e.g., Nunes, 2017). Moreover, other indicators have been used to assess the efficacy of behavioural modification programmes, such as behavioural diversity indexes (BDIs, e.g., the Shannon H Index) for environmental enrichment (Metter et al., 2008, Miller et al., 2020). Behavioural diversity indexes have not been used to measure changes in behaviour in the rehabilitation of trafficked wild animals before. However, they could be useful to measure behavioural diversity changes in these animals.

In this study, I assessed the behaviour of a group of trafficked bearded capuchin monkeys that were part of a rehabilitation programme run by a government wildlife rescue centre in Northeast Brazil to achieve the following aims: (1) to perform an initial assessment on the efficacy of the rehabilitation programme by analysing the changes in behaviour across time (i.e., activity budgets, space and substrate use and behavioural diversity indexes) of the rehabilitant individuals and comparing, where possible, with other rehabilitant and free-ranging individuals of the same species; (2) to analyse the behavioural effects of two husbandry/research practices, which included (a) placing the monkeys in the inner cages of their enclosures for approximately 30 minutes per day during the 3.5-month observational period and (b) placing the monkeys in individual cages to carry individual behavioural tests (i.e., for approximately five minutes per trial and no more than a total of 65 minutes per monkey in the 3.5-month observational period) on the exhibition of behaviours potentially indicative of stress (BPIS) of the capuchin monkeys that were part of the rehabilitation programme, (3) to utilise the individual behavioural profiles obtained to measure personality traits in the next Chapter of this dissertation (Chapter 5); finally, (4) to use the results as a basis for determining the type of environmental enrichment and/or pre-release training that rehabilitant capuchin monkeys need to enhance their survival skills before release and perform this as a controlled experiment in a subsequent Chapter of this dissertation. Please note that the fourth aim of this Chapter was not performed due to Covid-19 travel restrictions (refer to the Covid-19 Statement at the start of this dissertation).

My hypotheses were:

1. Genus normative behavioural patterns (GNB) (i.e., time spent in the categories of the activity budgets) of the rehabilitated individuals at the end of the 3.5-month

rehabilitation period would be similar to published behavioural data on wild, free-ranging bearded capuchin monkeys.

2. Time spent in foraging behaviour would increase when comparing the start and the end of the rehabilitation period.
3. Time spent in affiliative behaviour would increase if found as less than approximately 4% of the total activity budget or remain stable if found as approximately 4%, when comparing the start and the end of the rehabilitation period.
4. Behavioural diversity indexes would increase when comparing the start and the end of the rehabilitation period.
5. The rate per minute of agonistic behaviours would decrease if found over 0.6 or remain stable if found as less than 0.60 per minute of observation hour.
6. Time spent in behaviours potentially indicative of stress (BPIS) would decrease if found as over 3% of the total activity budget or remain stable if found as less than 3%, when comparing the start and the end of the rehabilitation period.
7. The monkeys would spend at least 40% of their total activity budget in the highest tier of their enclosure by the end of the rehabilitation period.
8. The monkeys would spend no more than 5% of their total activity budget in the lowest tier of their enclosure (i.e., on the ground) by the end of the rehabilitation period.
9. Adult females would spend more time grooming others than adult males by the end of the rehabilitation period.
10. Adult males would spend more time engaged in social play and vigilance than adult females by the end of the rehabilitation period.
11. Adult monkeys would spend more time engaged in vigilance than juveniles by the end of the rehabilitation period.
12. Juveniles would spend more time in social play and environmental manipulation than adults by the end of the rehabilitation period.
13. There would not be significant differences in the amounts of time engaged in behaviours potentially indicative of stress (BPIS) when comparing the three conditions related to husbandry and research practices as explained in the second aim of this Chapter.

4.2. Methodology

4.2.1. Ethical approval

The project received ethical approval by the Animal Welfare and Ethical Review Body of the University of Bristol in January 2019 (reference number UB/18/087) and complied with the NC3Rs Guidelines for Primate Accommodation, Care and Use (NC3Rs, 2017).

4.2.2. Study site and subjects

The study was performed in a government wildlife rescue centre located in Rio Grande do Norte, Brazil (CETAS–RN). This rescue centre receives primates and other wild animals that have been rescued/confiscated from the pet trade in Northeast Brazil. Sixteen capuchin monkeys (*Sapajus sp.*) were part of the study (see below). Most of these animals were born in the wild, captured and sold as pets illegally. Refer to Chapter 2: General Methodology for a full description of the origin of the individual as well as the rehabilitation programme carried out in the rescue centre. A summary of the individuals considered in this Chapter as well as the data available for analysis can be seen in Table 4.1.

Table 4.1

Individuals that were part of the study (n = 16)

	Name	Group (start of the study)	Group (end of the study)	Age category	Sex class	Origin
1	Dana (Da)	1	1	Adult	Female	Wild-born
2	Lombinho (Lm)	1	1	Adult	Male	Wild-born
3	Tino (Ti)	1	1	Adult	Male	Wild-born
4	Li (Li)	1	1	Juvenile	Female	Wild-born
5	Garrincha (Ga)	1	1	Juvenile	Male	Wild-born
6	V (V)	2	2	Adult	Female	Wild-born
7	Fúria (Fu)	2	2	Adult	Female	Wild-born
8	Galápagos (Gp)	2	2	Adult	Male	Wild-born
9	Café (Ca)	2	2	Adult	Male	Wild-born
10	Vitinho (Vi)	4	2	Juvenile	Male	Wild-born
11	Tapa (Ta)	3	3	Adult	Female	Wild-born
12	Arrebite (Ar)	3	3	Adult	Male	Wild-born
13	Tiquinho (Tq)	3	3	Juvenile	Male	Captive-born
14	Mãozinha (Mz)	3	3	Juvenile	Male	Captive-born
15	Capuccina (Cp)	4	4	Adult	Female	Wild-born
16	Linguinha (Lg)	4	4	Adult	Male	Wild-born

4.2.2.1. Social groups and changes

4.2.2.1.1. Group 1

This group was the only one that did not undergo any social changes throughout the study. It consisted of five individuals: an adult female (Dana), an adult male (Lombinho), a subadult male (Tino, which was counted as an adult as he was the only subadult in the study), and two juveniles; a female (Li) and a male (Garrincha). These individuals were not related and were all rescued or confiscated from the illegal pet trade. They lived in enclosure C at the start of the study (1 April 2019) and were moved to enclosure D on 22 May 2019. Both enclosures had the same dimensions and similar environmental enrichment structures and substrates (including a mobile bed); thus, this group did not experiment any social or environmental enrichment throughout the study except for the change of enclosure. For a full description of the enclosures, please refer to Chapter 2: General Methodology.

4.2.2.1.2. Group 2

At the start of the study, there were four unrelated adult monkeys in this group; two males, Café and Galápagos, and two females, V and Fúria. They were housed in enclosure D. On 20 May 2019, they were placed in enclosure A (which was approximately two times larger than enclosure D) along with Joana (an adult female) and Rabinha and Vitinho (a juvenile female and a juvenile male, respectively). All the monkeys from this group were rescued/confiscated from the illegal trade and were unrelated. This group stayed together in enclosure A from 20 May until the end of the study. A mobile bed was placed in this enclosure on that same day.

4.2.2.1.3. Group 3

At the start of the study, group 3 consisted of three adults (two females, Tapa and Joana, and a male, Arrebite) and two juvenile males, Mãozinha and Tiquinho. Tiquinho was Tapa’s and Arrebite’s son whilst Mãozinha was V’s (group 2) and Arrebite’s son. Hence, this was the only social group which included captive-born monkeys; however, the adults were wild-born and unrelated. The group occupied enclosure E, in which Joana was placed the day that the study started (1 April 2019). She spent most of the first week of the study hiding from the other monkeys inside the cage of the enclosure; hence, it was decided to exclude her observational data from this phase and to separate her and place her in enclosure A on 8 of April 2019. A mobile bed was placed in the enclosure of group 3 (enclosure E) on 3 June 2019. The remaining monkeys (Arrebite, Tapa, Mãozinha and Tiquinho) stayed in this enclosure throughout the whole study.

4.2.2.1.4. Group 4

This group consisted of two adults, Capuccina (female) and Linguinha (male); and two juveniles, Rabinha (female) and Vitinho (male). They were all unrelated and rescued/confiscated from the illegal trade and were housed in enclosure G (which was located in another building different to where enclosures A to F were located) at the start of the study.

Two days after the study started, Rabinha was taken out of the group for veterinary treatment (i.e. surgery for an umbilical hernia). After the surgery, she was placed alone in enclosure A for recovery. The rest of the group was moved to enclosure F on 12 April 2019 and stayed there together until Vitinho was moved to enclosure A on the 13 May 2019. Capuccina and Linguinha, the remaining members of group 4, were moved to enclosure B on the 17 June 2019. On 24 June 2019, two new (i.e., just received by the rescue centre) juvenile males (Fofo and Mazapán) were moved into this social group. They were all moved together to enclosure C on 1 July 2019, where they stayed until the end of the study.

4.2.2.1.5. Group 0

‘Group 0’ was a small group that was housed in enclosure A between 3 April and the 20 May 2019. At first, it was composed only by Rabinha (who was moved from group 4 on the 3 April 2019), then by Rabinha and Joana (moved from group 3 on the 8 April 2019), and finally by Rabinha, Joana and Vitinho (moved from group 4) from 13 to 20 May 2019. After this date, the monkeys from group 2 (Café, Galápagos, V and Fúria) were moved to this enclosure (A) and consequently the whole group was referred to as group 2 until the end of the study. Rabinha and Joana were excluded from analyses as their observations were incomplete; hence, the final sample size was 16 (n = 16).

4.2.3. Behavioural observations

I collected a total of 67 hours of behavioural observations for this study. The observations were divided into three phases: (1) baseline, (2) second, and (3) final, as seen in Table 4.2.

Table 4.2

Behavioural observation phases and dates

Phase		**Dates**	**Weeks since start of study**
1	Baseline	01/04/2019 – 10/04/2019	0
2	Second	06/05/2019 – 17/05/2019	3
3	Final	01/07/2019 – 17/07/2019	6

To record the behavioural data, I used Prim8 Mobile, an OS-based Android smartphone application designed to help in field behavioural data collection. I recorded the behaviour of the individuals using 5-min focal sampling; but space and substrate use as well as social proximity (i.e., to other individuals) were recorded every 30 seconds using scan sampling (see Section 2.5) (Altmann, 1974). The mean number of focal samplings per monkey was 44 (range 26 – 51). I performed the behavioural observations both in the morning, whilst the animals were contained in the inner cages for the caregivers to clean the enclosures (cage focal samplings or CFS), and in the afternoon, when the animals were free to move in their normal enclosures (enclosure focal samplings or EFS). All CFS were performed between 8:00 and 12:30, whereas all EFS were performed between 12:30 and 16:00. I followed a specific order to observe the individuals; each animal was assigned a number (i.e., 1 to 16) and on the first day of observation I observed animal number 1 first, then animal number 2, animal number 3 and so on. On the next day of observation I started with animal number 2 and animal number 1 was the last individual to be observed that day. I followed this pattern until all animals had been observed first and then repeated the pattern.

During the baseline phase, the number of cage and enclosure FS varied between individuals (CFS = 0 – 6; as a female juvenile was not placed in the inner cages during this time period), EFS = 7 – 10). Conversely, in the second and final phases, I aimed to observe all individuals for 6 CFS (total = 30 minutes) and 12 EFS (total = 60 minutes), for a total of 90 minutes per monkey, distributed through several days during each observational phase. Overall, the mean number of CFS per individual (i.e. for all phases) was 13 (range 2 – 18), whilst the mean number of EFS per individual was 32 (range 24 – 34). The ethogram utilised for the behavioural observations can be seen in Table 4.3. Behaviours registered as events are identified with an asterisk (*); all other behaviours were considered as states and measured in seconds.

Table 4.3

Ethogram utilised in the study

Behaviour	**Description**	**Reference**
Foraging		
Drinking water*	Water ingestion.	1
Eating	The individual approximates the food item to its mouth and ingests it.	1
Foraging	The animal moves in search of food, without manipulation or feeding during the sampling interval.	1, 2
Food manipulation	Manipulation of food in order to perform ingestion more efficiently (soften, open, hit, roll, etc.).	1, 2
Hunting insect*	Chasing and/or grabbing an insect to eat it.	1
Locomotion		
Vertical locomotion	Upwards or downwards movement in the environment, with limbs touching the trunks, mesh or other substrates. There is an end point for locomotion (the animal moves from A to B).	1
Horizontal locomotion	Horizontal locomotion in the environment, with limbs touching the ground / trunks. There is an end point for locomotion (the animal moves from A to B).	1, 2
Suspended locomotion	Horizontal locomotion at a certain height from the ground where the individual moves using 1 to 5 limbs whilst being suspended (normally in the "ceiling" of the enclosure but may be performed in trunks or other substrates).	1
Jumping*	Leaping from one surface or substrate to another.	1
Inactivity		
Inactivity with eyes closed	The individual is relatively static in a relaxed position. It may be lying down or sitting and usually has its arms and legs spread or hanging on a surface. Eyes are shut.	
Inactivity with eyes open	The individual is relatively static in a relaxed position. It may be lying down or sitting and usually has its arms and legs spread or hanging on a surface. Eyes are open.	
Affiliative		
Social play	The individual interacts with another by holding, grabbing, chasing, or biting without aggression.	1, 2
Mounting*	Individuals of the opposite sex or of the same sex mount on top of each other and there may be contact between their genital areas.	1, 2
Sexual behaviours	The animal opens and closes eyes repetitively, followed by contorted movement of the body, swinging from side to side (the behaviour is always directed to another individual). The arms are bent in his chest, his hands placed in the armpits. Usually presented by females in oestrus but can also occur outside the oestrus and by males. Manipulation of genitals or nipples may occur.	1, 2
Double masturbation	Simultaneous action of manipulating or stimulating one's genitals with another individual.	1
Affiliative display	The individual performs eyebrow lifting, contorted body movement, swinging from side to side, arms folded in his chest, showing teeth, to another individual.	1
Grooming	The animal manipulates the skin or fur of another individual with the hand or mouth.	1, 2
Receive grooming	Individuals have their hair slowly stimulated and/or touched by another individual with the hand or mouth.	1
Request grooming*	The individual approaches another who is self-grooming / resting / or in inactivity by lying down nearby in order to initiate social grooming (see receive grooming).	1
Observing individual	The animal observes attentively another individual from the same enclosure.	1
Observing individual(s) from other enclosure	The animal observes attentively one or more individuals from another enclosure through the mesh.	1
Interaction with individual(s) from other enclosure	The animal interacts with one or more individuals from another enclosure through the mesh. Sexual behaviour, food sharing and/or grooming may occur.	1
Agonistic		
Stare	The individual fixes its gaze on another individual i.e., establishes eye contact with another individual.	1
Withdrawal*	The individual withdraws with the approach of another.	1
Fight	The individual threatens (shouting and shows teeth) or is threatened, followed by beating, biting or attacks of another individual.	1, 2
Threat*	The individual threatens (if thrown towards the other, shouts and shows the teeth) or is threatened.	1, 2
Scream (at)	The animal begins to vocalise loudly, whether followed by any aggression performed by other individual or not.	
Environmental manipulation		
Manipulating the environment	The animal touches, moves, licks, bites, hits, digs or scrubs objects or part of the enclosure.	1, 2

Table 4.3 (continued)

Ethogram utilised in the study

Interaction with humans		
Threat to observer	Similar to incitement of aggression but directed to human.	1
Sexual display to observer	Similar to sexual behaviours but directed to human.	1, 2
Affiliative display to observer	Similar to affiliative display but directed to human.	
Vigilance		
Alarm	Acute vocalisation, followed by withdrawal, group dispersion and / or alert behaviour.	1
External vigilance	The individual is alert, eyes wide open, paying attention to the external environment (i.e. with its attention directed towards outside the enclosure).	1
Observing environment	The individual moves its head, looking and examining the environment and the other individuals of the group, without performing any other behaviour. In this case the individual is alert, not in a resting position, usually sitting on the floor or hanging from strings or the mesh.	1
Solitary play		
Solitary play	Pushes and hangs on ropes, twigs and strands alone. With limbs or tail.	1, 2
Behaviours potentially indicative of stress (BPIS)		
Self-directed behaviours		
Self-grooming	The individual repeatedly manipulates its hair with its hands, feet or mouth.	1, 2
Scratching*	The individual repeatedly scratches a part of its body.	1
Crouching	The individual holds itself with arms, feet and/or tail. Eyes are open but tend to look at the floor or to itself. It does not move or jump nor is it observing the environment. It differs from rest in that the animal is in a relaxed and slow position. It changes its position only to crouch again a few inches away.	1, 2
Self-clasping*	The individual holds itself with arms, feet and/or tail for a few seconds but does not stay in this position, as opposed to crouching.	1
Uncontrolled screaming	The animal vocalises loudly and without control with no apparent reason.	1
Masturbation	Stimulation and/or manipulation of the genital organs.	1, 2
Ingestion of urine, faeces, or sperm	Licking and eating/drinking urine, faeces and/or sperm. It can be of own origin or of another individual.	1, 2
Manipulation of urine, faeces, or sperm	Touching or rubbing urine, faeces and/or sperm in the environment or on its own body.	1
Urine washing	The individual urinates on the palms of its hands and rubs them on its feet and body hair.	1
Motor stereotypies		
Pacing	Walking or running covering the same circuit within the enclosure without an obvious goal for three or more repetitions.	2, 3
Shaking body	Shaking all the body for three or more repetitions.	1
Flipping	Performing three or more forward or backward somersaults.	3
Twirling*	Turning the body horizontally for three or more repetitions.	3
Swinging	Grasping a part of the mesh or another structure with one or two hands or feet, whilst moving in the exact pattern for three or more times.	3
Bouncing	Jumping up and down with a rigid posture for three or more repetitions.	3
Rocking	A rhythmic movement either side to side or forward and backward for at least three repetitions.	3
Head twirl*	Turning the head looking sideways and upwards for three or more repetitions.	1, 2
Self-injurious behaviour		
Self-injurious behaviours (SIB)	Chewing, biting, licking, and/or sucking a part of the body repeatedly; or pulling its own hair.	2, 4

(1) Nunes, 2017; (2) Ferreira et al. (2016); (3) Vandeleest et al. (2011); and (4) Honess et al. (2005).

Note that behaviours categorised as ‘affiliative’ were behaviours that could be interpreted (subjectively) as ‘friendly’ and were likely to produce cooperation and develop and maintain social (‘affiliative’) bonds between members of the social group in the wild (Sussman et al., 2005; Jasso del Toro and Nekaris, 2019). These may involve direct body contact, such as grooming (Fischer et al., 2017). However, certain behaviours do not require body contact to be considered as affiliative as long as they maintain their function of developing and/or maintaining social bonds (Jasso del Toro and Nekaris, 2019), such as affiliative display or sitting in close proximity (i.e., spatial proximity). In this study, I also considered ‘observing other individuals’ of the same and other social groups as *affiliative* because most groups could observe other individuals through the mesh of their enclosures, and it was considered as desirable for them to know each other and develop a social bond, if possible, before being released. Housing the animals close to each other was performed with the idea of allowing the groups to merge into one or two social groups after their reintroduction, as robust capuchin monkeys frequently live in medium-sized groups of approximately 10-17 individuals (Souvignet et al., 2019).

‘Agonistic’ behaviours, on the contrary, are likely to arise from competition of resources (e.g., food, sexual partners) among social group members and may either cause or try to reduce the possibility of injury (McGlone, 1986; Sussman et al., 2005). I included submissive (e.g., withdrawal) and aggressive (e.g., chasing) behaviours as ‘agonistic’, as proposed by Sussman et al. (2005).

4.2.3.1. Scan sampling for space use, substrate use, and social proximity

I recorded spatial and substrate use as well as social proximity using scan sampling every 30 seconds simultaneously to the enclosure focal samplings i.e., while I was focusing on one specific individual (Altmann, 1974). I performed a total of 5,188 scans throughout the three observational phases (mean per individual = 288, range = 214 – 316). I recorded the location of the monkey using the codes *G=ground, M=medium tier, H=high tier,* as well as the substrate s/he was utilising (*0=concrete, 1=fixed trunk, 2=mobile trunk, 3=vertical mesh, 4=horizontal mesh, 5=vertical cord/rope, 6=inner cage, 7=mobile bed*) and the name of the individual(s) s/he was in close proximity to (i.e., up to ~1m away from). Ground use was considered only when the monkey was directly on the concrete floor (i.e., at 0 metres); medium tier use was considered when the monkey was on top of any structure above the floor and below the top of the inner cage, which functioned as a ‘second concrete floor’ (above 0 m and below 1.5 m). Finally, I considered as high tier use when the monkey was in any structure located above the top of the inner cage (above 1.5 m and below the 2.8 m mesh ceiling) (see Figure 4.1).

Figure 4.1. Division of the enclosures utilised for recording spatial and substrate use.

4.2.4. Data analysis

Statistics were performed using SPSS 27 (SPSS Inc. Released 2020. SPSS for Windows, Version 27.0. Chicago, SPSS Inc). I used non-parametric statistics as the variables did not show normal distribution after performing Schapiro-Wilk analyses. Hence, the variables are shown as median (Mdn) and interquartile range (IQ).

4.2.4.1. Calculation of activity budgets

I organised the observational data according to the ten behavioural categories obtained from the ethogram (see Section 2.3): *foraging*, *locomotion*, *affiliative behaviour*, *agonistic behaviour*, *environmental manipulation*, *vigilance*, *solitary play*, *inactivity*, *human interaction*, and *behaviours potentially indicative of stress* (*BPIS*). To calculate the activity budgets (which were calculated per monkey), I added the time spent in every behaviour that belonged to the behavioural category of interest; for example, to calculate *time spent in locomotion* I followed the equation:

Total time spent in locomotion = *time spent in vertical locomotion* (seconds) + *time spent in horizontal locomotion* (seconds) + *time spent in suspended locomotion* (seconds)

Then, a percentage was calculated as follows:

$$\% = \frac{x}{y}\,(100)$$

In which *x* is the time observed in the behavioural category of interest (e.g., locomotion) and *y* is the total time observed minus the time the monkey was not visible, if any. This was performed for each behavioural category and observational phase: baseline (B), second (S), and final (F).

4.2.4.2. Behavioural events: calculation of rates per minute

Several behaviours were registered as behavioural events (e.g., drinking); thus, these behaviours were not included in the activity budgets as I did not have their duration. However, I calculated a rate per minute for each behaviour that belonged to this category (and for every observational phase) as follows:

$$Rate\ per\ minute = \frac{behavioural\ events\ observed\ (n)}{total\ time\ observed\ (minutes)}$$

4.2.4.3. Space and substrate use and social proximity

I measured space and substrate use as well as social proximity using scan sampling every 30 seconds. Therefore, I estimated the time spent in each of these by calculating the percentage of scans in performing the behaviours; in other words, using each level, substrate or being in social proximity.

This was performed as follows:

$$\% = \frac{scans\ observed\ performing\ the\ behaviour\ (n)}{total\ scans\ observed\ (n)}\ (100)$$

4.2.4.4. Analysis of changes in behaviour across time

I performed non-parametric Friedman tests to analyse the changes in behaviour between observational phases. I did this for the 10 behavioural categories considered in the ethogram: *foraging, locomotion, affiliative behaviour, agonistic behaviour, environmental manipulation, vigilance, solitary play, inactivity, human interaction,* and *behaviours potentially indicative of stress* (*BPIS*). If there were significant changes across time, I performed Wilcoxon signed-rank tests to identify which specific observational phase(s) showed changes (e.g., baseline vs second phase or baseline vs final phase). Furthermore, when there were significant changes, I analysed subcategories of the behavioural category separately to identify which specific behaviours changed (e.g., if changes in foraging occurred in the behaviours foraging (looking for food), eating or food manipulation).

4.2.4.5. Analysis of age and sex class differences

I used Mann-Whitney U tests to analyse the differences between sex class categories (males, n = 10 and females, n = 6) and age categories (juveniles, n = 5 and adults, n = 11).

4.2.4.6. Behavioural diversity indexes

I calculated the Shannon H indexes using the programme Past version 4.07. I considered the behavioural categories from genus normative behaviours (GNB): *foraging, locomotion, affiliative behaviour*, *agonistic behaviour*, *environmental manipulation*, *vigilance* and *solitary play* and excluded the categories *inactivity, human interaction*, and *behaviours potentially indicative of stress* (*BPIS*) (see ethogram, Section 2.3) (Miller et al., 2020).

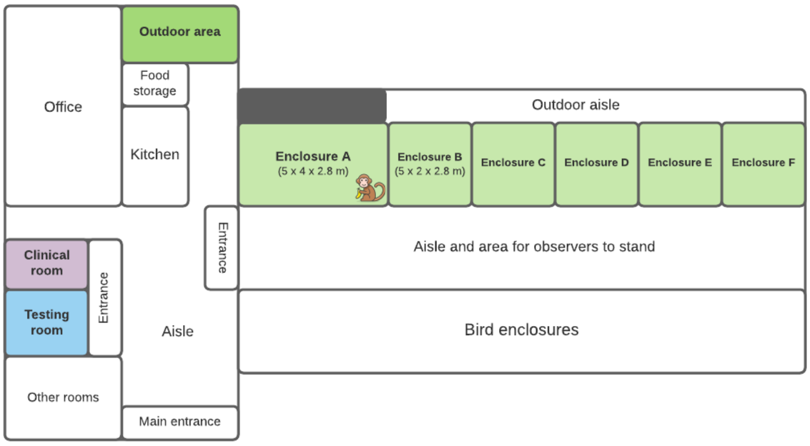
I performed a one sample t-test to evaluate if the difference between the final (F) and baseline (B) behavioural diversity indexes (F-B score) was significantly different from zero, as suggested by Metter (2008). A two-way analysis of variance (ANOVA) was used to evaluate differences related to age and/or sex class related to the Shannon H indexes. Furthermore, I performed a Pearson correlation to analyse the association between behavioural diversity and the exhibition of behaviours potentially indicative of stress (BPIS) (Miller et al., 2020).

4.2.4.7. Effect of husbandry and research practices on the exhibition of BPIS

In the rescue centre where this study was performed (CETAS–RN), the monkeys' enclosures were cleaned once per day in the early morning. During these procedure, the monkeys were placed in the concrete inner cage of their enclosures (see Figure 4.2(4) below) whilst the caregivers entered to clean. Moreover, various research projects were carried out with the monkeys in CETAS–RN. Several of these projects included performing individual behavioural tests with the monkeys; thus, the monkeys were placed in individual cages (1 m^2 cages) and transferred to a testing room (see Figure 4.3 below) several times per month.

Figure 4.2. Front (large) and back (small) views of enclosures C, D, E and F. (1) Metallic mesh ceiling, (2) concrete side wall, (3) back wall made of metallic mesh and concrete, (4) concrete inner cage (entrance seen in the small picture) and (5) concrete floor. Environmental enrichment structures can also be seen.

Figure 4.3. Floor plan of building I in CETAS–RN rescue centre.



To analyse the behavioural effects of these practices (if any), I analysed the exhibition of BPIS in the three different conditions: (1) when the animals were in their group enclosures (see Section 2.3), (2) when the animals were placed in the inner cages whilst the caregivers cleaned the enclosures for approximately 30 minutes (see paragraph above), and (3) when the monkeys were placed in individual cages and transferred to the testing room (i.e., when they were in social isolation) for up to five minutes during the behavioural tests performed for this dissertation (see Chapter 5). This was performed for a total of approximately 65 minutes per monkey for the complete observational period. For conditions 1 (group enclosures) and 2 (concrete inner cages), I calculated the time spent in BPIS separately for each of the three observational phases and then calculated a mean. For condition 3 (individual behavioural tests), I calculated the time spent in BPIS separately for each trial (i.e., behavioural test) and then calculated a mean. Finally, I performed a non-parametric Friedman test between the means obtained in each of the three conditions to analyse if there were significant differences in time spent in BPIS between them.

4.3. Results

4.3.1. Activity budgets

The percentage of time calculated for each behavioural category and phase can be seen in Table 4.4.

Table 4.4

Percentage of time (%) spent in each of the behavioural categories (median and interquartile range) across all individuals (n = 16)

Behavioural category	**Baseline**		**Second**		**Final**	
	Median	**IQ**	**Median**	**IQ**	**Median**	**IQ**
Foraging	34.71	12.55	45.84	8.40	32.17	12.55
Locomotion	18.97	10.40	16.77	10.19	16.59	10.66
Affiliative	1.45	3.59	5.76	9.43	9.57	21.85
Agonistic	0.00	0.00	0.00	0.00	0.00	0.00
Inactivity	0.00	0.91	0.68	1.99	2.03	3.41
Vigilance	20.76	18.43	15.75	19.61	22.25	9.29
Environmental manipulation	2.24	4.85	2.40	4.12	5.83	5.94
Solitary play	0.00	0.08	0.00	0.00	0.00	0.98
Interaction with humans	0.00	0.13	0.00	0.00	0.00	0.00
BPIS	1.45	2.45	1.60	3.97	2.18	5.36

4.3.1.1. Foraging

Overall, time engaged in foraging behaviours changed significantly across time (*Mdn* baseline = 34.71, second = 45.84, final = 32.17; $X^2_F = 18.500$, $p < 0.001$). These behaviours increased from the baseline to the second phase ($Z = -3.258$, $p < 0.001$) and then decreased from the second to the final phase ($Z = -3.258$, $p = 0.002$). Thus, there were no significant differences between the baseline and the final phase ($p = 0.918$).

The behavioural category *foraging* was divided into the three subcategories established in the ethogram: (a) *foraging–searching for food*, as in searching actively for food items, (b) *manipulating food*, which included grabbing, hitting, rubbing, throwing, squeezing, extracting, or any other type of manipulation with hands or feet that the monkeys performed with food items, and (c) *eating* i.e., ingesting food items. Percentage of time spent *foraging–searching for food* increased significantly from the baseline to the second phase ($Z = -2.095$ $p = 0.036$) and from the second to the final phase ($Z = -2.999$, $p = 0.004$). However, this effect was not observed when comparing the baseline and the final phase ($p = 0.196$). Furthermore, there was a significant increase in the percentage of time spent *eating* between the baseline and the second phase ($Z = -3.051$, $p = 0.002$) and between the second and final phases ($Z = -2.947$, $p = 0.003$) but no significant differences between the baseline and final phase ($p = 0.234$). Finally, time spent *manipulating food items* did not change significantly over time ($p = 0.074$). Foraging behaviours categorised as events (i.e., *drinking* and *hunting insects*) did not change significantly among observational phases ($p = 0.518$).

Figure 4.3. Box plot showing the changes between observational phases (baseline, B; second, S; final, F) in overall foraging behaviours.

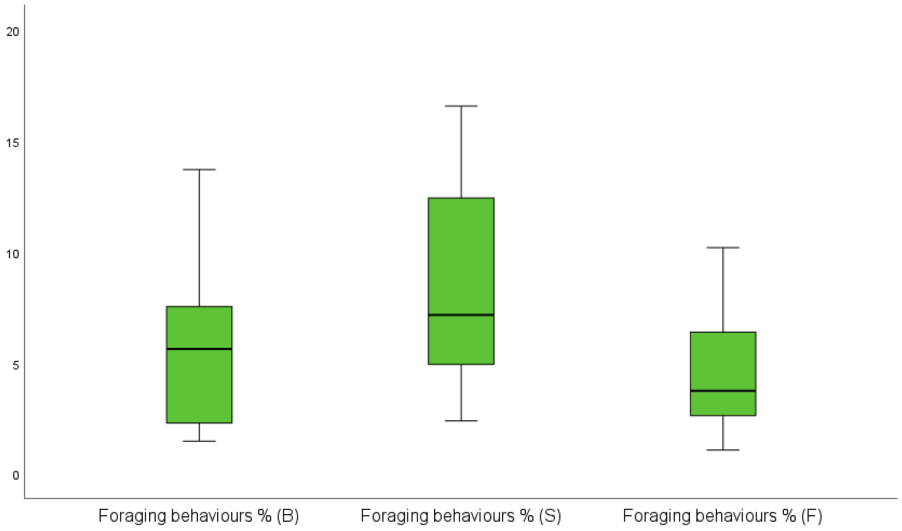
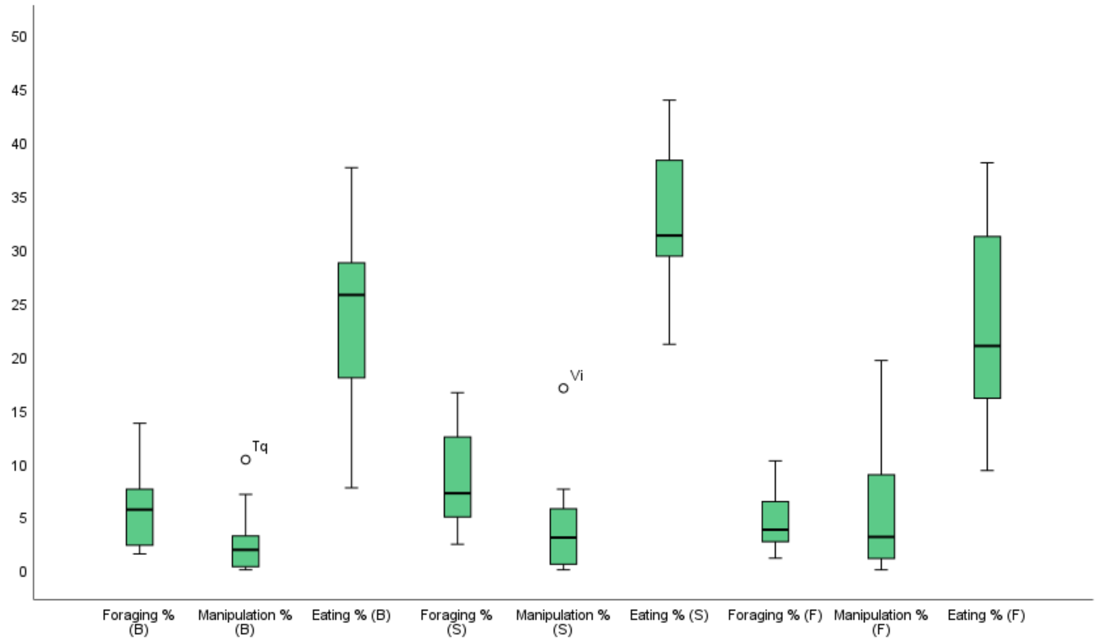


Figure 4.4. Box plot showing the changes between observational phases (baseline, B; second, S; final, F) in subcategories of foraging behaviours.



Age and sex class did not influence foraging behaviours during any of the observational phases ($p \geq 0.126$).

4.3.1.2. Locomotion

Time engaged in locomotor behaviours did not change significantly between observational phases (*Mdn* baseline = 18.97, second = 16.77, final = 16.59; $p = 0.472$). However, *jumping* (the only behaviour considered as event from this category and measured in rate per minute) changed significantly across time ($X_2 = 6.125$, $p = 0.047$). This behaviour increased significantly only between the second and the final phase ($Z = -3.009$, p = 0.003). There were no significant differences related to age or sex class regarding time spent in locomotion ($p \geq 0.104$). Nevertheless, the behaviour *jumping* showed significant differences related to age, with juveniles performing this behaviour significantly more than adults during the final phase ($U = 9.000$, $Z = -2.102$, $p = 0.036$; other phases, $p \geq 0.066$). This behaviour did not show a significant difference related to sex class in any of the observational phases ($p \geq 0.826$).

4.3.1.3. Affiliative

There was a significant increase in time spent in affiliative behaviours across observational phases ($X^2{}_F(2) = 10.889$, $p = 0.004$; *Mdn* baseline = 1.45, second = 5.76, final = 9.57). There were no significant changes between the baseline and the second phase ($p = 0.088$) nor between the second and the final phase ($p = 0.148$); however, there was a significant increase between the baseline and the final phase (Z = -3.258, p = 0.001). When analysing behaviours from this category separately, *social play* increased significantly across observational phases ($X^2_F(2)$ = 11.400, p = 0.003), with no significant changes between the baseline and second phases (p = 0.091) and between the baseline and final phase (p = 0.091) but with a significant increase between the second and the final phase (Z = -2.578, p = 0.010). There was a tendency towards a significant increase in time spent *grooming* (i.e., grooming another monkey) across observational phases ($X^2_F(2)$ = 5.722, p = 0.057), with a significant increase between the baseline and the second phase (Z = -1.988, p = 0.047) and between the baseline and the final phase (Z = -2.197, p = 0.028) and no significant differences between the second and the final phase (p = 0.953). Conversely, time spent *receiving grooming* did not change across observational phases (p = 0.388). Time spent performing *affiliative display*, however, increased significantly across time ($X^2_F(2)$ = 14.000, p = 0.001), with no significant changes between the baseline and second phases (p = 1.000) and significant increases between the second and final (Z = -2.366, p = 0.018) and baseline and final phases (Z = -2.366, p = 0.018). The remaining affiliative behaviours (*sexual behaviours*, *observing another individual*, *interaction with individuals from another enclosure*) did not change significantly across time ($p \geq 0.311$). Time spent *observing individuals from another enclosure* showed a tendency towards a significant change across time ($X^2_F(2)$ = 5.660, p = 0.057); nonetheless, when analysing separately between phases, there were no significant differences between them ($p \geq 0.225$).

Overall, time spent in affiliative behaviours was not significantly different between females and males ($p \geq 0.147$). Time spent in affiliative behaviours was significantly different between juveniles and adults during the baseline (U = 2.000, Z = -2.897, p = 0.004) and final (U = 10.000, Z = -1.983, p = 0.047) phases, with juveniles spending significantly more time than adults engaged in affiliative behaviours. These differences, however, were not observed during the second phase (p = 0.583).

Figure 4.5. Box plot showing the changes between observational phases (baseline, B; second, S; final, F) in overall affiliative behaviours.

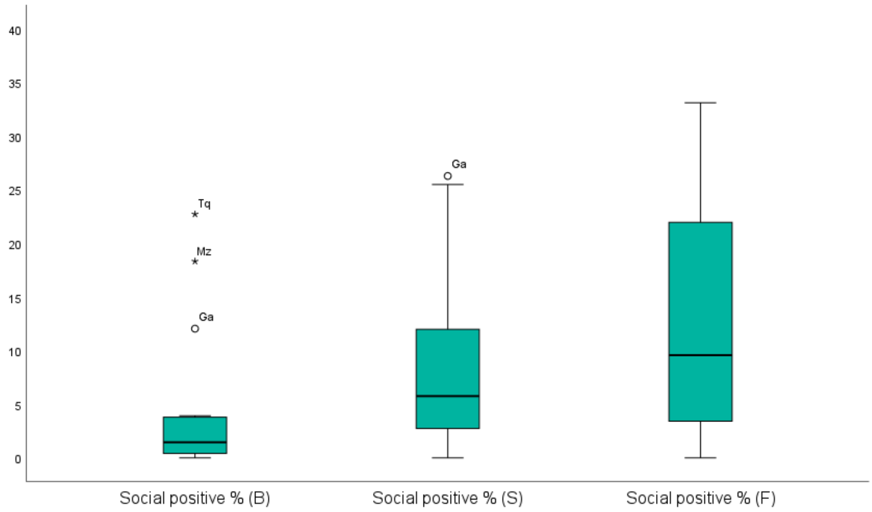
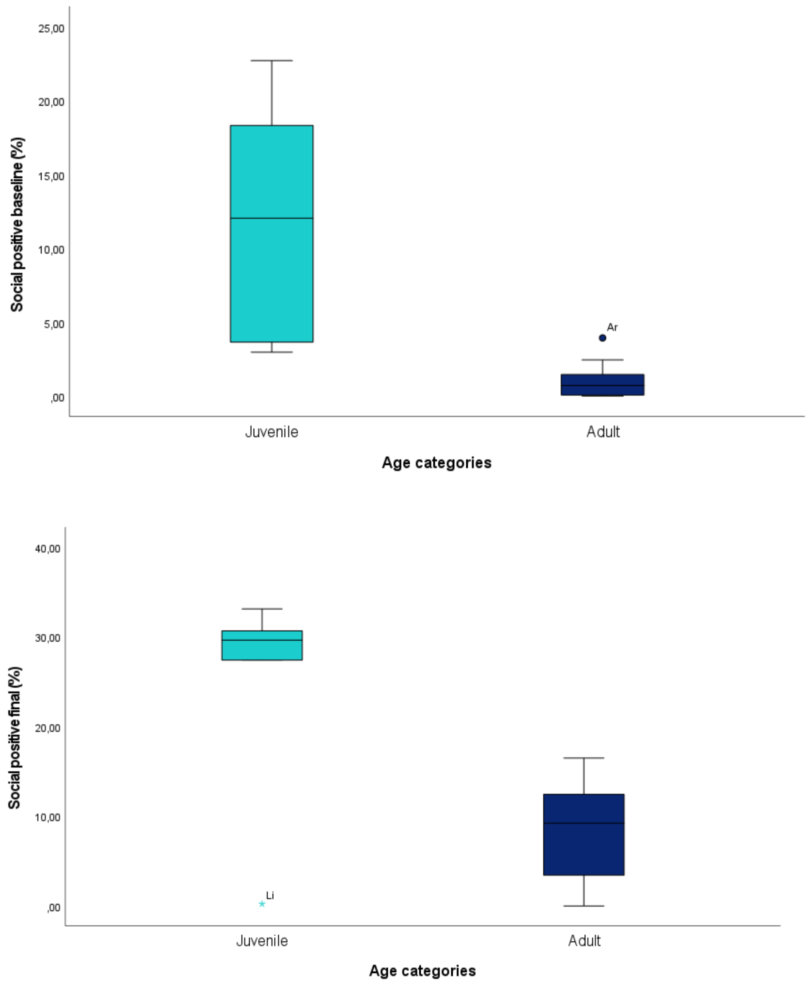


Figure 4.6. Box plots showing the significant difference between juveniles and adults in the exhibition of overall affiliative behaviours during the (1) baseline and (2) final observational phases.

(1)

(2)



4.3.1.4. Agonistic

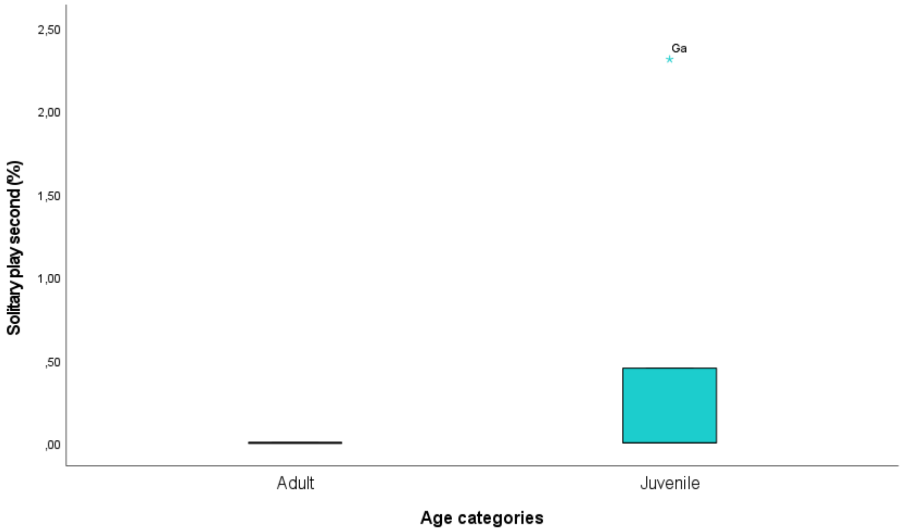
Time spent in agonistic behaviours did not change significantly across time and they were rarely observed (*Mdn* baseline = 0.00, second = 0.00, final = 0.00, p = 0.607). Nonetheless, there was a significant increase in the rate per minute of agonistic behaviours across time ($X^2_F(2) = 8.000$, p = 0.018) with a significant difference observed between the second (*Mdn* = 0.00) and the final (*Mdn* = 0.00) phases (Z = -2.121, p = 0.034), a tendency towards a significant difference between the baseline (*Mdn* = 0.00) and the final phase (Z = -1.890, p = 0.059) and no significant difference between the baseline and second phases (p = 1.000). However, no fights were observed during the whole observation period; the agonistic behaviours observed did not involve physical contact (*stare*, *scream at*, *threat*, and *withdrawal*). These behaviours were not influenced by age or sex class ($p \geq 0.197$).

