

Different responses of free-ranging wild guanacos (*Lama guanicoe*) to shearing operations: implications for better management practices in wildlife exploitation

P Taraborelli^{*†‡}, ME Mosca Torres[†], PF Gregorio[‡], P Moreno^{†§}, V Rago[#], A Panebianco[‡],
NM Schroeder^{‡¶}, R Ovejero^{‡¶} and P Carmanchah[‡]

[†] Grupo de Investigaciones de la Biodiversidad (GIB), Instituto Argentino de Investigaciones de Zonas Áridas, CCT-CONICET, Av Ruiz Leal s/n, Parque General San Martín, Mendoza ciudad CC 507, CP 5500, Argentina

[‡] Grupo de Investigación en Ecofisiología de Fauna Silvestre (GIEFAS), AUSMA - INIBIOMA - CONICET - UNCo, San Martín de los Andes, Neuquén, Argentina

[§] Lab de Ecología de Enfermedades, ICIVET, CONICET-UNL, Universidad Nacional del Litoral, Esperanza, Santa Fe, Argentina

[#] Wildlife Health and Health Policy Program, Wildlife Conservation Society (WCS), Buenos Aires, Argentina

[¶] Lab de Interacciones Ecológicas, IADIZA, CCT-Mendoza, CONICET, Argentina

* Contact for correspondence and requests for reprints: ptarabor@mendoza-conicet.gob.ar/paulataraborelli@gmail.com

Abstract

In certain areas of South America, free-ranging, wild guanacos (*Lama guanicoe*) are exploited for fibre by local people. This activity includes the capture and handling of animals which can adversely affect their behaviour and physiology. This study investigated the behavioural and physiological responses of guanacos to shearing and handling activities in order to obtain a better picture of the welfare state of individuals. Parameters that were assessed consisted of: time enclosed; handling time; sex; age; and vital signs (heart beat frequency per minute, respiratory rate per minute, body temperature and body condition). Blood samples were also collected to measure serum cortisol levels and neutrophil/lymphocyte ratios. Frequencies of spitting, kicking, escape attempts and vocalisations were recorded as behaviours considered indicative of stress. Our results showed that stress behaviour frequencies were higher with increased handling time, whereas serum cortisol and N/L levels were higher when body condition scores were low. Handling time should be kept as short as possible to minimise individual's stress levels, particularly when body condition is low. Stress behaviour rates and serum cortisol levels were higher in juvenile compared to adult guanacos. Finally, both physiological measures of stress — serum cortisol concentrations and N/L ratios — were higher during the management activities of 2010 than in 2009, which may have been as a result of more inclement weather in 2010. When managing guanacos, it is important to consider both animal traits and previous environmental conditions and to avoid shearing juveniles and individuals with poor body condition scores if weather conditions are severe. These management recommendations are likely to improve animal welfare, facilitating sustainable management of this wild and emblematic species from the desert biomes of South America.

Keywords: animal welfare, handling, *Lama guanicoe*, neutrophil/lymphocyte ratios, serum cortisol level, stress behaviours

Introduction

The economic exploitation of wild animals in a non-destructive manner might be a way of guaranteeing the long-term persistence of biodiversity (Giles 1971, 1978; Chardonnet *et al* 2002). It can also be a solution, in certain circumstances, for promoting the recovery of eroded populations (Smith & Wishnie 2000). If the people managing the wildlife resource are locals, standing to benefit from said exploitation, then there is a greater likelihood of the management being sustainable (Primack 2002; Baldi *et al* 2010). In effect, the cost of losing the resource is deemed too big to risk jeopardising it. Yet, developing sustainable exploitation programmes for endangered species and eroded populations requires a

sound scientific basis (Taylor & Dunstone 1996; Caro 1999a,b). By definition, exploitation of wildlife necessitates a degree of intervention by a population which may, in itself, already present a risk to the resource (Berger & Cunningham 1998; Williams *et al* 2002). However, the effects of such interventions on the wildlife remain poorly understood (Weber Nielsen & Bergfeld 2003).

Wildlife management based on the welfare of the individual animal encompasses behaviour, physiology, husbandry and even the cultural background of the keepers and managers. The latter is related, among other things, to the discomfort and pain that could be caused directly by humans due to the capture system, manipulation or captivity (Bonacic & Gimpel 2006). Animal capture and handling affect both the

behaviour (Vilá & Cassini 1993; Cook *et al* 2000; Tarlow & Blumstein 2007) and the physiology (Creel 2005) of animals. Among the variables that may respond immediately are: body temperature