4.3.1.5. Solitary play

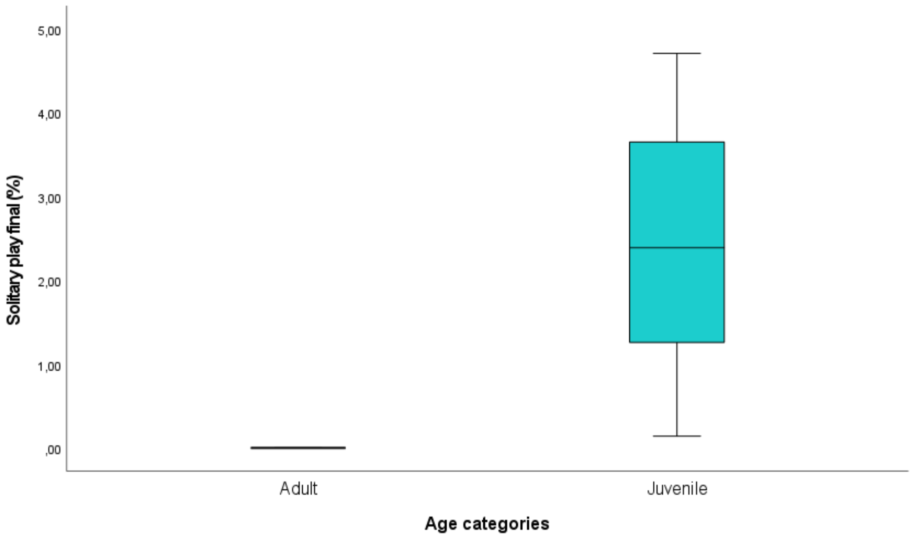
The percentage of time spent in solitary play did not change significantly across time (*Mdn* baseline = 0.00, second = 0.00, final = 0.00, p = 0.115). This behaviour was not affected by sex class ($p \geq 0.235$); however, juveniles dedicated significantly more time to this activity than adults during the second (U = 16.500, Z = -2.166, p = 0.030) and final phases (U = 0.000, Z = -3.788, p <0.001). This was not observed during the baseline phase (p = 0.113).

Figure 4.7. Box plots showing the significant difference between juveniles and adults in the exhibition of solitary play during the (1) second and (2) final observational phases.

(1)



(2)



4.3.1.6. Environmental manipulation

Time spent *manipulating the environment* did not change significantly across time (*Mdn* baseline = 2.24, second = 2.40, final = 5.83, $p = 0.156$). There were no significant differences between females and males regarding time spent in this category during the baseline and second phases ($p \geq 0.212$); however, there was a tendency towards a significant difference during the final phase ($U = 12.000$, $Z = -1.952$, $p = 0.051$). Furthermore, juveniles spent significantly more time manipulating the environment than adults in all phases (baseline $U = 9.000$, $Z = -2.102$, $p = 0.036$; second $U = 8.500$, $Z = -2.154$, $p = 0.031$; final $U = 7.000$, $Z = -2.322$, $p = 0.020$).

Figure 4.8. Box plot showing the significant difference between females and males in the exhibition of environmental manipulation during the final observational phase.

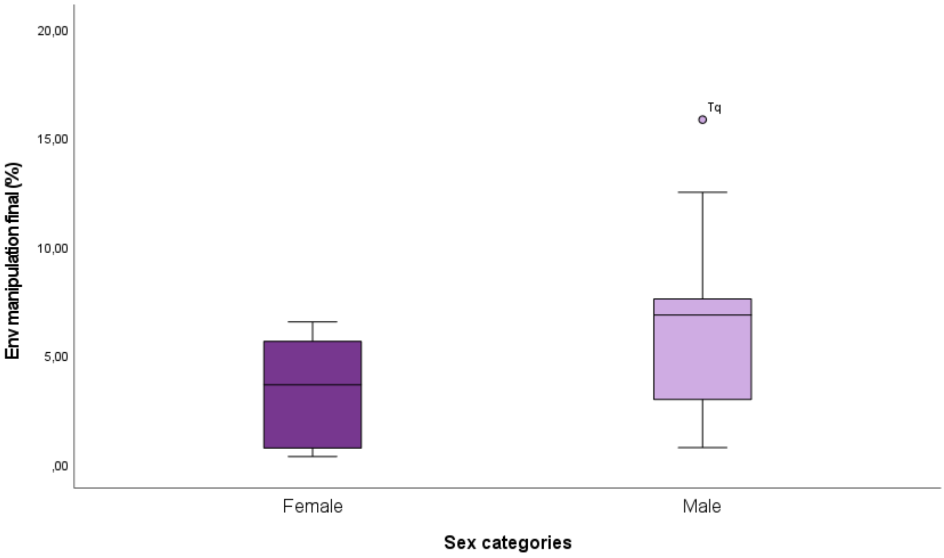
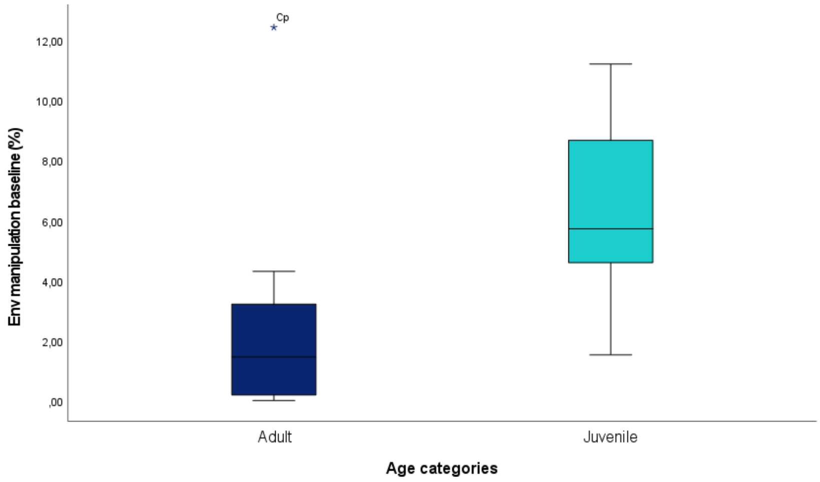
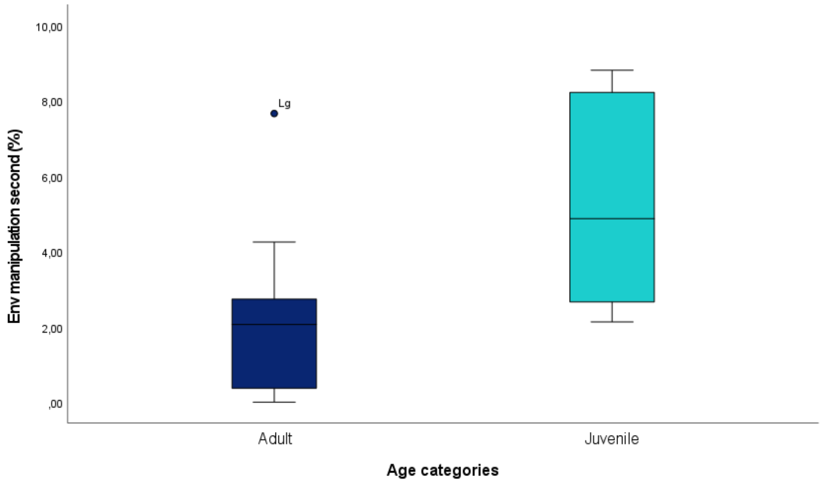


Figure 4.9. Box plots showing the significant difference between juveniles and adults in the exhibition of environmental manipulation during the (1) baseline, (2) second and (3) final observational phases.

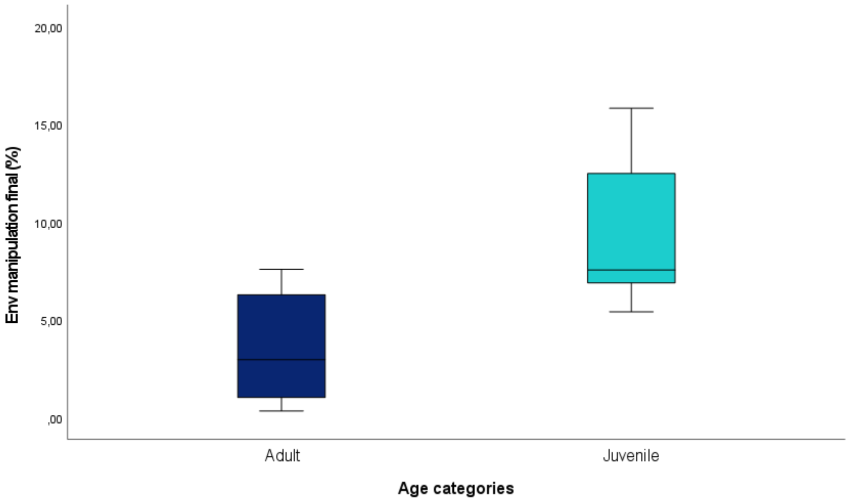
(1)



(2)



(3)

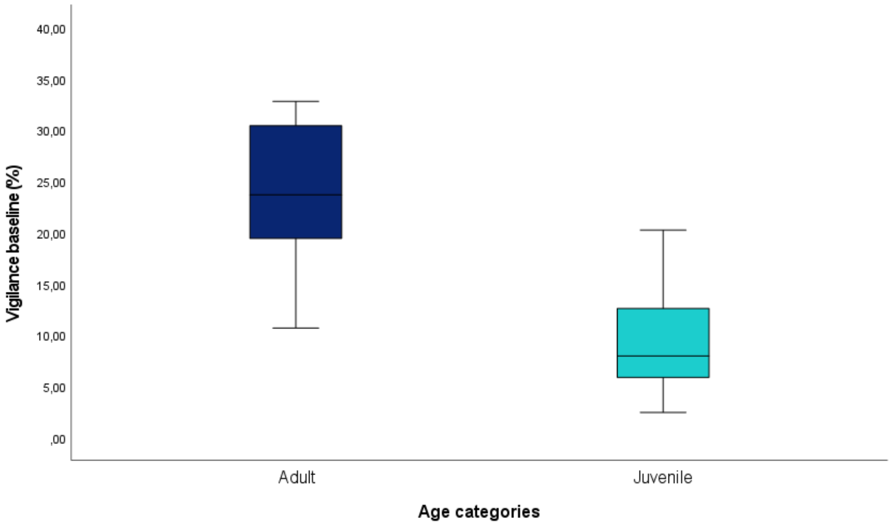


4.3.1.7. Vigilance

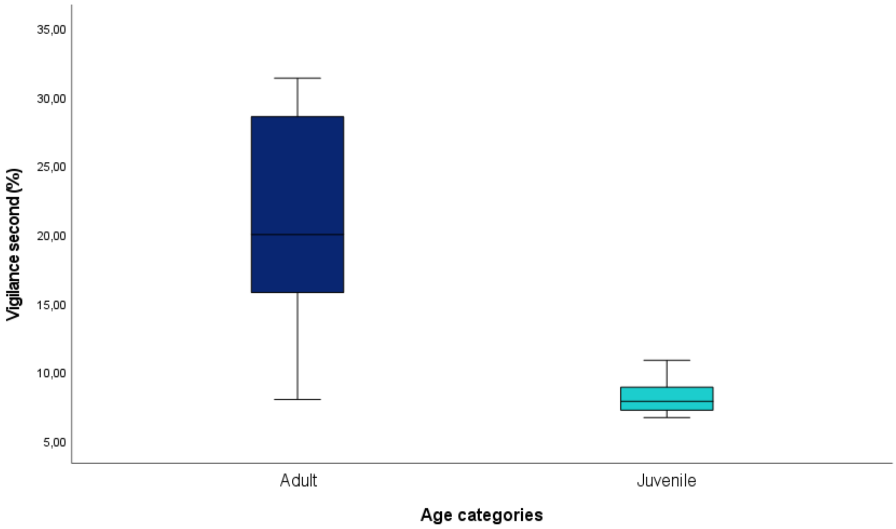
Overall, time engaged in *vigilance* behaviours did not change significantly over time (*Mdn* baseline = 20.76, second = 15.75, final = 22.25, $p = 0.939$). Moreover, adults spent significantly more time in vigilance behaviours than juveniles during the baseline ($U = 4.000$, $Z = -2.662$, $p = 0.008$), second ($U = 2.000$, $Z = -2.889$, $p = 0.004$) and final ($U = 10.000$, $Z = -1.983$, $p = 0.047$) phases. No differences related to sex class were found ($p \geq 0.448$).

Figure 4.10. Box plots showing the significant difference between juveniles and adults in the exhibition of vigilance behaviours during the (1) baseline, (2) second and (3) final observational phases.

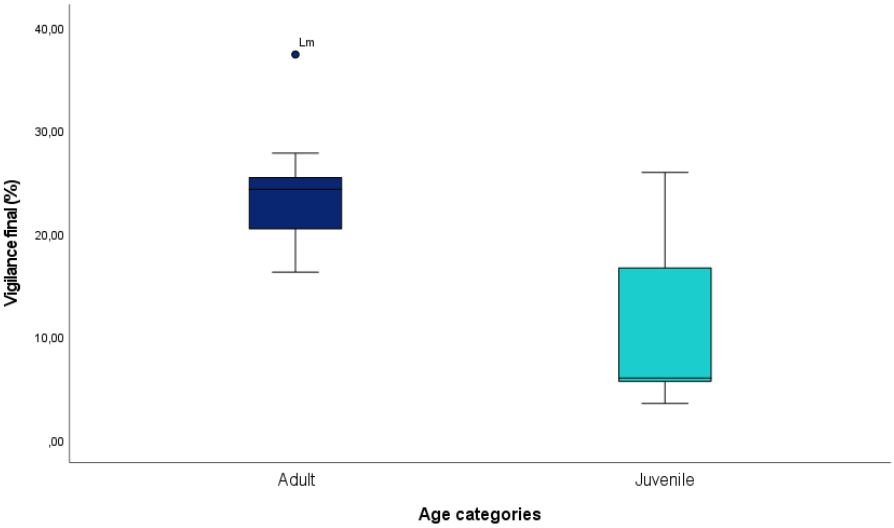
(1)



(2)



(3)



4.3.1.8. Inactivity

Time spent in *inactivity* increased significantly across time ($X^2_F(2) = 6.310$, $p = 0.043$; Mdn baseline = 0.00, second = 0.68, final = 2.03), with no significant differences between the baseline and the second ($p = 0.272$) nor between the second and the final phases ($p = 0.379$). However, there was a tendency towards a significant increase in the final phase when compared to the baseline ($Z = -1.852$, $p = 0.064$). When analysed subcategories separately, time spent in waking inactivity or *inactive with eyes open* increased significantly over time ($X^2_F(2) = 6.310$, $p = 0.043$) with a tendency to significant increase between the baseline and the final phase ($Z = -1.852$, $p = 0.064$) and no significant changes between the baseline and second phase and between the second and final phases ($p \geq 0.272$). Conversely, time spent *inactive with eyes closed* remained stable ($p = 0.867$). There were no significant differences related to sex class or age in time spent *inactive with eyes open* or *inactive with eyes closed* ($p \geq 0.073$).

Figure 4.11. Box plot showing the change between observational phases (baseline, B; second, S; final, F) in time spent in overall inactivity.

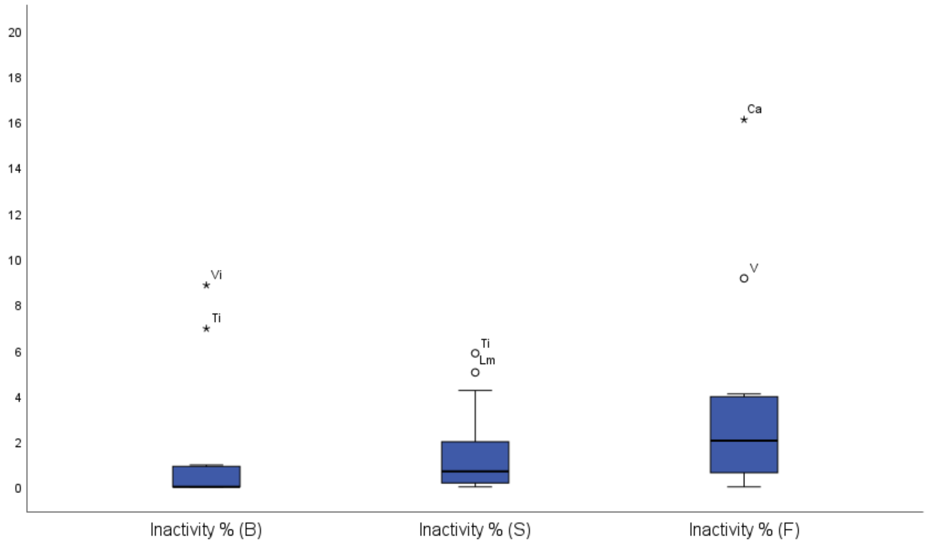
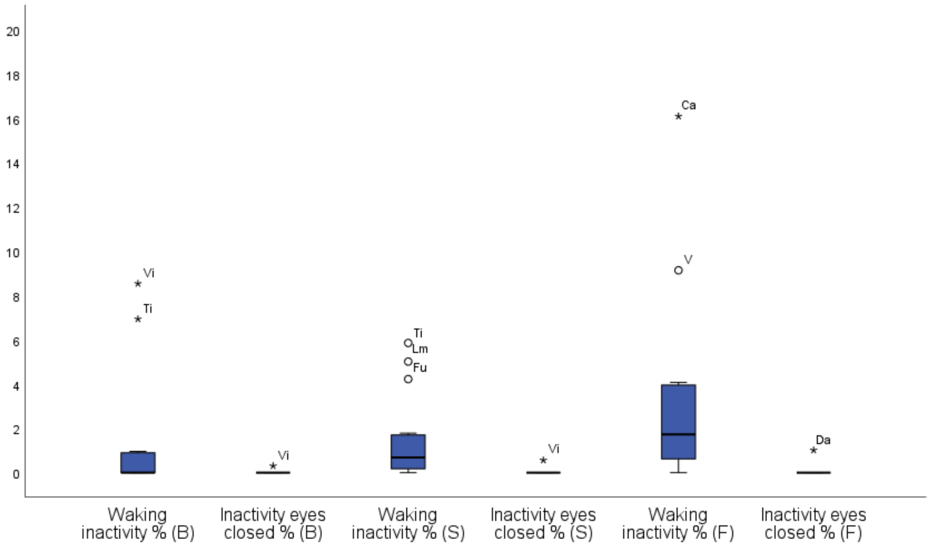


Figure 4.12. Box plot showing the change between observational phases (baseline, B; second, S; final, F) in time spent in waking inactivity and inactivity with eyes closed.

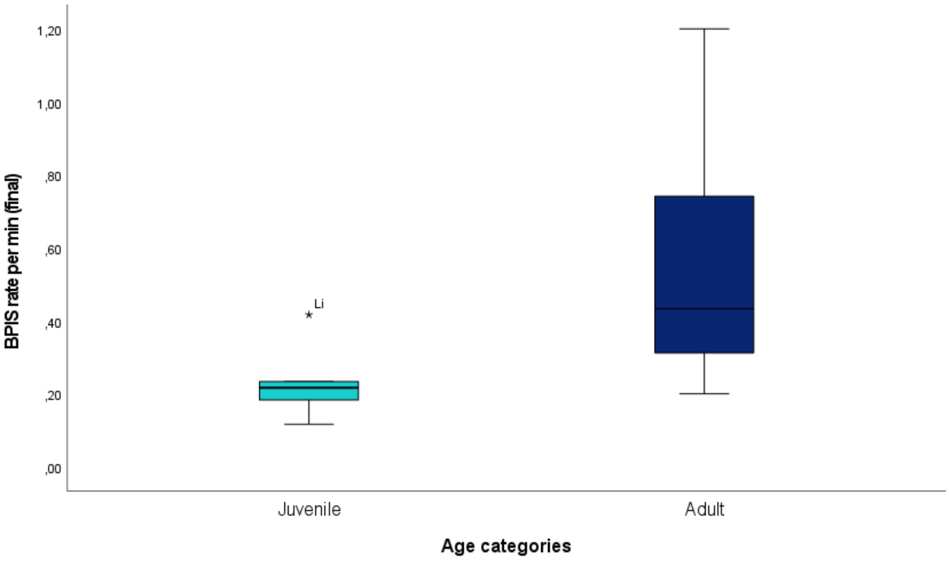


4.3.1.9. Behaviours potentially indicative of stress (BPIS)

Overall, behavioural states related to this category i.e., self-directed behaviours and motor stereotypies did not change significantly over time (*Mdn* baseline = 1.45, second = 1.60, final = 2.18, $p = 0.740$). However, when analysing separately, self-directed behavioural states (*self-grooming*, *crouching* and *uncontrolled screaming*) did not change over time ($p = 0.505$) whereas motor stereotypies (*pacing*, *rocking* and *shaking body*) significantly increased over time ($X^2_F(2) = 7.400$, $p = 0.025$). This effect was observed only between the baseline and the final phase ($Z = -2.521$, $p = 0.012$) and not between the baseline and second phases nor between the second and final phases ($p \geq 0.273$). Behavioural events related to BPIS, which also included several self-directed behaviours (*scratching* and *self-clasping*) and motor stereotypies (*head twirling* and *twirling*), did not change significantly across time ($p \geq 0.241$).

Furthermore, there were no differences related to sex class or age in BPIS-related behavioural states ($p \geq 0.234$). However, behavioural events related to BPIS were performed significantly more by adults than by juveniles during the final observational phase ($U = 8.500$, $Z = -2.154$, $p = 0.031$) albeit they were not influenced by age or sex class in the other phases ($p \geq 0.125$).

Figure 4.13. Box plot showing the significant difference between juveniles and adults in the exhibition of BPIS measured in rate per during the final observational phase.



4.3.1.10. Interaction with humans

Overall, behaviours related to *human interaction* were rarely observed (*Mdn* baseline = 0.00, second = 0.00, final = 0.00). However, there was a tendency towards a significant decrease over time ($X^2_F(2) = 5.818$, $p = 0.055$), with a significant difference observed only between the baseline and the final phase ($Z = -2.023$, $p = 0.043$; other phases $p \geq 0.109$). Moreover, behaviours from this category were divided into two subcategories during the behavioural observations: *affiliative* (i.e., *sexual/affiliative display*) and *aggressive* (i.e., *threat* and *scream at*). Hence, I analysed these subcategories separately as well. Affiliative behaviours towards humans did not increase or decrease significantly between phases ($p = 0.202$), whilst aggressive behaviour directed towards humans decreased over time ($X^2_F(2) = 8.000$, $p = 0.018$). This effect was observed between the baseline and the final phase ($Z = -1.826$, $p = 0.068$) but not between the baseline and second nor between the second and final phases ($p \geq 0.109$). No differences related to age nor sex class were found in behaviours directed towards humans ($p \geq 0.203$).

4.3.2. Spatial and substrate use

Results related to spatial and substrate use of each observational phase can be seen in Table 4.5.

Table 4.5

Spatial and substrate use calculated as percentages (median and interquartile range across all individuals) per observational phase (n = 16)

Phase	**Baseline**		**Second**		**Final**	
	Median	IQ	Median	IQ	Median	IQ
Space use						
Ground tier	16.95	16.87	18.41	27.23	14.23	18.68
Medium tier	32.33	14.23	36.61	32.21	36.87	17.91
High tier	50.68	29.84	38.32	48.36	45.10	25.25
Substrate use						
Concrete	25.67	27.30	32.73	25.75	31.19	22.56
Fixed trunk	53.63	25.79	50.89	26.17	42.47	26.96
Mobile trunk	1.35	6.06	4.95	12.51	2.27	14.67
Vertical mesh	7.65	5.19	2.76	5.29	6.32	6.70
Horizontal mesh	0.57	1.70	0.44	1.82	0.89	1.97
Vertical cord	0.00	5.03	0.00	0.96	1.78	2.96
Cage	0.58	3.36	0.00	1.27	0.44	1.17
Mobile bed[1]	0.00	0.86	0.00	0.21	1.82	6.78

[1] Group 1 was the only group that had a mobile bed throughout the whole observational period. Groups 2 and 3 had the mobile bed placed in their enclosures several days after the second observational phase. Hence, mobile bed use in the baseline and second phases was calculated using data only from group 1, whilst mobile bed use in the final phase was calculated using the data from groups 1, 2 and 3. Group 4 did not have a mobile bed in their enclosure at any time and was not included in the analysis of this substrate.

Overall, space use did not change significantly across time. There were no significant differences in time spent in the *ground*, *medium*, or *high tiers* of the enclosures among observational phases ($p \geq 0.305$). Moreover, the use of the substrates *concrete*, *fixed trunks*, *mobile trunks*, *vertical mesh*, *horizontal mesh*, and *vertical ropes/cords* remained stable throughout the observational period ($p \geq 0.099$). I analysed mobile bed use separately for group 1 and groups 2 and 3, as group 1 was housed in an enclosure with a bed through all observational phases, whilst the enclosures of groups 2 and 3 were enriched with a mobile bed after the end of the second phase (3 and 17 days after, respectively; group 4 was excluded as their enclosure did not have a bed at any time). The individuals of group 1 did not increase nor decrease the use of the mobile bed across time, whilst the individuals from groups 2 and 3 showed a significant increase after the bed was placed in their enclosures ($Z = -2.670$, $p < 0.01$).

Space and substrate use were not influenced by sex class during any of the observational phases ($p \geq 0.081$). Conversely, age influenced space and substrate use. Adults spent more time using the highest level of the enclosures than juveniles albeit this was observed only during the final phase ($U = 13.000$, $Z = -2.154$, $p = 0.031$). Furthermore, juveniles spent more time using the horizontal mesh during the baseline ($U = 10.000$, $Z = -2.118$, $p = 0.034$) and final phase ($U = 10.000$, $Z = -1.961$, $p = 0.050$) as well as the vertical ropes/cords placed on their enclosures during the second observational phase ($U = 15.000$, $Z = -2.161$, $p = 0.031$). There were no significant differences between adult and juvenile monkeys regarding the use of the remaining substrates (concrete, mobile trunks, mobile beds, and the inner cages) and phases ($p \geq 0.072$).

Figure 4.14. Box plot showing the changes in spatial use (ground, medium and high tiers of the enclosures) between observational phases (baseline, B; second, S; final, F).

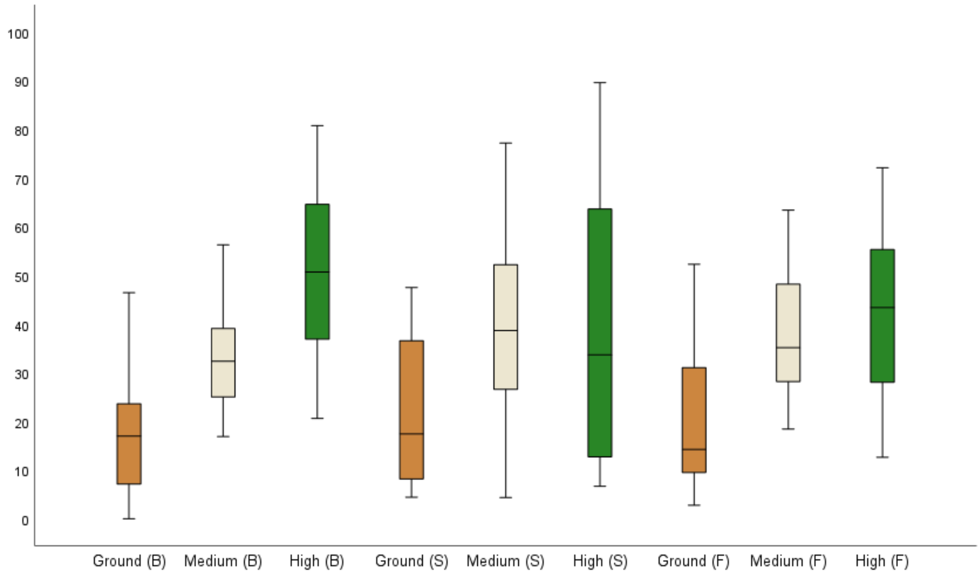
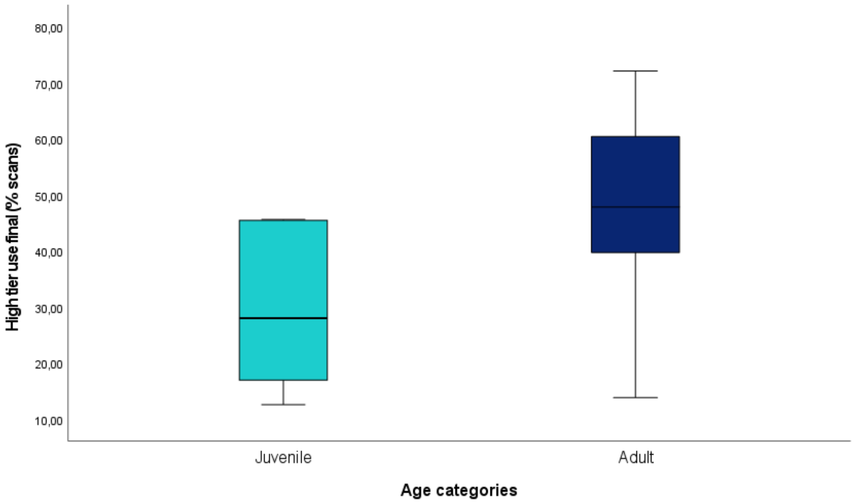
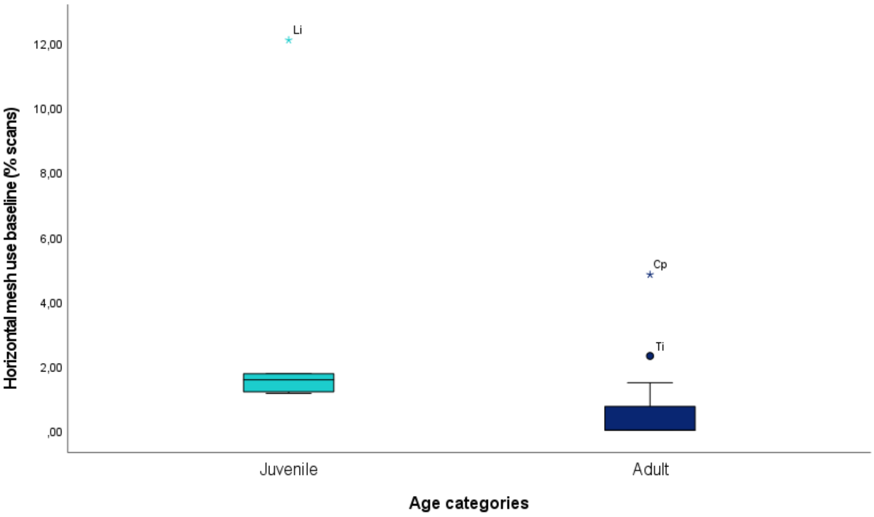


Figure 4.15. Box plots showing the significant difference between juveniles and adults in the use of the (a) high tier (final phase), (b) horizontal mesh (baseline), (c) horizontal mesh (final), and (d) ropes and cords (second phase) of their enclosures, measured in percentage of scans.

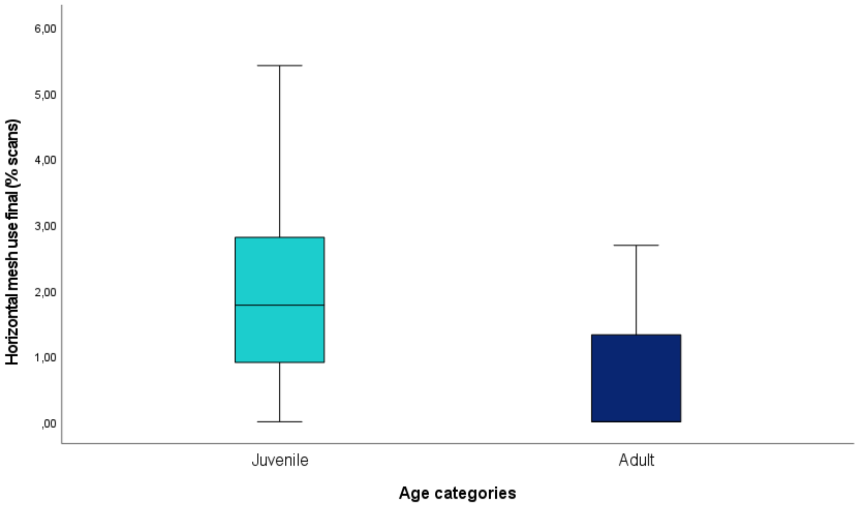
(a)



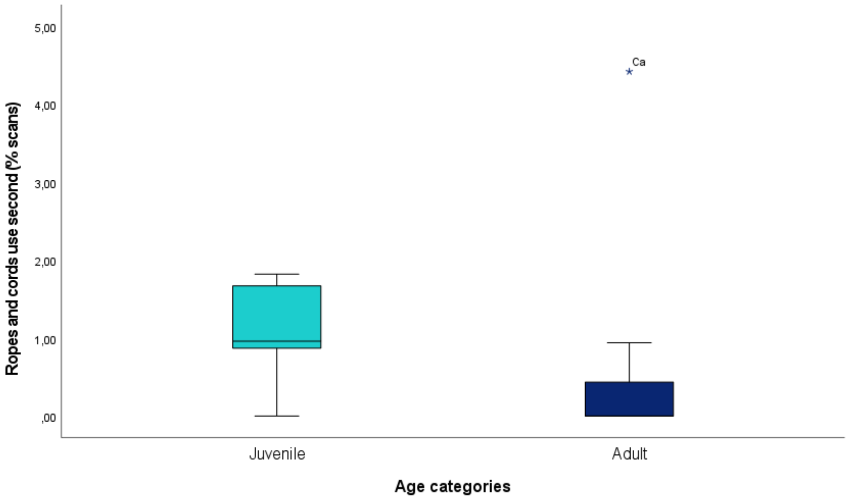
(b)



(c)



(d)

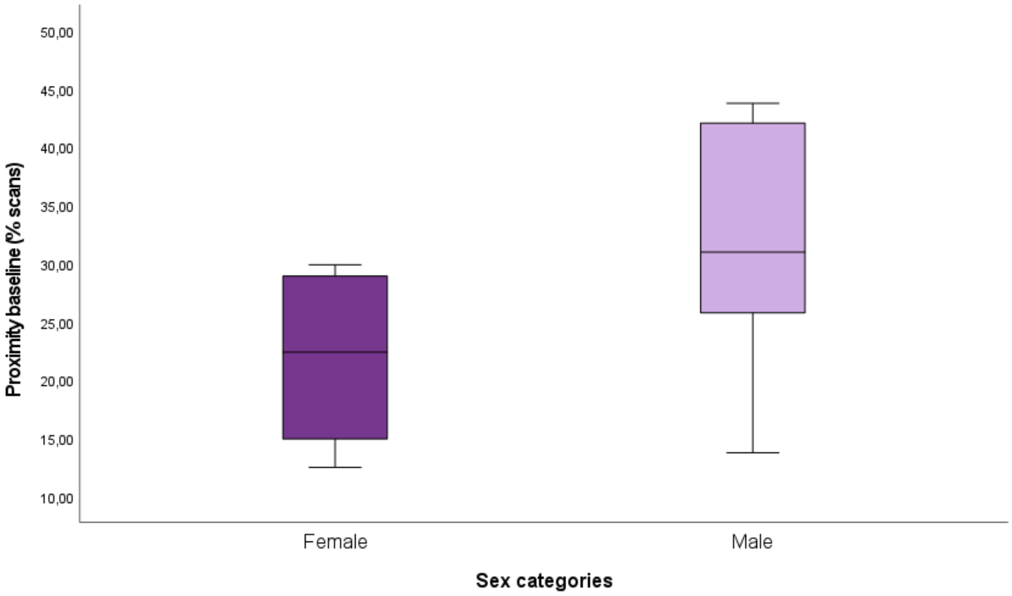


4.3.3. Social proximity

Of the 5,188 scans I performed, 1,586 included the focal individual in close proximity to another monkey (i.e., up 1 metre away from another individual) (*Mdn* = 30.67, IQ = 20.33). Thus, I calculated the proportion of scan samples (expressed as percentage, %) that every individual spent in close proximity to at least one other monkey, for every individual and observational phase. Individuals were observed spending *Mdn* = 28.15 (IQ = 10.68), *Mdn* = 30.91 (IQ = 26.32), and *Mdn* = 29.36 (IQ = 25.32) % of scans in proximity to other monkey(s) during the baseline, second, and final phases (respectively).

Overall, social proximity did not change significantly among observational phases ($p = 0.646$). When comparing between females and males, I found a tendency towards a significant difference during the baseline ($U = 12.000$, $Z = -1.952$, $p = 0.051$), with males spending more time in social proximity than females; nonetheless, this was not observed during the second or final phases ($p \geq 0.534$). There were no significant differences between juveniles and adults in any of the observational phases ($p \geq 0.100$).

Figure 4.16. Box plot showing the significant difference in percentage of scans spent in social proximity between males and females observed during the baseline phase.



4.3.4. Behavioural diversity indexes

The results from the calculations of the behavioural diversity indexes can be seen in Table 4.6.

Table 4.6

Baseline and final Shannon H diversity indexes calculated for every individual and difference between them (n = 16)

Name	Baseline (B)	Final (F)	F-B score
Li	1.30	1.11	-0.19
Garrincha	1.45	1.61	0.16
Tino	0.98	1.31	0.33
Lombinho	1.08	1.16	0.07
Dana	1.11	1.28	0.17
Café	1.14	1.52	0.38
Galápagos	1.03	1.41	0.38
V	1.16	1.06	-0.10
Fúria	1.12	1.48	0.36
Vitinho	1.42	1.48	0.06
Arrebite	1.31	1.48	0.17
Tapa	1.25	1.43	0.18
Tiquinho	1.33	1.57	0.24
Mãozinha	1.39	1.54	0.15
Linguinha	1.24	1.27	0.03
Capuccina	1.45	1.41	-0.04

A one sample t-test showed that F-B scores were significantly different from zero (t = -3.454, p = 0.004). A two-way ANOVA showed there were significant differences related to sex class (F = 5.649, p = 0.035), with males (n = 10) having significantly higher F-B scores than females (n = 6). No significant differences related to age nor age and sex were found ($p \geq 0.072$). Moreover, I did not find a significant correlation between the Shannon H indexes and behaviours potentially indicative of stress (BPIS) during the baseline or final phases ($p \geq 0.687$).

Figure 4.17. Box plot showing the difference between the calculated Shannon H indexes during the baseline and final observational phases.

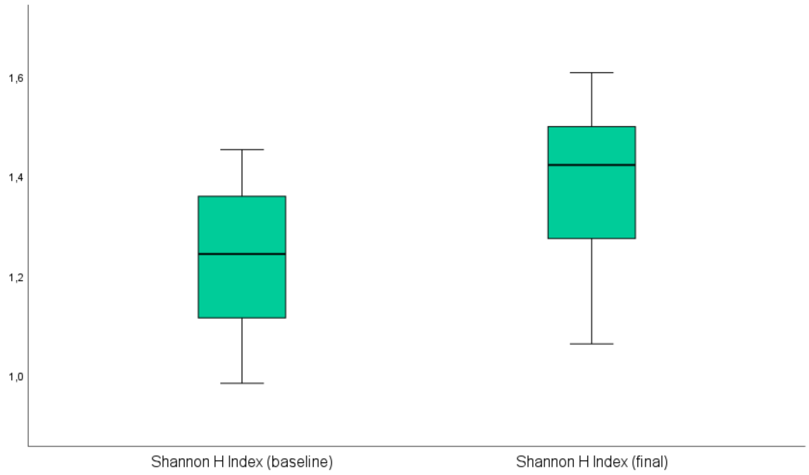
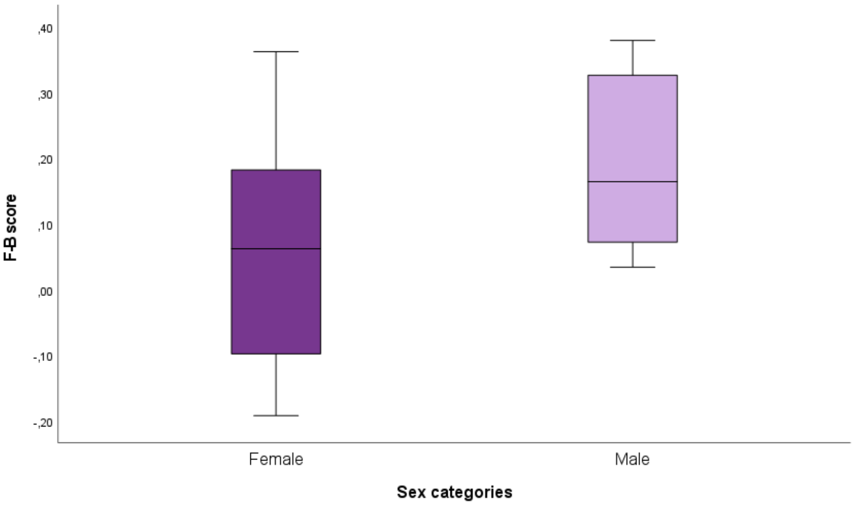


Figure 4.18. Box plot showing the significant difference in the calculated F-B scores between males and females.



4.3.5. Stress-related behaviours observed in the cage and in isolation

No significant differences were found in the exhibition of behaviours potentially indicative of stress (BPIS), including motor stereotypies and self-directed behaviours, among the three conditions tested ($p = 0.607$) as seen in Table 4.7.

Table 4.7

Comparison of percentage of time spent in behaviours potentially indicative of stress (BPIS) in the three conditions tested (group enclosure, inner cage, and individual cage) of all individuals (n = 14)

Group enclosure		Inner cage		Individual cage (isolation)	
Median	IQ	Median	IQ	Median	IQ
2.30	3.88	5.80	20.86	2.90	20.34

4.4. Discussion

Overall, there was a significant increase in time spent performing the behaviours from the general categories *foraging* and *affiliative* by the individuals throughout the rehabilitation programme. Furthermore, there was a significant decrease in time spent *interacting with humans*, and the behaviours related to *locomotion*, *vigilance*, *inactivity*, *solitary play*, *agonistic*, as well as *behaviours potentially indicative of stress* (BPIS), remained stable across time. Otherwise, the animals seem to be responding adequately to the rehabilitation programme: time interacting with humans, for example, decreased significantly; in fact, this behaviour was not observed during the final observational phase. Moreover, the increase in affiliative behaviours may be related to enrichment of the social environment (e.g., the addition of individuals to the social group). Interestingly, I did not find a significant increase in social (i.e., spatial) proximity in this study. Both grooming and proximity have been used to measure social integration in primates (Silk, 2003 in Fischer et al., 2017), and spatial proximity and affiliative behaviours have been associated in previous studies with brown capuchin monkeys (*Sapajus apella*) (Morton et al., 2015). In this sense, the significant increase in grooming observed in this study suggests an increase in social integration within social groups; however, the fact that social proximity did not increase throughout the rehabilitation period contradicts this. As suggested by Mendoza-Nakano (2016), small enclosures may promote the exhibition of social behaviours as a way of decreasing stress (Mendoza-Nakano, 2016). Thus, introducing new members to the social groups could be working as a form of ‘environmental enrichment’ for the monkeys in this study. If this was the case, it could mean that the social groups were less likely to stay together after release. As post-release monitoring was not performed in this study, I cannot confirm whether the monkeys stayed together after release or not. The rehabilitation programme does seem to be supporting the individuals' acquisition of intra-specific social skills; however, with the current data, I cannot determine if these skills were only temporary (i.e., an apparent effect of the current environment and not a true measure of social integration). A potential way to overcome this would be to collect more detailed data on the quality of social relationships of the individuals, namely affiliative behaviour (e.g., grooming) and social proximity. These could be used as measures of social integration in the reintroduction project in the future, including after the release of the individuals where possible.

When comparing to other *Sapajus libidinosus* captive and free-ranging groups (see Table 4.8 in the next page), the animals from the present study seem to have similar activity budgets, at least in some broad behavioural categories. Time spent in foraging and social behaviour was similar to other groups, both in the wild and in rehabilitation programmes (e.g. Vilela, 2003; Moura, 2004; Ferreira et al., 2016; Nunes, 2017). Conversely, time dedicated to locomotion was approximately half of the time observed in free-ranging groups for this activity (Vilela, 2003; Moura, 2004). A possible explanation for this could be the size of the enclosures in which the animals are kept during rehabilitation. In this study, most enclosures had an area of approximately 10 m^2, whilst the home ranges utilised by this species in the wild are much higher than this. For example, one study performed with two groups of free-ranging *S. libidinosus* estimated their home ranges to be between 250 and 350 ha (Izar et al., 2011). In this sense, less time spent in locomotion may be related to enclosure size rather than a lack of skills of the rehabilitant individuals. However, without the opportunity to use larger spaces during rehabilitation, such as pre-release enclosures used for other primate species rehabilitation before reintroduction (e.g., Mendoza-Nakano, 2016), it is difficult to assess the locomotion skills of the capuchins. This issue is also observed in the assessment of spatial and substrate use in this study.

Even though robust capuchin monkeys are considered arboreal primates, they rely heavily on the use of the ground and the medium strata of the canopy, spending up to 90% of the time below 10 m, particularly during the dry season (Oliveira et al., 2014). The individuals indeed seem to be utilising the space of their enclosures in a relatively similar way to this – up to 47% of the observations in the present study included the focal individual using the ground or the medium tier of the enclosure. However, as the enclosures in the present study are less than 3 m in height, it is not possible to know how the monkeys will use the canopy in their future release sites without the use of a pre-release enclosure and monitoring; in other words, without bigger enclosures where they can decide which tier to use. This is one of the potential disadvantages of hard releases, where animals are released without having much experience or training in a similar place to the release site.

Table 4.8

Comparison between activity budgets of *Sapajus libidinosus* in several published studies (means shown)

Behavioural category	**Present study (baseline)**	**Present study (final)**	**Nunes (2017) (n=13)**	**Ferreira et al. (2016) (n=113)**	**Ferreira et al. (2018) (n=25)**	**Sabbatini et al. (2008) (n=8)**	**Moura (2004) (n=10)**	**Vilela (2003) (n=7)**
Study site	Wildlife rescue centre(s)	Wildlife rescue centre(s)	Wildlife rescue centre(s)	Wildlife rescue centre(s)	Wildlife rescue centre(s) and zoos	Tropical forest (semi-deciduous)	Tropical forest (dry)	Tropical forest (semi-deciduous)
Origin of the animals	Wild-born	Wild-born	Wild-born	Wild-born	Wild-born and captive-born	Wild-born	Wild-born	Wild-born
	Captive-raised	Captive-raised	Captive-raised	Captive-raised	Captive-raised	Wild-grown	Captive-raised and wild-grown	Wild-grown
Foraging	32.75	32.64	40.00	27.32	36.83	34.00	41.90	47.50
Locomotion	19.15	18.30	20.00	11.41	17.10	41.00	37.10	38.00
Affiliative	4.46	13.27	10.00	10.34	9.38	<10.00	~6.5	-
Agonistic	0.03	0.14	<5.00	0.15	<10.00			-
Environmental manipulation	3.74	5.54	4.00	1.50	<10.00	-	-	-
Vigilance	19.53	19.97	15.00	25.12	20.06	-	<5.00	-
Solitary play	0.06	0.76	<5.00	0.67	<10.00	-	-	-
Inactivity	1.22	3.12	<5.00	6.18	<10.00	15.00	~5.5	14.50
Human interaction	1.03	0.00	<5.00	-	-	2.00	-	-
BPIS	1.60	3.09	6.00	17.72	10.02	-	-	-
Ground use	(0 m) 17.63	(0 m) 20.78	-	-	-	(<5 m) 51.86	(<1 m) 22.50	-
Medium strata use	(>0-1.5 m) 32.42	(>0-1.5 m) 37.71	-	-	-	(5-10 m) 44.17	(4-6 m) 55.00	-
High strata use	(>1.5 m) 49.94	(>1.5 m) 41.51	-	-	-	(>10 m) 3.97	(>6 m) 22.50	-

Regarding substrate use, individuals in the present study relied primarily on the use of fixed trunks and concrete. This, along with their apparent ability to jump efficiently (i.e., I observed only one fall from a juvenile and none from adult individuals during the complete observational period) and use mobile substrates as expected (e.g., using tree branches to rest and eat), suggests their already acquired locomotion skills are *relatively* developed and may prepare them for reintroduction, albeit the small enclosures in which they are housed. Of course, this does not mean the monkeys should be housed in small enclosures nor that they should not undergo training in pre-release enclosures, as there are other concerns related to their survival skills that the housing systems and husbandry practices maintained to this date in the CETAS–RN rescue centre may not be providing. One example of these concerns are the manipulation and foraging skills of the individuals, which may be enhanced in reintroduction projects by applying environmental enrichment techniques (Reading et al., 2013). Capuchin monkeys need to practice and perfect their manipulation and foraging skills, including tool use, in order to survive in the wild, as they rely heavily on plant and animal species that are inaccessible for other wild animals (e.g., nuts), including other primates (Fragaszy et al., 2004). Thus, they must learn to identify and manipulate the food items available on the release sites –their future home environments– adequately. Individuals in CETAS–RN rescue centre are given the opportunity to learn to open nuts with the use of heavy rocks as they would in the wild (personal observation). As this did not happen during my observations, I was not able to use this data. Hence, the individuals may be gaining some abilities related to the manipulation of food items that I was unable to observe. Indeed, tool use is an important aspect of foraging and manipulation skills observed in bearded capuchins monkeys both in captivity and in the wild (Fragaszy et al., 2004). Even though I did observe a general increase in foraging and manipulation behaviours, including juveniles and adults, the animals are being fed mainly with fruits for human consumption and not with fruits that they will encounter in the wild (i.e., at the release sites). Moreover, the way in which the food is provided (i.e., cut in small pieces and placed directly on the floor) limits the possibilities of the monkeys to spend more time foraging and perfecting their manipulation skills. Thus, these may be areas for further research to be performed in the future.

Regarding sex class and age-related comparisons, the individuals of the present study appear to behave accordingly to what is found in the scientific literature. Juveniles, for example, dedicated more time to social and solitary play as well as food and environmental manipulation than adults, whilst adults spent more time performing vigilance behaviours than younger monkeys. These differences are consistent with what is normally observed in captive and free-ranging robust capuchin monkey (*Sapaju*s spp.) groups (Fragaszy et al., 2004).

The use of comparisons between activity budgets of wild (free-ranging) versus captive primates is a matter of much debate, as many studies have proposed to use them mainly for wellbeing assessment (Powell and Cheyne, 2019). However, the present study aimed to use these comparisons to provide an initial assessment of the rehabilitation programme and to propose improvements for it, and not to draw any conclusions regarding the welfare of the subjects. The same can be applied to the use of the Shannon H index as a behavioural diversity index in this study, as this has been used as an indicator of animal welfare in previous studies (Millet et al., 2020) and not as an indicator of rehabilitation progress or 'success'. In this sense, the significant difference observed between the baseline and final behavioural diversity indexes of the rehabilitant capuchins suggests that the rehabilitation programme is helping the individuals to increase their behavioural diversity. Further research is needed on the use of behavioural diversity indexes to assess rehabilitation outcomes in capuchins and other primate species.

4.5. Conclusion

In this study, I analysed the behaviour of 16 trafficked (captive-raised or raised by humans) bearded capuchin monkeys that were part of a rehabilitation programme run by a government wildlife rescue centre, CETAS–RN, in Northeast Brazil. I aimed to perform an initial assessment on the efficacy of the rehabilitation programme by analysing the changes in behaviour across time (i.e., activity budgets, space and substrate use and behavioural diversity indexes) of the rehabilitant individuals and comparing with other individuals of the same species. I hypothesised that genus normative behavioural patterns of the rehabilitated individuals (i.e., activity budgets) at the end of the rehabilitation period would be similar to published data on wild, free-ranging bearded capuchins. I also hypothesised that the time the individuals spent engage in foraging as well as the behavioural diversity indexes would increase and the time spent in affiliative behaviour would increase if found as less than 4% or remain stable if found as 4% or more of the overall activity budget. Conversely, the rate per minute of agonistic interactions and the time spent in behaviours potentially indicative of stress (BPIS) would decrease if found over 0.6 rate per minute and 3% of the total activity budget, respectively, or remain stable if found as less than 0.60 rate per minute and as less than 3%, when comparing the start and the end of the rehabilitation period.

As predicted, the activity budgets of the individuals were similar to previous studies, including several made with other trafficked individuals in Brazilian rescue centres and with free-ranging conspecifics (See Table 4.4). Furthermore, there was a significant increase in affiliative behaviours (i.e., from *Mdn* = 1.45 to *Mdn* = 9.57) and behavioural diversity indexes (as predicted) and a decrease in human interaction behaviours (which was not predicted). The rate per minute for agonistic behaviours, as predicted, was below 0.60 and remained stable across observational phases. BPIS were found below 3% of the overall activity budget and remained stable throughout the rehabilitation period, as I predicted and as recommended by Cheyne et al. (2012) for rehabilitant primates.

These are positive findings in a rehabilitation programme, as group formation can be challenging in capuchin monkeys (Souvignet et al., 2019) and detachment from human beings and is desirable for the animals when released, to prevent human-wildlife conflicts. Moreover, the low and stable level of stress-related behaviours observed suggests that the general welfare level of the animals is adequate, albeit I did not confirm this with the use of a physiological indicator, such as faecal glucocorticoids (e.g., Ferreira et al., 2018). I consider that the rehabilitation programme was successful in providing the animals with key survival skills, mainly species-specific social skills, and was also promoting detachment from humans. However, it is yet to be found if these skills would be maintained after release and contrary to my predictions, foraging behaviours did not increase significantly when comparing the start and the end of the rehabilitation programme.

Regarding space use, I predicted that the monkeys would spend at least 40% in the highest tier of their enclosure and no more than 5% in the lowest tier of their enclosure (i.e., on the ground) of their total activity budget by the end of the rehabilitation period. As predicted, the capuchins spent more than 40% of their time in the highest tier of the enclosures in all observational phases; however, they spent more than 5% of the time on the ground in the first, second and final observational phases. As mentioned in the Discussion, free-ranging robust capuchin monkeys (*Sapajus* spp.) rely heavily on the use of the ground and medium strata (Oliveira et al., 2014); hence, this does not seem to be an issue that could compromise their future survival.

Regarding age and sex class-related differences, I hypothesised that adult females would spend more time engaged grooming others than adult males, whereas adult males would spend more time engaged in social play and vigilance than adult females. Moreover, adult monkeys would spend more time engaged in vigilance than juveniles, and juveniles would spend more time in social play and environmental manipulation, by the end of the rehabilitation period. As predicted, juveniles dedicated more time to social play as well as food and environmental manipulation than adult monkeys, whilst adults spent more time performing vigilance behaviours than juveniles. These differences are consistent with what is normally observed in captive and free-ranging robust capuchin monkey groups (Fragaszy et al., 2004), as mentioned above. However, there were no significant differences between adult females and adult males in time spent in affiliative behaviours, including allogrooming and social play.

The second aim of this Chapter was to analyse the behavioural effects of two husbandry/research practices, which included (a) placing the monkeys in the inner cages of their enclosures for approximately 30 minutes per day during the 3.5-month observational period and (b) placing the monkeys in individual cages to carry out individual behavioural tests (i.e., for approximately five minutes per trial and no more than a total of 65 minutes per monkey in the 3.5-month observational period) on the exhibition of behaviours potentially indicative of stress (BPIS) of the capuchin monkeys that were part of the rehabilitation programme. I hypothesised that there would not be significant differences in the amounts of time engaged in BPIS when comparing the three conditions considered (i.e., (a) placing the monkeys in the inner cages of their enclosures, (b) individual behavioural tests and (c) usual group enclosures). As expected, I did not find significant differences in the exhibition of BPIS in the three conditions considered, which suggests these practices were not negatively impacting the well-being of the individuals.

The third aim of this Chapter was to utilise the behavioural profiles (activity budgets) to measure personality traits as part of the next Chapter of this dissertation, which I was able to do. The fourth aim of this Chapter was to use these results, in terms of survival skills, as a basis for determining the type of environmental enrichment and/or pre-release training that rehabilitant bearded capuchin monkeys would need to enhance these skills before being released back to the wild. Unfortunately, I was not able to perform a second fieldwork trip to Brazil because of the Covid-19 pandemic, which prevented me from designing an environmental enrichment programme or pre-release training as part of this dissertation. Nevertheless, the animals were not released until they were considered behaviourally fit to survive by the staff of the rescue centre. I suggest that the establishment of an environmental enrichment specifically targeted at improving foraging skills could enhance the current rehabilitation/reintroduction project in the future. This could include, for example, providing the animals with food items (e.g., fruits) that will be available in their future release sites and/or foraging puzzles. However, this may be difficult to perform in government wildlife rescue centres with high quantities of incoming animals, overcrowded facilities and limited resources, as is the case of many rescue centres in Latin America (Mitman et al., 2021).

Chapter 5: Assessment of Personality Structure in Rehabilitant Capuchin Monkeys (*Sapajus* sp.)

Chapter 5: Assessment of Personality Structure in Rehabilitant Capuchin Monkeys (*Sapajus* sp.)

5.1. Introduction

In the last decades, several studies have been performed with the aim of exploring the personality structure of capuchin monkeys (*Cebus* spp. and *Sapajus* spp.). The fact that capuchins are interesting from an ethological perspective could be explained by several reasons; they have a high encephalization quotient (i.e., large brains) when compared to other Neotropical primates, they live long lives, use tools, manipulate their environment, and are behaviourally flexible (Fragaszy et al., 2004; Uher and Visalberghi, 2016). Unfortunately (and possibly because of similar same reasons), they are frequently kept by humans as pets, leading to large numbers of individuals of these species ending up in wildlife rescue centres in Latin America and being reintroduced back to the wild every year (Levacov et al., 2011; Mitman et al., 2021), as has been reviewed throughout this dissertation. Here, I will briefly review the studies made in the last decade which have investigated the personality structure of capuchin monkeys.

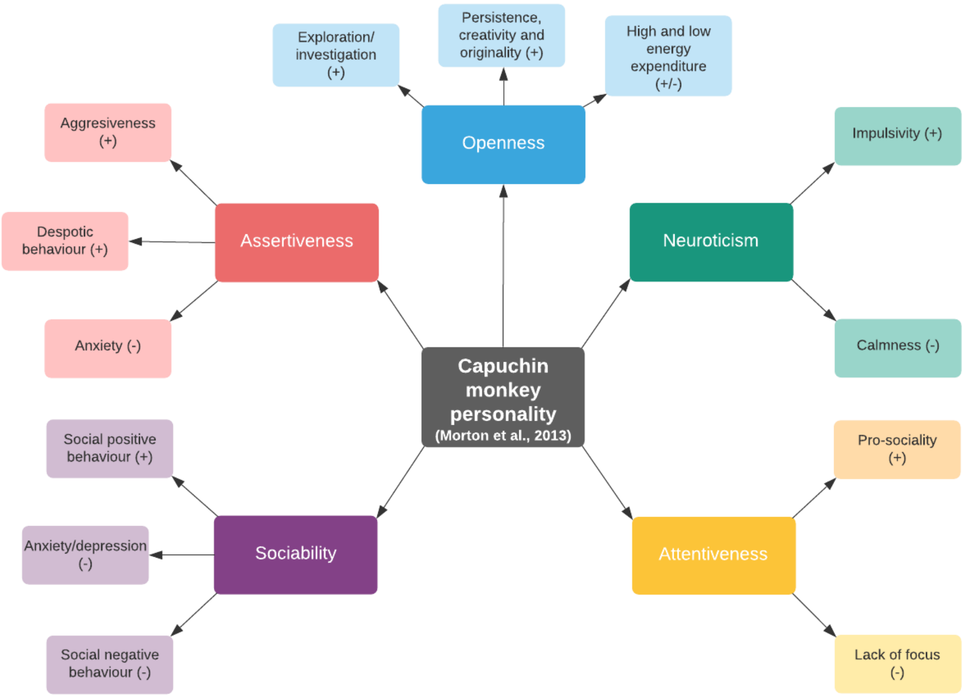
Overall, there are three methods to assess animal personality: observer traits ratings (e.g., questionnaires), behavioural tests, and behavioural observations. Manson and Perry (2013) used observer trait ratings (i.e., a personality questionnaire comprised of 26 items) to investigate the personality structure of 240 wild, free-ranging Colombian white-faced capuchin monkeys (*Cebus capucinus*). They found that white-faced capuchin personality was composed by five dimensions: Extraversion, Openness, Neuroticism, Agreeableness, and Eccentricity (Manson and Perry, 2013). All dimensions, except Eccentricity, closely resembled four of the 'Big Five' found in human personality (Extraversion, Openness, Neuroticism and Agreeableness. See Chapter 1: General Introduction and Aims for a review of the 'Big Five' Personality Model). Eccentricity was characterised by high loadings of items related to Openness and Conscientiousness. Several of the traits found in the questionnaire, such as 'sociability', were validated using behavioural observations; in other words, they were highly correlated with observable behaviours (Manson and Perry, 2013).

In the same year, another study was published aimed at exploring capuchin personality; in this case, of brown capuchin monkeys (*Sapajus apella*). Morton et al. (2013) assessed the personality of 127 captive brown capuchin monkeys using the Hominoid Personality

Questionnaire (Weiss et al., 2009) and proposed five personality dimensions for this genus: Assertiveness, Openness, Neuroticism, Sociability, and Attentiveness (Morton et al., 2013). The dimensions Assertiveness, Openness and Neuroticism closely resembled those found in humans. The dimension Assertiveness was characterised positively by traits related to aggressive behaviour and negatively by anxiety-related items. Openness was characterised positively by items related to high energy expenditure, exploration, investigation, and creativity, originality, and persistence; and negatively by traits reflecting low energy expenditure. Moreover, Neuroticism was characterised positively by traits related to impulsivity and volatility and negatively by items related to calmness and tranquillity. Sociability was characterised positively by items describing affiliative behaviour such as friendliness, and negatively by anxiety and depression-related items and those related to agonistic behaviour. Finally, Attentiveness was characterised positively by traits related to pro-sociality and negatively by traits that described a lack of focus (Morton et al., 2013). A summary of the traits that characterise each dimension can be seen in Figure 5.1 in the next page.

Morton et al. (2013) identified the five personality dimensions using the Hominoid Personality Questionnaire (HPQ) from Weiss et al. (2009) and validated these dimensions using behavioural observations. This was achieved by performing correlation analyses between the scores obtained through the use of the HPQ and specific behaviours that could reflect these personality traits (for example, a correlation analysis could be performed between Neuroticism scores and percentage of time spent in stereotypic behaviours). Using this method, they found several significant correlations between the dimensions proposed and the behavioural profiles of the individuals. For example, it was found that monkeys with higher scores on Assertiveness spent more time engaged in aggressive behaviours and less time in social isolation than monkeys with lower scores in this dimension. Similarly, Sociability was negatively correlated with time spent alert and in social isolation (Morton et al., 2013).

Figure 5.1. Capuchin monkey (Sapajus apella) personality structure (adapted from Morton et al., 2013).



Interestingly, a study performed by Robinson et al. (2016), which compared the personality structure of white-faced capuchin monkeys (*Cebus capucinus*; n = 240) and brown capuchin monkeys (*Sapajus apella*, n = 100), found that the dimensions Assertiveness, Openness and Neuroticism were shared by these taxa and suggested they may have been shared by a common evolutionary ancestor (Robinson et al., 2016). However, it is important to consider that the dimensions obtained in a specific study are partly related to the instrument being used (Manson and Perry, 2013). In this sense, the Hominoid Personality Questionnaire (Weiss et al., 2009), which is comprised of 54 items and was used to assess the personality of the brown capuchins, could have been more able to detect certain personality traits than the 26-item questionnaire used to assess the personality of white-faced capuchins because it had more items and not necessarily because white-capuchins *lack* certain personality traits. Hence, utilising the same instrument could be useful to perform comparisons between different taxa. As mentioned by the authors, further research is needed to fully understand the evolution of personality traits in capuchin species (Robinson et al., 2016).

The use of observer trait ratings to assess primate personality may be considered as a matter of debate. It has been suggested that 'personality' is a human *construct* i.e., an abstract idea which is constructed by humans; thus, it cannot be observed directly (Uher and Visalberghi, 2016).

In fact, personality is frequently assessed by using observer trait ratings and/or tests in addition to direct behavioural observations. Moreover, it may be influenced by personal and social knowledge of the observer (Uher and Visalberghi, 2016) and the specific questionnaire used (e.g., Manson and Perry, 2013). Because of this, the specific traits measured in a given questionnaire or test may not be ecologically relevant for the species in question. To overcome these challenges, Uher et al. (2013) utilised an approach called the 'Behavioural Repertoire x Environmental Situations Approach' (BRxES-Approach) to assess the personality of 26 captive brown capuchin monkeys (*Sapajus apella*). This approach was used to generate non-lexical personality constructs with high ecological validity (Uher et al., 2013). The constructs were generated by assuming, hypothetically, the possible individual patterns that could be observed in a given situation or context; and were referred to as 'working constructs'. These working constructs were labelled *before* the experiments were carried out and then reviewed after analysis and given a new label if necessary.

To apply this approach, Uher et al. (2013) used a combination of 15 behavioural tests, performed individually or in dyads, planned to elicit specific behaviours as well behavioural observations performed in two different settings: (1) in a pre-feeding situation (i.e., before the animals were fed) and (2) in a social situation, where the animals were in their normal enclosure with the other members of their social group. Ten-minute focal sampling was used as well as scan sampling and event sampling for behavioural observations (Uher et al., 2013). A brief description of Uher et al. (2013) behavioural tests and observations as well as the personality constructs measured in each situational context can be seen in Table 5.1.

Table 5.1

Behavioural tests and observations and personality constructs proposed by Uher et al. (2013) using the BRxES-Approach

Behavioural test/ observation (situational context)	**Description**	**Personality constructs measured**
Conveyor belt test	Different food items were placed in a conveyor belt close to the experiment cage. The monkey could reach it with his/her hand by moving a wheel.	Food orientation
Conveyor belt disconnected test	Different food items were placed in a conveyor belt close to the experiment cage. However, the monkey could not move the conveyor belt in this case as it was disconnected from the wheel.	Arousability Impulsiveness
Food competition test	One food item was offered to two individuals in the experiment cage.	Aggressiveness Competitiveness Dominance
Hidden food test	Small food items or honey were hidden/spread in the experiment cage.	Arousability Anxiousness Physical activity Social orientation to conspecifics Social orientation to humans Vigilance

Table 5.1 (continued)

Behavioural tests and observations and personality constructs proposed by Uher et al. (2013) using the BRxES-Approach

Yoghurt grid test	Plain yoghurt was spread in a platform close to the experiment cage.	Distractibility
Human interaction test	The experimenter talked to the monkey and encouraged him/her to approach, then offered food and stopped. Then called them again, encouraging them to approach.	Arousability Aggressiveness to humans Social orientation to humans
Masked human test	The experimenter entered the test room, disguised with a wig, a mask and a dress. They offered food to the monkey through the cage to enable direct contact if desired by the monkey.	Aggressiveness to humans Anxiousness Arousability Social orientation to humans
Novel food test	Four unfamiliar food items and four familiar food items were given to the monkey in the experiment cage.	Curiousness Food orientation
Multiple objects test	Several familiar and unfamiliar objects were placed inside the experiment cage.	Aggressiveness Anxiousness Arousability Creativeness/inventiveness Curiousness Physical activity
Tunnel basket test	A laundry PVC basket was placed inside the experiment cage. One of the open ends was covered with a dark cloth (but the monkey could still go inside).	Aggressiveness Anxiousness Arousability Creativeness/inventiveness Curiousness Physical activity Social orientation to humans
Large cloth test	A large bed sheet was placed inside the experiment cage, so that it was hanging over transversally.	Aggressiveness Anxiousness Arousability Curiousness Creativeness/inventiveness Physical activity Social orientation to humans
Furry animal test	A small, soft plush toy was placed in front of the experiment cage. The toy’s eyes were covered to diminish the perceived threat.	Aggressiveness Anxiousness Arousability
Blocked food tube test	A tube with preferred food items was fixed at a 45° angle inside the experiment cage.	Arousability Food orientation Impulsiveness
Foraging box test	A foraging box with wood shavings and three pumpkin seeds was placed inside the experiment cage. The monkey could reach the seeds by putting the hand inside the box.	Persistency Vigilance
Sudden noise test	A foreign news programme suddenly started to play back in the experiment room for 10 seconds, twice with a break of 20 seconds.	Aggressiveness Anxiousness Arousability Vigilance
Pre-feeding observation	The individuals were observed before being fed. They could hear the caregivers preparing their food and see and hear the neighbouring enclosures being fed.	Arousability Food orientation Social orientation to conspecifics
Social group observation	The individuals were observed in their outdoor enclosures in their normal social groups.	Aggressiveness Aggressiveness to humans Anxiousness Arousability Dominance Food orientation Gregariousness Physical activity Playfulness Self-cleanliness Sexual activity Social orientation to conspecifics Social orientation to humans

These tests and observations were performed in two blocks of approximately two weeks, and a two-week pause was performed between each block. Behavioural observations were performed during the same weeks as the behavioural tests. Intraclass correlations were used to assess consistency between trials. Latencies as well as durations of specific behaviours were measured, z-standardised and analysed to produce a score in each personality construct. The results showed that '*capuchin monkeys have pronounced and stable individual differences in a broad range of behaviours, comparable to those described in great apes and humans'* (Uher et al., 2013; p. 12).

Even though one could argue against (or in favour) of using Uher et al. (2013) BRxES-Approach or the approach taken by Morton et al. (2013) and Manson and Perry (2013) (i.e., using observer trait ratings and behaviours to validate them) to assess the personality structure of capuchins, both methods have advantages and disadvantages and may ultimately complement each other (refer to Chapter 1: General Introduction and Aims for a discussion on this issue). In fact, a study performed by Nunes (2017) aimed to assess the personality of 13 bearded capuchin monkeys (*Sapajus libidinosus*) by using a combination of these methods (proposed by Morton et al., 2013 and Uher et al., 2013). Seven behavioural tests were slightly modified to be applied in a wildlife rescue centre, with the objective of promoting the expression of certain behaviours in the subjects. These behaviours were **previously** categorised into nine personality constructs: (1) Curiosity/Playfulness, (2) Neophilia, (3) Creativity, (4) Persistence, (5) Aggressiveness, (6) Distractibility, (7) Sociability to humans, (8) Boldness and (9) Exploration, based on the BRxES-Approach proposed by Uher et al. (2013). Furthermore, observer trait ratings were performed using the Hominoid Personality Questionnaire (HPQ) (Weiss et al., 2009) as proposed by Morton et al. (2013). Five pre-established personality dimensions (obtained from Morton et al., 2013) were used for the analysis of data obtained from the observer trait ratings, with each animal having a score on Assertiveness, Openness, Neuroticism, Sociability and Attentiveness. Several significant correlations were found between the personality constructs obtained from the behavioural tests (BT), the observer trait ratings (HPQ) and the behavioural observations (BO). For example, Openness and Neuroticism (HPQ) were positively correlated with time spent interacting with the environment (BO). Moreover, Openness (HPQ) was negatively correlated with vigilance (BO) and Openness and Sociability (HPQ) were positively correlated with solitary play (BO) (Nunes, 2017).

Interestingly, Nunes (2017) did not find any significant correlations between the personality constructs obtained from the behavioural tests based on Uher et al. (2013) and the dimensions obtained from the HPQ, based on Morton et al. (2013). However, it must be noted that in this study (Nunes, 2017), the constructs and dimensions that characterised capuchin personality were established *a priori*, based on ethological knowledge. This is known as an *expert-based method* (Mazzamuto et al., 2019). Thus, a variable reduction method was not used. Using a reduction method, such as Principal Component Analysis (PCA), Factor Analysis (FA), or Regularised Exploratory Factor Analysis (REFA) is a common practice in animal behaviour and personality research and has the advantages of potentially reducing the number of statistical tests by reducing the number of variables in a relatively objective way (Budaev, 2010; Morton and Altschul, 2019). This is a potential limitation of the Nunes (2017) study. Moreover, the sample size was small (n = 13) and the monkeys utilised in the study were born in the wild and raised by humans as pets, which could have influenced the development of their personality. It is known that traumatic experiences related to the illegal pet trade may influence the behavioural development of primates (Soulsbury et al., 2009). Unfortunately, little is known about the effects of the illegal pet trade on the ontogeny of capuchin monkeys' personality in particular.

In a further study by Uher and Visalberghi (2016), the personality of 150 captive brown capuchin monkeys (*Sapajus apella*) was assessed using observer ratings and behavioural coding (both in naturalistic and testing contexts). Five factors were obtained from observer ratings: (1) 'dominant-competitive-aggressive', (2) 'curious-inventive-persistent', (3) 'playful-active-impulsive', (4) 'gregarious-prosocial', and (5) 'excitable-vigilant'. Constructs related to aggressiveness towards conspecifics and humans, creativity and inventiveness, curiousness, dominance, food orientation, gregariousness, playfulness, self-cleanliness, social orientation to humans, and sexual activity were similar between methods, meaning that the subjective assessments performed by the observers (raters) were significantly correlated with behaviours coded from testing and naturalistic contexts (Uher and Visalberghi, 2016).

More recently, Fernández-Bolaños et al. (2020) explored the personality structure of yellow-breasted capuchin monkeys (*Sapajus xanthosternos*). In this study, 26 wild, free-ranging monkeys were assessed using behavioural observations and observer trait ratings using the Hominoid Personality Questionnaire (HPQ) (Weiss et al., 2009). Three personality dimensions were obtained from observer trait ratings: Openness-Neuroticism, Assertiveness, and Attentiveness-Sociability, similar to those found by Morton et al. (2013). Moreover, three dimensions were obtained from behavioural observations: Pro-sociality, which was correlated positively with Attentiveness-Sociability and Assertiveness and negatively with Openness-

Neuroticism; Aggressiveness, which was correlated positively with Assertiveness and negatively with Attentiveness-Sociability; and Reactivity to Humans, which was correlated positively with Openness-Neuroticism (Fernández-Bolaños et al., 2020). Interestingly, vigilance behaviours were highly (positively) correlated with Assertiveness in this study. Conversely, in captive capuchin monkeys, vigilance behaviours are frequently highly correlated with Neuroticism or Openness, probably related to stress and/or curiosity (Morton et al., 2013; Fernández-Bolaños et al., 2020).

Summarising, relatively few studies have investigated the personality of capuchin monkeys (*Cebus* spp. and *Sapajus* spp.). The dimensions Openness, Neuroticism and Assertiveness have been found in both genus (*Cebus* sp., Manson and Perry, 2013; *Sapajus* sp., Morton et al., 2013), suggesting that these dimensions existed in their common ancestor (Robinson et al., 2016). Uher and Visalberghi (2016) found similar (i.e., 'curious-inventive-persistent', 'excitable-vigilant' and 'dominant-competitive-aggressive') using the Behavioural Repertoire x Environmental Situations Approach, suggesting these dimensions are ecologically relevant for these taxa (Uher and Visalberghi, 2016). Even though most of these studies were performed with captive individuals (Morton et al., 2013; Uher et al., 2013; Uher and Visalberghi, 2016), several studies have supported the existence of similar personality dimensions in wild-born, captive-raised individuals (Nunes, 2017; n = 13) and in wild-born, free-ranging individuals (Fernández-Bolaños et al., 2020; n = 26) from this genus (*Sapajus* spp.). Nevertheless, these studies have been made with relatively small sample sizes, presumably due to the practical and conservation-related challenges of working with wild individuals, either in a free-ranging context or with animals destined for reintroduction.

Thus, the main aim of this Chapter was to broaden the current scientific knowledge on capuchin personality structure by replicating a previously described study (Nunes, 2017) in several groups of wild-born, captive-raised bearded capuchin monkeys (*Sapajus libidinosus*, n = 18) who were part of a rehabilitation and reintroduction programme run by a Brazilian wildlife rescue centre. To achieve this, I assessed the personality of the subjects using observer trait ratings (the Hominoid Personality Questionnaire from Weiss et al., 2009 and based on Morton et al., 2013 and several behavioural tests as modified by Nunes (2017) and based on Uher et al., 2013). I also performed behavioural observations to validate these assessments by performing correlation analyses.

My hypotheses were:

1. The personality structure of the subjects would closely resemble the capuchin personality structure found in previous studies, namely Morton et al. (2013) (observer trait ratings) and Uher et al. (2013) (behavioural tests) if assessed following the method proposed by Nunes (2017).
2. There would be a high correlation between the resulting personality traits (i.e., after using observer trait ratings and behavioural tests and analysing using a reduction method) and the behaviours observed in a 'naturalistic' context (e.g., *sociability,* if found, would be positively correlated with *time spent grooming others* and/or *time spent in close proximity to other monkeys*; *openness* would be positively correlated with *time spent manipulating the environment*; *aggressiveness* would be positively correlated with *time spent in agonistic behaviour,* etc.).

5.2. Methodology

5.2.1. Ethical approval

The project received ethical approval by the Animal Welfare and Ethical Review Body of the University of Bristol in January 2019 (reference number UB/18/087) and complied with the NC3Rs Guidelines for Primate Accommodation, Care and Use (NC3Rs, 2017).

5.2.2. Study site and subjects

The study was performed in a government wildlife rescue centre (Centro de Triagem de Animais Salvagens, CETAS) located in Natal, Rio Grande do Norte, Brazil (hereafter, CETAS–RN). This rescue centre receives primates and other wildlife that have been rescued/confiscated from the illegal pet trade or surrendered voluntarily by their 'owners'. Eighteen capuchin monkeys (*Sapajus libidinosus*) were part of the study. Most of these animals were born in the wild, captured and sold as pets illegally, except for two juveniles that were born in the rescue centre to confiscated wild-born adult females (Table 5.1). Refer to Chapter 2: General Methodology for a complete description of the origin of the individuals as well as the rehabilitation programme of the rescue centre.

Table 5.1

Individuals that were part of the study and data available for each individual (n = 18)

Name/ID	**Age category**	**Sex class**	**Group**	**Origin**	**Personality assessment method**		
					Observer trait ratings	**Behavioural tests**	**Behavioural observations**
Dana	Adult	Female	1	Wild-born	Yes	Yes	Yes
Lombinho	Adult	Male	1	Wild-born	Yes	Yes	Yes
Tino	Adult	Male	1	Wild-born	Yes	Yes	Yes
Li	Juvenile	Female	1	Wild-born	Yes	Yes	Yes
Garrincha	Juvenile	Male	1	Wild-born	Yes	Yes	Yes
V	Adult	Female	2	Wild-born	Yes	No	Yes
Fúria	Adult	Female	2	Wild-born	Yes	Yes	Yes
Joana	Adult	Female	2	Wild-born	Yes	Yes	Yes
Galápagos	Adult	Male	2	Wild-born	Yes	Yes	Yes
Café	Adult	Male	2	Wild-born	Yes	Yes	Yes
Rabinha	Juvenile	Female	2	Wild-born	Yes	Yes	Yes
Vitinho	Juvenile	Male	2	Wild-born	Yes	Yes	Yes
Tapa	Adult	Female	3	Wild-born	Yes	Yes	Yes
Arrebite	Adult	Male	3	Wild-born	Yes	Yes	Yes
Tiquinho	Juvenile	Male	3	Captive-born	Yes	Yes	Yes
Mãozinha	Juvenile	Male	3	Captive-born	Yes	Yes	Yes
Capuccina	Adult	Female	4	Wild-born	Yes	No	Yes
Linguinha	Adult	Male	4	Wild-born	Yes	No	Yes

5.2.3. Personality assessment

I used two different methods to assess personality in this study: (1) behavioural coding in a set of behavioural tests (n = 15) based on Uher et al., 2013 and (2) observer trait ratings (n = 18) using the Hominoid Personality Questionnaire (Weiss et al., 2009; Morton et al., 2013) as described by Nunes (2017). Behavioural observations were performed to validate the observer trait ratings and the behavioural tests, as described above.

A timeline of the application of these assessments can be seen in Table 5.2.

Table 5.2

Methods and timeline used for personality assessment in this study

Method	Group	Dates (phase 1)	Dates (phase 2)	Dates (phase 3)
Behavioural observations	All groups (n=18)	01/04/2019–10/04/2019	06/05/2019–17/05/2019	01/07/2019–17/07/2019
Behavioural tests	Group 1 (n=5)	30/04/2019–06/05/2019	21/05/2019–27/05/2019	NA
	Group 2 (n=6)	23/04/2019–26/04/2019[1]	30/05/2019–04/06/2019	NA
	Group 3 (n=4)	07/05/2019–13/05/2019	25/06/2019–01/07/2019	NA
	Group 4 (n=2)	NA	NA	NA
Behavioural test (tunnel)	Groups 1,2,3 (n=15)	02/07/2019–15/07/2019	NA	NA
	Group 4 (n=2)	NA	NA	NA
Hominoid Personality Questionnaire	All groups (n=18, *k*=4)	1 to 17 August 2020	NA	NA

[1] The first set of behavioural test trials for Vitinho (from Group 2, see Table 1) were applied between 13/05/2019–20/05/2019.

5.2.3.1. Behavioural observations

I used the behavioural observations described in the previous chapter (see Chapter 4 for a full description of the methodology) to obtain a behavioural profile for every individual in this study. A total of 47.5 hours of behavioural observations were included.

To record the behavioural data, I used Prim8 Mobile, an OS-based Android smartphone application designed to help in field behavioural data collection. I recorded the behaviour of the individuals using 5-min focal sampling (Altmann, 1974). The average number of focal samplings per monkey over the three observational phases was 32 (range 24 – 34). I performed the behavioural observations after the animals had been fed and were free to move in their familiar enclosures, between 10:30 and 16:00. The complete ethogram utilised for the behavioural observations can be seen in Chapter 4. Overall, I considered ten behavioural categories: *foraging*, *locomotion*, *affiliative behaviour*, *agonistic behaviour*, *environmental manipulation, vigilance, solitary play, inactivity, human interaction, and behaviours potentially indicative of stress* (BPIS).

5.2.3.2. Behavioural tests

A total of seven behavioural tests based on Uher et al. (2013) and adapted to be used in the wildlife rescue centre by Nunes (2017) were performed in two sets, according to the following timetable:

Table 5.3

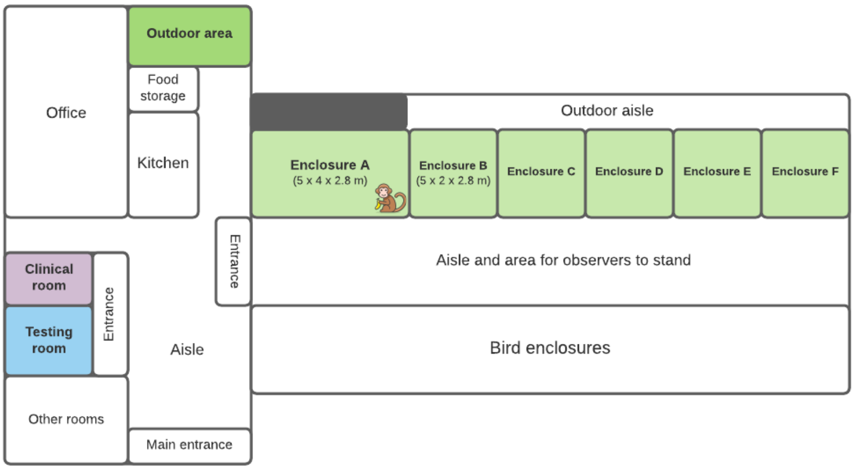
Timetable used for behavioural tests in this study

Phase 1		
Week 1	Days 1, 3, 5	Food neophilia and novel object tests trials 1, 2, 3 respectively
	Days 2 and 4	Hidden food, human interaction, and plush monkey toy tests; trials 1 and 2 respectively
14 days pause[1]		
Phase 2		
Week 2	Days 6, 8, 10	Food neophilia and novel object tests trials 4, 5, 6 respectively
	Day 7	Hidden food, human interaction, and plush monkey toy tests; trial 3
All tunnel tests performed once all groups finished the trials of the above tests[2]		
Week 3	Day 11	Tunnel test
	Day 12	Covered tunnel test

[1] I aimed to perform all behavioural tests with a 14-day pause between blocks of trials; however, this was possible only in three (75%) of the groups studied (see Table 2 above). [2] The tunnel tests were performed to all groups at the end of the rest of the trials, except for group 1, for which we performed these tests several weeks before.

The tests were performed by the author, a master's student, and several undergraduate students. We offered the individual a piece of food (mostly banana) for them to get inside an individual transport cage (made of metal bars) of approximately 1 m^3. The individuals entered this cage voluntarily and all 16 individuals participated as planned. However, one female adult (‘V’) did not complete the tests for reasons unrelated to this study. Thus, I excluded this individual from analysis (corrected n = 15). Once inside the cage, the monkey to be tested was carried to another room (i.e., the testing room) outside the view of other monkeys and any other visual disturbances. Then, the test was performed and the behaviour of the individual was recorded in video. The monkeys were left alone during the tests; however, one of the experimenters was outside the room and entered occasionally to make sure the monkey was not distressed. If the individual was severely distressed the test was cancelled and the individual was returned to his/her enclosure. Figure 5.2 shows the floor plan of the main building of CETAS–RN, in which the testing room was located.

Figure 5.2. Figure showing the floor plan of the main building of CETAS–RN. In each individual behavioural test, the monkey was moved from his/her enclosure in an individual $1m^3$ cage, to which s/he entered voluntarily, to the testing room.



5.2.3.2.1. Description of behavioural tests

1) Measuring ‘food neophilia’: food neophilia tests

In this study, *food neophilia* was defined as the *willingness to try novel food items* (Ristic et al., 2016). In food neophilia tests, we presented a novel (unknown) food item to the individual being tested whilst s/he was inside the testing cage, along with a preferred known food item (a piece of banana or mango) and a less preferred known food item (a piece of chayote, carrot or cucumber). Then, the monkey was recorded for five minutes. We measured the latency (in seconds) for first manipulation and ingestion of these items (novel item, most preferred known food item and less preferred known food item). Moreover, we recorded the diversity of behaviours displayed. For these tests, we used several ‘novel’ food items that were not part of the monkeys' diet in the rescue centre, such as pineapple, avocado and tomato. However, we could not be completely certain these items were unknown to the monkeys as they had lived with humans before being confiscated or surrendered to the rescue centre.

2) Measuring 'creativity': novel objects tests

In this study, the objective was to assess *creativity* by measuring several traits and behaviours related to creative processes (Kaufman et al., 2011; Uher et al., 2013): neophilia, risk-taking and openness (if the animal would touch a novel object, how quickly and for how long) and innovation (if the animal attempted to use the object in different ways, i.e., the diversity of behaviours observed). Thus, in the novel object tests, a novel object was placed inside the cage (in each trial), and the monkey was video-recorded for five minutes. We used several objects that had the potential to elicit some type of manipulation whilst being attractive and safe for the monkeys, such as plastic toys (e.g., pieces of Lego for toddlers) and wooden objects. Unfortunately, the objects were not available for all trials; hence, different objects were presented to different individuals and in different order, which could have influenced the results of these tests.

3) Measuring 'persistence': hidden food tests

We performed 'hidden food tests', in which a plastic bottle filled with paper (e.g., toilet paper, paper towels) with peanuts hidden in the bottom was placed inside the testing cage. In this test, the monkey had to pull out all the paper in order to reach the hidden food. As with the novel object tests, we video-recorded the individual for five minutes. The purpose of this test was to persistence (i.e., *continuing and/or repeating a behaviour or activity*; APA, 2022), including if the animal would attempt to manipulate the plastic bottle in different ways to obtain the peanuts, how many times, and for how long. This was based on Uher et al. (2013). Note that I did not know if the monkey was acquainted with plastic bottles before the first trial, as these animals have been raised by humans as pets. Therefore, some of them could have seen and/or manipulated plastic bottles before.

4) Measuring 'sociability to humans': human interaction test

We assessed social orientation to humans in the 'human interaction test'. In each trial, a person who was known to the animal (e.g., the author) approached the testing cage and said the monkey's name out loud and called him/her in a playful, friendly tone for 30 seconds, inviting them to interact. After this time, the person moved away from the monkey's view for 30 seconds, and then returned offering food (e.g., peanuts) for an additional 30 seconds. After offering the food, the person approached the monkey again in a playful and friendly manner for the last 30 seconds, without offering food. The objective was to assess if the animal approached the familiar person, how quickly, and for how long with and without offering food to them. This was based on Uher et al. (2013) and Nunes (2017).

5) Measuring ‘aggressiveness’ to conspecifics

In the ‘plush monkey toy test’, a soft monkey toy (i.e., a ‘teddy’) was placed approximately one metre away from the testing cage for 60 seconds, with the objective of eliciting a response from the monkey. We recorded the behaviour of the individual as in the previous tests. The objective of this test was to assess the aggressiveness of the individual towards conspecifics. We included agonistic behaviours such as threatening or vocalisations (e.g., screaming), behaviours that were not aggressive or were not directed towards the soft toy, and for how long these were performed. This was based on Uher et al. (2013) and Nunes (2017).

6) Measuring ‘risk-taking’

We performed two type of tests to measure risk-taking: the ‘tunnel test’ and the ‘covered tunnel test’. In the ‘tunnel test’, the monkey was moved (in the testing cage) to an empty enclosure, where the cage was positioned with an open door in front of a wooden tunnel that led to an unknown enclosure. Once the door was removed (and the monkey was free to go inside the enclosure), the individual was recorded for five minutes. The individual could choose to pass through the tunnel and explore the unknown enclosure, with all its behaviours and location recorded on the five-minute video, or could stay inside the testing cage. After the five minutes, the individual was taken back to his/her familiar enclosure. The ‘covered tunnel test’ was similar to the tunnel test, except that in this case, the tunnel was modified with a black cover to simulate an apparent ‘no way out’. The remaining of the test was identical to the tunnel test. We assessed if the individuals passed through the tunnel (and how long it took them to pass through it) and explored the unknown enclosure (and for how long). *Risk-taking* was defined as *performing a behaviour which outcome is uncertain* (Trimpop, 1994). This was based on Uher et al. (2013) and Nunes (2017).

7) Measuring stereotypic behaviour: all tests

We assessed the time spent in motor stereotypies in all test trials. This was performed with the objective of evaluating if the animals became aroused or anxious during the tests, evidenced by time spent in motor stereotypies (e.g., pacing, bouncing).

Table 5.4 summarises the possible results of the tests, which variables were then grouped into ‘**working’ personality constructs defined *a priori*** based on Uher et al. (2013) and Nunes (2017) (Table 5.5). Note that these are **not** the final personality constructs. The final constructs were interpreted after performing the data analysis, including the variable reduction method.

Table 5.4

Variables measured in each type of behavioural test, based on Uher et al. (2013) and Nunes (2017)

Food neophilia	Novel object	Hidden food	Plush monkey toy	Human interaction	Tunnel	Covered tunnel
Latency for first manipulation of new food item (seconds)	Latency for first manipulation (seconds)	Latency for first manipulation (seconds)	Aggressive behaviour (yes/no)	Latency for first approach (seconds)	Latency for first entrance to tunnel (seconds)	Latency for first entrance to tunnel (seconds)
Latency for first manipulation of most preferred known food item (seconds)	Total time manipulating (seconds)	Number of manipulation bouts (number)	Time in aggressive behaviour (seconds)	Duration of first approach (seconds)	Exploration of the unknown enclosure (number of quadrants)	Time spent in motor stereotypies (seconds)
Latency for first manipulation of less preferred known food item (seconds)	Different types of manipulation (number)	Average time of every attempt (seconds)	Affiliative/sexual display (yes/no)	Latency for first approach for food (seconds)	Time exploring (seconds)	
Latency for first ingestion of new food item (seconds)	Average duration of every manipulation bout (seconds)	Total time manipulating (seconds)	Time in affiliative/sexual display (seconds)	Latency for first approach after food (seconds)	Latency for first return through the tunnel (seconds)	
Latency for first ingestion of most preferred known food item (seconds)	Number of manipulation bouts (N)	Time spent in motor stereotypies (seconds)	Time performing other behaviours (not including BPIS) (seconds)	Time spent in motor stereotypies (seconds)	Number of times the individual passed through the tunnel (number)	
Latency for first ingestion of less preferred known food item (seconds)	Time spent in motor stereotypies (seconds)		Time in alert/alarm (seconds)		Time spent in motor stereotypies (seconds)	
Different types of manipulation before ingesting new food item (N)			Time spent in motor stereotypies (seconds)			
Time spent in motor stereotypies (seconds)						

Table 5.5

Description of ‘working’ personality constructs defined *a priori* based on Uher et al. (2013) and Nunes (2017) and corresponding tests, variables and measurement units

‘Working’ personality construct	**Tests**	**Variables measured**	**Measurement unit**
Neophilia/ Creativity	Novel object test	Latency for first manipulation	Seconds
		Total time manipulating	Seconds
		Number of attempts	Seconds
		Diversity of behaviours displayed	Number
Food neophilia	Food neophilia test	Latency for first manipulation of most preferred known food item	Seconds
		Latency for first manipulation of less preferred known food item	Seconds
		Latency for first manipulation of new food item	Seconds
		Diversity of behaviours displayed	Number
Persistence	Hidden food test	Average time (duration) of every attempt	Seconds
Aggressiveness to conspecifics	Plush monkey toy test	Time in aggressive behaviour	Seconds
Social orientation humans	Human interaction test	Latency for first approach without food	Seconds
		Duration of first approach	Seconds
Exploration/ Boldness	Covered tunnel test	Latency for first entrance in the dark tunnel towards unknown enclosure	Seconds
	Tunnel test	Number of different quadrants explored in unknown enclosure	Number
		Latency for first entrance to unknown enclosure	Seconds
Stereotypic	All tests	Time spent performing motor stereotypies	Seconds

5.2.3.3. Observer trait ratings

I used a version of the Hominoid Personality Questionnaire (Weiss et al., 2009) translated to Portuguese to obtain the observer trait ratings. The questionnaire consisted of 54 adjectives that described the individuals, such as ‘friendly’, ‘aggressive’ and ‘decisive’, with a Likert scale of 7 points ranging from 0 (‘the individual does not show any trace of the adjective or shows it in negligible amounts’) to 6 (‘the individual shows an excessive amount of the adjective’). The questionnaire was answered by four raters who knew the animals for at least one year (range 1-2 years). The glossary of the terms used in Portuguese and translated to English and can be seen in Appendix 2.

5.2.4. Data analysis

All statistical analyses as well as figures (graphs) were performed using SPSS 27 (SPSS Inc. Released 2020. SPSS for Windows, Version 27.0. Chicago, SPSS Inc).

5.2.4.1. Normality tests

Schapiro-Wilk tests were performed to investigate whether the data obtained followed a normal distribution. Since most of the results followed a non-normal distribution, I performed non-parametric tests. Hence, results are expressed in median (Mdn) and interquartile range (IQ) if not specified otherwise.

5.2.4.2. Behavioural coding from naturalistic observations

I organised the naturalistic observational data according to the ten behavioural categories obtained from the ethogram: feeding, locomotion, affiliative behaviour, agonistic behaviour, environmental manipulation, vigilance, solitary play, inactivity, human interaction, and behaviours potentially indicative of stress (BPIS) (see Chapter 4). To produce a behavioural profile for every individual, I added the time spent in every behaviour that belonged to the behavioural category of interest e.g., *time spent in locomotion* = time spent in vertical locomotion (seconds) + time spent in horizontal locomotion (seconds) + time spent in suspended locomotion (seconds). Then, a percentage was calculated as follows:

$$\% = \frac{x}{y}\ 100$$

In which x is the time observed in the behavioural category of interest and y is the total time observed. This was performed for each category and observational phase (baseline, second, and final). After this, I calculated the mean between the three observational phases to obtain an overall behavioural profile for every monkey.

5.2.4.3. Behavioural coding from behavioural tests

I analysed the video recordings from the behavioural tests using BORIS software. The variables measured and obtained from the videos can be seen in Table 5.5 above.

5.2.4.3.1. Consistency between trials

I used intraclass correlation analysis (ICC) to analyse the internal consistency between all trials performed for the same type of behavioural test (i.e., novel object, food neophilia, hidden food, plush monkey toy and human interaction) as suggested by Uher et al. (2013) and Nunes (2017). This is performed to assess if the behavioural responses observed in the tests are consistent across time. Variables that were not consistent (i.e., whose intraclass correlation analysis resulted in a non-significant *p*-value) were excluded from further analysis. Moreover, I measured the time spent in behaviours potentially indicative of stress (BPIS) in every trial of the behavioural tests and then analysed the consistency between trials for each type of test. Variables from tests performed only once could not be examined for inter-trial consistency.

5.2.4.3.2. Data reduction and interpretation

Principal Component Analysis (PCA) with varimax rotation was used for data reduction of significantly consistent variables (see Section 2.5.3.1).

5.2.4.4. Personality questionnaires

5.2.4.4.1. Inter-rater reliability

I used intraclass correlation analysis (ICC) to analyse the reliability between raters of the 54 adjectives that were part of the personality questionnaire. ICC(3,1) is used to estimate the reliability of single ratings and ICC(3,*k*) is used to evaluate the reliability across mean ratings, based on *k* number of raters (Shrout and Fleiss, 1979; Robinson et al., 2016). I considered as acceptable all the results with ICC(3,*k*) significant levels i.e., $p < 0.05$.

5.2.4.4.2. Data reduction and interpretation

I calculated the mean between raters for every adjective considered as reliable (see above section). The results were then analysed using Principal Component Analysis (PCA) as performed in previous capuchin studies (e.g., Morton et al., 2013; Manson and Perry, 2013; Fernández-Bolaños et al., 2020). The components were rotated with Varimax and Promax procedures, with loadings ≥ 0.40 considered as salient. If an item was part of more than one component, it was assigned to the component with the highest loading, following the methodology by Morton et al. (2013). I interpreted the results based on previous studies on robust capuchin monkey personality, mainly Morton et al. (2013) and Fernández-Bolaños et al (2020).

5.2.4.5. Correlations between personality assessment methods

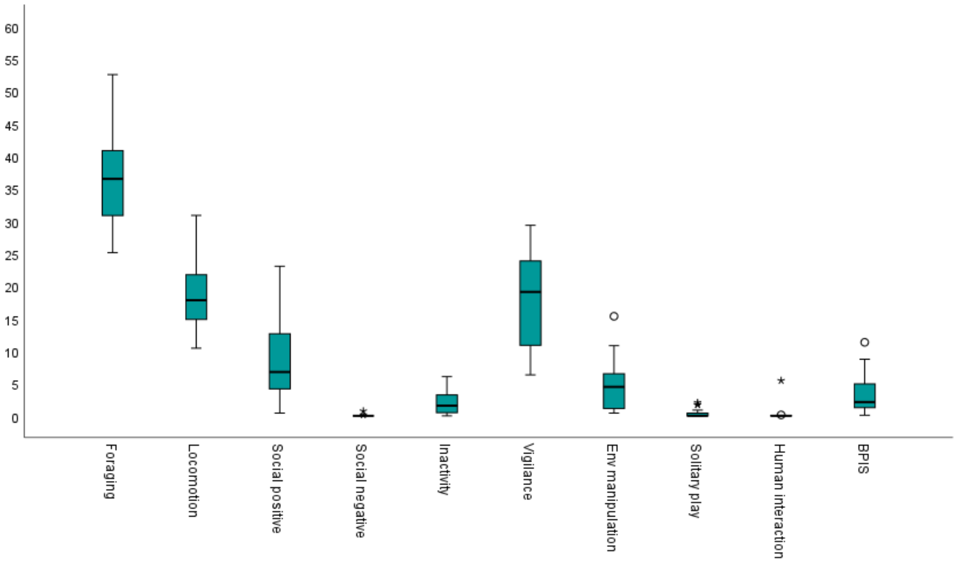
Spearman correlations were performed between the behavioural profiles, behavioural tests, and personality questionnaires. I used Holm-Bonferroni corrections as correction factors for multiple comparisons.

5.3. Results

5.3.1. Behavioural coding in naturalistic context: behavioural profiles

Overall, the individuals spent 36.51% (Mdn) of the time foraging (IQ = 10.27), 17.80% in locomotion (IQ = 7.26), 6.77% in affiliative behaviours (IQ = 8.65), 4.46% in environmental manipulation (IQ = 5.51), 19.10% in vigilance (IQ = 13.26), 1.57% in inactivity (IQ = 2.90), and 2.12% in BPIS (IQ = 3.68). Time spent in solitary play and human interaction was noticeably low (Mdn = 0.00, IQ = 0.54 and Mdn = 0.00, IQ = 0.06, respectively), as well as time spent in agonistic behaviours, which was almost negligible (Mdn = 0.00, IQ = 0.00). Figure 5.2 shows a graph of the overall behavioural profiles calculated.

Figure 5.2. Boxplot showing the percentage of time (%) spent in each of the behavioural categories considered.



5.3.2. Behavioural tests

5.3.2.1. Consistency between test trials

The results from the intraclass correlation analyses to assess the internal consistency between trials can be seen in Table 5.6. Note that only the variables in **bold** had significant levels of inter-trial consistency; hence, the rest of the variables were excluded from further analysis (i.e., because they were not consistent between trials). Even though the variable *latency for first manipulation of most preferred known food item* had a significant level of inter-trial consistency, I decided to exclude it because the variables *latency for first manipulation of most preferred known food item* and *latency for first manipulation of new food item* did not have a significant level of (inter-trial) consistency. This last variable (*latency for first manipulation of new food item*) was the variable of interest to measure ‘food neophilia’.

The variables (1) *latency for first entrance in the dark tunnel towards unknown enclosure*, (1) *number of different quadrants explored in unknown enclosure*, (3) *latency for first entrance to unknown enclosure* and (4) *time spent in stereotypic behaviour* were included because inter-trial consistency could not be measured i.e., as only **one** trial of the tunnel and covered tunnel tests were performed.

Table 5.6

Consistency across trials (ICC 2,*k*) of variables measured in the behavioural tests

Test	**Variable**	**ICC (2,*k*)**	***p***	**Inclusion in further analyses**
Novel object	Latency (time) for first manipulation	0.35	0.118	No
	Total time manipulating	**0.77**	**<0.001***	**Yes**
	Number of attempts	**0.75**	**<0.001***	**Yes**
	Diversity of behaviours displayed	**0.80**	**<0.001***	**Yes**
	Average time (duration) of every attempt	0.36	0.113	No
Food neophilia	Latency (time) for first manipulation of most preferred known food item	0.84	<0.001*	No
	Latency (time) for first manipulation of less preferred known food item	0.12	0.342	No
	Latency (time) for first manipulation of new food item	0.32	0.154	No
	Diversity of behaviours displayed	**0.53**	**0.016***	**Yes**
Hidden food	Average time (duration) of every attempt	-0.20	0.637	No
Plush monkey toy	**Time spent in aggressive behaviour**	**0.99**	**<0.001***	**Yes**
	Time spent in other behaviours not directed towards the soft monkey toy (not including BPIS)	**0.77**	**<0.001***	**Yes**
Human interaction	**Latency (time) for first approach without food**	**0.69**	**0.005***	**Yes**
	Duration of first approach	**0.82**	**<0.001***	**Yes**
Covered tunnel	Latency (time) for first entrance in the dark tunnel towards unknown enclosure	NA	NA	Yes
Tunnel	Number of different quadrants explored in unknown enclosure	NA	NA	Yes
	Latency (time) for first entrance to unknown enclosure	NA	NA	Yes

In Table 5.7, the results from the intraclass correlation analysis of time spent in motor stereotypies) are shown.

Table 5.7

Inter-trial consistency of time spent in motor stereotypies (MS) during the behavioural tests

Test	**(ICC 2,*k*)**	***P***	**Inclusion in further analyses**
Novel object	0.95	<0.001	Yes
Food neophilia	0.94	<0.001	Yes
Hidden food	0.89	<0.001	Yes
Plush monkey toy	0.87	<0.001	Yes
Human interaction	0.79	<0.001	Yes
Tunnel	NA	NA	Yes
Covered tunnel	NA	NA	Yes

5.3.2.2. Data reduction

Principal Component Analysis (PCA) was used for data reduction of the variables which were significantly consistent across all trials, the three final variables which were measured only once and time spent in motor stereotypies (total = 18 variables). Figure 5.3 shows the scree plot obtained.

Figure 5.3. Scree plot obtained from the PCA of the 18 variables selected from the behavioural tests.

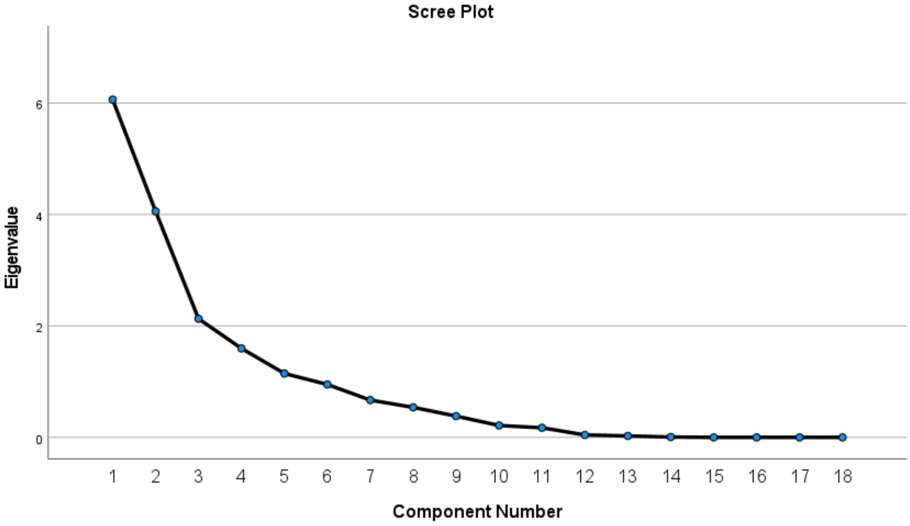


Table 5.8 shows the variance of the components obtained from the analysis.

Table 5.8

Variance of components obtained from the Principal Component Analysis (PCA) from the behavioural tests

	Initial Eigenvalue		Rotated Square Loadings	
Component	Total	Variance (%)	Total	Variance (%)
1	6.065	33.693	5.636	31.310
2	4.059	22.550	2.998	16.653
3	2.130	11.833	2.715	15.086
4	1.597	8.872	2.220	12.332
5	1.147	6.373	1.429	7.940

The structure matrix of the rotated component loadings can be seen in Table 5.9 in the next page.

Table 5.9

Structure Matrix of Varimax Rotated Component Loadings

	Component				
	1	2	3	4	5
Time spent in MS during 'plush monkey toy test' trials	0.971				
Time spent in MS during 'hidden food test' trials	0.899				
Time spent in MS during 'novel object test' trials	0.897				
Time spent in MS during 'human interaction test' trials	0.857				
Time spent in MS during 'food neophilia' trials	0.823				
Time spent in other behaviours not directed towards the soft monkey toy (mean across trials)[1]	-0.774				
Time spent in MS during 'covered tunnel test'	0.692				
Latency to enter the covered tunnel	-0.498				
Number of attempts manipulating object (mean across trials)		0.873			
Total time manipulating object (mean across trials)		0.864			
Diversity of object manipulation behaviours observed (mean across trials)		0.789			
Diversity of food manipulation behaviours observed (mean across trials)		0.697			
Latency to enter tunnel			0.957		
Number of quadrants explored in unknown enclosure			-0.763		
Time spent in MS during 'tunnel' trials			0.734		
Latency to approach familiar human (mean across trials)				-0.879	
Time in proximity to familiar human (mean across trials)				0.859	
Time in aggressive behaviour towards soft monkey toy (mean across trials)					0.793

[1] Time spent in other behaviours did not include time spent engaged in motor stereotypies.

5.3.2.3. Component interpretation

Component 1 was characterised positively by time spent in motor stereotypies during six of the seven types of tests (all tests except tunnel test), and negatively by time spent in other behaviours not directed towards the soft monkey toy (in the plush monkey toy tests) and latency to enter the covered tunnel (in the covered tunnel test). I named it 'stereotypic'. Thus, an individual with a higher score would be 'more stereotypic' and an individual with a lower score would be 'less stereotypic'. Component 2 was characterised positively by more attempts at manipulating objects, more time spent manipulating objects, and a higher diversity of behaviours observed when presented with novel objects and food items. I named it 'creative', following the model of animal creativity of Kaufman et al. (2011) and the behaviours used in the Behavioural Repertoire x Environmental Situations Approach from Uher et al. (2013). Individuals with higher scores would be considered as 'more creative' and individuals with lower scores as 'less creative'. Note that even though I intended to measure latency to manipulate novel objects and food items, these variables were not consistent between trials; hence, they were excluded from further analyses. Because of this, the labels 'neophilia' and ‘food neophilia’ were not used in this study, even though neophilia is considered as part of creativity (Kaufman et al., 2011).

Component 3 was characterised positively by larger latencies to enter the tunnel and more time spent in motor stereotypies during the tunnel test. Moreover, this factor was characterised negatively by the number of quadrants explored. I named this component ‘risk-averse’. Therefore, monkeys with higher scores would be ‘more risk-averse’ and monkeys with lower scores would be ‘less risk-averse’. Component 4 was characterised positively by more time spent in close proximity to humans and negatively by larger latencies to approach a familiar human; thus, I named it ‘sociable to humans’. Animals with higher scores would be ‘more sociable to humans’. Conversely, animals with lower scores would be ‘less sociable to humans’. Finally, Component 5 was characterised positively by more time being aggressive to the plush monkey toy. Hence, I named it ‘aggressive’. Animals with higher scores would be ‘more aggressive’, and animals with lower scores would be ‘less aggressive’.

5.3.3. Observer trait ratings

5.3.3.1. Inter-rater reliability

The results of the intraclass correlations (ICCs) to assess the inter-rater reliability of the adjectives that composed the Hominoid Personality Questionnaire can be seen in Table 5.10.

Table 5.10

Inter-rater reliability of the 54 adjectives that compose the Hominoid Personality Questionnaire

Adjective (Portuguese)	**Adjective (English)**	**ICC (3,1)**	**ICC (3,*k*)**	***P***
Brincalhão	Playful	0.763	0.928	<0.001
Vulnerável	Vulnerable	0.695	0.901	<0.001
Dominante	Dominant	0.685	0.897	<0.001
Ativo	Active	0.636	0.875	<0.001
Submisso	Submissive	0.629	0.872	<0.001
Depressivo	Depressed	0.627	0.871	<0.001
Imitativo	Imitative	0.570	0.842	<0.001
Ansioso	Anxious	0.543	0.826	<0.001
Medroso	Fearful	0.534	0.821	<0.001
Desafiante	Defiant	0.517	0.811	<0.001
Solitário	Solitary	0.516	0.810	<0.001
Tranquilo	Cool	0.513	0.808	<0.001
Estereotipado	Stereotypic (Autistic)	0.492	0.795	<0.001
Dependente/Seguidor	Dependent/Follower	0.490	0.794	<0.001
Curioso	Curious	0.481	0.788	<0.001
Agressivo	Aggressive	0.470	0.780	<0.001
Afetuoso	Affectionate	0.445	0.762	<0.001
Desajeitado	Clumsy	0.438	0.757	<0.001
Preguiçoso	Lazy	0.431	0.752	<0.001
Descuidado	Thoughtless	0.428	0.750	<0.001
Inconsequente	Reckless	0.423	0.746	<0.001
Sensível	Sensitive	0.393	0.721	<0.001

Table 5.10 (continued)

Inter-rater reliability of the 54 adjectives that compose the Hominoid Personality Questionnaire

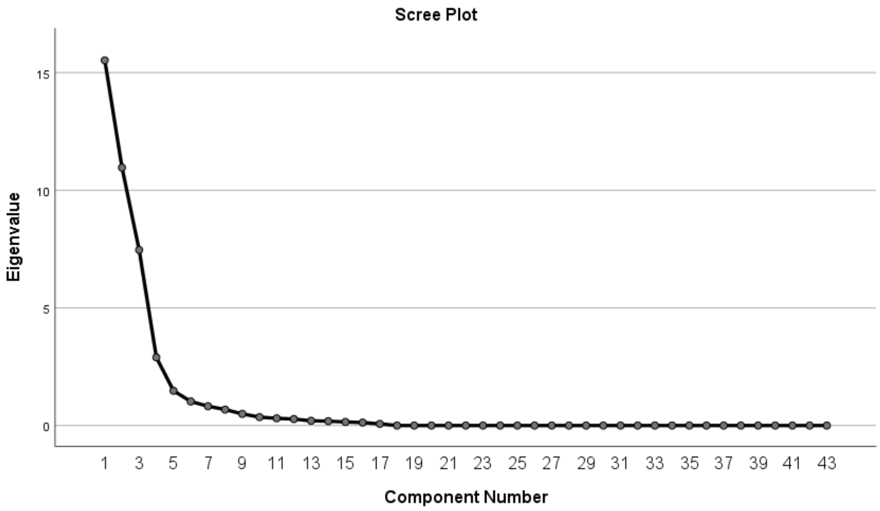
Inventivo/Criativo	Inventive	0.385	0.715	<0.001
Impulsivo	Impulsive	0.385	0.714	<0.001
Prestativo	Helpful	0.373	0.704	<0.001
Sociável	Sociable	0.366	0.697	<0.001
Avarento/Ganancioso	Stingy/Greedy	0.356	0.688	0.001
Bullying	Bullying	0.352	0.685	0.001
Inquisitivo	Inquisitive	0.347	0.680	0.001
Protetor	Protective	0.326	0.660	0.002
Amigável	Friendly	0.318	0.651	0.002
Ciumento	Jealous	0.300	0.632	0.003
Irritável	Irritable	0.291	0.621	0.004
Persistente	Persistent	0.282	0.611	0.005
Cauteloso	Cautious	0.272	0.600	0.006
Excitável	Excitable	0.257	0.580	0.009
Gentil	Gentle	0.255	0.578	0.009
Decidido	Decisive	0.250	0.572	0.010
Errático	Erratic	0.250	0.571	0.010
Complacente	Sympathetic	0.248	0.568	0.011
Independente	Independent	0.239	0.557	0.013
Estável	Stable	0.214	0.522	0.022
Tímido	Timid	0.197	0.495	0.031
Inovador	Innovative	0.150	0.413	0.073
Insensível	Unemotional	0.135	0.385	0.092
Desorganizado	Disorganised	0.132	0.379	0.096
Manipulador	Manipulative	0.124	0.361	0.110
Convencional	Conventional	0.063	0.212	0.250
Previsível	Predictable	0.023	0.085	0.386
Individualista	Individualistic	0.021	0.081	0.390
Distraído	Distractible	-0.005	-0.019	0.493
Desapercebido	Unperceptive	-0.008	-0.032	0.505
Inteligente	Intelligent	-0.046	-0.211	0.657
Desistente	Quitting	-0.087	-0.471	0.807

Indicators *k* = 4.

5.3.3.2. Data reduction

Figure 5.4 in the next page shows the scree plot obtained from the Principal Component Analysis (PCA) of the 43 reliable adjectives.

Figure 5.4. Scree plot obtained from the PCA of the 43 reliable adjectives from the Hominoid Personality Questionnaire.



The first six components had eigenvalues ≥ 1.0 and explained more than 90% of the rotated variance, as observed in Table 5.11:

Table 5.11

Variance of components obtained from the Principal Component Analysis (PCA) of reliable HPQ items

	Initial Eigenvalue		Rotated Square Loadings	
Component	Total	Variance (%)	Total	Variance (%)
1	15.522	36.097	12.149	28.255
2	10.964	25.497	8.679	20.185
3	7.461	17.350	8.385	19.500
4	2.896	6.734	6.509	15.137
5	1.476	3.432	2.068	4.809
6	1.023	2.378	1.550	3.605

I excluded components 5 and 6 from further analysis as their rotated variance was low (less than 5%) and they were only composed by two and four adjectives, respectively, whose loadings were lower than 0.50. Furthermore, those adjectives were already part of the first four components. Thus, components 1, 2, 3 and 4 jointly explained more than 80% of the total variance observed. The structure matrix of the Varimax and Promax rotated component loadings can be seen in Table 5.12 in the following page. As the correlations between Promax-rotated components were weak or moderate at best (Table 5.13), which is related to significant orthogonality between components (Fernández-Bolaños et al., 2020), I kept the Varimax-rotated components for analysis instead, as suggested in previous capuchin monkey personality studies (Morton et al., 2013; Fernández-Bolaños et al., 2020).

5.3.3.3. Component interpretation

The first component was characterised by adjectives that had positive loadings on behaviours related to exploration, creativity and high energy expenditure; and by adjectives with negative loadings describing low energy expenditure, vigilance, and anxiety/depression. Since this component was considerably similar to the Openness dimension described by Morton et al. (2013) for brown capuchin monkeys (*Sapajus apella*), I labelled it the same. The second component was loaded positively by items that described self-control, stability and independence, and negatively by items describing fearfulness and vulnerability. If I reflected these items by multiplying each item by –1 as performed by Morton et al. (2013), this dimension and their dimension Neuroticism would be similar to each other, sharing several items, particularly those related to emotional stability (e.g., cool, stable). Therefore, I gave it the same name, ‘Neuroticism’. The third component was loaded positively by adjectives that described aggressive behaviour, dominance, and impulsivity. This was also considerably similar to one capuchin personality dimension proposed by Morton et al. (2013); hence, it was given the same name: ‘Assertiveness’. Finally, the fourth component was loaded positively by items that described positive social skills and friendliness/amicableness. In a similar way to the first and third components, it resembled a capuchin personality dimension proposed by Morton et al. (2013) and was given the same name, ‘Sociability’. Table 5.12 in the next page shows the structure matrix of Varimax and Promax rotated component loadings from the PCA, obtained from analysis of the reliable items from the Hominoid Personality Questionnaire.

Table 5.12

Structure Matrix of Varimax and Promax Rotated Component Loadings from the PCA obtained from analysis of reliable HPQ items

Item	Varimax-rotated components				Promax-rotated components			
	OP[1]	**NE**	**AS**	**SO**	**OP**	**NE**	**AS**	**SO**
Active	**0.925**	0.183	-0.053	0.009	**0.931**	0.171	-0.223	-0.123
Affectionate	0.376	0.304	0.079	**0.708**	0.114	0.124	0.246	**0.655**
Aggressive	0.131	0.414	**0.811**	-0.328	-0.002	0.220	**0.815**	-0.213
Anxious	**-0.792**	-0.344	-0.143	-0.141	**-0.720**	-0.232	-0.093	-0.054
Bullying	0.106	0.255	**0.775**	-0.322	-0.127	-0.006	**0.878**	-0.244
Cautious	-0.423	**-0.550**	-0.370	0.329	-0.347	**-0.473**	-0.207	0.382
Clumsy	**0.652**	-0.335	-0.093	-0.194	**0.665**	-0.242	-0.328	-0.265
Cool	0.062	**0.690**	-0.508	0.185	0.033	**0.782**	-0.660	-0.049
Curious	**0.910**	0.008	0.153	0.125	**0.994**	0.009	0.004	0.135
Decisive	0.411	**0.650**	0.496	-0.119	0.400	**0.614**	0.294	-0.076
Defiant	0.143	0.339	**0.896**	-0.131	-0.022	0.150	**0.906**	0.042
Dependent	**0.632**	-0.543	-0.046	0.166	0.404	**-0.668**	0.046	0.074
Depressed	**-0.694**	-0.485	-0.241	-0.276	**-0.542**	-0.353	-0.201	-0.211
Dominant	0.019	**0.738**	0.616	0.056	-0.089	**0.641**	0.545	0.157
Erratic	-0.178	-0.107	**0.693**	-0.066	-0.117	-0.162	**0.736**	0.218
Excitable	**0.855**	0.115	0.230	0.030	**0.757**	0.109	-0.003	-0.013
Fearful	-0.041	**-0.913**	-0.273	-0.036	-0.008	**-0.965**	-0.019	-0.036
Friendly	0.344	-0.124	-0.230	**0.834**	0.280	-0.144	-0.114	**0.828**
Gentle	0.063	-0.051	-0.558	**0.751**	0.209	0.058	-0.485	**0.723**
Greedy	0.172	0.398	**0.794**	-0.251	-0.030	0.214	**0.767**	-0.154
Helpful	0.189	-0.064	-0.280	0.875	0.165	-0.047	-0.170	0.884
Imitative	**0.832**	-0.389	-0.116	0.020	**0.962**	-0.415	-0.117	-0.024
Impulsive	0.645	0.041	**0.656**	-0.093	0.551	-0.085	**0.577**	0.012
Independent	-0.324	**0.752**	0.362	-0.108	-0.283	**0.754**	0.268	-0.027
Inquisitive	**0.952**	-0.035	0.165	0.166	**0.931**	-0.093	0.071	0.142
Inventive	**0.922**	0.185	0.144	0.232	**0.912**	0.133	0.034	0.194
Irritable	0.049	0.361	**0.858**	-0.188	-0.071	0.218	**0.825**	-0.009
Jealous	0.319	0.065	**0.838**	-0.266	0.152	-0.145	**0.869**	-0.127
Lazy	**-0.856**	0.304	-0.191	-0.132	**-0.846**	0.431	-0.251	-0.144
Persistent	0.331	**0.519**	0.469	-0.480	0.313	**0.540**	0.159	-0.471
Playful	**0.911**	-0.160	-0.076	0.012	**0.979**	-0.214	-0.108	-0.075
Protective	-0.209	0.568	-0.110	**0.730**	-0.318	0.571	-0.063	**0.722**
Reckless	**0.671**	0.129	0.655	-0.163	**0.644**	0.026	0.538	-0.055
Sensitive	0.007	-0.261	-0.201	**0.839**	-0.090	-0.360	0.105	**0.868**
Sociable	0.589	0.261	0.075	**0.719**	0.447	0.158	0.133	**0.701**
Solitaire	**-0.714**	-0.283	-0.049	-0.293	**-0.505**	-0.177	-0.001	-0.166
Stable	-0.060	**0.893**	-0.034	0.145	-0.060	**0.916**	-0.145	0.067
Stereotypic	-0.343	-0.239	0.264	-0.026	**-0.494**	-0.184	0.176	0.089
Submissive	-0.003	**-0.919**	-0.322	0.060	0.101	**-0.901**	-0.138	0.088
Sympathetic	-0.011	0.118	-0.396	**0.873**	0.015	0.181	-0.308	**0.864**
Thoughtless	**0.715**	0.044	0.414	-0.284	**0.782**	0.046	0.190	-0.229
Timid	**-0.556**	-0.547	-0.288	-0.395	-0.432	**-0.520**	-0.126	-0.399
Vulnerable	-0.032	**-0.901**	-0.317	0.020	0.109	**-0.853**	-0.170	0.061

[1] OP = Openness, NE = Neuroticism, AS = Assertiveness, SO = Sociability.

Table 5.13

Component Correlation Matrix obtained (Promax rotation)

Component	Openness	Neuroticism	Assertiveness	Sociability
Openness	1.000	0.109	0.363	0.074
Neuroticism		1.000	0.453	0.041
Assertiveness			1.000	-0.302
Sociability				1.000

5.3.4. Correlations between personality assessment methods

5.3.4.1. Behavioural profiles obtained from observations (BO) and behavioural tests (BT)

There were negative correlations between time spent in affiliative behaviour (BO) and the trait risk-averse (BT) (r_s = -0.721, n = 15, p = 0.002, p' = 0.257) (*p*' = *p*-value obtained after performing Holm-Bonferroni sequential correction). Moreover, there were positive correlations between time spent in human interaction (BO) and the trait aggressive (BT) (r_s = 0.642, n = 15, *p* = 0.010, *p*' = 1.000), time spent in environmental manipulation (BO) and the trait sociable to humans (BT) (r_s = 0.600, n = 15, *p* = 0.018, p' = 1.000) and time spent in vigilance behaviours (BO) and the trait risk-averse (BT) (r_s = 0.554, n = 15, *p* = 0.032, *p*' = 1.000). None of these correlations were significant after performing Holm-Bonferroni sequential correction (see Appendix 3 for non-significant *p*-values).

5.3.4.2. Behavioural tests (BT) and personality questionnaires (HPQ)

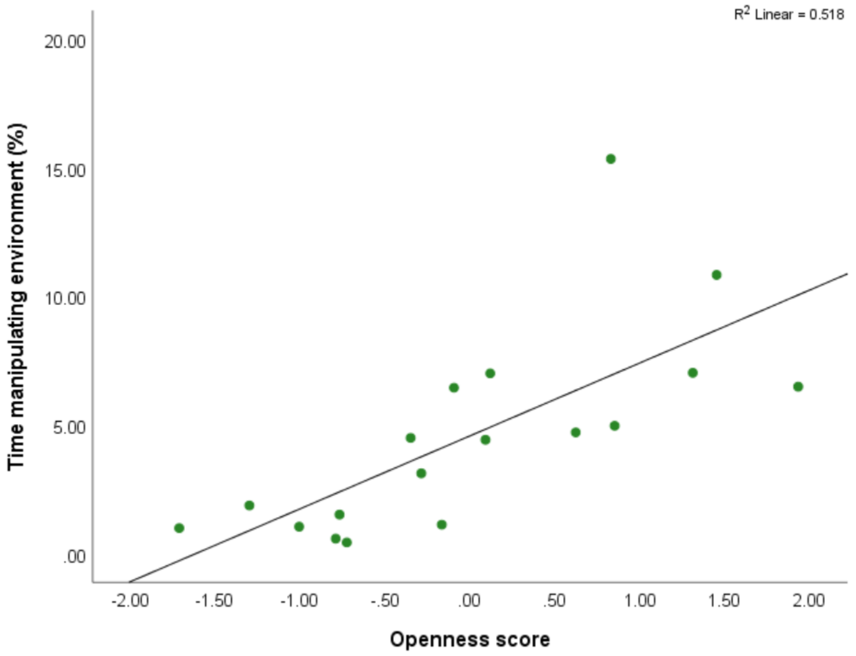
There was a negative correlation between the trait stereotypic (BT) and the trait assertive (HPQ) (r_s = -0.518, n = 15, *p* = 0.048). This correlation was not significant after applying Holm-Bonferroni sequential correction (*p*' = 1.000).

5.3.4.3. Behavioural profiles obtained from observations (BO) and personality questionnaires (HPQ)

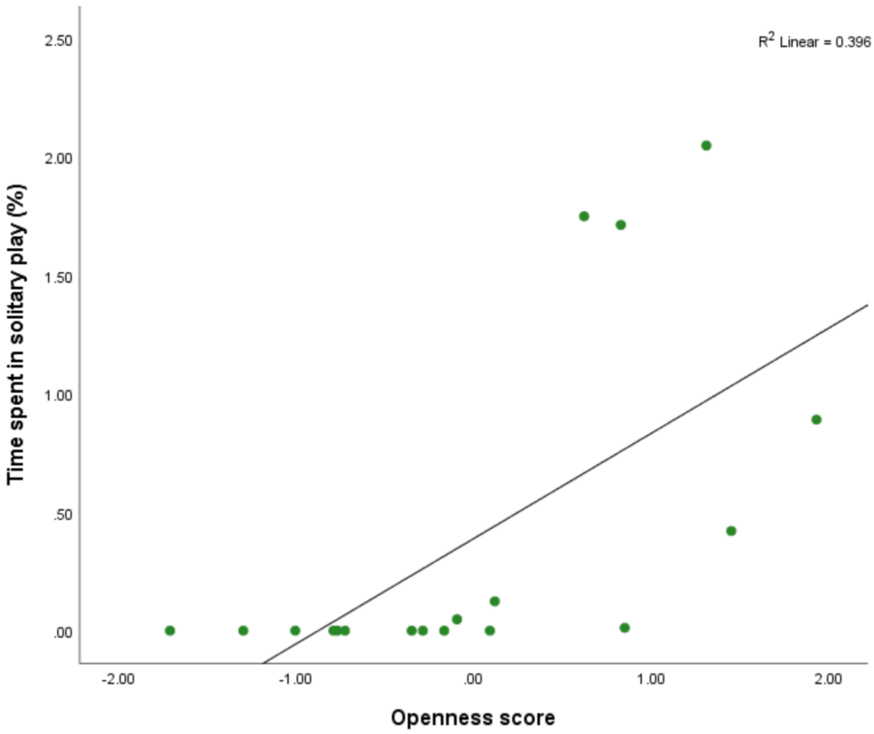
There was a significant positive correlation between the Openness scores of the individuals (HPQ) and time spent manipulating the environment (BO) (r_s = 0.843, n = 18, *p* < 0.001, *p*' = 0.001) and time spent in solitary play (BO) (r_s = 0.828, n = 18, *p* < 0.001, *p*' = 0.002) after applying Holm-Bonferroni sequential correction (Figure 6). I did not find any other significant correlations between the other personality components (i.e., Assertiveness, Neuroticism, and Sociability) and the behavioural profiles of the individuals.

Figure 5.5. Scattergrams showing the significant positive correlation between Openness scores and (a) time spent manipulating the environment and (b) time spent in solitary play.

(a)



(b)



5.4. Discussion

5.4.1. Behavioural observations

The profiles obtained from the behavioural observations were similar to those described by other authors for bearded capuchin monkeys (*Sapajus libidinosus*). Over several studies, bearded capuchins have been found spending most of their time foraging (between 27 and 50% of the time observed), followed by time spent in locomotion (11 to 41%), vigilance (<5 to 25%) and affiliative behaviours (4 to 10%). Furthermore, time spent in inactivity (resting), solitary play, environmental manipulation, agonistic behaviours, and stress-related behaviours was less than 10% for each category in all studies (Vilela, 2003; Moura, 2004; Sabbatini et al., 2008; Ferreira et al., 2016; Nunes, 2017; Ferreira et al., 2018). The behavioural profiles of the monkeys in the present study fall within these ranges for the categories considered albeit the context was different from previous studies. In this study, the monkeys were born in the wild, raised by humans as pets and transferred to a rescue centre, whilst several of the previous studies were performed with free-ranging and captive-born bearded capuchin monkeys.

5.4.2. Behavioural tests

Based on the behavioural tests, I identified five capuchin personality constructs: ‘stereotypic’, ‘creative’, ‘risk-averse’, ‘sociable to humans’, and ‘aggressive’. In comparison, Nunes (2017) described nine capuchin personality constructs based on the same behavioural tests, broadly based on Uher et al. (2013): Curiosity/Playfulness, Food Neophilia, Creativity, Persistence, Aggressiveness, Distractibility, Sociability to Humans, Boldness, and Exploration (Nunes, 2017). The differences observed in the constructs obtained in both studies may be related to several issues. For a start, the analysis of the present study considered less variables as consistent between trials when compared to Nunes (2017). Nunes (2017) found an ICC(2,*k*) >0.70 in 13/14 variables, whereas I found an ICC(2,*k*) >0.70 in 9/14 variables. The variables (1) latency for first manipulation of novel object, (2) latency for first manipulation of most preferred known food item, (3) latency for first manipulation of less preferred known food item, (4) latency for first manipulation of novel food item, (5) average duration of every attempt (novel object test) and (5) average duration of every attempt (hidden food test) ICC 2,*k* results were <0.70 and had non-significant *p*-values. Because fewer variables were deemed as consistent across time, fewer personality constructs characterised the personality of the capuchins in the present study (n = 5), obtained by using behavioural tests, when compared to Nunes (n = 9) (2017).

Furthermore, the present study used Principal Component Analysis to extract the variables into components i.e., the final personality constructs. In comparison, Nunes (2017) did not use a reduction factor method; instead, they used pre-defined constructs, based on those described by Uher et al. (2013) as their final constructs. This is called an expert-based method. Interestingly, previous studies have found similar animal personality traits using expert-based methods, Principal Component Analysis and Factor Analysis (Mazzamuto et al., 2019). Indeed, the personality constructs obtained in the present study do resemble some of those described by Nunes (2017). I also found similarities between the personality constructs obtained via the Behavioural Repertoire x Environmental Situations Approach in brown capuchins (*Sapajus apella*) by Uher et al. (2013) and the personality constructs obtained in the present study (Table 5.14). This could support the existence of these constructs across robust capuchin species (*Sapajus* spp.).

However, both the present study and Nunes (2017) included behavioural tests based on Uher et al. (2013), in which constructs and/or variables measured were defined *before* performing the behavioural tests using a different species (*Sapajus apella*) and not the species used in this study (*Sapajus libidinosus*). Thus, the results from these studies could resemble the results from Uher et al. (2013) mainly because they were based on it. In other words, they could be biased towards certain personality traits observed in brown capuchins and simultaneously overlooking other personality traits that may be ecologically relevant for bearded capuchins (and were not measured). Further studies could include behavioural tests specifically designed for bearded capuchin monkeys and their most ecologically relevant behavioural traits.

Table 5.14

Personality constructs found in different studies for robust capuchin monkeys (*Sapajus* spp.) using the Behavioural Repertoire x Environmental Situations Approach based on Uher et al. (2013)

Uher et al., 2013 (*Sapajus apella*, n = 26)	Nunes, 2017 (*Sapajus libidinosus*, n = 13)	Present study (*Sapajus libidinosus*, n = 15)
Curiousness, Creativeness/Inventiveness	Curiosity/Playfulness	Creative
Arousability, Anxiousness	–	Stereotypic
Social orientation to humans	Sociability to humans	Sociable to humans
Aggressiveness	Aggressiveness	Aggressive

Another aspect to consider regarding the behavioural tests is if the personality traits which were supposedly being assessed were actually identified. For example, if food neophilia tests were actually measuring ‘the willingness to try novel foods’ (Ristic et al., 2016). Carter et al. (2013) have argued that animal personality definitions are not always consistent, and that (personality) tests sometimes measure too many traits, or that a single trait is measured in too many tests, which leads to confusing or inconsistent results (Carter et al., 2013). In the present study, food neophilia was measured by providing ‘novel’ food items to the subjects. However, as the monkeys were kept as pets by people before being received by the rescue centre, I did not know if they were completely naïve to the ‘novel’ food items offered. There was the possibility that they had tried these food items before, which could have influenced their food preferences (Heuberger et al., 2020). Furthermore, different fruits were available for different trials; thus, some of the monkeys did not get the same novel food items as the other monkeys (e.g., an individual could have been offered carrot as *less preferred food item* in most of the trials and another individual could have been offered cucumber instead). This may have influenced the results of the food neophilia tests as well, as different fruits/vegetables have different taste and macronutrient contents, which has been found to influence capuchin food preferences (Heuberger et al., 2020).

In general, capuchins and other primates are considered as *food neophobic*, meaning they tend to be cautious when encountering novel food items (Sabbatini et al., 2007; Heuberger et al., 2020). However, capuchins, like humans, are omnivores –they eat a variety of foods including fruits, small insects, etc.– which means they must balance the risks of eating novel (unknown) foods with the benefits this may provide; mainly, a nutritionally rich and varied diet. This is known as the ‘omnivore dilemma’ (Rizer, 1976 in Heuberger et al., 2020). In addition to this, the food preferences of capuchin monkeys can be influenced by their past experiences and the macronutrient content of novel foods. In fact, capuchins tend to select novel food items which are high in fat, but not when they have a high sugar content (Heuberger et al., 2020). Because of these reasons, assessing food preferences –and how this relates to individual differences in personality– can be difficult in capuchin monkeys.

Moreover, ‘food neophilia’ and ‘food neophobia’ may be seen as opposing extremes of the same construct (Lenglet, 2018) or as different, separate constructs (Sabbatini et al., 2007), which further complicates measuring personality traits related to food preferences. In this study, I could not measure ‘food neophilia’ albeit I performed ‘food neophilia tests’ because the variables *latency for first manipulation of most preferred known food item, latency for manipulation less preferred known food item* and most importantly *latency for first manipulation of new food item* were not consistent between trials (Table 5.7) and were excluded from further analyses. Thus, I considered I did not have consistent data to score the individuals as more or less ‘food neophilic’.

Regarding the assessment of *creativity*, I used a behavioural test modified by Nunes (2017) for its use in the wildlife rescue centre (where both studies were performed; the present study and Nunes, 2017) and based on the Behavioural Repertoire x Environmental Situation Approach by Uher et al. (2013). I utilised the neurobiological model of animal creativity proposed by Kaufman et al. (2011) as a base to interpret the results obtained. According to this model, animal creativity is composed by a distinct set of cognitive processes. These processes include (1) understanding that an experience or object is novel, known as *novelty recognition*; (2) *novelty seeking*, which includes *neophilia* (i.e., a willingness to try novel objects or experiences; Lenglet, 2018), certain risk-taking behaviours, and openness to new experiences; (3) *observational learning*, which involves adding a novel behaviour to the animal's own behavioural repertoire, learned by observing others; and (4) *innovation*, or focusing on the product of the creative process i.e., a novel use (Kaufman et al., 2011). These processes require complex cognitive abilities and measure personality traits highly related to creativity, namely risk-taking, novelty recognition and seeking, and openness (Kaufman et al., 2011). Indeed, the variables which characterised this trait i.e., 'creative' (*number of attempts manipulating object, total time manipulating object*, *diversity of object manipulation behaviours observed* and *diversity of food manipulation behaviours observed*), reflected creative cognitive processes and behaviours and creativity-related personality traits as described by Kaufman et al. (2011), such as innovation (e.g., diversity of behaviours) and novelty seeking (e.g., attempts at manipulating objects) (Kaufman et al., 2011).

The 'hidden food tests' were performed to assess *persistence*. Note that I did not know if the monkeys were acquainted with the objects to measure 'persistence' in these tests (i.e., plastic bottles and toilet paper) as these animals had been raised as pets by humans. Therefore, some of them could have seen and/or manipulated plastic bottles before. Thus, it was unclear if I was actually measuring persistence, or if, in reality, I was measuring 'how well the monkeys were acquainted with and/or learning to use a human plastic bottle'. Furthermore, the variable measured in the trials of this test (*mean duration of every attempt at manipulating object*) was not consistent between trials and I excluded it from further analysis. Because of this, 'persistence' was not considered as one of the final personality traits, derived from behavioural tests, measured in this study.

The 'plush monkey toy test' aimed to investigate *aggressiveness* by measuring time spent in aggressive behaviours (towards a soft monkey toy), such as screaming or threatening. This test was based on Uher et al. (2013) and Nunes (2017). In the original test (Uher et al., 2013), the soft toy's eyes were covered to decrease the perception of threat. Conversely, I did not cover the eyes of the soft toy. Only adult males reacted aggressively in these tests in the present study. However, it is unclear how much these soft toys were perceived as a threat by these animals (i.e., because I did not cover the eyes of the soft toys). It could be the case that the adult males were only 'defending' themselves to what they perceived as a threat rather than intending to attack the soft toy. Further tests could be made, this time with the soft toy's eyes covered and/or with an unknown conspecific, to explore aggressiveness in capuchin monkeys.

The 'human interaction test' assessed *social orientation to humans* and was based on Uher et al. (2013) and Nunes (2017). In the original test from Uher et al. (2013), the person who approached the subjects was wearing a costume, whereas in Nunes (2017) and in the present study, the person who approached the monkeys was not wearing a costume and was completely familiar with them. Hence, the present study and Nunes (2017) were measuring social orientation to *familiar* humans, whilst the original test (Uher et al., 2013) was measuring social orientation to *unfamiliar* humans, supposing the monkeys did not recognise the disguised person. Further studies could be made where a completely unfamiliar person approaches the subjects to test for social orientation to humans. However, it must be considered that capuchin monkeys (or other animals) that have been trafficked could be reacting to an unknown person based on their previous negative experiences with humans, as these are common during captivity (Soulsbury et al., 2009; personal observation). In this case, it would be unclear if the test is measuring 'social orientation' or 'sociability' to humans or behavioural signs related to the animals' previous traumatic experiences, for example. A careful justification would be needed for experiments which expose rehabilitant monkeys to conditions that may be detrimental for their well-being and/or post-release survival.

Finally, the 'tunnel' and 'covered tunnel' tests were performed with the objective of measuring *risk-taking* and were based on Uher et al. (2013) and Nunes (2017). Risk-taking was understood as *performing a behaviour which outcome is uncertain* (Trimpop, 1994) and *risk-averse* was considered as the opposite of risk-taking (i.e., a more risk-averse individual would be less risk-taking and vice-versa). To measure this trait, the monkeys were placed outside an unknown enclosure in front of a tunnel (or a covered tunnel) and I measured the latency to enter such tunnel. Even though some animal personality studies use risk-taking and boldness as equivalents, I consider only 'risk-taking' was assessed in the present study. Boldness is difficult to define and measure in animals, as this trait is sometimes treated as neophilia, by performing tests in which animals are exposed to novel objects, and sometimes as risk-taking plus neophilia, by exposing animals to novel objects in ‘risky’ situations, such as predation risks (Carter et al., 2013). To avoid confusion, the present study considered ‘neophilia’ as part of creativity (Kaufman et al., 2011) and risk-taking as a separate construct and avoided the term ‘boldness’.

5.4.3. Observer trait ratings

In the present study, four personality dimensions were obtained using the Hominoid Personality Questionnaire (HPQ; Weiss et al., 2009): Openness, Neuroticism, Assertiveness, and Sociability. Morton et al. (2013) proposed five dimensions for brown capuchin monkeys (*Sapajus apella*): Assertiveness, Openness, Neuroticism, Sociability and Attentiveness (Morton et al., 2013). The dimensions Openness, Neuroticism, Assertiveness and Sociability found in the present study closely resembled those described by Morton et al. (2013). Moreover, Fernández-Bolaños et al. (2020) found three personality dimensions in yellow-breasted capuchin monkeys (*Sapajus xanthosternos*) obtained from the application of the Hominoid Personality Questionnaire (Weiss et al., 2009): Openness-Neuroticism, Assertiveness, and Attentiveness-Sociability (Fernández-Bolaños et al., 2020). These are also similar to those found by Morton et al. (2013) for *Sapajus apella*. Moreover, these dimensions also resemble those found in the present study. It may be possible that these personality dimensions are present in all or several species of robust capuchin monkeys.

However, it must be noted that the personality traits found in this study by using observer trait ratings could be biased towards the 54 items described in the Hominoid Personality Questionnaire (Weiss et al., 2009). I could have overlooked other personality traits that are ecologically relevant for bearded capuchin monkeys if they were not included in this questionnaire. Because of this, it could be useful including both methods –behavioural tests and observer trait ratings specifically performed for the species in question– to assess capuchin personality structure, and validating them using behavioural observations.

5.4.4. Data reduction method

In the present study, I used Principal Component Analysis (PCA) as data reduction method for the variables obtained from the behavioural tests and the Hominoid Personality Questionnaire. Furthermore, I used Kaiser’s criterion to determine which components to extract from such analyses (Kaiser, 1960 in Morton and Altschul, 2019). I utilised these methods to be consistent with previous studies aimed at investigating capuchin personality structure, such as those performed by Morton et al. (2013), Manson and Perry (2013) and more recently, Fernández-

Bolaños et al. (2020). PCA and Factor Analysis (FA) are considered as *exploratory analyses*, meaning they are performed to summarise data and generate hypotheses, and not to confirm specific hypotheses (Budaev, 2010). When comparing PCA and FA, PCA is considered as the best option when the objective is only to reduce the number of dimensions. Furthermore, PCA may be used in small sample sizes (e.g., n ~ 25) as long as the original measures are highly reliable and the communalities are high (Budaev, 2010). In the present study, it was considered that the data complied with these requirements albeit the small sample size of the study (n = 18); therefore, PCA was selected as the data reduction method.

Nunes (2017) found similar results when using an expert-based method with individuals from the same species (*Sapajus libidinosus*) maintained in captivity in the same wildlife rescue centre. The present study aimed to replicate the findings of this study (Nunes, 2017), using a statistical data reduction method instead of an expert-based method. In this sense, I consider the present study was successful at replicating and strengthening the results from Nunes (2017). This was achieved by using PCA instead of selecting the personality dimensions *a priori*, based on ethological knowledge, thus identifying more specific personality constructs and dimensions than in the original study. However, this was not achieved without limitations. Even though these methods (PCA, FA and EB) have been used to reduce variables into personality traits in animal studies before, yielding similar results (Mazzamuto et al., 2019), other practices for data reduction such as using automated methods (e.g., parallel analysis instead of Kaiser's criterion) and using Regularised Exploratory Factor Analysis (REFA) are now considered more adequate for certain studies, such as those performed with small sample sizes (Morton and Altschul, 2019). Hence, utilising REFA combined with an automated method (e.g., parallel analysis) could be more adequate for the present study, particularly considering its small sample size. These analyses could be performed in the future to improve the results from the present study and its scientific robustness.

5.4.5. Correlations between assessment methods and behavioural observations

In the present study, I found several significant correlations between the personality constructs and dimensions found by using the two different methodological approaches and the behavioural observations. Even though several correlations were not significant after applying the Holm-Bonferroni sequential correction for multiple comparisons, I will still discuss them in this Section.

5.4.5.1. Correlations between traits obtained from the behavioural tests and behavioural observations

There was a negative correlation between time spent in affiliative behaviour (found in the behavioural observations, BO) and the trait *risk-averse*, derived from the behavioural tests (BT). Individuals that spent more time engaged in affiliative behaviours (BO) were considered as less risk-averse (or more *risk-taking*) in the behavioural tests. Furthermore, there was a positive correlation between time spent in vigilance behaviours (BO) and risk-averse scores (BT). Thus, individuals that spent more time engaged in vigilance behaviours were also rated as more risk-averse. This makes sense as more cautious individuals would be less likely to enter an unknown enclosure and more easily distracted by the environment in the room in which the tunnel and covered tunnel tests were performed.

Moreover, there was a positive correlation between time spent in human interaction (BO) and the trait aggressive (BT). In Chapter 4, it was shown that in many cases, behaviours directed towards humans are aggressive. Hence, it may be the case that more aggressive individuals are also more prone to be socially oriented towards humans, even becoming aggressive towards them. Additionally, there was a positive correlation between time spent in environmental manipulation (BO) and the trait sociable to humans (BT). It could be the case that individuals that spent more time manipulating the environment are also drawn out to humans out of ‘curiosity’. Interestingly, I did not obtain a correlation between time manipulating the environment (BO) and the trait creative (BT).

5.4.5.2. Correlations between HPQ-derived traits and behavioural observations

There was a significant positive correlation between Openness (HPQ) and time spent manipulating the environment (BO) and time spent in solitary play (BO). Similarly, Morton et al. (2013) found a correlation between time spent in play and Openness (Morton et al., 2013). As opposed to Morton et al. (2013) and what I predicted at the start of this study, I found few correlations between the HPQ-derived traits and the behavioural observations despite most of them (43/54) being considered as reliable between raters. For example, Sociability was not (highly) positively correlated with time spent grooming or social play; moreover, Neuroticism was not positively correlated with time spent in behaviours potentially indicative of stress (BPIS) during the behavioural observations.

5.4.5.3. Correlations between traits obtained from the behavioural tests and HPQ-derived traits

There was a negative correlation between the trait stereotypic (BT) and the dimension Assertiveness (HPQ). This is consistent with previous studies; in fact, Morton et al. (2013) found that Assertiveness was negatively loaded by items related to anxiety and time spent in social isolation during behavioural observations (Morton et al., 2013). However, I did not find a correlation between this trait (stereotypic) and Neuroticism (HPQ). Furthermore, other possibly related traits were not correlated with each other in this study. For example, I did not find a positive correlation between creative (BT) and Openness (HPQ), or between aggressiveness (BT) and Assertiveness (HPQ) as I would have expected. Interestingly, Nunes (2017) also failed to find strong correlations between the traits derived from the behavioural tests and the HPQ-derived items (Nunes, 2017). Hence, further research is needed to investigate the validity of utilising these methods to assess personality in bearded capuchin monkeys, and their complementarity.

5.5. Conclusion

In the present study, I aimed to broaden the current scientific knowledge on capuchin personality structure by replicating a previously described study (Nunes, 2017) in a set of 18 wild-born, captive-raised bearded capuchin monkeys (*Sapajus libidinosus*), maintained in temporary captivity in a Brazilian wildlife rescue centre. To achieve this, I assessed the personality of the subjects using two methods: observer trait ratings and behavioural tests. These methods have been described and used to assess capuchin personality structure before (Uher et al., 2013; Morton et al., 2013) and have been deemed as reliable and valid. I also performed behavioural observations to validate these assessments. I predicted that the personality structure of the subjects would closely resemble the personality structure found in previous studies (Uher et al., 2013; Morton et al., 2013; Nunes, 2017) by following the method proposed by Nunes (2017) for trafficked capuchins maintained in the same wildlife rescue centre in which the present study was performed. Moreover, I predicted that there would be a strong correlation between the personality traits derived from these methods and the behaviours observed in a ‘naturalistic’ context that were likely to reflect those traits. For example, sociability (if found) would be positively correlated with time spent grooming others and/or time spent in close proximity to other monkeys, openness (if found) would be positively correlated with time spent manipulating the environment, assertiveness (if found) would be positively correlated to agonistic behaviour, etc.

Most of the variables measured in the behavioural tests were considered as consistent across time (18/24) and were statistically reduced to five components (stereotypic, creative, aggressive, risk-averse and sociable to humans) which resembled the personality traits found in previous capuchin studies using the same methodological approach (Uher et al., 2013; Uher and Visalberghi, 2016; Nunes, 2017). Nonetheless, I found few strong associations between these traits and the behaviours observed during the 'naturalistic' observations. Similarly, the dimensions obtained using the 43/54 significantly reliable items from the Hominoid Personality Questionnaire from Weiss et al. (2009) (i.e., Assertiveness, Sociability, Openness and Neuroticism) closely resembled those found by Morton et al. (2013) and Fernández-Bolaños et al. (2020) for *Sapajus apella* and *Sapajus xanthosternos* (respectively) using the same methodological approach, except for Attentiveness (Morton et al., 2013; Fernández-Bolaños et al., 2020), which was not found in the present study. However, I also found few strong correlations between these dimensions and the behaviours observed during the naturalistic observations. In addition to this, the sample size of the present study was small (n = 18), the subjects used were wild-born, captive-raised monkeys, and Principal Component Analysis was used. Even though the data set complied with the most basic requirements for the use of PCA (Budaev, 2010), the use of Kaiser's criterion could have been replaced by a more scientifically robust method, such as an automated method (e.g., parallel analysis; Morton and Altschul, 2019). Similarly, PCA could have been replaced by a more adequate, scientifically robust data reduction method, particularly in light of the small sample size of the present study. Thus, the use of Regularised Exploratory Factor Analysis (instead of PCA) combined with the use of parallel analysis (instead of Kaiser's criterion) for data reduction could strengthen the results of this and similar animal personality studies (Morton and Altschul, 2019). Because of these limitations –the use of Kaiser's criterion and PCA plus the use of trafficked subjects– I suggest considering the findings of the present study as preliminary. More research is needed to understand the implications of the illegal pet trade in the ontogeny of personality in bearded capuchin monkeys and other primates, as well as the replicability of this study.

Chapter 6: Personality and Health of Rehabilitant Capuchin Monkeys (*Sapajus* sp.)

Chapter 6: Personality and Health of Rehabilitant Capuchin Monkeys (*Sapajus* sp.)

6.1. Introduction

In recent years, the relationship between stable inter-individual differences (i.e., 'personality') and health has gained much interest, both in humans and animals. In humans, research has shown the strong association between personality and health; in fact, personality traits are considered an integral part of human health psychology (Gosling et al., 2008; Ferguson, 2013). Higher scores in personality traits associated with high levels of negative affect, social isolation and Neuroticism, may lead to poor health outcomes (Gosling et al., 2008; Kupper et al., 2013; Jandackova et al., 2017). Even though the causal link between certain personality traits and poor health (e.g., risk of certain diseases) is not entirely clear, it has been hypothesised that personality traits influence the way in which individuals react to the environment, which in turn influence the way the immunological and other physiological systems respond (Gosling et al., 2008).

The study of animal personality and its relationship with health may be equally important and complement human studies, as animal models have several advantages over human models (Cavigelli, 2005). Studies with animals allow for greater control over experimental variables and longitudinal research is easier to conduct with animals with short lifespans (Cavigelli, 2005; Uher, 2011). Furthermore, the study of animal personality and health may improve animal welfare, as physical health is strongly associated with overall well-being (Finkemeier et al., 2018).

There are several physiological indicators that have been used to study the link between animal health and personality. Immunological (e.g., antibodies) and physiological responses to stress (e.g., stress hormones) are among those frequently used. In a study with domestic pigs (*Sus scrofa*), for example, it was found that 'coping style' influenced the immune responses of the individuals: 'low-resister' pigs had higher levels of IgM antibodies and CD8+ cytotoxic cells than 'high-resister' pigs (Luo et al., 2020) In this study, coping style was assessed by applying a test in which the piglets were restrained in supine position for one minute; in this test, latency to escape as well as escape attempts and vocalisations are recorded and piglets are categorised as 'high-resisters' (i.e., piglets that had longer latencies to escape and less escape attempts/vocalisations) or 'low-resisters' (piglets that had shorter latencies to escape and more escape attempts and vocalisations) (Luo et al., 2020). In another study, the relationship between 'behavioural types' and responses to chronic stress in domestic birds (i.e., laying hens, *Gallus gallus*) was analysed by subjecting the animals to unpredictable feeding schedules. It was found that 'proactive' (brown) hens had significantly higher heterophil-to-lymphocyte ratios (i.e., the ratio between heterophiles in birds and neutrophiles in mammals and lymphocytes; this ratio tends to be higher when animals are subjected to chronic stress) than 'reactive' (white) hens after 14 days of treatment. Corticosterone levels, however, were not significantly different between strains/behavioural types (Pusch et al., 2018). Even though some studies can detect this type of associations –e.g., between certain diseases and personality traits– it may still remain unclear whether these associations are true causal relationships.

In the present study, I aimed to investigate the association, if any, between personality and health in several groups of trafficked (i.e., born in the wild and raised by humans as pets) capuchin monkeys in Brazil. Most of these monkeys were raised in social isolation (from conspecifics) and were received by the rescue centre as juveniles or adults. Originally, I planned to use three health indicators: faecal glucocorticoids (i.e., cortisol), neutrophil-to-lymphocyte ratios and body condition scores. Even though faecal glucocorticoids and neutrophil-to-lymphocyte ratios (or heterophil-to-lymphocyte ratios in birds) can both be used to measure the effects of stress, they do not achieve this in the same way. Glucocorticoids measure the activity of the hypothalamic-pituitary-adrenal axis (HPA) whilst neutrophil/heterophil-to-lymphocyte ratios (NLR/HLR) measure the activity of the immune system. Glucocorticoids, specifically cortisol, will tend to increase as the HPA becomes activated as a stress response in human and non-human primates (Novak et al., 2013). Neutrophil/heterophil-to-lymphocyte ratios, on the other hand, will increase due to an increase in neutrophils (or heterophils) and a decrease in lymphocytes in peripheral blood as a response to stress (Pusch et al., 2018). Thus, both may be used to measure stress. NLR/HLR have the advantage of being less sensitive to handling stressors (i.e., than cortisol taken from a blood sample), but do require handling the animals for blood sampling (Pusch et al., 2018). Conversely, faecal glucocorticoids may be collected non-invasively. Unfortunately, the faecal samples obtained for this research project could not be analysed to determine cortisol levels due to issues related to the Covid-19 pandemic. Hence, I only used neutrophil-to-lymphocyte ratios and body condition scores for analysis. Considering this, I hypothesised that:

1. Less neurotic and less stereotypic individuals would have higher scores in body condition (BCS) when compared to more neurotic and more stereotypic monkeys.

2. More sociable, less neurotic and less stereotypic would have lower neutrophil-to-lymphocyte ratios (NLR) when compared to less sociable, more neurotic and more stereotypic monkeys.

Please note that as the data available on the relationship between personality and health of trafficked capuchin monkeys and other non-human primates is scarce in the scientific literature, I considered this study as exploratory (as opposed to confirmatory or hypothesis-testing) and proposed two *working hypotheses* i.e., ‘h*ypotheses that are subject to change, are provisional and the possibility of finding contradictory evidence is real*’ (Casula et al., 2021, p. 1709), rather than highly specific predictions, typical of confirmatory or hypothesis-testing research (Casula et al., 2021).

6.2. Methodology

6.2.1. Ethical approval

The project received ethical approval by the Animal Welfare and Ethical Review Body of the University of Bristol in January 2019 (reference number UB/18/087) and complied with the NC3Rs Guidelines for Primate Accommodation, Care and Use (NC3Rs, 2017).

6.2.2. Study site and subjects

The study was performed in a government wildlife rescue centre located in Rio Grande do Norte, Brazil (CETAS–RN). This rescue centre receives primates and other wildlife that have been trafficked and rescued in Northeast Brazil. Thirteen capuchin monkeys (*Sapajus libidinosus*) were part of the study (see below). Most of these animals were born in the wild, captured and sold as pets illegally. Refer to Chapter 2: General Methodology for a full description of the origin of the individuals as well as the rehabilitation procedures carried out in the rescue centre. A summary of the individuals considered in this Chapter as well as the data available for analysis can be seen in Table 6.1.

Table 6.1

Individuals considered in this study and data available for each animal

Name/ID	Age category	Sex class	Arrived at rescue centre[1]	Group	Personality assessment	Health Assessment
Dana	Adult	Female	≥ 12 months	1	Yes	Yes
Lombinho	Adult	Male	< 12 months	1	Yes	Yes
Li	Juvenile	Female	< 12 months	1	Yes	Yes
Fúria	Adult	Female	< 12 months	2	Yes	Yes
Café	Adult	Male	< 12 months	2	Yes	Yes
Galápagos	Adult	Male	< 12 months	2	Yes	Yes
Vitinho	Juvenile	Male	< 12 months	2	Yes	Yes
Rabinha	Juvenile	Female	< 12 months	2	Yes	Yes
Joana	Adult	Female	< 12 months	2	Yes	Yes
Tapa	Adult	Female	≥ 12 months	3	Yes	Yes
Arrebite	Adult	Male	≥ 12 months	3	Yes	Yes
Mãozinha	Juvenile	Male	Born at rescue centre	3	Yes	Yes
Tiquinho	Juvenile	Male	Born at rescue centre	3	Yes	Yes

[1] The exact date in which the individuals arrived at the rescue centre was not known for every animal; hence, I divided them into two groups: (1) more or (2) less than a year before the start of the study.

6.2.3. Health assessment

6.2.3.1. Neutrophil-to-lymphocyte ratios

Blood samples were obtained and analysed as described in Chapter 2: Health of Capuchin Monkeys (*Sapajus* sp.) in Northeast Brazilian Wildlife Rescue Centres. Neutrophil-to-lymphocyte ratios (NLR) were calculated by dividing the absolute neutrophil count by the total lymphocyte count of each individual.

6.2.3.2. Body condition

Body condition was assessed using a 5-point body condition score (BCS) developed and validated for this dissertation and based on Clingerman and Summers (2005). See Chapter 3: Health of Capuchin Monkeys (*Sapajus* sp.) in Northeast Brazilian Wildlife Rescue Centres for a complete description of the score as well as its development and validation.

6.2.4. Personality assessment

Personality was assessed using a combination of behavioural observations, behavioural tests (based on Uher et al., 2013) and observer trait ratings (Hominoid Personality Questionnaire; Weiss et al., 2009). This method (i.e., the HPQ) had acceptable levels of inter-rater reliability (ICC 3,*k*). For a full description of personality assessment methods, refer to Chapter 5: Assessment of Personality Structure in Rehabilitant Capuchin Monkeys (*Sapajus* sp.). The personality traits considered in this study were:

Table 6.2

Personality traits and behaviours considered in this Chapter

Derived from the Hominoid Personality Questionnaire	**Derived from behavioural tests**	**Derived from behavioural observations**
Openness Neuroticism Assertiveness Sociability	stereotypic creative risk-averse aggressive sociable to humans	time spent in behaviours potentially indicative of stress (BPIS)

Every individual had a score (or percentage, in the case of time spent in BPIS) for each personality trait.

As mentioned in the introduction, the data available on the relationship between personality and health of trafficked capuchin monkeys is scarce in the scientific literature. Hence, I considered this study as exploratory (as opposed to confirmatory or hypothesis-testing) and proposed two working hypotheses i.e., '*hypotheses that are subject to change, are provisional and the possibility of finding contradictory evidence is real*' (Casula et al., 2021, p. 1709), rather than highly specific predictions, typical of confirmatory or hypothesis-testing research (Casula et al., 2021). For the analyses, I focused on the aim (research objective) and used all personality data available i.e., all the traits that were assessed using the Hominoid Personality Questionnaire and behavioural tests (see Chapter 5 for a complete description). I also included the percentage of time spent in behaviours potentially indicative of stress (BPIS), as the exhibition of motor stereotypies and self-directed behaviours may be related to stress in capuchin monkeys (Ferreira et al., 2016), which, in turn, has been related to poor health outcomes in many species, including humans (e.g., Gosling et al., 2008; Kupper et al., 2013).

6.2.5. Statistical analysis

Statistics were performed using SPSS 27 (SPSS Inc. Released 2020. SPSS for Windows, Version 27.0. Chicago, SPSS Inc). Variables are shown as mean and standard deviation if not specified otherwise. I performed Schapiro-Wilk analyses on all dependent variables to test for normality deviations and converted results with non-normal distribution to normal distribution using log10 function in SPSS. An analysis of variance (univariate ANOVA) was used to investigate if there were significant differences in health parameters related to age and/or sex class and category of permanence in rescue centre. For this, I divided the animals into two groups based on the time they had spent in the wildlife rescue centre (CETAS-RN) previous to the start of the study. Animals were assigned to group A if they had spent <12 months at the rescue centre or to group B if they had spent ≥ 12 months or were born at the rescue centre. I refer to these groups as ‘categories of permanence in rescue centre’. Unfortunately, the exact date in which each individual arrived at rescue centre was unknown. Therefore, I divided the individuals into these two groups (i.e., A and B).

Moreover, I tested for correlations between personality traits and health variables using Pearson correlation analysis. I considered *p*-values ≤ 0.05 as significant and Pearson correlation coefficient values as very weak (0.00–0.19), weak (0.20–0.39), moderate (0.40–0.59), strong (0.60–0.79) and very strong (0.80–1.00) (Cardenio et al., 2020). Holm-Bonferroni sequential corrections were used for multiple comparisons. I performed simple regression analysis or multiple regression analysis with backward elimination for moderate, strong and very strong correlations. Multiple regression analyses are used when there are two or more predictor variables (Pagano et al., 2013). Backward elimination, also known as backward deletion, is a method in which all predictor variables are entered into the regression model simultaneously, and then are eliminated one by one depending on their contribution to the regression equation (Statistics Solutions, 2021). This method has been used to analyse personality traits in non-human primates in previous studies (e.g., Wergård et al., 2016). The personality traits considered (see Table 6.2) were used as the independent (predictor) variables and the health indicators (i.e., body condition scores and NLR) were used as the dependent (outcome) variables.

6.3. Results

6.3.1. Health

6.3.1.1. Age and sex-related differences

There were no significant differences related to age and/or sex class in body condition nor in neutrophil-to-lymphocyte ratios (NLR) ($n = 13$, $p \geq 0.213$).

6.3.1.2. Differences associated with time spent in the rescue centre

I did not find any significant difference associated with category of permanence in rescue centre in the parameters body condition and neutrophil-to-lymphocyte ratios ($n = 13$, $p \geq 0.305$).

6.3.1.3. Correlation between personality traits and health parameters

The results obtained from the Pearson correlations analyses between neutrophil-to-lymphocyte ratios (NLR) and body condition scores (BCS) and the personality traits considered (see Table 6.2) can be seen in the following table:

Table 6.3

Correlation between health parameters (body condition score and neutrophil-to-lymphocyte ratio) and personality traits of rehabilitant capuchin monkeys (*Sapajus libidinosus*) in order of descending size of correlation coefficient (n = 13)

Health parameter	Personality trait	Pearson coefficient	*p*-value	*p*-value after Holm-Bonferroni correction (*p*')
BCS	Time spent in BPIS	-0.642	0.018*	0.360
NLR	Sociability	-0.638	0.019*	0.361
NLR	Sociable to humans	-0.540	0.057	1.000
NLR	Assertiveness	0.395	0.181	1.000
NLR	Neuroticism	0.365	0.220	1.000
BCS	Neuroticism	0.356	0.233	1.000
BCS	Openness	0.344	0.250	1.000
NLR	Time spent in BPIS	-0.332	0.268	1.000
BCS	Sociability	-0.313	0.298	1.000
BCS	Assertiveness	-0.255	0.400	1.000
BCS	Stereotypic	-0.207	0.498	1.000
NLR	Stereotypic	0.137	0.655	1.000
NLR	Openness	-0.130	0.671	1.000
NLR	Creative	-0.115	0.709	1.000
BCS	Sociable to humans	0.077	0.802	1.000
NLR	Aggressive	0.057	0.854	1.000
NLR	Risk-averse	-0.048	0.877	1.000
BCS	Risk-averse	0.018	0.953	1.000
BCS	Aggressive	-0.016	0.958	1.000
BCS	Creative	-0.003	0.991	1.000

Summarising, I found negative associations between BCS and time spent in BPIS ($r = -0.642$), NLR and Sociability ($r = -0.638$) and NLR and sociable to humans ($r = -0.542$). These were all non-significant after applying Holm-Bonferroni corrections (see Table 6.3 for *p*-values obtained).

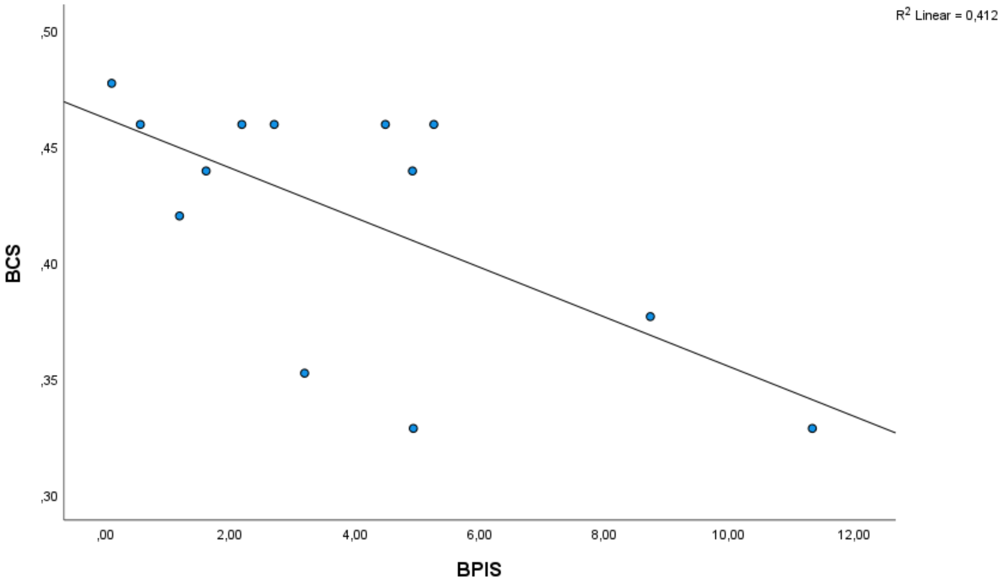
6.3.1.4. Linear regression analyses

Since NLR and BCS did not differ significantly by age and/or sex nor category of permanence in rescue centre (see Sections 3.1.1 and 3.1.2), I performed linear regression analysis without considering these variables. I performed these analyses only to Pearson correlations with moderate, strong and very strong coefficients (≥ 0.40) and did not consider correlations with very weak or weak coefficients (≤ 0.39) (Cardenio et al., 2020). These correlations were: BCS and time spent in BPIS (r = -0.642), NLR and Sociability (r = -0.638) and NLR and sociable to humans (r = -0.542).

6.3.1.5. Simple regression analysis

I performed a simple regression analysis using time spent in behaviours potentially indicative of stress (BPIS) as the independent (predictor) variable and body condition score (BCS) as the dependent (outcome) variable. I found that time spent in BPIS significantly predicted body condition score (BCS), $F(1,11) = 7.703$, p = 0.018, $R^2 = 0.412$ as shown in Figure 6.1.

Figure 6.1. Scatterplot matrix showing the negative correlation between body condition score (BCS) and time spent in behaviours potentially indicative of stress (BPIS)



6.3.1.6. Multiple regression analysis

I used multiple regression with backward elimination with NLR as the outcome variable and Sociability and sociable to humans as predictors. A summary of the model utilised can be seen in Table 6.4.

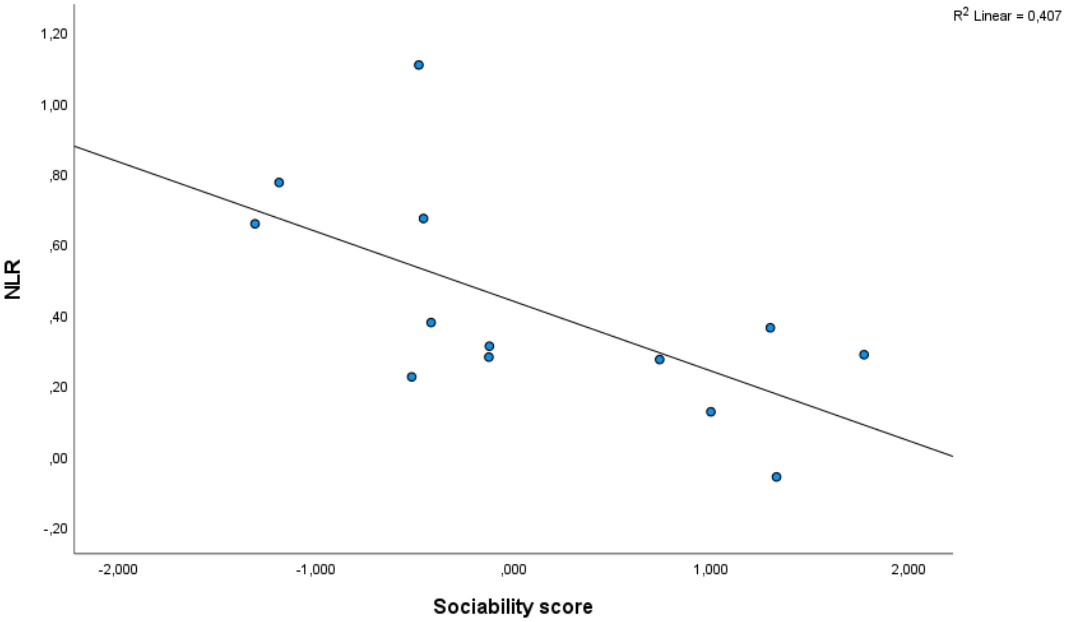
Table 6.4

Model summary of multiple regression with backward elimination

Model	R	Adjusted R^2	Standard error of estimate	Predictors (constant)
1	0.758	0.490	0.71389	Sociability, sociable to humans
2	0.638	0.354	0.80400	Sociability

The analysis (Table 6.4) showed that Sociability significantly predicted neutrophil-to-lymphocyte ratios (NLR), $F(3,9) = 7.564$, $p = 0.019$, $R^2 = 0.407$ as shown in Figure 6.2.

Figure 6.2. Scatterplot matrix showing the negative correlation between neutrophil-to-lymphocyte ratios (NLR) and Sociability scores



6.4. Discussion

6.4.1. Personality and health

6.4.1.1. Behaviours potentially indicative of stress (BPIS) predict body condition

In this study, percentage of time spent in BPIS (i.e., motor stereotypies and self-directed behaviours) significantly predicted body condition scores. Individuals that spent more time engaged in these behaviours had lower body condition scores (BCS), whilst individuals that spent less time performing BPIS had higher BCS. Nonetheless, the causal direction of this relationship is difficult to establish from this type of cross-sectional study.

The effect that stress has on the body and its functions is well known. High stress levels have been associated with neurological, cognitive, immune, gastrointestinal, cardiovascular, and endocrine dysfunction (Yaribeygi et al., 2017). Furthermore, high stress levels may have an impact on weight gain and/or maintenance by negatively affecting appetite, nutrition patterns and general gastrointestinal function (Yaribeygi et al., 2017).

In the present study, I did not find an association between neutrophil-to-lymphocyte ratios and time spent in BPIS. This finding suggests that individuals who displayed higher rates of BPIS were not necessarily more physiologically stressed than individuals who displayed lower rates of these behaviours. Furthermore, all individuals were provided with the same diet. Thus, a possible explanation for the association observed between time spent in BPIS and body condition could be early-life stress. Monkeys that have been experimentally subjected to maternal deprivation often show an increase in stress-related hormones (e.g., glucocorticoids) coupled with severe behavioural abnormalities when compared to individuals reared by their mothers (Pryce et al., 2002). Moreover, primates raised by stressed, aggressive mothers have higher corticotropin-releasing hormone levels than individuals raised by less aggressive, more nurturing females (Pryce et al., 2002). In the present study, most of the individuals were raised by humans, and it was not known at what age they were captured and sold as pets or from which primate populations they were subtracted. Neotropical primate populations with high levels of human impact, such as habitat fragmentation and hunting, show high levels of faecal glucocorticoids (Rimbach et al., 2013). Hence, it is likely that individuals in the present study came from stressed populations and the fact that they were not reared by their mothers only added to the development of behaviours potentially indicative of stress (BPIS). Unfortunately, it is not possible to investigate differences in early-life experiences in this study as it is not known in which specific conditions the monkeys were kept before being surrendered to the rescue centre. Nonetheless, I hypothesise that differences in early-life stress are likely to be the cause of certain individuals displaying higher levels of BPIS than others. However, the reason behind the negative correlation between time spent in BPIS and body condition observed in this study remains unclear. Other differences in personality traits were not associated with body condition and because all animals had the same diet, they should have had access to the same nutrients. However, the fact that they were spending more time engaged in repetitive behaviours, such as motor stereotypies (e.g., pacing) could have led to these individuals either having more physical activity and more energy expenditure as a consequence and/or spending less time foraging because they were distracted by the BPIS. Further research is needed to understand the relationship between BPIS and body condition in trafficked primates, considering the effect of early-life experiences, stress-related hormones, and the diet, amount of food, calories and/or physical activity each animal is actually having.

6.4.1.2. Sociability predicts neutrophil-to-lymphocyte ratios

In the present study, I found that individuals with higher Sociability scores (i.e., more sociable monkeys) had lower neutrophil-to-lymphocyte ratios (NLR). As explained before, NLR ratios tend to increase as neutrophils increase and lymphocytes decrease in peripheral blood when the immune system becomes activated (Pusch et al., 2018), particularly in chronic conditions (i.e., over several days). This could be, for example, when there is an infectious agent in the body. Because of this, NLR are frequently used to assess severity of infectious diseases in humans and animals, such as Covid-19 (e.g., Kong et al., 2020). In this sense, the results from the present study suggest that more sociable individual may be less chronically stressed (and have lower NLR) when compared to less sociable individuals.

This is consistent with previous studies on the relationship between stress and Sociability in capuchin monkeys (*Sapajus* spp.). In a study by Ferreira et al. (2018), increased values in genus normative behaviours (e.g., affiliative behaviours) related to Sociability predicted lower median, mean and minimum values of faecal glucocorticoids metabolites in bearded capuchin monkeys (*S. libidinosus*). (Ferreira et al., 2018). Furthermore, in a study by Robinson et al. (2016), in which the relationship between personality, welfare and subjective well-being of capuchin monkeys (*S. apella*) was studied, Sociability had the largest effect on both welfare and subjective well-being ratings (Robinson et al., 2016). Thus, the results obtained in the present study are consistent with what is known regarding Sociability in primates, as in, more sociable individuals tend to have lower stress levels as evidenced by physiological indicators such as faecal glucocorticoids and neutrophil-to-lymphocyte ratios than less sociable individuals.

Nevertheless, it is not possible to investigate to which extent the maternal and social isolation experienced by the subjects in the present study impacted the development of their social skills and consequently the occurrence of affiliative behaviours in their daily life, which were used to ‘quantify’ or score their Sociability. In other words, it is difficult to understand why individual differences in Sociability arise in rehabilitant i.e., wild-born, captive-raised capuchin monkeys. Most of the monkeys considered in the present study were raised by humans (except for two male juveniles, Tiquinho and Mãozinha, who were born in the rescue centre to hand-reared mothers); hence, the specific conditions in which they were raised were unknown, including their age at capture and if they were raised with other capuchin monkeys, other primates and/or other animals. It is well known that maternal and social deprivation can be considerably damaging for non-human primates. Individuals who have been raised in total social isolation frequently display abnormal behaviours such as impaired locomotion, high levels of aggression, and low interest for novel situations, environments or objects (Novak et al., 2006); furthermore, maternal and social deprivation can lead to physiological and health-related issues, including immunosuppression and high levels of stress hormones (Soulsbury et al., 2009).

Primates reared by humans often display the highest rates of behaviours potentially indicative of stress when compared to individuals raised by conspecifics (Marriner and Drickamer, 1994; Vandeleest et al., 2011). Conversely, primates who have the opportunity to develop high quality social relationships show higher levels of physical and psychological well-being, with lower stress levels and even reduced parasite loads (Robinson et al., 2016). Because of this, it could be useful to perform a thorough investigation of the social and environmental conditions in which primates are kept before they arrive to wildlife rescue and rehabilitation centres, albeit this is likely to be unsuccessful due to the lack of information given in most cases by the ‘owners’ when an animal is surrendered or confiscated (personal observation).

Even though age, sex class and ‘category of permanence in the rescue centre’ did not seem to influence neutrophil-to-lymphocyte ratios in this study, the sample size considered was small (n = 13) and other indicators of physiological stress were not analysed (e.g., faecal glucocorticoids). Further research is needed to fully understand the association between early-life social experiences, stress, and differences in personality, particularly those related to Sociability, in wild-born, captive-raised primates, especially if the objective is to enhance the post-release survival of the individuals and the success of the reintroduction programme.

6.5. Conclusion

In this Chapter, I aimed to investigate the association between personality and health of trafficked bearded capuchin monkeys that were part of a rehabilitation and reintroduction programme. I used body condition scores (BCS) and neutrophil-to-lymphocyte ratios (NLR) as health indicators. Firstly, I hypothesised that less neurotic and less stereotypic individuals would have higher scores in body condition (BCS) when compared to more neurotic and more stereotypic monkeys.

As I predicted, more ‘stereotypic’ individuals, or individuals who spent more time engaged in behaviours potentially indicative of stress (BPIS) (i.e., namely motor stereotypies and self-directed behaviours) had lower body condition scores (BCS), whilst individuals that spent less time performing BPIS had higher BCS. Interestingly, individuals who had higher scores on Neuroticism or scored more highly on the trait ‘stereotypic’ in the behavioural tests, as assessed in Chapter 5, did not have higher BCS. As mentioned in the discussion, the causal direction of this association (i.e., time spent in BPIS was associated with the BCS, but Neuroticism and the trait ‘stereotypic’ were not associated with the BCS) is difficult to establish from this type of cross-sectional study. Particularly considering that I did not find an association between neutrophil-to-lymphocyte ratios (NLR) and time spent in BPIS. This suggests that individuals who spent more time engaged in BPIS were more physically active and had a higher energy expenditure as a consequence and/or they spent less time foraging (and consequentially fed less) because they were distracted by the BPIS. Further research is needed to understand the association of motor stereotypies and self-directed behaviours with body condition of trafficked capuchin monkeys, considering the effects of early life experiences.

Finally, I hypothesised that more sociable, less neurotic and less stereotypic individuals would have lower neutrophil-to-lymphocyte ratios (NLR) when compared to less sociable, more neurotic and more stereotypic monkeys, suggesting they would be more chronically stressed. As I predicted, individuals with higher scores on Sociability had lower NLR when compared to less sociable monkeys. As mentioned above, this is consistent with previous studies on the relationship between physiological stress and Sociability in capuchin monkeys (e.g., Robinson et al., 2016; Ferreira et al., 2018). Further research is needed to understand more deeply the relationship between individual differences in Sociability and stress in trafficked capuchin monkeys. This is particularly important to improve the survival prospects of these animals in reintroduction projects, as social skills play a vital role in the survival of primates and other animals (Melfi and Marples, 2000, Cheyne et al., 2012).

Chapter 7: Personality and Behaviour in Rehabilitation of Capuchin Monkeys (*Sapajus* sp.)

Chapter 7: Personality and Behaviour in Rehabilitation of Capuchin Monkeys (*Sapajus* sp.)

7.1. Introduction

Primate rehabilitation is defined as the process by which non-human primates raised by humans are treated medically, are helped to regain physical and psychological health, and are provided with the opportunity to acquire and/or develop the necessary skills to survive in the wild with little human provision, or none (Beck et al., 2007). Even though the scientific literature on primate rehabilitation is growing, little is known about the relationship between individual differences on personality and rehabilitation of trafficked primates. In humans, research into rehabilitation and personality has found that certain individuals achieve better rehabilitation outcomes, particularly those with high scores on Extraversion (e.g., Herbert and Powell, 1989). More recent research has shown that post-traumatic stress disorder (PTSD), for example, is positively associated with high levels of negative affect, Neuroticism, anxiety and novelty-seeking, and negatively associated with high levels of Extraversion and Conscientiousness (Jakšić et al., 2012). Indeed, there is a growing interest in the relationship between personality and psychopathology (and rehabilitation from it, where applicable), particularly on those related to trauma exposure, in human and non-human primates (Miller et al., 2003; Richter-Levin et al., 2019).

As mentioned in previous Chapters, the impact of maternal and social deprivation on the cognitive, behavioural and psychological development of non-human primates has been widely documented. Primates reared solely by humans (i.e., in complete isolation from conspecifics) often develop severe ‘abnormal’ or pathological behaviours such as impaired locomotion, excessive aggression, and repetitive behaviours (e.g., motor stereotypies) and self-injurious behaviours (Novak et al., 2006; Vandeleest et al., 2011). Early environmental experiences help to determine emotional, behavioural, and physical development; thus, early traumatic experiences may alter future coping skills and behavioural/emotional regulation (Sánchez et al., 2001). Primates separated from their social group at infancy and reared by humans in captivity will tend to develop psychopathologies, observable in behavioural abnormalities and poor well-being (Soulsbury et al., 2009). Hence, these issues need to be resolved during rehabilitation and before releasing the animals back into the wild and pose an opportunity to study the association between personality, psychopathology, and rehabilitation.

Unfortunately, the scientific literature on the association between personality traits and rehabilitation of trafficked primates is extremely scarce. In a non-published study performed by Sita (2016), environmental enrichment was used to support the rehabilitation of ten rescued black capuchin monkeys (*Sapajus nigritus*). It was found that more neophilic individuals showed a significantly higher increase in foraging behaviours, whilst less neophilic individuals exhibited an increase in stereotypies and other stress-related behaviours. Moreover, more active individuals exhibited a decrease in locomotion, and individuals with higher scores in exploration showed a decrease in investigation-related behaviours and an increase in locomotion and stress-related behaviours. Furthermore, individuals with higher scores in Sociability increased exploratory behaviours more when compared to less sociable monkeys (Sita et al., 2016).

Conversely, studies on reintroduction outcomes and personality traits are more common among the scientific literature. It is important to consider and understand these studies as well, as reintroduction is the ultimate goal of many primate rehabilitation projects, including the rehabilitation programme of the present study. Overall, the current scientific literature suggests that more risk-taking, explorative and bolder individuals have better post-release survival outcomes than more risk-averse, less explorative and shyer individuals (Bremner-Harrison et al., 2004; de Azevedo and Young, 2021).

The relationship between personality and space use, which is another important aspect to be considered in wildlife rehabilitation, has been widely studied both in captivity and in the wild (Spiegel et al., 2017). A study with 21 wild, free-ranging bank voles (*Myodes glareolus*), for example, found that bold individuals travel longer distances, occupy larger areas including home ranges, prefer more diverse microhabitats and have less spatial overlap with conspecifics when compared to shy individuals (Schirmer et al., 2019). Similarly, common brushtail possums (*Trichosurus vulpecula*) with higher scores in exploration and Eurasian red squirrels (*Sciurus vulgaris*) with higher scores on boldness use larger core areas when compared to individuals with lower scores in these personality traits (Wat et al., 2020; Wauters et al., 2021). As individual variations in space use may influence foraging success, utilisation of home ranges, social networks and habitat preferences (Spiegel, et al., 2017) they could, in turn, influence the survival of reintroduced primates and their well-being whilst in captivity and after release.

As discussed thoroughly in Chapter 1: General Introduction and Aims, bolder, more explorative and/or risk-taking individuals are considered as more prone to exhibit behaviours considered as risky for their post-release survival than shyer, less explorative and/or risk-averse individuals but may also be more successful at establishing large core areas, home ranges, and finding food sources (Bremner-Harrison et al., 2004, de Schirmer et al., 2019; Wat et al., 2020; Azevedo and Young, 2021; Wauters et al., 2021). Interestingly, in reintroduction projects, 'success' is generally equated to 'post-release survival'. Hence, studies which explore the relationship between personality and reintroduction success often consider individuals as 'more successful' if the animals survive for longer (than 'less successful' individuals). There are several examples of this in the scientific literature related to reintroduction biology, such as 'bold' swift foxes (*Vulpes velox*; Bremner-Harrison et al., 2004), 'bold' Tasmanian devils (*Sarcophilus harrissii*), 'explorative' Blanding's turtles (*Emydoidea blandingii*) and 'bold' and 'explorative' European minks (*Mustela lutreola*), surviving for longer than less bold and/or explorative individuals in several studies (Bremner Harrison et al., 2004; de Azevedo and Young, 2021).

However, there are several issues that need to be addressed regarding the assumption that risk-taking, explorative and/or bold individuals are actually 'more successful' after release than individuals with lower scores on these personality traits. Firstly, it is not entirely clear what these studies are actually measuring. In fact, animal personality studies seem to be inconsistent regarding their definition of 'boldness' (Carter et al., 2013). Some of these studies (e.g., Bremner-Harrison et al., 2004) have assessed boldness by measuring latency to approach a novel object, which may be argued that it is neophilia or novelty-seeking, a part of creativity (Kaufman et al., 2011) and not boldness. Moreover, it has been argued that there is a need to focus on all personality traits that may be ecologically relevant for the species in question and not only on the most frequently studied traits; boldness, exploration, aggressiveness, activity and sociability (Koski, 2014).

Even if there is consensus on the personality traits being measured across wildlife reintroduction projects, it is known that social groups composed by individuals with different personality traits may be more stable and fare better against environmental variations than groups composed by individuals with similar personalities (Watters and Meehan, 2007). Individuals with different personality traits not only respond differently in terms of behaviour to distinct scenarios, but also possess different physiological responses to pathogens and environmental stressors (Watters and Meehan, 2007). Hence, it is necessary to construct diverse groups, in terms of personality traits, to promote resilience to environmental change in populations aimed for reintroduction to the wild (Watters et al., 2003; Watters and Meehan, 2007).

In addition to this, equating reintroduction success with post-release survival does not take into account the well-being of the animals whilst they are being rehabilitated and after they are reintroduced. Because the final goal of many reintroduction projects which include trafficked (i.e., wild-born, captive-raised) wildlife, particularly those that include non-human primates, is to release animals back to the wild to improve their welfare level (Guy et al., 2014), it does not make sense to consider post-release survival as the only rehabilitation/reintroduction success indicator. Indeed, other behavioural and physiological indicators such as the exhibition of abnormal behaviours and faecal glucocorticoids have been used in rehabilitant primates to assess their welfare state (e.g., Cheyne, 2006; Ferreira et al., 2018). In this sense, more sociable, assertive and neophilic and less neurotic individuals would be likely to have a better welfare level during rehabilitation when compared to less sociable, assertive, neophilic and more neurotic individuals, as has been shown in previous studies made with captive Neotropical primates (Robinson et al., 2016; Sita, 2016).

Nevertheless, it is not entirely clear how this would relate to post-release survival. More sociable and neophilic individuals could be more likely to engage in affiliative and exploratory behaviours, such as establishing relationships with resident conspecifics and exploring novel objects, food sources or larger areas after release (de Azevedo and Young, 2021). However, these behaviours could also put them at risk (of predation or poisoning, for example). Additionally, less neurotic individuals could have a stronger immune system and respond better against environmental stressors, including infectious pathogens, than more neurotic conspecifics (Watters and Meehan, 2007; Capitanio, 2011). Even though different scores in neophilia (or broader creativity), assertiveness, sociability, exploration and boldness could lead to mixed results in terms of post-release survival and welfare during rehabilitation and after release, individuals who are more easily stressed (i.e., more neurotic) would probably have lower well-being levels both before and after release. This association –between Neuroticism and poor well-being outcomes– is very well known in humans, and has been observed in primate personality studies (Jakšić et al., 2012; Robinson et al., 2016).

In the present study, I aimed to investigate the association, if any, between individual differences in personality (as measured in Chapter 5) and behaviour in rehabilitation (as measured in Chapter 4) of several groups of trafficked capuchin monkeys i.e., that were rescued from the illegal pet trade in Brazil. Unfortunately, it was not possible to assess the personality of the individuals in this study prior to their rehabilitation, which would have been ideal. Personality was assessed during or after the rehabilitation period, which could have led to unclear results, as the associations that may have been found could be confusing. It may not have been clear if personality traits influenced rehabilitation or vice-versa. Furthermore, as mentioned throughout this dissertation, the scientific literature on the association between personality traits and rehabilitation of trafficked primates is extremely scarce, and, to my knowledge, there is only another non-published study performed which aimed to analyse the relationship between individual differences in personality and rehabilitation of trafficked capuchin monkeys (Sita et al., 2016).

Because of this, I used *working hypotheses* i.e., '*hypotheses that are subject to change, are provisional and the possibility of finding contradictory evidence is real*' (Casula et al., 2021, p. 1709), rather than highly specific predictions, typical of confirmatory or hypothesis-testing research (Casula et al., 2021).

Considering this, I hypothesised that:

1. Less 'neurotic' individuals would spend more time engaged in behaviours that could be considered as positive for their welfare (e.g., affiliative behaviours) and less time in behaviours considered as negative for their welfare (e.g., motor stereotypies) during rehabilitation by the end of the rehabilitation period when compared to more 'neurotic' individuals.
2. Less 'neurotic' individuals would spend more time engaged in behaviours that could be considered as 'risky' for their survival after release back into the wild by the end of the rehabilitation period when compared to more 'neurotic' individuals.
3. Less 'risk-averse' and more 'assertive' and 'aggressive' individuals would spend more time engaged in behaviours that could be considered as risky for their survival after release back into the wild by the end of the rehabilitation period when compared to more 'risk-averse' and less 'assertive' and 'aggressive' individuals.

7.2. Methodology

7.2.1. Ethical approval

The project received ethical approval by the Animal Welfare and Ethical Review Body of the University of Bristol in January 2019 (reference number UB/18/087) and complied with the NC3Rs Guidelines for Primate Accommodation, Care and Use (NC3Rs, 2017).

7.2.2. Study site and subjects

The study was performed in a government wildlife rescue centre located in Rio Grande do Norte, Brazil (CETAS–RN). This rescue centre receives primates and other wild animals that have been rescued/confiscated from the pet trade in Northeast Brazil. Thirteen capuchin monkeys (*Sapajus libidinosus*) were part of the study (Table 7.1) . Most of these animals were born in the wild, captured and sold as pets illegally. For a full description of the origin of the individuals as well as the rehabilitation procedures carried out in the rescue centre see Chapter 2: General Methodology. It is important to consider that the present study was a performed with a small sample size due to limitations related to Covid-19 restrictions and personality was assessed during or after the rehabilitation of the individuals, as mentioned above. Thus, it is an exploratory study and must be considered as an initial attempt to investigate the research questions proposed and not as a definitive (confirmatory) study.

Table 7.1

Individuals considered in this Chapter and data available for each animal

Name	Age category	Sex class	Group	Personality assessment	Rehabilitation assessment
Dana	Adult	Female	1	Yes	Yes
Lombinho	Adult	Male	1	Yes	Yes
Tino	Adult	Male	1	Yes	Yes
Li	Juvenile	Female	1	Yes	Yes
Garrincha	Juvenile	Male	1	Yes	Yes
Furia	Adult	Female	2	Yes	Yes
Café	Adult	Male	2	Yes	Yes
Galápagos	Adult	Male	2	Yes	Yes
Vitinho	Juvenile	Male	2	Yes	Yes
Tapa	Adult	Female	3	Yes	Yes
Arrebite	Adult	Male	3	Yes	Yes
Mãozinha	Juvenile	Male	3	Yes	Yes
Tiquinho	Juvenile	Male	3	Yes	Yes

7.2.3. Assessment of rehabilitation outcomes

‘Rehabilitation outcomes’ were assessed by analysing the changes in behaviour across the four-month rehabilitation period in the ten behavioural categories used in Chapter 4 (using the same data): *foraging, locomotion, affiliative behaviour, agonistic behaviour, inactivity, vigilance,* *environmental manipulation*, *solitary play*, *human interaction* and *behaviours potentially indicative of stress* (BPIS). I compared the changes in activity budgets and space and substrate use as well as the changes in behavioural diversity using the Shannon H index. In this context, 'change' was considered as the difference observed between the baseline and final observational phases (i.e., the changes observed between the start and the end of the 3.5-month rehabilitation period). Each individual was observed for a total of approximately 180 minutes using 5-minute focal sampling in three observational phases performed in April, May and July 2019 (please refer to Chapter 4 for a complete description of the methodology of the behavioural observations).

The activity budgets were calculated using the percentage of time spent in each behavioural category (e.g., locomotion) considered in the ethogram. Space (i.e., ground, medium and high tier(s) use) and substrate use (i.e., concrete, fixed trunks, mobile trunks, vertical mesh, horizontal mesh, vertical cords and inner cages of the enclosures) were measured using scan sampling every 30 seconds whilst the 5-minute focal samplings were performed (i.e., simultaneously). Behavioural diversity was analysed using the Shannon H index as proposed by other authors (e.g., Metter, 2008; Miller et al., 2020). Please refer to Chapter 4: Behaviour of Rehabilitant Capuchin Monkeys (*Sapajus* sp.) for a complete description of these methods.

7.2.4. Assessment of personality

Personality was assessed using a combination of behavioural tests and observer trait ratings (Hominoid Personality Questionnaire; Weiss et al., 2009). For a full description of these assessments, refer to Chapter 5: Assessment of Personality Structure in Rehabilitant Capuchin Monkeys (*Sapajus* sp.). The personality traits considered in this study were:

Table 7.2

Personality traits considered in this study

Derived from the HPQ	Derived from behavioural tests
Openness Neuroticism Assertiveness Sociability	stereotypic creative risk-averse aggressive sociable to humans

Every individual had a score for each personality trait, which was derived from the PCA obtained in Chapter 5: Personality Structure in Rehabilitant Capuchin Monkeys (*Sapajus* sp.).

As mentioned in the introduction, the data available on the relationship between personality traits and behavioural rehabilitation of trafficked capuchin monkeys is scarce in the scientific literature. Hence, I considered this study as exploratory (as opposed to confirmatory or hypothesis-testing) and proposed two *working hypotheses* i.e., ‘*hypotheses that are subject to change, are provisional and the possibility of finding contradictory evidence is real*’ (Casula et al., 2021, p. 1709), rather than highly specific predictions, typical of confirmatory or hypothesis-testing research (Casula et al., 2021). For the analyses, I focused on the aim (research objective) and used all personality data available i.e., all traits that were assessed using the Hominoid Personality Questionnaire and behavioural tests (see Chapter 5 for a complete description).

Table 7.3

Timeline used for personality and rehabilitation assessment in this study (n = 13)

	2019																2020
Group	Week 1 1–5 April	Week 2 8–12 April	Week 3 15–19 April	Week 4 22–26 April	Week 5 29 April to 3 May	Week 6 6–10 May	Week 7 13–17 May	Week 8 20–24 May	Week 9 27–31 May	Week 10 3–7 June	Week 11 10–14 June	Week 12 17–21 June	Week 13 24–28 June	Week 14 1–5 July	Week 15 8–12 July	Week 16 15–19 July	August 1–17
1 (n=5)	OBS1	OBS1				OBS2	OBS2	BT2						OBS3 BT(t)	OBS3 BT(t)	OBS3	HPQ
2 (n=4)[2]	OBS1	OBS1		BT1	BT1	OBS2 BT1	OBS2			BT2				OBS3 BT(t)	OBS3 BT(t)	OBS3	HPQ
3 (n=4)	OBS1	OBS1				OBS2	OBS2						BT2	OBS3 BT(t)	OBS3 BT(t)	OBS3	HPQ

[1] The first trials of the behavioural tests of Vitinho (from Group 2, see Table 1) were performed between 13/05/2019 and 20/05/2019.

OBS1 = baseline behavioural observations
OBS2 = second behavioural observations
OB3 = final behavioural observations
BT1 = first trials of behavioural tests
BT2 = second trials of behavioural tests
BT(t) = tunnel behavioural tests
HPQ = application of the Hominoid Personality Questionnaire

7.2.5. Statistical analysis

Statistics were performed using SPSS 27 (SPSS Inc. Released 2020. SPSS for Windows, Version 27.0. Chicago, SPSS Inc). In the preliminary analysis, I tested for correlations between personality traits and rehabilitation variables using Pearson correlation analysis. I considered p-values ≤ 0.05 as significant and Pearson correlation coefficient values as very weak (0.00–0.19), weak (0.20–0.39), moderate (0.40–0.59), strong (0.60–0.79) and very strong (0.80–1.00) (Cardenio et al., 2020). Holm-Bonferroni sequential corrections were used for multiple comparisons (i.e., when more than two tests were performed to the same variables); furthermore, regression analyses were planned for moderate, strong and very strong correlations. However, preliminary analyses showed there were no significant nor strong correlations/regressions between the personality traits (independent/predictor variables) and the rehabilitation (dependent/outcome) variables considered.

Hence, to investigate the association between personality traits and changes in behaviour during rehabilitation, I used a median cut-off, also known as 'median split approach', following the methodology explained by DeCoster et al. (2011). The animals with scores below the median were assigned to group 1 ($n = 6$) and animals above the median were assigned to group 2 ($n = 6$); finally, the animal with the median score was excluded (DeCoster et al., 2011). Repeated measures analysis of variance (ANOVA RM) was then used to analyse changes in rehabilitation outcomes between observational phases (i.e., baseline and final). To analyse if the differences between personality traits were already present at the baseline observational phase, I conducted a univariate analysis of variance (ANOVA). Behavioural differences related to age and/or sex class were not described in this Chapter as they have been covered in Chapter 4: Behaviour of Rehabilitant Capuchin Monkeys (*Sapajus* sp.).

7.3. Results

7.3.1. Rehabilitation

7.3.1.1. ANOVA RM: Activity budgets

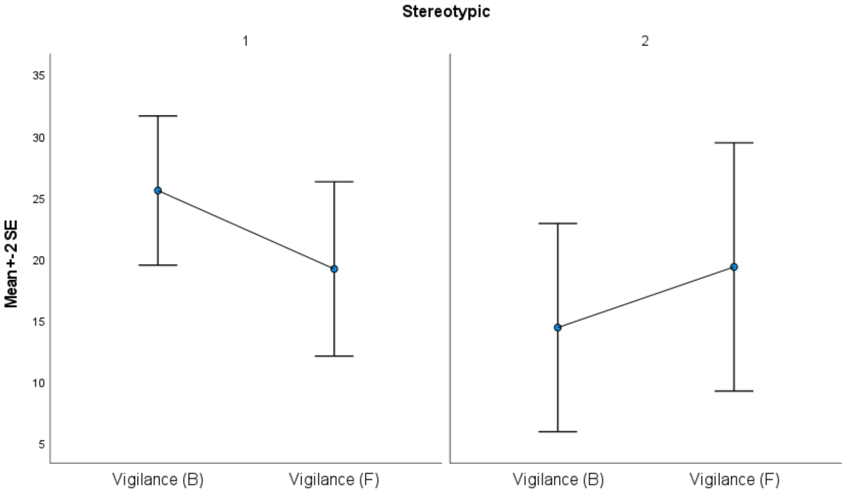
I did not find any significant differences in any of the behavioural categories *foraging*, *locomotion*, *affiliative*, *agonistic*, *inactivity*, *environmental manipulation*, *human interaction* and *behaviours potentially indicative of stress* related to personality traits (ANOVA RM; all tests: $n = 12$, $p \geq 0.083$; see Appendix 4 for complete p-values obtained from analyses).

However, I found significant differences associated with personality traits in the behavioural categories *vigilance* and *solitary play* as detailed below.

7.3.1.1.1. Vigilance and *stereotypic* (ANOVA RM)

Individuals with scores above the median (i.e., more stereotypic, n = 6) tended to increase the time they spent engaged in vigilance behaviours over the study period (from 14.38% at the baseline to 19.32% during the final phase), whilst less stereotypic individuals (n = 6) decreased the time engaged in these behaviours over time (25.53% to 19.32%). Thus, there was a significant interaction between the trait stereotypic and changes in time engaged in vigilance behaviours (*F* = 15.110, n = 12, *p* = 0.003). When performing an ANOVA between baseline stereotypic groups, these significant differences were not observed; more stereotypic and less stereotypic individuals did not spend significantly different time in vigilance-related behaviours during the baseline observational phase, although they showed a non-significant trend to do so (*p* = 0.058)

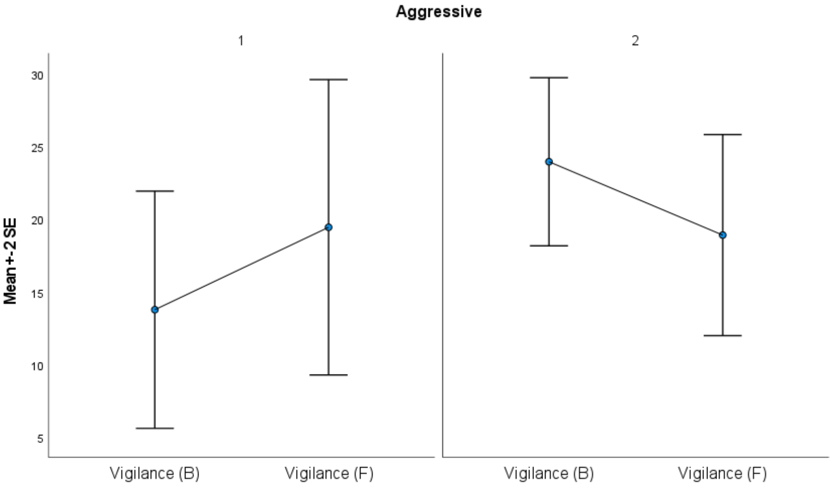
Figure 7.1. Mean and SE in vigilance level in baseline (B) and final (F) phases for individuals scored as low (1) and high (2) on the trait stereotypic.



7.3.1.1.2. Vigilance and *aggressive* (ANOVA RM)

There was a significant difference in change in vigilance behaviours between animals scoring differently for the trait aggressive (*F* = 10.451, *p* = 0.009). Less aggressive individuals (n = 6) tended to increase the time spent in vigilance-related behaviours (from 13.76% to 19.43%), whilst more aggressive monkeys (n = 6) decreased the time engaged in these behaviours (from 23.93% to 18.89%). When comparing the groups during the baseline phase, there was no significant difference at the 5% level (*p* = 0.069).

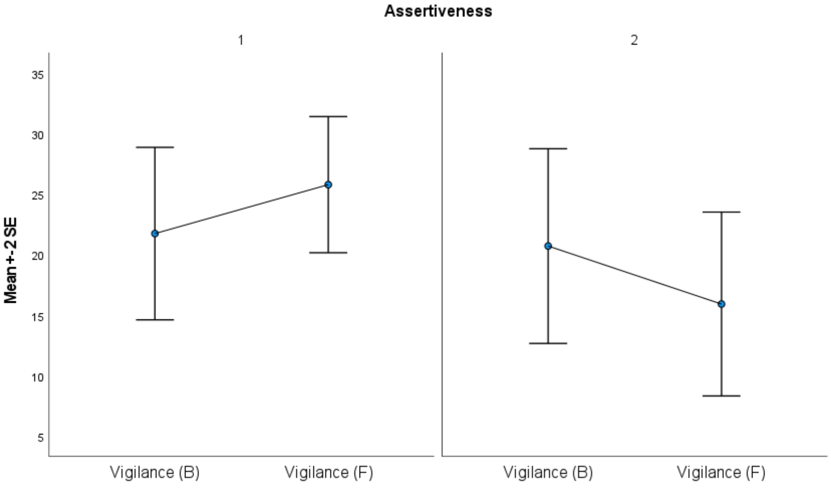
Figure 7.2. Mean and SE in vigilance level in baseline (B) and final (F) phases for individuals scored as low (1) and high (2) on the trait aggressive.



7.3.1.1.3. Vigilance and Assertiveness (ANOVA RM)

Similarly to the findings on the traits stereotypic and aggressive, I found a significant difference in vigilance when comparing individuals scoring high and low for Assertiveness using the median as cut-off (*F* = 5.155, *p* = 0.047). Overall, less assertive individuals (n = 6) increased the time spent in vigilance behaviours over time (from 21.73% to 25.77%) whereas more assertive individuals (n = 6) decreased the time spent in vigilance-related behaviours in the final (15.92%) when compared to the baseline (20.70%) phases. When comparing vigilance between assertive groups at baseline, I did not find a significant difference (*p* = 0.852).

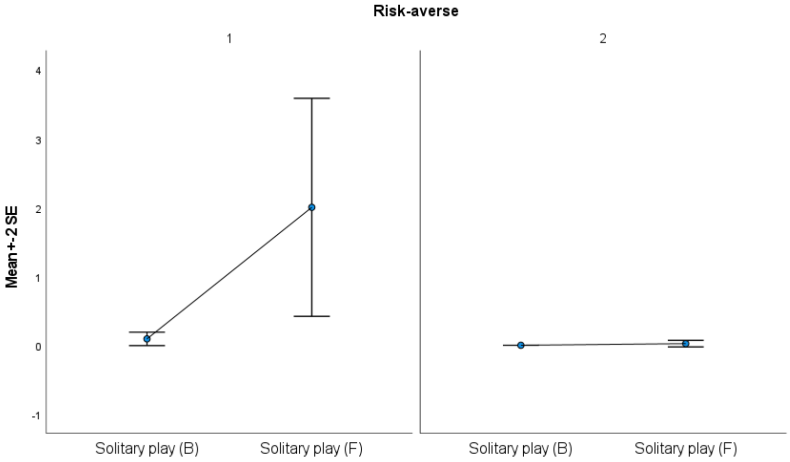
Figure 7.3. Mean and SE in vigilance level in baseline (B) and final (F) phases for individuals scored as low (1) and high (2) on Assertiveness.



7.3.1.1.4. Solitary play and *risk-averse* (ANOVA RM)

I found a significant difference in solitary play associated with the trait risk-averse when comparing individuals using the median score as cut-off (*F* = 6.074, *p* = 0.033). In general, less risk-averse individuals significantly increased the time spent in solitary play (from 0.09% in the baseline to 2.01% during the final phase) whilst individuals scoring lower for this trait changed little in their solitary play level (from 0.00% to 0.02%). When I performed an ANOVA between risk-averse baseline groups, no significant difference was found (*p* = 0.087).

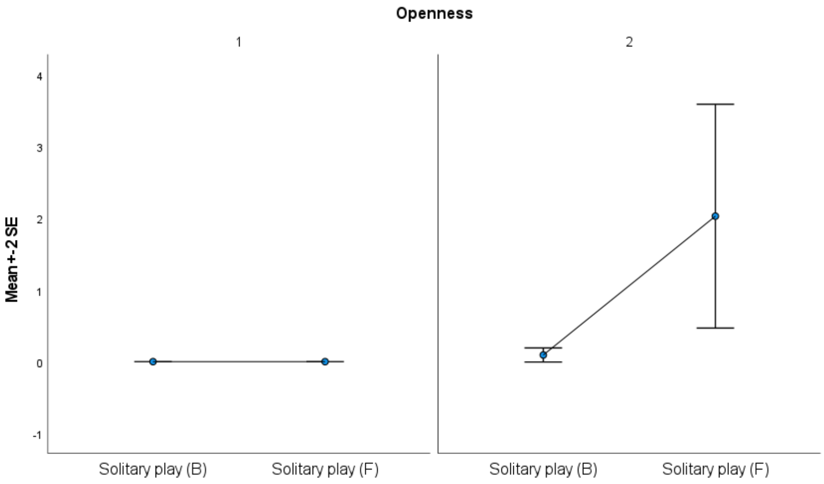
Figure 7.4. Mean and SE in solitary play level in baseline (B) and final (F) phases for individuals scored as low (1) and high (2) on the trait risk-averse.



7.3.1.1.5. Solitary play and Openness

I found a significant difference in solitary play between Openness groups when using the median as group cut-off (*F* = 6.590, *p* = 0.028). In general, individuals scoring higher for Openness increased the time spent in solitary play, from 0.09% during the baseline to 2.03% during the final phase. Conversely, less open individuals did not spent time performing this behaviour in any of the observational phases (i.e., baseline and final). When performing an ANOVA, no significant difference was observed between baseline Openness groups (*p* = 0.087).

Figure 7.5. Mean and SE in solitary play level in baseline (B) and final (F) phases for individuals scored as low (1) and high (2) on Openness.

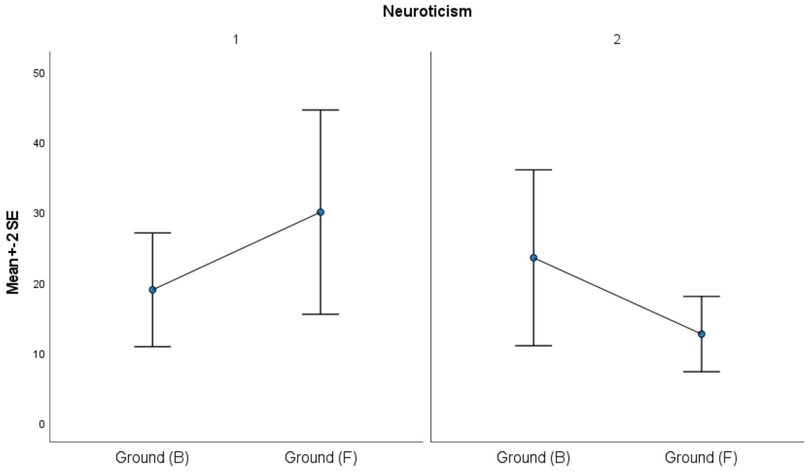


7.3.1.2. ANOVA RM: Space use

7.3.1.2.1. Ground use

I did not find a significant difference in the use of the ground when comparing the baseline and final phases associated with the personality traits stereotypic, creative, risk-averse, sociable to humans, aggressive, Openness, Assertiveness and Sociability ($p \geq 0.076$). There was a tendency towards a significant difference; however, on the use of the ground associated with Neuroticism ($F = 4.492$, $p = 0.060$). In general, individuals scoring higher for Neuroticism increased their time spent on the ground from 18.90% during the baseline to 29.94% in the final phase, whilst less neurotic individuals decreased the time spent on this level of their enclosure from 23.45% to 12.60%.

Figure 7.6. Mean and SE in ground use level in the baseline (B) and final (F) phases for individuals scored as high (1) and low (2) on Neuroticism.



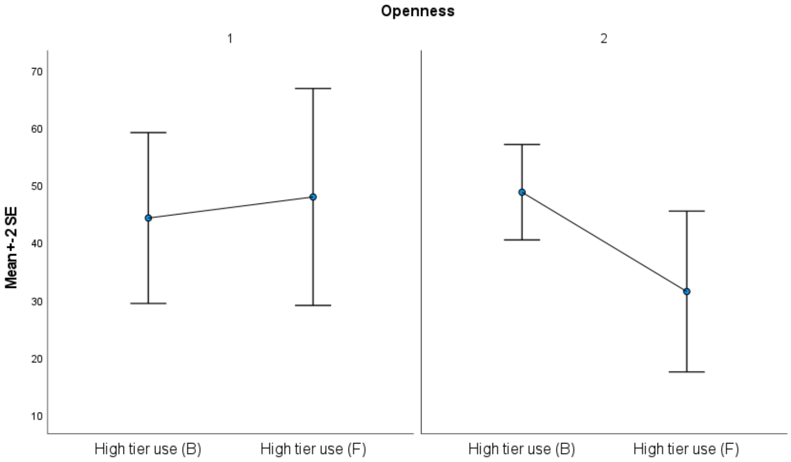
7.3.1.2.2. Medium tier use

Regarding the use of the medium tier of the enclosures, I did not find a significant difference in its use when comparing change between the baseline and final phases and its association with the personality traits considered in this study ($p \geq 0.151$).

7.3.1.2.3. High tier use

I did not find a significant difference in the use of the high tier over time associated with the personality traits stereotypic, creative, risk-averse, sociable to humans, aggressive, Neuroticism, Assertiveness and Sociability ($p \geq 0.263$). There were significant differences, however, associated with Openness ($F = 5.177$, $p = 0.046$). Individuals scoring lower for Openness slightly increased time spent in the high tier, from 44.19% to 47.85%, whilst those scoring higher decreased the time spent in this tier over time, from 48.67% to 31.42%.

Figure 7.7. Mean and SE in high tier use level in the baseline (B) and final (F) phases for individuals scored as low (1) and high (2) on Openness.



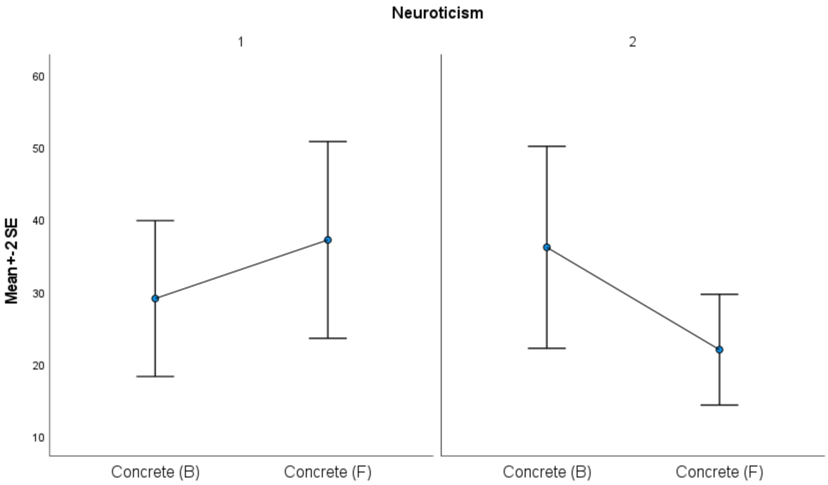
7.3.1.3. ANOVA RM: Substrate use

7.3.1.3.1. Concrete

When comparing the baseline and final phases, I did not find a significant difference in the change in the use of concrete associated with the personality traits stereotypic, creative, risk-averse, sociable to humans, aggressive, Openness, Assertiveness and Sociability ($p \geq 0.093$).

Nonetheless, there was a tendency towards a significant difference associated with Neuroticism ($F = 4.591$, $p = 0.058$). More neurotic monkeys tended to increase the time spent in this substrate (concrete) from 29.01% in the baseline to 37.11% during the final phase, whereas less neurotic individuals decreased the time spent in this substrate, from 36.10% to 21.95% (i.e., baseline vs. final phase).

Figure 7.8. Mean and SE in concrete use level in the baseline (B) and final (F) phases for individuals scored as high (1) and low (2) on Neuroticism.

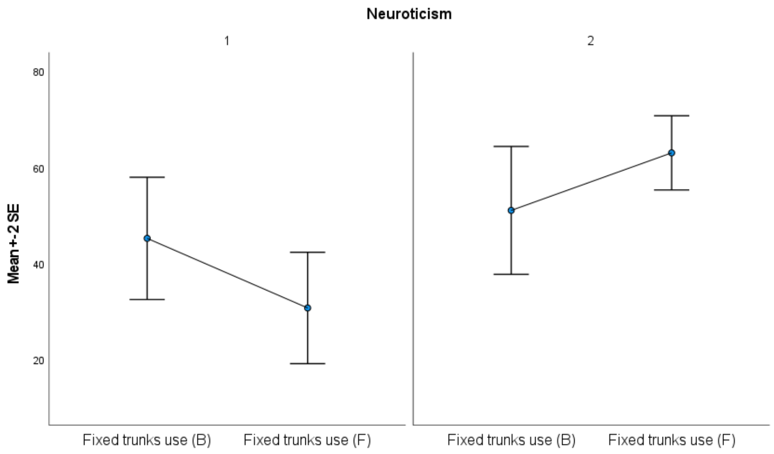


7.3.1.3.2. Fixed trunks

I did not find a significant difference in the use of the fixed trunks related to the personality traits stereotypic, creative, risk-averse, sociable to humans, aggressive, Openness, Assertiveness and Sociability ($p \geq 0.197$).

Nonetheless, there was a significant difference associated with Neuroticism ($F = 7.610$, $p = 0.020$). More neurotic individuals generally decreased the time spent on fixed trunks over time, from 45.09% in the baseline to 30.58% during the final phase. Conversely, less neurotic monkeys increased the time spent in this type of substrate from 50.92% between the baseline to 62.92% during the final phase.

Figure 7.9. Mean and SE in ground use level in the baseline (B) and final (F) phases for individuals scored as high (1) and low (2) on Neuroticism.



7.3.1.3.3. Mobile trunks and vertical mesh and horizontal mesh

When comparing the baseline and final phases, I did not find a significant difference in the change in use of the mobile trunks associated with the personality traits considered in this study ($p \geq 0.118$).

7.3.1.3.4. Vertical cords

I did not find a significant difference in the use of the cage related to the personality traits stereotypic, sociable to humans, aggressive, Openness, Neuroticism, Assertiveness, Sociability ($p \geq 0.158$).

There were significant differences, however, related to the trait creative ($F = 7.031$, $p = 0.024$), and risk-averse ($F = 6.079$, $p = 0.033$). More creative animals decreased the time spent in vertical cords over time (2.59% to 1.37%) whilst less creative animals increased the time spent using these substrates (0.38% to 2.92%). Similarly, individuals who scored lower in the trait risk-averse decreased the time spent on vertical cords over time (2.97% to 1.83%) whilst individuals with higher scores increased the time spent in these substrates (0.00% to 2.46%).

Figure 7.11. Mean and SE in vertical cords use level in the baseline (B) and final (F) phases for individuals scored as low (1) and high (2) on the trait creative.

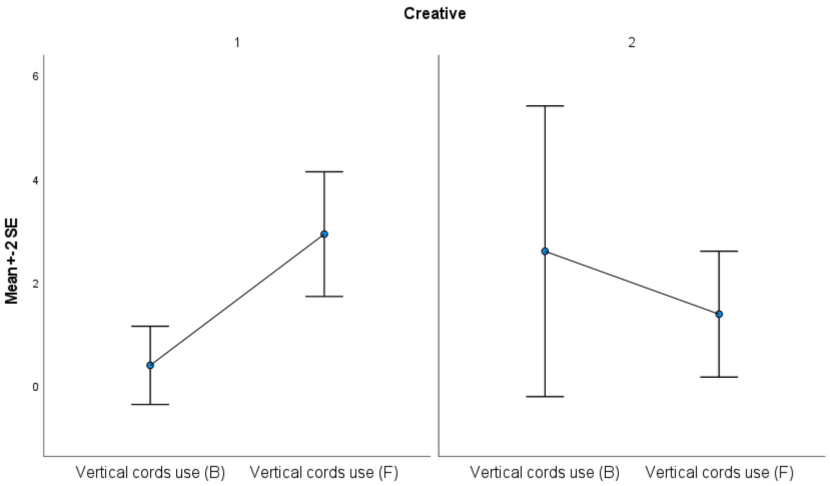
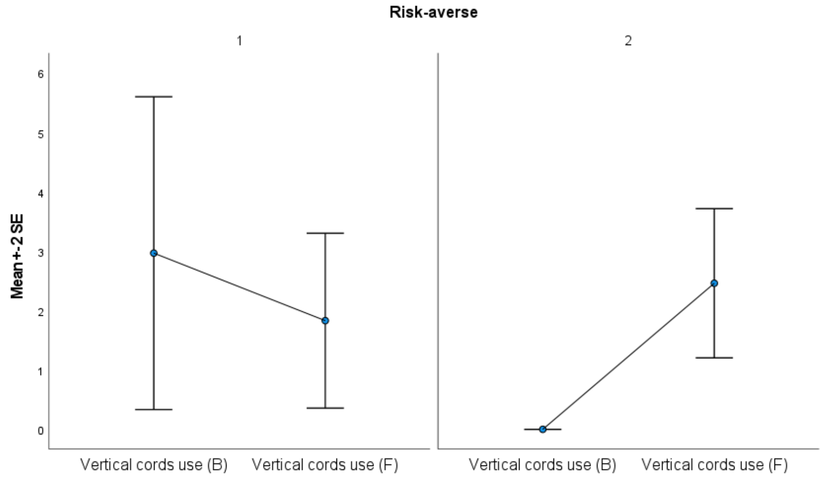


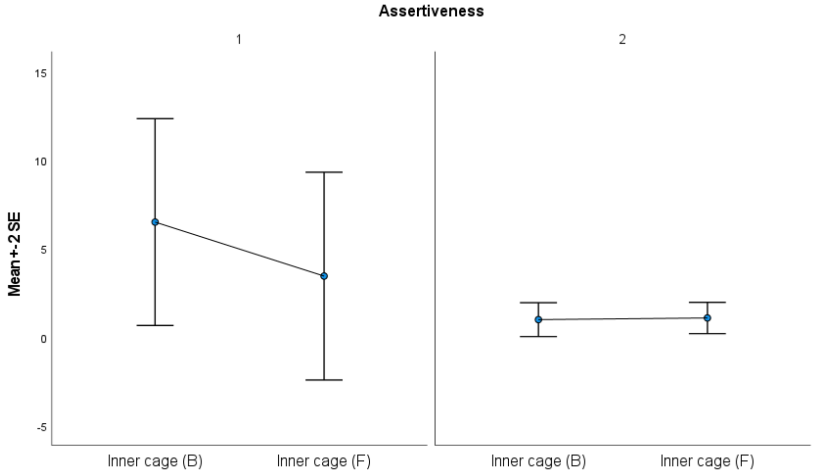
Figure 7.12. Mean and SE in vertical cords use level in the baseline (B) and final (F) phases for individuals scored as low (1) and high (2) on the trait risk-averse.



7.3.1.3.5. Cage

When comparing the baseline and final phases, I did not find a significant difference in the use of the cage associated with the personality traits stereotypic, creative, risk-averse, sociable to humans, aggressive, Openness, Neuroticism and Sociability ($p \geq 0.282$). Nonetheless, there was a tendency towards a significant difference associated with Assertiveness ($F = 4.474$, $p = 0.061$). Less assertive individuals generally decreased the time spent in the inner cage of their enclosures from 6.49% during the baseline to 3.44% during the final phase. Conversely, more assertive individuals slightly increased the time spent in this substrate over time, from 0.99% during the baseline to 1.08% during the final observational phase.

Figure 7.13. Mean and SE in inner cage use level in the baseline (B) and final (F) phases for individuals scored as low (1) and high (2) on Assertiveness.



7.3.1.4. ANOVA RM: Behavioural diversity indexes (BDI)

I did not find any significant differences when comparing the baseline and final indexes associated with the personality traits considered in this study ($p \geq 0.105$).

7.4. Discussion

Even though I found several significant results in the present study, it is important to consider that, as mentioned in the Introduction and the Methodology of this Chapter, this was an exploratory study with a small sample size, partly due to limitations and restrictions related to the Covid-19 pandemic. Hence, I suggest considering these findings as preliminary. In this Section (Discussion), I will discuss in relation to previous studies and propose several possible explanations for the results obtained. Further research is needed to fully understand the relationship between personality traits and rehabilitation of trafficked capuchin monkeys, as well as the replicability of this study.

7.4.1. Personality and rehabilitation: activity budgets

In the present study, I found significant associations between time spent in vigilance-related behaviours during rehabilitation and the traits ‘stereotypic’, ‘aggressive’, and ‘Assertiveness’. Individuals with higher scores on the trait stereotypic and lower scores on the trait aggressive and Assertiveness increased the time they engaged in vigilance behaviours when comparing the start and the end of the 3.5-month rehabilitation period; conversely, individuals with lower scores on the trait stereotypic and higher scores for the trait aggressive and Assertiveness decreased the amount of time they spent in vigilance behaviours across observational phases.

These results are consistent with previous studies with capuchin monkeys (*Sapajus* spp.) in captivity. Morton et al. (2013) reported that, in captivity, more open and neurotic capuchin monkeys tend to spend more time in vigilance-related behaviours when compared to less open and neurotic conspecifics, suggesting this may be related to curiosity or stress (Morton et al., 2013). Conversely, wild (i.e., free-ranging) capuchin monkeys with higher scores on Assertiveness and aggressiveness spend more time in vigilance behaviours when compared to less assertive/aggressive individuals (Fernández-Bolaños et al., 2020). In this case, vigilance-related behaviours may be seen as anti-predator behaviours, leading to bolder individuals expressing them (Fernández-Bolaños et al., 2020).

Moreover, I found that less risk-averse and more open individuals increased the time spent in solitary play, whilst more risk-averse and less open monkeys decreased the time engaged in this behavioural category. In terms of rehabilitation and post-release survival, I hypothesise this could lead to mixed results: more open and less risk-averse individuals could potentially engage more in solitary play after release which could work as a coping skill in a new environment, for example, by allowing the monkey to learn to use a substrate or food item found at the release site. It could also be potentially endangering to the individual if s/he was to be distracted by this activity. However, I predict this would be unlikely to happen as most monkeys spend less than 2% of the time in solitary play, regardless of their personality.

Behavioural differences between captive and free-ranging wild animals are well known, and Neotropical primates are not the exception. Captive-born capuchin monkeys require extensive practice to achieve the level of proficiency that free-ranging conspecifics display in essential activities for their survival such as tool use, locomotion and travel, and location of food and water resources, even if they are a naturally assertive and flexible species (Fragaszy et al., 2004). The results obtained from the present study suggest that the rehabilitant individuals are still behaving as other captive capuchin monkeys would do by the end of the 3.5-month rehabilitation period. Ideally, the behaviour of individuals who are going to be reintroduced back into the wild in the short-term would resemble more the behaviour of wild, free-ranging individuals than the behaviour of captive-born individuals. As mentioned above, by the end of this study, more aggressive and assertive individuals were spending less time engaged in vigilance behaviours than less aggressive and assertive individuals.

Conversely, wild, free-ranging capuchin monkeys with higher scores on Assertiveness and aggressiveness tend to spend more time in vigilance behaviours when compared to less assertive and aggressive individuals. In this context, vigilance-related behaviours are interpreted as anti-predator behaviours, leading to ‘bolder’ individuals expressing them (Fernández-Bolaños et al., 2020). Nonetheless, it must be considered that the individuals in this study were housed in relatively small enclosures outside the release area. Hence, moving them to a pre-release enclosure at the release site for several weeks or months, as it has been performed in other trafficked primates reintroduction studies (e.g., Arango-Guerra et al., 2013), could give them the opportunity to express these behaviours in a more similar way, including in terms of personality traits, to their wild counterparts.

The results of this study seem to be consistent with previous research on personality and reintroduction in which ‘bold’ individuals tend to exhibit more risky behaviours than shy individuals (Bremner-Harrison et al., 2004; Lopes et al., 2017; de Azevedo and Young, 2021). Indeed, more assertive and aggressive individuals decreased over time the time spent in vigilance-related behaviours that could potentially help them (e.g., by helping them to identify a predator quickly) when released, whilst less aggressive and assertive monkeys increased the time spent in vigilance behaviours over time. Unfortunately, it was not possible to monitor the individuals after their release as part of the present study; thus, I could not assess the behavioural changes or post-release survival of the individuals and their relationship to individual differences. Furthermore, the two methods of personality assessment considered in the present study (i.e., behavioural tests and the use of the Hominoid Personality Questionnaire, Weiss et al., 2009) were applied more than 12 months apart (see Table 7.3 in Section 2). In addition to this, the behavioural tests were applied to the different social groups in different times and in the same months in which the rehabilitation assessments were performed. Ideally, the personality of the monkeys would have been assessed prior to the rehabilitation assessment and not during and after, albeit using behavioural observations and personality ratings done as much as one year apart as well as their relation to each other has been used in other studies (e.g., Morton et al., 2013), presumably due to the apparent stability of personality traits over time (e.g., Réale et al., 2007).

7.4.2. Personality and rehabilitation: space and substrate use

In this study, I found several differences related to personality traits and changes in space and substrate use over time. More neurotic animals decreased the time spent in fixed trunks over time whilst less neurotic individuals increased the time spent in this tier over time. Moreover, less open individuals slightly increased time spent in the highest tier of their enclosures;

conversely, more open individuals decreased the time spent in this tier over time. In addition to this, more assertive individuals increased the time spent in the inner cage of their enclosures, whereas less assertive monkeys decreased the time spent in this part of the enclosures.

This seems to be relatively similar to previous findings of studies on personality and reintroduction, where ‘bold’ individuals tend to engage in more risky behaviours when compared to shy individuals (Bremner-Harrison et al., 2004). Boldness has been positively associated with medium to high scores on Openness and low scores on Neuroticism in previous studies in humans, for example (e.g., Poy et al., 2014; Donnellan and Burt, 2016). If these associations between personality traits –boldness, Openness and Neuroticism– exist in capuchin monkeys, utilising the ground and medium tier as well as the fixed trunks and the inner cage of the enclosure could be interpreted as risky behaviours performed more frequently by less neurotic and more open and assertive individuals, as the predominant substrate in the enclosure of the monkeys was concrete (there were less fixed trunks than concrete surfaces) and descending to the ground (i.e., in this case, descending to the concrete floor and/or entering the inner cage) could be risky for arboreal primates due to the presence of potential predators. Note that ‘boldness’ was not assessed in this study. Thus, further research could be made to investigate the association between boldness, Openness and Neuroticism and spatial use in the rehabilitation of trafficked capuchin monkeys.

Interestingly, the traits creative and risk-averse seemed to influence substrate use in a different way. I found that more creative animals decreased the time spent in vertical cords over time, whilst less creative animals increased the time spent using these substrates over time. Similarly, monkeys scoring lower for the trait risk-averse (i.e., more ‘risk-taking’) decreased the time spent in vertical cords over time, whereas individuals with higher scores in the trait risk-averse increased the time spent in these substrates. I hypothesise this finding was related to the specific characteristics of these substrates. Vertical cords were short pieces of metal or rope cords that the enclosures had which purpose were only to connect one substrate to another (e.g., a fixed trunk to a mesh wall). Hence, only one or two monkeys could use them simultaneously and did not seem to be places for social activities such as grooming or playing (i.e., because their surface was quite small) but were rather used by individuals to sit, eat and/or observe the environment. Conversely, fixed trunks had large surfaces which several animals could use simultaneously to perform almost any activity, including social interaction, eating, resting and travel (i.e., locomotion).

As with the section on personality and activity budgets, I was not able to investigate the differences among individuals related to space and substrate use after release as post-release monitoring was not performed. I hypothesise more open monkeys would have explored larger areas, established a larger core area and/or home range after release and possibly finding food or water resources would have been easier for them, when compared to less open monkeys, similarly to what happens with free-ranging individuals of other species (e.g., Wat et al., 2020; Wauters et al., 2021). This may seem like an advantage at first, but these monkeys would have also been at more risk of predation if there were to use larger areas of the forest or lowest areas of the canopy, for example, when compared to less neurotic or open animals. In fact, Sita et al. (2016) found that more active capuchin monkeys survived less longer after release when compared to less active conspecifics (Sita et al., 2016). This supports the idea proposed by Watters and Meehan (2007), where it is important to include animals with different personality traits in reintroduction projects as different behavioural or personality traits provide distinct advantages for the post-release survival of the individuals (Watters and Meehan, 2007).

7.4.3. Association between rehabilitation outcomes and personality

Finally, an important aspect to consider in this Chapter is the relationship that may exist between personality traits and ‘rehabilitation outcomes’, or behaviour during rehabilitation. Indeed, both of them are related to individual behaviour. Furthermore, in this study, they were both assessed during the same months (April–July 2019). Ideally, all personality assessments would have been performed before assessing behaviour during rehabilitation. Unfortunately, this was not possible in the present study.

To control for potential issues that may have arisen because of this, I included an analysis of the baseline phase to compare between the two groups (i.e., below and above the median cut-off) when a specific personality trait seemed to be influencing a certain behaviour by the end of the rehabilitation period. For example, I compared between (1) more and (2) less assertive, aggressive and stereotypic groups regarding their time spent in vigilance-related behaviours at the start of the rehabilitation programme, and they did not show significant differences. They did show, however, significant differences in time spent in vigilance behaviour at the end of the 3.5-month rehabilitation programme. These suggests that the differences observed at the end of the rehabilitation programme were in fact related to personality traits (e.g., Assertiveness). In other words, if the individuals had already shown these differences, for example, in time spent in vigilance related to Assertiveness at the start of their rehabilitation, it would have been more difficult to argue that the differences observed in vigilance behaviours at the end of the programme –the rehabilitation outcome– were associated with Assertiveness, because they would have been already present *before* the rehabilitation started. However, this did not happen in the present study.

In fact, previous studies have found variations in time budgets related to personality traits in primates. In a study performed with 13 captive lion-tailed macaques (*Macaca silanus*), Kluiver et al. (2022) found that sociability, persistence and anxiety influenced individual time budgets related to foraging, resting and activity (Kluiver et al., 2022). Persistency positively predicted time spent in activity and foraging-related behaviours and negatively predicted time spent resting. Moreover, anxiety negatively predicted time spent foraging and sociability positively predicted time spent active (Kluiver et al., 2022). Kluiver et al. (2022) used behavioural tests and behavioural observations to assess personality traits, and then analysed their relationship with time budgets calculated from behavioural observations performed *separately*. In the present study, I assessed personality traits using observer trait ratings (the Hominoid Personality Questionnaire or HPQ; Weiss et al., 2009) and behavioural tests (based on Uher et al., 2013 and Nunes, 2017), used behavioural observations to validate them (i.e., the mean across the three observational phases: baseline, second, and final), and then analysed if the personality traits were associated with differences in time budgets in the final observational phase of the rehabilitation programme.

Note that Kluiver et al. (2022) used different behavioural observations to assess personality and calculate time budgets, whereas I used the same behavioural observations. However, I did not use *exactly* the same data. To validate the personality traits derived from the HPQ and the behavioural tests, I used the mean across observational phases of each behavioural category (e.g., inactivity). To investigate the association between rehabilitation outcomes and personality traits, I used the time budgets obtained from the baseline and final observational phases and analysed using Repeated Measures Analysis of Variance (ANOVA RM). Only when there was a difference related to personality traits, I used the activity budgets from the baseline phase (separately) to investigate if that difference already existed at the start of the rehabilitation programme, as explained above. Thus, I consider the present study as capable of identifying the association, if any, between individual differences in personality and behaviour at the end of a rehabilitation programme or ‘rehabilitation outcomes’. Nevertheless, it must be considered that this was an exploratory study, aimed at generating hypotheses for future research (please refer to the Introduction of this Chapter for a complete explanation) and not at confirming highly specific predictions. Further studies could be made in which behavioural observations aimed at assessing rehabilitation outcomes and those aimed at identifying personality traits are performed separately, and, if possible, at different times (i.e., personality assessments could be performed before rehabilitation assessments).

7.5. Conclusion

In this Chapter, I aimed to investigate the association between personality and behaviour in rehabilitation or ‘rehabilitation outcomes’ of bearded capuchin monkeys as part of a rehabilitation and reintroduction programme. I hypothesised that less neurotic and risk-averse and more open, assertive and/or aggressive individuals would exhibit more risky behaviours by the end of the 3.5-month rehabilitation period when compared to more neurotic and risk-averse and less open, assertive and/or aggressive individuals. Overall, less neurotic and open individuals were spending more time utilising the fixed trunks of the enclosures (than more neurotic and less open monkeys) by the end of the rehabilitation period. Moreover, less open individuals slightly increased the time they spent in the highest tier of their enclosures; conversely, more open individuals decreased the time spent in this tier over time.

Furthermore, more assertive monkeys spent slightly more time in the inner cage of the enclosures by the end of the rehabilitation period when compared to less assertive individuals. When considering these behaviours –utilising the fixed trunks as well as the ground and the inner cage of the enclosures– as ‘risky’ and hypothesising a positive association exists in this species between Openness and boldness and a negative association between Neuroticism and boldness, it could be suggested that these results are ‘similar’ to previous studies performed in wildlife reintroduction projects, where bold animals exhibit more risky behaviours than shy animals (e.g., Bremner-Harrison et al., 2004). Further studies could investigate the association between boldness, Openness and Neuroticism in *Sapajus* spp. and how these personality factors relate to risky behaviours during rehabilitation or, even better, after release.

I also hypothesised that less neurotic monkeys would engage less time in behaviours that could be considered as negative for their welfare (e.g., motor stereotypies) and/or more time in behaviours that could be considered as positive for their welfare (e.g., affiliative behaviours). However, I did not find any correlation between Neuroticism and the behavioural categories considered in the ethogram (foraging, locomotion, affiliative, agonistic, inactivity, environmental manipulation, solitary play, vigilance, human interaction and behaviours potentially indicative of stress). Hence, this personality trait (Neuroticism) did not seem to be influencing the welfare, negatively or positively, during the rehabilitation of the individuals in this study. Further research could be made to address this specific research question i.e., which personality traits are related to the pre- and post-release welfare level of rehabilitant capuchin monkeys, ideally with a larger sample size.

In a rehabilitation and reintroduction programme, ideally all personality assessments, including the behavioural observations to validate them, would be performed before assessing rehabilitation outcomes and not simultaneously or after, as rehabilitation in itself could potentially change personality traits or their assessment. To avoid this, it would be better to perform personality assessments before the rehabilitation programme starts. This would have the added benefit of not needing to handle the animals during the rehabilitation period. Nonetheless, this is difficult to achieve in a government wildlife rescue centre, as most decisions are made by the competent authorities and not by the principal investigator or other researchers. In the present study, it was not possible to measure personality traits before measuring rehabilitation outcomes nor assessing post-release survival. Thus, this study must be considered only as an exploratory study.

Chapter 8: Ethical Assessment of the Rehabilitation and Reintroduction of Trafficked Neotropical Primates

Chapter 8: Ethical Assessment of the Rehabilitation and Reintroduction of Trafficked Neotropical Primates

8.1. Introduction

Neotropical primates, or New World monkeys, are non-human primates inhabiting Central and South America (NPC, 2021). They are also known as platyrrhines and include 139 species that occupy a wide range of tropical environments in the American continent (Püschel et al., 2017; NPC, 2021). These species are commonly traded illegally in Central and South America (Duarte-Quiroga and Estrada, 2003). The illegal wildlife trade is one of the top five illegal international trades and one of the major drivers for the endangerment of Neotropical primate species (Esmail et al., 2020; Shostell and Ruíz-García, 2016).

Even though Neotropical primates are commonly kept as pets in Latin American countries, they are not suitable pets as they can become aggressive and be a hazard for human beings and domestic animals (Soulsbury et al., 2009). Moreover, their wellbeing is frequently compromised in these situations as the ‘owners’ do not have the necessary knowledge of their behavioural and welfare needs to keep them in adequate environments and they are often kept in social isolation (Soulsbury et al., 2009). This leads to many animals eventually being rescued or confiscated by environmental authorities (where illegal) and taken to wildlife rescue centres. In Brazil, for example, approximately 4,600 monkeys were received in government wildlife rescue centres between 1999 and 2006 (Levacov et al., 2011). After the animals are received, there are three options for management: (1) euthanasia, (2) long-term captivity or (3) rehabilitation and reintroduction back into the wild. Currently, euthanasia is not often performed and most government wildlife rescue centres cannot keep the animals for long periods of time due to the constant influx of other wildlife. In certain cases, animals will end up in zoos; however, some animals will eventually be released back into the wild (Mitman et al., 2021).

Releasing Neotropical primates back into the wild may have positive and negative consequences for the environment as well as for resident wildlife species and even for humans (Mitman et al., 2021). Because of this, the International Union for the Conservation of Nature (IUCN) has proposed a set of guidelines for potential reintroduction cases. These include the Guidelines for the Management of Confiscated Live Organisms (Maddison, 2019) and the IUCN/SSC Re-Introduction Specialist Group: Guidelines for Nonhuman Primate Re-

Introductions (Baker, 2002). These guidelines include specific procedures that should be undertaken before attempting any non-human primate reintroduction, such as disease screening, behavioural and genetic assessment (Baker, 2002; Maddison, 2019). Unfortunately, only a handful of primate rehabilitation and reintroduction projects around the world actually follow these guidelines (Guy et al., 2014), and this may also be true for Latin American countries performing rehabilitation and reintroduction of Neotropical primates (Mitman et al., 2021), albeit this has not been widely studied.

Irrespective of the level of compliance with the IUCN guidelines among Latin American primate rehabilitation and reintroduction projects, the ethical aspects have not been explored. Thus, we aimed to investigate the ethical issues that may arise from the reintroduction of trafficked Neotropical primates back into the wild. To achieve this, we used a revised version of the Ethical Matrix (Mepham, 1996) for the ethical analysis of conservation-related issues (Biasetti and de Mori, 2021).

The Ethical Matrix (hereafter, EM) is an analytical tool originally proposed by Mepham in 1994 (Mepham et al., 2006). Methodologically speaking, the EM seeks to develop the principles encompassed by common morality, which refer to the ethical code of a society. Thus, the aim of the EM is to use principles that represent two major ethical theory traditions: consequentialism and deontology. Overall, consequentialism focuses mainly on the outcomes or consequences of choices (Mullan et al., 2017; Card and Smith, 2020). In other words, if a choice is morally right based solely on its consequences (Sinnott-Armstrong, 2021). In this sense, a certain choice could be deemed as morally right under a consequentialist perspective as long as the outcome is beneficial, even if the way of achieving said outcome is morally wrong. Conversely, deontology focuses on the way choices (or decisions) are made i.e., if they are made following a moral rule or norm (Mullan et al., 2017). Certain choices can be deemed as morally wrong even if the outcomes or consequences of these choices are beneficial; thus, only choices that conform to moral rules will be morally right (Alexander and Moore, 2021). The EM encompasses consequentialism and deontology in two of its three standard principles. The principle of *respect for wellbeing* represents utilitarianism, a consequentialist theory that seeks to ‘maximise the good’ (i.e., obtaining the most beneficial outcome for the greatest number of individuals) (Mepham et al., 2006, Mullan et al., 2017). The principle of *autonomy* represents deontological theories by following the moral rule of treating individuals as ‘ends’ that are important in themselves and not purely as ‘means’ to achieve the desired outcomes (Mepham et al., 2006).

The three standard principles of the EM are *respect for wellbeing*, *autonomy* and *fairness* (Mepham et al., 2006). These principles are arranged in columns whereas the ‘groups of interest’ (i.e., the groups that may be affected by the issue in question) are placed in rows. The EM expected outcomes include raising awareness on ethical issues, providing a basis for ethical decision-making and explaining the reasoning that led to specific ethical decisions (Mepham et al., 2006). The EM has been used to address various ethical issues, such as the use of biotechnology (Forsberg, 2004), genetically-modified fish (Kaiser et al., 2007) and fisheries (Kaiser and Forsberg, 2001), quality and ethics in educational research (Tangen, 2013) and ethical analysis in veterinary science (Miller, 2012). More recently, a revised version of the EM has been published to address conservation-related issues (Biasetti and de Mori, 2021) which I applied to the analysis of the ethical issues related the rehabilitation and reintroduction of trafficked Neotropical primates.

8.2. Methodology

8.2.1. Gathering information

Overall, building an EM involves three steps: gathering information, identifying the relevant stakeholders, and identifying the value demands (Biasetti and de Mori, 2021). In the context of building an EM for conservation, the *stakeholders* are potential ecological entities (e.g., ecosystems, taxa), individual animals or humans that may be affected by the (conservation) issue in question (Biasetti and de Mori, 2021). The *value demands* are the requirements that must be met to protect the ethical principles –i.e., respect for wellbeing, autonomy, and fairness– of the stakeholders (Biasetti and de Mori, 2021).

I gathered the relevant information for this study by conducting a search of the available scientific literature regarding the reintroduction of rescued and confiscated Neotropical primates performed in Latin America as well as the welfare of these animals in captivity (i.e., during rehabilitation and before being rescued or confiscated) and the related financial costs and views of human stakeholders involved in this issue. Studies related to the rehabilitation and reintroduction of trafficked primates are scarce in the scientific literature (Ongman et al., 2013). I conducted this scientific literature search using online scientific and open search engines, such as Web of Science, Science Direct, Google Scholar and Google. I used the words ‘primate’ + ‘rehabilitation’, ‘primate’ + ‘reintroduction’, ‘primate’ + ‘translocation’ and ‘primate’ + ‘trafficked’. This was performed in English and Spanish. I also reviewed the available International Union for the Conservation of Nature (IUCN) information on reintroduction such as the IUCN Global Reintroduction Perspective Series edited by Soorae (2002, 2008, among others), the IUCN Guidelines for Nonhuman Primate Re-introductions (Baker, 2002).For primate reintroduction, the review entitled ‘A History of Primate Reintroduction’ by Beck (2017) was particularly useful.

I found ten studies performed between 2000 and 2018. These studies included five performed in Latin American countries (i.e., Colombia, Belize, Brazil, Argentina and French Guiana) involving several Neotropical primate species; howler monkeys (*Alouatta* spp.), spider monkeys (*Ateles* sp.), woolly monkeys (*Lagothrix* sp.), capuchin monkeys (*Cebus* sp. and *Sapajus* sp.), squirrel monkeys (*Saimiri* sp.) and tamarins (*Saguinus* sp.). The studies included between five and 78 individuals and most were conducted primarily with wild-born monkeys that were rescued or confiscated from the illegal pet trade (90%, 9/10). One study included monkeys born in captivity along with the wild-born animals. A summary of the studies considered can be seen in Table 8.1 below. I used these studies to help me build the EM; however, the objective was to develop a generic EM that could be applied to the reintroduction of any Neotropical primate species.

Table 8.1

Summary of studies

Case study	Species	Sample size (n)	Age categories	Rehabilitation duration	Post-release monitoring	Health screening	Genetic testing	Post-release survival
Tricone, 2018	Black howler monkeys (*Alouatta pigra*)	20	Juveniles and adults	Not reported	3 months	Yes (pathogens not reported)	Not reported	70%
Sita, 2016	Bearded capuchin monkeys (*Sapajus libidinosus*)	78	Juveniles and adults	Not reported	6 months	Yes (arbovirus)	No	35%
Arango Guerra et al., 2013	Cotton-top tamarins (*Saguinus oedipus*)	5	Juveniles and adults	18 weeks	1 month	Not reported	Not reported	Not reported
Bennett et al., 2013	Brown woolly monkeys (*Lagothrix lagothrica*)	11	Juveniles and adults	Not reported	6 months	Not reported	Not reported	Not reported
de Palomino, 2013	Peruvian spider monkey (*Ateles chamek*)	9	Juveniles and adults	6 years	12 months	Yes (pathogens not reported)	Not reported	100%
Bruno et al., 2005	Brown howler monkeys (*Alouatta caraya*)	34	Juveniles and adults	NA (long-term semi-captivity)	NA	Not reported	Not reported	NA
Centro de Primatología Araguatos, 2004	White-fronted capuchin monkeys (*Cebus albifrons*)	17	Juveniles and adults	12 weeks	Yes	Yes (TB, hepatitis B, toxoplasmosis, intestinal parasites)	Yes	Not reported
Vogel et al., 2002	Common squirrel monkeys (*Saimiri sciureus*)	14	Adults	7 months	15 weeks	Not reported	Not reported	Not reported
Suárez et al., 2001	Brown capuchin monkeys (*Sapajus apella*)	9	Adults	5 months	6.5 months	Yes (TB, toxoplasmosis, intestinal parasites)	Not reported	Not reported
Brockett and Clark, 2000	Black howler monkeys (*Alouatta pigra*)	2	Juveniles	15 months	Not reported	Yes (TB, intestinal parasites)	Not reported	Not reported

8.2.2. Identification of stakeholders

Following the recommendations published by Biasetti and de Mori (2021) for building ethical matrices related to conservation issues, I selected seven stakeholders: (1) *ecosystem*, (2) *primate species of concern*, (3) *individual rescued/reintroduced primates*, (4) *individual free-ranging primates living at the release site*, (5) *local communities living in or in close proximity to the release site*, (6) *staff involved in the reintroduction project* and (7) *local society* (e.g., society in the city or country where the issue is taking place). Stakeholders were selected based on their involvement in the ethical issue and suitability for assessment (Mepham et al., 2006) and belonged to the three categories proposed by Biasetti and de Mori (2021) for conservation-related issues: (1) ecosystem, (2) animals, or (3) humans involved (Biasetti and de Mori, 2021).

The first stakeholder category, the *ecosystem*, refers to all the living organisms in the release site or the area where the monkeys are released as part of the reintroduction project. This includes other animals, plants, fungi, etc., and the way they interact with each other (Cambridge Dictionary, 2021). The second stakeholder category, the *primate species of concern*, refers to the Neotropical primate species that is part of the rehabilitation and reintroduction project (e.g., mantled howler monkey, *Alouatta palliata*). If more than one species were part of the reintroduction project, this would need to be considered to build the EM.

The third stakeholder category are the *individual rescued/reintroduced primates* that are part of the reintroduction project and will be released. The fourth stakeholder category are the *individual free-ranging primates living at the release site*, if any. The inclusion of this stakeholder category may be relevant or not depending on the specific characteristics of the rehabilitation and reintroduction project. Some of these projects could be performed in areas without free-ranging primates; hence, including this stakeholder category may not be necessary. However, if there are no free-ranging monkeys living at the release site, but the geographic area is historically known to be part of the natural area of occurrence of the species, then it is worth including this stakeholder category as changes may occur in the future. Note that Neotropical primates are represented in three different stakeholder categories: *primate species of concern, individual rescued/reintroduced primates* and *individual free-ranging primates living at the release site*. This is done intentionally to reflect the fact that in certain situations, particularly those related to conservation, the interests of individual animals may be in conflict with the interests of their species as a whole; hence, it is recommended to place them in multiple stakeholder categories (Biasetti and de Mori, 2021).

The fifth stakeholder category, the *local communities living in or in close proximity to the release site* refers to human communities living at the release site or in close proximity to it. These could be small communities such as rural communities with few residents or larger communities such towns and villages. Similarly to the case of the fourth stakeholder category, human communities may or may not be present at the release site, but their inclusion as a stakeholder category may be important if the likelihood of such communities establishing in the release area in the future is high.

The sixth stakeholder category, the *staff involved in the rehabilitation and reintroduction project* refers to all the people who work directly or indirectly with the rescued/confiscated primates that will be released. This could include veterinarians, biologists, academic researchers, wildlife rehabilitators, caregivers, undergraduate students and all the staff of the rescue centre as well as the people involved in the rescue or confiscation of the animals, such as environmental agency workers, police officers and fundraisers.

Finally, the seventh stakeholder category is composed by the *local society*. This refers to humans communities that may be interested in the rehabilitation and reintroduction project, such as people interested in animal welfare, wildlife conservation or environmental education, that may be affected by decisions made regarding the rehabilitation and reintroduction project.

8.3. An Ethical Matrix for the Rehabilitation and Reintroduction of Neotropical Primates

We filled this EM (Table 8.2) following an ethical matrix tailored for conservation-related issues (Biasetti and de Mori, 2021).

Table 8.2

Ethical Matrix of the rehabilitation and reintroduction of trafficked Neotropical primates

	1. Wellbeing	2. Autonomy	3. Fairness
A. Ecosystem	*Conservation of the ecosystem*: Biodiversity and the ecosystem where the monkeys will be released must be conserved. Hence, appropriate measures should be taken to minimise risks for the conservation of all species living in the ecosystem.	*Freedom from human intervention*: The reintroduction of rehabilitated monkeys should not compromise the species living in the release site, e.g., they should not compete for resources (i.e., overexploit resources) or introduce pathogens to naïve environments.	*Respect for the worth of every species*: There should be respect for the existence and value of all species living in the ecosystem where the rehabilitated monkeys will be reintroduced, i.e., no hierarchy of species irrespective of the human lens of interest such as aesthetics, genetic proximity or usefulness to humans.
B. Neotropical primate species of concern	*Conservation of the species:* Health and genetic risks could be associated with decreased long-term survival and/or damage to the viability of the species. This could also be the case if there was an increase in competition for food or other resources. There could also be a benefit for the species if population numbers were decreasing and rescued individuals were to be reintroduced to boost population numbers. In any case, genetic testing would be recommended for the conservation of the species as a whole.	*Free from human intervention:* Conservation efforts as well as rehabilitation and reintroduction projects should not compromise the primate species.	*Respect for the worth of the species:* There should be respect for the existence and value of the species. Many Neotropical primate species are considered umbrella species and have important ecosystem functions.
C. Reintroduced individual monkeys	*Health and physical and psychological welfare:* These are heavily dependent on the release method and the level of post-release monitoring and provisioning. The reintroduction project may lead to long-term survival and a good quality of life, or it may lead to a poor quality of life and even death. A thorough assessment of the risks and benefits of the reintroduction project for the wellbeing of the rescued individuals should be conducted before their release.	*Living natural lives and exercising species-specific behaviours:* Expression of complete, natural behavioural repertoire	*Respect for the worth of every individual:* Not living in worse conditions than those of captive conspecifics.

Table 8.2 (continued)

Ethical Matrix of the rehabilitation and reintroduction of trafficked Neotropical primates

D. Free-ranging individual monkeys living at the release site	*Health and physical and psychological welfare:* Health and welfare risks for free-ranging monkeys living at the release site could include infectious diseases and competition for food, territory, or other resources.	*Living natural lives and exercising species-specific behaviours:* This may depend on the size and resources available at the release site. Once the rescued individuals are released, if the population is too big for the ecosystem to sustain, the free-ranging individuals could be prevented from leading natural lives or exercising species-specific behaviours.	*Respect for the worth of every individual:* Not living in worse conditions after the rescued monkeys are released (i.e., conditions should be the same or better for the free-ranging monkeys after the rescued individuals are released).
E. Local communities living in or close to the release site	*Health and physical, psychological, economic and social welfare:* The release of rescued monkeys into the area should not pose a health or safety risk for local human communities, such as an increased risk of infection with zoonotic pathogens, injuries from aggression of released monkeys, or crop-raiding behaviours. Moreover, it could also offer wellbeing opportunities, such as ecotourism and pride in the environment.	*Freedom of choice:* The local human communities should be informed and consulted about the reintroduction project. Ideally, they should be actively involved in the design of the project and approve it before releasing the monkeys into the area.	*Fair treatment:* Right to be involved, informed, considered and consulted regarding the reintroduction project. Right to have an economic benefit from the reintroduction project, if possible (e.g., ecotourism).
F. People involved in the rehabilitation and reintroduction project	*Health and physical and psychological welfare*: Assessment of health and safety risks (e.g. injuries and zoonotic pathogens) and prevention of psychological discomfort and/or stress. Offer of wellbeing opportunities, such as financial, educational, pride in involvement in a project that promotes environmental education and/or conservation.	*Self-determination:* Having access to proper practical training (e.g., handling monkeys) and being stimulated-allowed to express their own concerns and interests.	*Fair treatment*: Working under clear instructions and in a safe environment and benefit from the research, if any is being carried out and according to their participation (e.g., authorship in scientific publications or conferences). Staff involved, such as veterinarians, biologists, psychologists and caregivers should have the freedom to act according to the codes of conduct of their profession or institution.
G. Local society	*Psychological welfare*: prevention of psychological discomfort and/or stress and promotion of wellbeing opportunities such as involvement in ecotourism, environmental education, etc.	*Freedom of choice*: The local society should be informed about the reintroduction project. Ideally, an environmental education programme would be in place to involve the local society and, if possible, receive their support.	*Fair treatment:* Right to be informed and have the opportunity to vote and give their opinions regarding the reintroduction project if possible and if not, at least for a democratic government with policies related to this aspect.

8.4. Analysis of situation and potential conflicts

In this Section, I will explain more thoroughly certain aspects of the relevant studies (Table 8.1) to perform a deeper situation analysis and to provide a frame for potential conflicts between categories of stakeholders considered in the EM.

8.4.1. Situation and conflicts relative to the conservation of the species

As stated by Biasetti and de Mori (2021), practices aimed at benefitting the conservation of the species of interest may harm individuals from the same species; thus, individual monkeys and their species as a whole should stand as separate stakeholders in this EM (Biasetti and de Mori, 2021). Moreover, individual monkeys may come from two sources: rescued and confiscated monkeys that are part of the reintroduction project, and free-ranging monkeys that already live in the release area (if any). In fact, four studies reviewed for this Chapter reported releasing monkeys in areas with free-ranging conspecifics (Brockett and Clark, 2000; Vogel et al., 2002; de Palomino, 2013; Tricone, 2018). In those cases, there could be potential for conflicts to arise between the stakeholders: *(B) Neotropical primate species of concern, (C) reintroduced individual monkeys* and *(D) free-ranging individual monkeys living at the release site*. These potential conflicts mainly include those associated with a risk of harming the long-term viability of the species such as the accidental introduction of pathogens or individuals of a different species or subspecies. Indeed, the reintroduction of trafficked individuals from non-native primate species could threaten the conservation of other primate or wildlife species native to the release site. This is the case of the buffy-tufted-ear marmoset (*Callithrix aurita*) and the buffy-headed marmoset (*Callithrix flaviceps*). These marmoset species are native to the Atlantic rainforest in southeast Brazil; however, they are currently endangered and listed in the IUCN Red List. This is partly due to the introduction of black-tufted marmosets (*Callithrix penicillata*) and common marmosets (*Callithrix jacchus*) which are native to the northeast of Brazil. These species were severely trafficked during the 1980s and 1990s leading to abandonment in the Atlantic rainforest, reproduction and genetic mixing with *C. aurita* and *C. flaviceps* (Zanon, 2020).

To mitigate these risks, health screening and genetic testing could be performed; in fact, they are a requirement of the IUCN for the reintroduction of non-human primates (Baker, 2002). Several Neotropical primate reintroduction studies reported carrying out health screening; specific tests included studies for tuberculosis, hepatitis B, toxoplasmosis, intestinal parasites and arboviruses (Brockett and Clark, 2000; Suárez et al., 2001; Centro de Primatología Araguatos, 2004; Sita, 2016; Tricone, 2018) and one study reported using genetic testing (Centro de Primatología Araguatos, 2004). A recent study performed by Oklander et al. (2020) reported that only four of 17 howler monkeys originated from the same genetic cluster to which they were reintroduced (i.e., to their native populations) (Oklander et al., 2020). Introducing animals from different genetic clusters may lead to the introduction of non-local genetic variability, as these individuals are not native to those primate populations (Oklander et al., 2020). This may result in an artificial, human-led mixture of different evolutionary lineages and homogenisation of diversity and biogeographic patterns, which could negatively affect the viability of the species in the long-term (Oklander et al., 2020).

8.4.2. Situation and conflicts relative to the wellbeing of free-ranging individual monkeys living at the release site

In addition to the potential of introducing pathogens, there could be other instances in which the wellbeing of the stakeholder categories *(C) reintroduced individual monkeys* and *(D) free-ranging individual monkeys living at the release site* could be in conflict. These could include competition or fights for resources such as food or access to females. I have not found evidence of this happening in any published study; nonetheless, several studies reported contact between free-ranging and reintroduced Neotropical monkeys as well as female dispersal, but competition or agonistic behaviours were not reported (Brockett and Clark, 2000; Vogel et al., 2002; de Palomino, 2013; Tricone, 2018).

8.4.3. Situation and conflicts relative to local human communities living in or close to the release area

Close contact between non-human primates and humans may lead to the transmission of zoonotic pathogens, and the potential for occurrence of this type of events is growing as the human-primate interface grows (Lappan et al., 2020). It is known that Neotropical primates may carry zoonotic pathogens such as *Leptospira* spp. (Aliaga-Samanez, 2021), *Mycobacterium tuberculosis* (Mitman et al., 2021), *Trypanosoma cruzi* (Mitman et al., 2021), hepatitis B virus (Mitman et al., 2021), simian foamy viruses (Muniz et al., 2017; Santos et al., 2019) and several intestinal parasites (e.g., *Ancylostoma* spp. in this dissertation, see Chapter 2). Moreover, other pathogens carried by primates have the potential to produce spill-overs or infection in human populations, such as *Plasmodium* *malariae* (Pereira-Figueiredo, 2017; Sousa, 2018), Zika virus (Han et al., 2019) and coronaviruses, potentially including, SARS-CoV-2 (Guimarães et al., 2020). Even though four of the reviewed reintroduction studies reported carrying out specific health tests for zoonotic pathogens, these were limited to tuberculosis, hepatitis B, toxoplasmosis, intestinal parasites and arboviruses (Centro de Primatología Araguatos, 2004; Suárez et al., 2001; Brockett and Clark, 2000; Sita, 2016). Thus, reintroducing primates without knowing their origin (as it may happen when rescuing or confiscating individuals from the illegal pet trade) may be detrimental for human health, as the people in charge of the reintroduction project would not know what pathogens to test for. Genetic testing may have the added advantage of providing accurate information on the species and/or subspecies of the individual and allow for adequate disease and pathogen screening.

Another situation in which the wellbeing of these categories of stakeholders (i.e., local human communities and reintroduced monkeys) could be in conflict is if the reintroduced individuals pose a safety risk for the local human communities. Centro de Primatología Araguatos (2004), for example, reported that the reintroduced monkeys kept coming back to the release site to try to interact with humans. Similarly, Suárez et al. (2001) reported aggression towards unfamiliar humans (Centro de Primatología Araguatos, 2004; Suárez et al., 2001). These situations may be prevented by performing long-term post-release monitoring and provisioning (e.g., 12 months as proposed by Baker, 2002) in which the injured, isolated and/or ill individuals are removed and taken back to captivity where possible, and by involving the relevant members of the human communities in the planning and execution of the reintroduction project.

Furthermore, Neotropical primate reintroduction projects could lead to economic damages for human communities. Centro de Primatología Araguatos (2004) reported that the reintroduced monkeys confused agricultural areas with the forest during the first few weeks (Centro de Primatología Araguatos, 2004). This could lead to economic loses for the human communities if the monkeys engage in behaviours such as crop-raiding. However, these behaviours could also be prevented by performing post-release monitoring. In reintroduction projects where the local human communities are involved, this could lead to economic benefits for them, such as revenue from ecotourism. Bennett et al. (2013), for example, reported economic benefits for the community, even though it was not explained how this was achieved (Bennett et al., 2013).

8.5. Conclusions and recommendations

The rehabilitation and reintroduction of rescued and/or confiscated Neotropical primates may lead to several potential ethical conflicts, mainly those related to the conservation of other primate or wildlife species as well as conflicts with resident free-ranging monkeys of the same or other species and human communities living closely or at the release site.

After reviewing the available scientific data on Neotropical primate rehabilitation and reintroduction projects and developing the EM specifically tailored for this issue, I conclude that there are certain situations in which Neotropical primates should not be released back into the wild:

(1) When the reintroduction of trafficked primates represents a substantial or significant risk for the health of resident primates, other wild animal populations, the ecosystem or humans, such as in cases where the health status of the trafficked monkeys has not been adequately assessed, ideally, by following the IUCN and/or government guidelines.

(2) When the reintroduction of trafficked primates has the potential of producing damage to the long-term viability of resident primate or other wildlife species living at the release site by introducing genes from another species or subspecies i.e., when the genetic status of the trafficked primates has not been assessed adequately, , by following the IUCN and/or government guidelines.

(3) When the reintroduction of trafficked primates has a significant potential of producing competition for resources with resident primates or with other wildlife and the habitat’s capacity has not been assessed adequately by following IUCN and/or government guidelines.

(4) When the reintroduction project has not secured sufficient funding to be able to provide long-term monitoring and provisioning of the released primates to ensure their wellbeing will not be compromised during the first weeks or months after release. This could include food provisioning, removal of ill, injured and/or isolated animals, etc. Ideally, this would be performed for a minimum of 12 months as proposed by the IUCN guidelines.

(5) When the reintroduction of trafficked primates has not been fully disclosed to and approved by the local human communities living in or closely to the release site, if any. Ideally, these communities should be actively involved in the process of

planning the reintroduction and could gain financial and/or other type of benefits from the reintroduction project, such as ecotourism, environmental education, etc.

Unfortunately, IUCN guidelines are seldom used in Latin American primate reintroduction projects (Mitman et al., 2021). In the studies found for this Chapter, only 10% (1/10) used genetic testing, 60% (6/10) did some form of disease screening before releasing the monkeys, and 40% (4/10) mentioned releasing monkeys in areas with resident monkeys of the same species without any agonistic interaction between the released and resident monkeys occurring during the post-release monitoring period. Encouragingly, 90% (9/10) of studies mentioned carrying out some form of post-release monitoring, ranging from one to 12 months (Centro de Primatología Araguatos, 2004; Arango Guerra, 2013; Suárez et al., 2001; Brockett and Clark, 2000; Tricone, 2018; de Palomino, 2013; Vogel et al., 2002; Sita, 2016; Bennett et al., 2013).

Even though practitioners, such as wildlife rehabilitators, agree with the IUCN Guidelines for the Re-introduction of Non-human Primates (Baker, 2002) which encourage measures such as long-term monitoring and disease screening, the lack of funding, government support and public awareness limit their application in Latin American countries, as evidenced by a recent study performed in Peru (Mitman et al., 2021). Hence, there needs to be an adaptation of these guidelines to the reality of Latin American countries (Mitman et al., 2021).

Rehabilitation and reintroduction projects of Neotropical primates should involve the local human communities who live in the release site or closely to it (if any) to respect their freedom of choice and treat them fairly. This needs to be done before the animals are released, and it should be clear how the reintroduction project will affect the human communities as well as any potential risks and/or benefits for them. Similarly, the local society must be informed as well and could benefit by the establishment of an environmental education programme, for example.

Finally, the staff involved in the rehabilitation and reintroduction project, such as undergraduate students, caregivers, researchers and wildlife rehabilitators, must have the sufficient training to work safely with the animals and the freedom to express their concerns and interests. They should be treated fairly and be allowed to act in accordance with their professional codes of conduct.

In conclusion, the rehabilitation and reintroduction of trafficked Neotropical primates may be a viable option to manage these animals as long as the potential for ethical conflicts between stakeholders has been addressed, there is adherence to IUCN guidelines (ideally) or government guidelines, and the five points mentioned above have been met: (1) health assessment, (2) genetic testing, (3) habitat survey, (4) long-term monitoring and (5) local community involvement (ideally) and approval (minimally).

It must be taken into account that this EM was built using exclusively the scientific evidence obtained from the 10 studies considered for this Chapter and related to the reintroduction of Neotropical trafficked primates (i.e., I did not consider the reintroduction or translocation of captive-born Neotropical primates). It may be the case where potentially beneficial outcomes of the rehabilitation and reintroduction of trafficked New World monkeys have been overlooked simply because this information or data has not been published. Thus, further research is needed to understand the outcomes of trafficked Neotropical primate rehabilitation and reintroduction projects, and to which the above mentioned conclusions and other guidelines (e.g., the IUCN guidelines) can be adapted to the reality of Latin American countries.

Chapter 9: General Discussion

Chapter 9: General Discussion

The main aim of this dissertation was to investigate the association of individual differences in personality with health and behaviour during the rehabilitation of trafficked bearded capuchin monkeys (*Sapajus libidinosus*). There are few examples in the scientific literature regarding the rehabilitation of trafficked Neotropical primates and its association with personality traits albeit thousands of these animals are received every year in Latin American wildlife rescue centres and then released back into the wild (Levacov et al., 2011; Mitman et al., 2021).

The first objective of this dissertation (Chapter 3) was to investigate the health of rehabilitant bearded capuchin monkeys (*Sapajus libidinosus*) maintained in Northeast Brazilian wildlife rescue centres by (1) using non-invasive health indicators (a body condition score and a coat condition score developed for this purpose) and (2) performing physical examinations and blood collections to obtain physiological and haematological values as well as morphometry and parasitological status of these individuals and compare them with previously published data.

The inter-observer reliability of the body condition score was variable and deemed as acceptable (i.e., with a significant p-value) for the scorings obtained during the physical examinations and non-invasively (i.e., remotely). Nonetheless, the correlation coefficients between body mass index and the aspects of the body condition score obtained separately (body condition – general; body condition – face/head; body condition – body) were moderate at best (ICC 3,k = 0.42 – 0.65, $p \leq 0.03$). Body mass index seemed to be strongly correlated to body weight ($rs \geq 0.74$, $p \leq 0.04$) as reported in other primate species (e.g., macaques, Berman and Schwartz, 1988). Hence, I considered it appropriate to use it as the 'gold standard' to validate the body condition score. Even though other methods can be used to validate body condition scores in primates such as x-ray absorptiometry (e.g., Summers et al., 2005), this was not possible in the present study due to lack of funding and resources. The coat condition score had strong inter-observer reliability (ICC 3,k ≥ 0.72, $p < 0.01$) in all applications and a good level of validity (i.e., correlation, $r = 0.55$, $p < 0.01$) when comparing the score used during the physical examinations and non-invasively (remotely). However, both scores could benefit from being used in studies with larger sample sizes, as the one used in this study was relatively small ($n = 26$).

Physiological and haematological parameters of bearded capuchin monkeys in the present study were similar to those observed in previous published studies, except for cardiac frequency. This was probably related to the use of the anaesthetic xylazine, which seems to promote bradycardia in this and other animals species. More research is needed on the effect of alpha2-agonists on the cardiovascular parameters of bearded capuchins. Even though I did not find any age or sex-related differences in the haematological values considered, previous studies suggest some of these differences exist and may not have been observed due to the small sample size of the present study. Thus, more research is needed on the haematological and physiological parameters of bearded capuchin monkeys, both in the wild and in captivity. This study was the first to investigate the relationship between infection by *Ancylostoma* spp. and haematological values of rescued capuchins. I found that ancylostomiasis leads to disruption in haematological values in bearded capuchins similar to those observed in other animal species (e.g., dogs) and humans (Ngui et al., 2012; Lappin, 2013). Similarly, more research could be performed to understand the effect of parasites and other pathogens on the haematology and general health of capuchin monkeys in reintroduction programs, so that they may have the best chance of survival after release and do not pose a risk to other wildlife or humans.

The second objective (Chapter 4) was to analyse the behaviour of rehabilitant bearded capuchin monkeys to provide an initial assessment of the efficacy of the rehabilitation programme and propose potential improvements (n = 16). This was performed by using changes in behaviour across time (activity budgets), space and substrate use and a behavioural diversity index (the Shannon H index) and comparing with previously published data, both in captivity and in the wild, where applicable.

The analysis of the activity budgets revealed that there was an increase in affiliative behaviours and a decrease in affiliative and aggressive behaviours directed towards humans when comparing the start and the end of the 3.5-month rehabilitation period. I considered these results as positive (i.e., suggesting ‘rehabilitation success’). Social skills and social cohesion are vital for primates and other wild animals to survive in the wild (Melfi and Marples, 2000). Moreover, the extinction of behaviours directed towards humans (as they were not observed during the last observation phase) is another positive finding of this study, because these behaviours could potentially endanger the animals when released by making them prone to re-capture by humans or to develop undesirable behaviours such as crop-raiding. Space use did not change significantly across time and substrate use changed only related to the use of the mobile bed, which increased after it was placed in the enclosures after the second observational phase. I considered this as a positive finding, as the monkeys seemed to be responding to a new environmental enrichment device by using it. Even though space use did not change nor the use of the other substrates, I did not consider this as a negative finding as the enclosures of the monkeys were quite small (as in, much smaller than their home ranges would be in the wild) and this could have prevented them from expressing their full natural behavioural repertoire. Furthermore, bearded capuchins have been observed spending up to 90% on the ground and below 10 meters of the canopy during the dry season in certain parts of their natural area of occurrence (Oliveira et al., 2014). Hence, the use of the lower parts of the canopy and the ground does not seem to be a disadvantage for this species as much as for other arboreal Neotropical primate species.

I used the Shannon H index in this study to measure behavioural diversity as used in previous studies. I hypothesised that there would be a significant change when comparing the start and the end of the rehabilitation period. On average, there was a significant change and when analysed individually, 13 of the 16 individuals considered showed an increase in their calculated Shannon H indexes. This suggests that the rehabilitation programme was helping the individuals to increase their behavioural diversity. However, I did not find a correlation between the Shannon H indexes and the exhibition of stress-related behaviours as has been observed in previous studies with other animal species (Millet et al., 2020). Further research is needed to understand the association between behavioural diversity and ‘rehabilitation success’ (i.e., addressing research questions such as if animals with higher behavioural diversity indexes survive for longer after release than animals with lower indexes) as well as between behavioural diversity and wellbeing of bearded capuchin monkeys.

The third objective of this dissertation (Chapter 5) was to broaden the current scientific knowledge of capuchin personality structure by replicating the findings from Nunes (2017), utilising two methodological approaches to measure personality: behavioural tests and observer trait ratings (the Hominoid Personality Questionnaire or HPQ; Weiss et al., 2009). By using the HPQ (n = 18), I found four personality dimensions: Openness, Neuroticism, Assertiveness and Sociability. These dimensions closely resembled those obtained in previous studies with larger sample sizes, such as the study performed by Morton et al. (2013) with captive brown capuchin monkeys (*Sapajus apella*, n = 127) and the study performed by Fernández-Bolaños et al. (2020) with free-ranging yellow-breasted capuchin monkeys (*Sapajus xanthosternos,* n = 26). Like them, I used Principal Component Analysis (PCA) with Varimax and Promax rotations to analyse the data and correlated the dimensions obtained with behavioural observations. These results support the existence of the personality dimensions obtained in these studies, possibly across the whole genus *Sapajus,* and the use of PCA for lexical personality assessment even with small sample sizes.

Similarly, I obtained five personality constructs (stereotypic, aggressive, risk-averse, sociable to humans and creative) by using the behavioural tests as described by Nunes (2017) and based on Uher et al. (2013) (n = 15). These resembled the constructs ‘Curiousness’, ‘Creativeness–Inventiveness’, ‘Anxiousness’, ‘Arousability’, ‘Social orientation to humans’ and ‘Aggressiveness’ found by Uher et al. (2013) in captive brown capuchin monkeys (*Sapajus apella*) and supports the existence of these constructs in other species of the genus *Sapajus*. Nonetheless, there could be certain concerns when using an ‘invasive approach’, such as behavioural tests, that require manipulation and social isolation of the individuals in a rehabilitation and reintroduction programme. In this sense, there may be advantages of using a ‘non-invasive approach’ such as the HPQ and behavioural observations instead of behavioural tests for individuals aimed for reintroduction. Even though highly standardised tests provide greater control over the variables measured (Gosling, 2008; Uher, 2011), they could potentially disrupt the rehabilitation of the individuals, particularly those tests that require close interaction with humans and/or in monkeys that have a strong attachment to humans. Even though I did not find a significant difference in time spent in motor stereotypies during the behavioural tests when comparing with behavioural observations done in the enclosure where the animals were normally housed or in pre-feeding conditions, I did not test if the individuals that underwent the behavioural tests had differences regarding rehabilitation outcomes. Thus, analysing the benefits and costs as well as advantages and disadvantages of performing behavioural tests with the objective of assessing personality in rehabilitant capuchin monkeys must be performed for ethical and practical reasons, particularly if the behavioural tests are performed close to the release of the monkeys back to the wild. Even though certain personality traits such as boldness and neophilia have been associated with high survival prospects for reintroduced animals (Bremner-Harrison et al., 2004; Sita, 2016), other studies have failed to find such association (Lopes et al., 2017). Nevertheless, social groups composed of individuals with different personality traits may be more stable and fare better against environmental variations (than groups composed by animals with similar personality traits) as the variability of these groups relies not only on individual differences in behaviour but also in different physiological responses to pathogens and environmental stressors (Watters et al., 2003; Watters and Meehan, 2007).

The fourth objective (Chapter 6) was to investigate the association between personality scores (obtained from Chapter 5) and health parameters (obtained from Chapter 2) of rehabilitant bearded capuchin monkeys. I found that time spent in stress-related behaviours significantly predicted body condition score (BCS) (F(1,11) = 7.703, p = 0.018, R2 = 0.412, n = 13) and Sociability significantly predicted neutrophil-to-lymphocyte ratios (NLR) (F(3,9) = 7.564, p = 0.019, R2 = 0.407, n = 13). Individuals that spent more time engaged in motor stereotypies and self-directed behaviours were more likely to have a lower body condition score when compared to individuals that spent less time engaged in these behaviours. Furthermore, more sociable monkeys had lower NLR when compared to less sociable individuals. These findings were consistent with the previous studies with capuchins. NLR tend to be higher in stressed individuals and act as indicators of immune function, and more sociable capuchin monkeys have been found to be less stressed and have higher levels of wellbeing when compared to less sociable conspecifics (Robinson et al., 2016). The association between body condition and time spent in motor stereotypies and self-directed behaviours was less clear and straightforward. I hypothesised that capuchins who spent more time engaged in these behaviours could have been more distracted and less interested in the food provided and/or were spending more energy by performing these behaviours. Further research is needed to understand the association between stress-related behaviour and body condition in bearded capuchin monkeys.

The fifth objective (Chapter 7) was to investigate the association between personality scores (obtained from Chapter 5) and rehabilitation outcomes (obtained from Chapter 4) of bearded capuchins (n = 13). In this study, I found significant associations between time spent in vigilance-related behaviour during rehabilitation and the personality traits stereotypic, aggressive, and Assertiveness. More stereotypic individuals, as well as less aggressive and assertive monkeys, increased the time spent in vigilance behaviours when comparing the start and the end of the 3.5-month rehabilitation period. Conversely, less stereotypic and more aggressive and assertive capuchins decreased the time spent in vigilance behaviours across the rehabilitation period. These results are consistent with the results reported in previous studies with captive capuchin monkeys (Morton et al., 2013) and different to those observed with capuchins in the wild (Fernández-Bolaños et al., 2020). As proposed by Fernández-Bolaños et al. (2020), vigilance behaviours can be seen as anti-predator behaviours, leading to bolder individuals expressing them (Fernández-Bolaños et al., 2020). Thus, the results from the present study suggested that, by the end of the 3.5-month rehabilitation period, the individuals were still behaving as other captive capuchin monkeys would do and not as wild, free-ranging monkeys would. Ideally, the behaviour of primates who are going to be reintroduced back into the wild should resemble the behaviour of wild, free-ranging conspecifics. However, the enclosures where the individuals were housed in the present study were quite different in terms of size (home range), predator threats and general environmental conditions when compared to the home environments of capuchin monkeys in the wild. As mentioned above, this could have prevented the individuals from expressing their full natural behavioural repertoire. Thus, I suggest being careful when interpreting these results and comparing them with free-ranging conspecifics in both Chapters (4 and 5).

I found that more neurotic capuchins decreased the time spent in fixed trunks (trunks placed in the capuchin enclosures) when comparing the start and the end of the 3.5-month rehabilitation period. Conversely, less neurotic capuchins increased the time spent in this tier. Less open individuals slightly increased the time spent in the highest tier of their enclosures whilst more open individuals decreased the time spent in this tier. This seems to be relatively similar to previous findings of studies on personality and reintroduction, where bold individuals tend to engage in more risky behaviours when compared to shy (less bold) individuals (Bremner-Harrison et al., 2004). I suggested this based on the fact that boldness has been positively associated with medium to high scores on Openness and low scores on Neuroticism in previous studies performed with humans (e.g., Poy et al., 2014; Donnellan and Burt, 2016). If these associations exist in capuchin monkeys, using the ground and medium tier of the enclosure as well as the fixed trunks could be interpreted as risky behaviours performed more frequently by less neurotic and more open individuals, as the predominant substrate in the enclosure of the monkeys was concrete and there were less fixed trunks than concrete surfaces. Similarly, descending to the ground could be ‘risky’ for arboreal primates, such as capuchins, because of potential predators. Further research could be made to study the personality trait boldness (e.g., performing specific tests where boldness can be measured, such as those proposed by Nunes, 2017) and its association with Openness and Neuroticism in trafficked capuchin monkeys that are part of reintroduction programmes.

Finally, the sixth objective (Chapter 8) was to investigate the ethical issues that may arise from the reintroduction of trafficked Neotropical primates by using a revised version of the Ethical Matrix for conservation-related issues from Biasetti and de Mori (2021).

I found only a handful of studies (10) in which trafficked Neotropical primates were rehabilitated and reintroduced back to the wild. It was evident that the rehabilitation and reintroduction of trafficked Neotropical primates may lead to several potential ethical conflicts, such as the conservation of other primate or wildlife species, conflicts with resident wild, free-ranging monkeys of the same or other species, and human-wildlife conflicts, such as crop-raiding or other conflicts with human communities living at or close to the release site.

After reviewing the available scientific data on this theme and developing the Ethical Matrix, I concluded that there are certain situations in which Neotropical primates should not be reintroduced back into the wild. These mainly include situations (1) where there is a significant risk of damaging the health, long-term viability and/or producing competition with resident human, primate or other wildlife communities by reintroducing the trafficked monkeys; (2) when the reintroduction project has not secured sufficient funding to provide long-term monitoring and provisioning of the released monkeys to ensure their wellbeing will not be compromised during the first weeks or months after their release; finally, (3) when the reintroduction of trafficked Neotropical primates has not been fully disclosed to and approved by the local human communities living in or close to the release site if any.

It must be considered that the Ethical Matrix is a framework that uses principles of utilitarianism, consequentialism and deontology (Mepham et al., 2006); thus, this framework seeks to find the most beneficial outcome for the largest number of individuals. This may not always apply to every individual case or may lead to conflicts of opinion among wildlife conservationists, animal welfare scientists, etc. Hence, I suggest using both the Ethical Matrix and the set of recommendations I proposed as tools to guide decision-making in Neotropical primate rehabilitation and reintroduction projects and not as conclusive, inflexible rules.

There may be cases where potentially beneficial outcomes of the rehabilitation and reintroduction of trafficked Neotropical primates have been overlooked simply because this information or data has not been published. Hence, further research is needed to fully understand the outcomes of rehabilitation and reintroduction programmes of trafficked Neotropical primates and the ethical issues related to them.

Overall, I suggest that the rehabilitation and reintroduction programme of which the individuals of the present study were part could benefit from an extension of the rehabilitation period (from four months to 12 months, for example, as proposed by the IUCN Guidelines for Nonhuman Primate Re-introductions; Baker, 2002) and certain modifications to the rehabilitation protocol.

As the current rehabilitation programme does not include any type of anti-predator training and/or ‘soft-release’, I suggest the inclusion of an anti-predator training programme (if deemed beneficial and ethically viable) and the provision of a naturalistic environment where the capuchins can learn survival skills such as the location of food and water and general use of the canopy, or the specific environment in which they will live and learn to travel, forage, and reproduce. This could be achieved by releasing the animals and providing long-term (e.g., 12-18 months) food and monitoring or by using a pre-release enclosure at the rescue centre and/or at the release site. Furthermore, personality could be assessed in a similar way to the present study, to further investigate the association between personality traits and rehabilitation and reintroduction of trafficked bearded capuchin monkeys.

The overall aim of this dissertation was to study the relationship between individual differences in personality and health and behaviour during the rehabilitation of trafficked bearded capuchin monkeys that were part of a government reintroduction programme in Northeast Brazil. Even though I found significant results, it must be considered consider that, as mentioned throughout the dissertation, the experimental studies (Chapters 3, 4, 5, 6 and 7) were performed with small sample sizes, partly due to the limitations and restrictions related to the Covid-19 pandemic. Hence, further research is needed to fully understand the relationship between personality traits and health and behaviour during the rehabilitation of bearded capuchin monkeys, as well as the replicability of these studies.

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Appendix 1: Health data (Chapter 3)

Physiological parameters obtained from all individuals examined during July 2019 (n = 27)

Name	Age category	Sex class	Rescue centre	Temperature (°C)	Heart rate (bpm)	Respiratory rate (rpm)
Acara	Adult	Female	CETAS/CE	36.1	120	32
Aclara	Adult	Female	CETAS/CE	35.9	64	40
Angélica	Adult	Female	CETAS/CE	38.6	112	52
Fénix	Adult	Female	CETAS/CE	37.3	104	56
Florentina	Adult	Female	CETAS/CE	37.6	120	36
Silvana	Adult	Female	CETAS/CE	38.3	80	40
Dana	Adult	Female	CETAS/RN	38.7	80	44
V	Adult	Female	CETAS/RN	35.9	128	48
Fúria	Adult	Female	CETAS/RN	38.3	116	32
Tapa	Adult	Female	CETAS/RN	38.4	72	52
Melequinha	Adult	Male	CETAS/CE	37.6	108	20
Peruco	Adult	Male	CETAS/CE	38.4	120	60
Walber	Adult	Male	CETAS/CE	38.2	104	28
Lombinho	Adult	Male	CETAS/RN	38.3	92	60
Café	Adult	Male	CETAS/RN	38.3	88	52
Galápagos	Adult	Male	CETAS/RN	37.3	116	60
Arrebite	Adult	Male	CETAS/RN	39.5	80	52
Magrela	Juvenile	Female	CETAS/CE	39.7	80	72
Li	Juvenile	Female	CETAS/RN	38.8	128	88
Rabinha	Juvenile	Female	CETAS/RN	36.1	92	88
Piolho	Juvenile	Male	CETAS/CE	39.0	108	32
Garrincha	Juvenile	Male	CETAS/RN	NA	112	72
Vitinho	Juvenile	Male	CETAS/RN	37.2	92	60
Mãozinha	Juvenile	Male	CETAS/RN	39.1	104	62
Tiquinho	Juvenile	Male	CETAS/RN	39.0	96	56
Joana	Adult	Female	CETAS/RN	37.8	116	64
Amadeu	Adult	Male	CETAS/CE	39.0	108	68
Tino	Adult	Male	CETAS/RN	38.2	100	56

Weight, crown-rump length (CRL) and body mass index (BMI) of individuals sampled during July 2019 (n = 26)

Name	Age category	Sex class	Rescue centre	Weight (kg)	Crown-rump length (CRL) (cm)	Body mass index (BMI)
Acara	Adult	Female	CETAS/CE	2.40	34.00	20.72
Aclara	Adult	Female	CETAS/CE	1.72	28.50	21.11
Angelica	Adult	Female	CETAS/CE	1.64	29.00	19.44
Fenix	Adult	Female	CETAS/CE	2.44	32.50	23.10
Florentina	Adult	Female	CETAS/CE	1.65	31.50	16.58
Silvana	Adult	Female	CETAS/CE	2.49	30.50	26.77
Dana	Adult	Female	CETAS/RN	2.40	35.00	19.59
V	Adult	Female	CETAS/RN	1.65	31.50	16.63
Furia	Adult	Female	CETAS/RN	1.85	32.50	17.51
Tapa	Adult	Female	CETAS/RN	2.20	31.00	22.89
Melequinha	Adult	Male	CETAS/CE	4.32	36.00	33.33
Peruco	Adult	Male	CETAS/CE	3.00	34.00	25.95
Walber	Adult	Male	CETAS/CE	2.83	31.50	28.47
Lombinho	Adult	Male	CETAS/RN	3.20	33.50	28.51
Café	Adult	Male	CETAS/RN	2.90	35.00	23.67
Galapagos	Adult	Male	CETAS/RN	3.20	33.00	29.38
Arrebite	Adult	Male	CETAS/RN	3.15	34.50	26.47
Magrela	Juvenile	Female	CETAS/CE	1.78	28.00	22.64
Li	Juvenile	Female	CETAS/RN	1.40	25.75	21.11
Rabinha	Juvenile	Female	CETAS/RN	1.15	25.00	18.40
Piolho	Juvenile	Male	CETAS/CE	1.84	27.50	24.33
Garrincha	Juvenile	Male	CETAS/RN	1.30	26.00	19.23
Vitinho	Juvenile	Male	CETAS/RN	1.85	28.00	23.60
Mãozinha	Juvenile	Male	CETAS/RN	1.30	25.50	19.99
Tiquinho	Juvenile	Male	CETAS/RN	1.50	27.00	20.58
Joana	Adult	Female	CETAS/RN	1.80	32.00	17.58
Amadeu	Adult	Male	CETAS/CE	2.36	27.00	32.37
Tino	Adult	Male	CETAS/RN	2.65	33.75	23.26

Red cell blood counts of individuals sampled during July 2019 (n = 26)

Name/ID	**Age category**	**Sex class**	**Erythrocytes ($x10^{12}$/L)**	**Packed cell volume**	**Haemoglobin (g/L)**	**Mean corpuscular volume (fL)**	**Mean corpuscular haemoglobin concentration (%)**
Acara	Adult	Female	4.85	0.37	115.00	76.20	31.00
Aclara	Adult	Female	5.08	0.37	115.00	72.20	31.30
Angélica	Adult	Female	4.74	0.36	112.00	75.90	31.10
Fénix	Adult	Female	5.79	0.42	127.00	72.10	30.30
Florentina	Adult	Female	4.56	0.35	104.00	75.80	30.00
Silvana	Adult	Female	5.09	0.40	123.00	78.30	30.80
Dana	Adult	Female	6.52	0.48	116.00	73.00	24.20
V	Adult	Female	5.41	0.35	86.00	65.00	24.50
Fúria	Adult	Female	5.96	0.40	98.00	67.00	24.20
Tapa	Adult	Female	6.60	0.48	121.00	72.00	25.30
Joana	Adult	Female	3.85	0.27	65.00	70.00	24.20
Melequinha	Adult	Male	5.72	0.41	122.00	71.10	29.90
Peruco	Adult	Male	4.99	0.37	116.00	73.70	31.50
Walber	Adult	Male	5.57	0.43	142.00	76.40	33.30
Lombinho	Adult	Male	5.78	0.43	104.00	75.00	24.00
Café	Adult	Male	5.46	0.39	94.00	71.00	24.20
Galápagos	Adult	Male	7.11	0.51	123.00	71.00	24.30
Arrebite	Adult	Male	7.19	0.51	129.00	71.00	25.10
Magrela	Juvenile	Female	4.76	0.37	111.00	76.80	30.30
Li	Juvenile	Female	6.56	0.42	103.00	64.00	24.40
Rabinha	Juvenile	Female	5.39	0.40	102.00	74.00	25.80
Piolho	Juvenile	Male	4.80	0.33	106.00	69.10	31.90
Vitinho	Juvenile	Male	5.33	0.39	94.00	72.00	24.20
Mãozinha	Juvenile	Male	6.20	0.43	110.00	70.00	25.50
Tiquinho	Juvenile	Male	6.13	0.42	102.00	68.00	24.30
Amadeu	Adult	Male	5.02	0.38	125.00	75.40	32.90

White cell blood counts of all individuals sampled during July 2019 (n = 26)

Name/ID	Age category	Sex class	Leucocytes (x10^{9}/L)	Band neutrophils (x10^{9}/L)	Segmented neutrophils (x10^{9}/L)	Eosinophils (x10^{9}/L)	Basophils (x10^{9}/L)	Lymphocytes (x10^{9}/L)	Monocytes (x10^{9}/L)	Platelets (x10^{9}/L)
Acara	Adult	Female	17.60	0.00	11.09	0.53	0.00	4.93	1.06	206.00
Aclara	Adult	Female	18.20	0.00	5.64	2.55	0.00	9.10	0.91	179.00
Angélica	Adult	Female	7.90	0.00	2.69	0.32	0.00	4.42	0.47	404.00
Fénix	Adult	Female	10.90	0.00	5.34	0.55	0.00	4.47	0.55	350.00
Florentina	Adult	Female	20.40	0.00	16.32	0.20	0.00	3.26	0.61	247.00
Silvana	Adult	Female	7.30	0.00	3.58	0.51	0.00	2.70	0.51	176.00
Dana	Adult	Female	10.41	0.00	5.62	1.15	0.00	2.91	0.73	290.00
V	Adult	Female	18.81	0.00	13.73	0.11	0.00	2.82	1.13	296.00
Fúria	Adult	Female	11.88	0.00	8.91	0.48	0.00	1.90	0.59	318.00
Tapa	Adult	Female	8.60	0.00	5.33	0.00	0.00	2.32	0.95	482.00
Joana	Adult	Female	12.00	0.00	6.84	0.00	0.00	5.16	0.00	347.00
Melequinha	Adult	Male	11.90	0.00	4.05	0.24	0.00	7.02	0.60	214.00
Peruco	Adult	Male	13.00	0.00	7.41	0.39	0.00	4.55	0.65	296.00
Walber	Adult	Male	10.60	0.00	5.41	0.53	0.00	4.03	0.64	306.00
Lombinho	Adult	Male	17.37	0.00	11.81	1.56	0.00	2.61	1.39	351.00
Café	Adult	Male	10.83	0.00	6.39	0.43	0.00	3.36	0.65	462.00
Galápagos	Adult	Male	15.30	0.00	13.46	0.46	0.00	1.07	0.31	831.00
Arrebite	Adult	Male	8.36	0.00	4.26	0.75	0.00	2.09	1.25	335.00
Magrela	Juvenile	Female	21.00	0.00	7.14	0.42	0.00	11.97	1.47	471.00
Li	Juvenile	Female	18.27	0.00	10.05	0.37	0.37	6.03	1.46	293.00
Rabinha	Juvenile	Female	21.66	0.00	12.56	0.22	0.00	6.71	2.17	389.00
Piolho	Juvenile	Male	16.80	0.00	5.71	1.18	0.00	8.90	1.01	292.00
Vitinho	Juvenile	Male	7.62	0.00	3.12	0.69	0.00	3.58	0.23	266.00
Mãozinha	Juvenile	Male	14.89	0.15	8.49	0.74	0.00	3.57	1.94	493.00
Tiquinho	Juvenile	Male	13.40	0.00	11.12	0.40	0.00	1.88	0.00	384.00
Amadeu	Adult	Male	17.20	0.00	7.05	0.86	0.00	8.08	1.20	295.00

Appendix 2: Glossary of terms used in English and translated to Portuguese from the Hominoid Personality Questionnaire (Weiss et al., 2009) (Chapter 5)

Fearful: Subject reacts excessively to real or imagined threats by displaying behaviours such as screaming, grimacing, running away or other signs of anxiety or distress.

Medroso: O indivíduo reage de forma excessiva a ameaças reais ou imaginárias, apresentando comportamentos como gritar, fazer careta de medo, fugir ou outros sinais de ansiedade, angústia ou aflição

Dominant: Subject is able to displace, threaten, or take food from other monkeys. Or subject may express high status by decisively intervening in social interactions.

Dominante: O indivíduo é capaz de afastar, ameaçar ou tomar comida de outros macacos. O indivíduo pode também expressar seu status superior intervindo de forma decisiva em interações sociais.

Persistent: Subject tends to continue in a course of action, task, or strategy for a long time or continues despite opposition from other monkeys.

Persistente: Indivíduo tende a continuar em uma mesma ação, tarefa ou estratégia por muito tempo e continua independentemente de outros estímulos ao seu redor.

Cautious: Subject often seems attentive to possible harm or danger from its actions.

Cauteloso: Indivíduo costuma aparentar ser cauteloso e atento a possíveis ameaças ou consequências de suas ações. Aparenta evitar situações que possam ser perigosas.

Stable: Subject reacts to its environment including the behaviour of other monkeys in a calm, equable, way. Subject is not easily upset by the behaviours of other monkeys.

Estável: O indivíduo reage de forma calma e uniforme ao seu entorno, inclusive a comportamentos de outros macacos. O indivíduo não é perturbado facilmente pelo comportamento de outros macacos.

Stereotypic (original *Autistic*): Subject often displays repeated, continuous, and stereotyped behaviours such as rocking or self-clasping.

Estereotipado: Indivíduo recorrentemente apresenta comportamentos repetidos, contínuos e estereotipados como movimentos pendulares, auto-mutilação, auto-enganche (puxar/abraçar de forma aberrante os próprios membros do corpo como perna, rabo, etc), locomoção aberrante (ir para um lado e para o outro do recinto sem motivo aparente).

Curious: Subject has a desire to see or know about objects, devices, or other monkeys. This includes a desire to know about the affairs of other monkeys that do not directly concern the subject.

Curioso: O indivíduo apresenta desejo de ver ou saber sobre objetos, dispositivos ou outros macacos. Isso inclui um desejo de saber sobre eventos envolvendo outros macacos que não são de interesse direto para o indivíduo.

Reckless: Subject is rash or unconcerned about the consequences of its behaviours.

Inconsequente: Indivíduo comumente se comporta de forma imprudente ou inconsequente.

Stingy/greedy: Subject is excessively desirous or covetous of food, favored locations, or other resources. Subject is unwilling to share these resources with others.

Avarento/ganancioso: O indivíduo é excessivamente desejoso ou ávido por comida, lugares favoritos ou outros recursos. O indivíduo é relutante em compartilhar esses recursos com outros macacos.

Jealous: Subject is often troubled by others who are in a desirable or advantageous situation such as having food, a choice location, or access to social groups. Subject may attempt to disrupt activities of advantaged conspecifics.

Ciumento: O indivíduo fica frequentemente incomodado quando outros estão em uma situação desejável ou vantajosa, como ter comida, lugar preferido ou acesso a grupos sociais. O indivíduo pode tentar interromper as atividades dos macacos favorecidos.

Individualistic: Subject's behaviour stands out compared to that of the other individuals in the group. This does not mean that it does not fit or is incompatible with the group.

Individualista: O comportamento do indivíduo se sobressai em relação ao comportamento dos outros indivíduos do grupo. Isso não significa que o indivíduo não se adapte ou que seja incompatível com o grupo.

Sociable: Subject seeks and enjoys the company of other monkeys and engages in amicable, affable, interactions with them.

Sociável: O sujeito busca e desfruta da companhia de outros macacos e participa de interações amigáveis e afáveis com eles.

Distractible: Subject is easily distracted and has a short attention span.

Distraído: Indivíduo é facilmente distraído e passa pouco tempo prestando atenção a uma coisa só.

Timid: Subject lacks self-confidence, is easily alarmed and is hesitant to venture into new social or non-social situations.

Tímido: Indivíduo tem pouca auto-confiança, é facilmente assustado e hesita em entrar em situações sociais com outros indivíduos.

Sympathetic: Subject seems to be considerate and kind towards others as if sharing their feelings or trying to provide reassurance.

Complacente: O indivíduo parece ser atencioso e preocupado com os outros, como se compartilhasse os seus sentimentos ou tentasse dar segurança.

Playful: Subject is eager to engage in lively, vigorous, sportive, or acrobatic behaviours with or without other monkeys.

Brincalhão: Indivíduo aparenta gostar de se envolver em comportamentos esportivos, acrobáticos (tal como se pendurar pelo rabo de cabeça para baixo), balançar troncos, brincadeiras (com outros macacos ou sozinho).

Solitary: Subject prefers to spend considerable time alone not seeking or avoiding contact with other monkeys.

Solitário: O indivíduo prefere passar muito tempo sozinho, sem procurar ou evitando o contato com outros macacos.

Vulnerable: Subject is prone to be physically or emotionally hurt as a result of dominance displays, highly assertive behaviour, aggression, or attack by another monkey.

Vulnerável: O indivíduo tende a ser fisicamente ou emocionalmente ferido como resultado de demonstrações de dominância, comportamentos assertivos ou de ataques por parte de outros macacos.

Innovative: Subject engages in new or different behaviours that may involve the use of objects or materials or ways of interacting with others.

Inovador: O indivíduo engaja em comportamentos novos ou diferentes que podem envolver o uso de objetos ou materiais, assim como formas de interagir com os outros.

Active: Subject spends little time idle and seems motivated to spend considerable time either moving around or engaging in some overt, energetic behaviour.

Ativo: Indivíduo passa pouco tempo parado e aparenta gostar de passar o tempo se movendo, andando pelo recinto ou se engajando em um comportamento ativo que envolva gasto de energia.

Helpful: Subject is willing to assist, accommodate, or cooperate with other monkeys.

Prestativo: O indivíduo se mostra disposto a ajudar, acomodar ou cooperar com outros macacos.

Bullying: Subject is overbearing and intimidating towards younger or lower ranking monkeys

Bullying: Indivíduo pratica bullying com outros macacos (intimida-os, ameaça, geralmente macacos mais novos ou em posições mais baixas na hierarquia)

Aggressive: Subject often initiates fights or other menacing and agonistic encounters with other monkeys.

Agressivo: O indivíduo muitas vezes inicia brigas ou encontros ameaçadores e agonísticos com outros macacos.

Affectionate: Subject seems to have a warm attachment or closeness with other monkeys. This may entail frequently grooming, touching, embracing, or lying next to others.

Afetuoso: O indivíduo parece ter um vínculo caloroso ou de proximidade com outros macacos. Isso pode implicar em catações, toques e abraços frequentes, ou deitar-se frequentemente junto a outros.

Excitable: Subject is easily aroused to an emotional state. Subject becomes highly aroused by situations that would cause less arousal in most monkeys.

Excitável: O indivíduo é facilmente estimulado para um estado emocional. O indivíduo se torna altamente instigado por situações que causariam menor entusiasmo em outros macacos.

Inquisitive: Subject seems drawn to new situations, objects, or animals. Subject behaves as if it wishes to learn more about other monkeys, objects, or persons within its view.

Inquisitivo: O indivíduo parece ser atraído para situações, objetos ou animais novos. O indivíduo se comporta como se desejasse aprender mais sobre outros macacos, objetos ou pessoas no seu campo de visão.

Submissive: Subject often gives in or yields to another monkey. Subject acts as if it is subordinate or of lower rank than other monkeys.

Submisso: Indivíduo comumente apresenta um comportamento submisso, tem sua comida/lugar roubado, geralmente por indivíduos mais dominantes,

Cool: Subject seems unaffected by emotions and is usually undisturbed, assured, and calm.

Tranquilo: O indivíduo parece não ser afetado pelas emoções e é normalmente imperturbável, seguro e calmo.

Dependent/follower: Subject often relies on other monkeys for leadership, reassurance, touching, embracing and other forms of social support.

Dependente/seguidor: O indivíduo habitualmente depende de outros macacos para liderança e reconforto, procurando contato, abraços e outras formas de apoio social.

Irritable: Subject often seems in a bad mood or is impatient and easily provoked to anger exasperation and consequent agonistic behaviour.

Irritável: O indivíduo parece habitualmente mal-humorado ou impaciente, sendo fácil provocar raiva, exasperação e, consequentemente, comportamentos agonísticos.

Unperceptive: Subject is slow to respond or understand moods, dispositions, or behaviours of others.

Desapercebido: O indivíduo demora para compreender ou responder a estados de ânimo, motivações ou comportamentos dos outros.

Appendix 3: Results from Spearman correlation analyses (Chapter 5)

Personality trait/behaviour	**Personality trait/behaviour**	**Spearman Rho**	***p*-value**	***p*-value after Holm-Bonferroni sequential correction**	**N**
Environmental manipulation	Openness	0.843	0.000*	0.001*	18
Solitary play	Openness	0.828	0.000*	0.002*	18
Vigilance	Openness	-0.678	0.002*	0.214	18
Risk-averse	Affiliative	-0.721	0.002*	0.257	15
Aggressive	Human interaction	0.642	0.010*	1.000	15
Sociable to humans	Environmental manipulation	0.600	0.018*	1.000	15
Risk-averse	Vigilance	0.554	0.032*	1.000	15
Assertiveness	Stereotypic	-0.518	0.048*	1.000	15
Affiliative	Openness	0.445	0.064	1.000	18
Sociability	Sociable to humans	0.454	0.089	1.000	15
Assertiveness	Aggressive	0.443	0.098	1.000	15
Creative	Human interaction	0.435	0.105	1.000	15
Locomotion	Assertiveness	0.383	0.117	1.000	18
Openness	Sociable to humans	0.407	0.132	1.000	15
Affiliative	Sociability	0.366	0.135	1.000	18
Human interaction	Assertiveness	0.358	0.145	1.000	18
Risk-averse	Solitary play	-0.391	0.150	1.000	15
Sociable to humans	Vigilance	-0.382	0.160	1.000	15
Inactivity	Openness	-0.340	0.168	1.000	18
Aggressive	Agonistic	0.374	0.169	1.000	15
Foraging	Openness	-0.329	0.182	1.000	18
Aggressive	Solitary play	-0.363	0.184	1.000	15
Stereotypic	Locomotion	-0.361	0.187	1.000	15
Vigilance	Neuroticism	0.323	0.191	1.000	18
Assertiveness	Risk-averse	-0.354	0.196	1.000	15
Human interaction	Neuroticism	-0.318	0.198	1.000	18
Risk-averse	Agonistic	-0.338	0.218	1.000	15
Creative	Affiliative	0.336	0.221	1.000	15
Sociable to humans	Agonistic	-0.332	0.227	1.000	15
Aggressive	BPIS	0.325	0.237	1.000	15
Openness	Creative	0.321	0.243	1.000	15
Agonistic	Openness	-0.285	0.251	1.000	18
Locomotion	Sociability	0.284	0.254	1.000	18
Vigilance	Sociability	-0.282	0.257	1.000	18
Assertiveness	Creative	-0.311	0.260	1.000	15
Stereotypic	Solitary play	0.310	0.260	1.000	15
Inactivity	Neuroticism	-0.276	0.268	1.000	18
Sociability	Creative	0.300	0.277	1.000	15
Foraging	Assertiveness	-0.269	0.280	1.000	18
BPIS	Sociability	0.261	0.295	1.000	18
Sociability	Stereotypic	-0.286	0.302	1.000	15
Neuroticism	Risk-averse	0.282	0.308	1.000	15
Stereotypic	Foraging	0.271	0.328	1.000	15
Solitary play	Neuroticism	-0.244	0.330	1.000	18
Risk-averse	Human interaction	-0.266	0.338	1.000	15
Sociable to humans	Solitary play	0.254	0.361	1.000	15
Stereotypic	BPIS	-0.254	0.362	1.000	15
Creative	BPIS	-0.254	0.362	1.000	15
Foraging	Sociability	-0.228	0.363	1.000	18
Environmental manipulation	Sociability	0.228	0.363	1.000	18
Creative	Vigilance	-0.243	0.383	1.000	15
Risk-averse	Locomotion	0.239	0.390	1.000	15

Personality trait/behaviour	**Personality trait/behaviour**	**Spearman Rho**	***p*-value**	***p*-value after Holm-Bonferroni sequential correction**	**N**
Risk-averse	Environmental manipulation	-0.239	0.390	1.000	15
Assertiveness	Sociable to humans	-0.232	0.405	1.000	15
Stereotypic	Vigilance	-0.229	0.413	1.000	15
Risk-averse	BPIS	0.225	0.420	1.000	15
Stereotypic	Environmental manipulation	0.221	0.428	1.000	15
Foraging	Neuroticism	0.199	0.428	1.000	18
Creative	Environmental manipulation	0.218	0.435	1.000	15
Openness	Risk-averse	-0.211	0.451	1.000	15
Stereotypic	Inactivity	-0.207	0.459	1.000	15
Sociable to humans	BPIS	0.207	0.459	1.000	15
Affiliative	Sociability	0.183	0.467	1.000	18
Affiliative	Neuroticism	-0.179	0.478	1.000	18
Sociable to humans	Inactivity	-0.196	0.483	1.000	15
Aggressive	Vigilance	0.196	0.483	1.000	15
Aggressive	Locomotion	-0.182	0.516	1.000	15
Human interaction	Sociability	0.147	0.561	1.000	18
Locomotion	Openness	-0.146	0.565	1.000	18
Agonistic	Assertiveness	0.146	0.565	1.000	18
Neuroticism	Stereotypic	-0.157	0.576	1.000	15
Sociability	Aggressive	0.157	0.576	1.000	15
Creative	Locomotion	-0.150	0.594	1.000	15
Aggressive	Affiliative	-0.143	0.612	1.000	15
Risk-averse	Foraging	0.139	0.621	1.000	15
Creative	Affiliative	0.139	0.622	1.000	15
Creative	Foraging	-0.132	0.639	1.000	15
BPIS	Assertiveness	0.117	0.645	1.000	18
Stereotypic	Affiliative	0.127	0.653	1.000	15
BPIS	Neuroticism	-0.102	0.687	1.000	18
Inactivity	Sociability	-0.100	0.693	1.000	18
Creative	Inactivity	0.111	0.694	1.000	15
Sociable to humans	Foraging	-0.111	0.694	1.000	15
Sociable to humans	Locomotion	0.111	0.694	1.000	15
Aggressive	Inactivity	0.111	0.694	1.000	15
Creative	Solitary play	0.101	0.721	1.000	15
Neuroticism	Sociable to humans	-0.100	0.723	1.000	15
Human interaction	Openness	-0.086	0.735	1.000	18
Neuroticism	Creative	-0.093	0.742	1.000	15
Stereotypic	Affiliative	-0.089	0.752	1.000	15
Risk-averse	Inactivity	0.089	0.752	1.000	15
Locomotion	Neuroticism	0.079	0.754	1.000	18
BPIS	Openness	-0.069	0.785	1.000	18
Inactivity	Assertiveness	-0.059	0.817	1.000	18
Environmental manipulation	Neuroticism	-0.051	0.842	1.000	18
Openness	Aggressive	-0.050	0.860	1.000	15
Affiliative	Assertiveness	0.042	0.868	1.000	18
Sociable to humans	Human interaction	0.037	0.897	1.000	15
Solitary play	Assertiveness	-0.033	0.897	1.000	18
Aggressive	Foraging	-0.036	0.899	1.000	15
Vigilance	Assertiveness	-0.032	0.900	1.000	18
Stereotypic	Human interaction	0.032	0.910	1.000	15
Environmental manipulation	Assertiveness	0.028	0.913	1.000	18
Sociable to humans	Affiliative	-0.029	0.919	1.000	15
Sociability	Risk-averse	-0.029	0.919	1.000	15
Neuroticism	Aggressive	0.029	0.919	1.000	15
Aggressive	Environmental manipulation	-0.021	0.940	1.000	15

Personality trait/behaviour	**Personality trait/behaviour**	**Spearman Rho**	***p*-value**	***p*-value after Holm-Bonferroni sequential correction**	**N**
Agonistic	Neuroticism	0.019	0.941	1.000	18
Solitary play	Sociability	0.008	0.975	1.000	18
Openness	Stereotypic	0.000	1.000	1.000	15

Appendix 4: Results (*p*-values) obtained from the Analyses of Variance of Repeated Measures (Chapter 7)

***p*-values obtained from the Analysis of Variance of Repeated Measures (ANOVA RM) (n = 12)**									
	Stereotypic	Creative	Risk-averse	Sociable to humans	Aggressive	Openness	Neuroticism	Assertiveness	Sociability
Foraging	0.751	0.462	0.238	0.777	0.547	0.552	0.926	0.083	0.197
Locomotion	0.786	0.202	0.411	0.628	0.787	0.389	0.741	0.597	0.125
Affiliative	0.263	0.260	0.150	0.973	0.512	0.509	0.531	0.172	0.088
Agonistic	0.272	0.341	0.341	0.341	0.453	0.341	0.272	0.453	0.272
Inactivity	0.859	0.563	0.406	0.645	0.785	0.771	0.750	0.191	0.581
Environmental manipulation	0.739	0.166	0.260	0.544	0.706	0.602	0.567	0.206	0.282
Solitary play	0.103	0.456	0.033*	0.755	0.103	0.028*	0.195	0.954	0.551
Vigilance	0.003*	0.332	0.717	0.349	0.009*	0.521	0.936	0.047*	0.482
Human interaction	0.358	0.329	0.321	0.358	0.321	0.350	0.329	0.321	0.350
BPIS	0.492	0.670	0.818	0.289	0.790	0.478	0.301	0.974	0.324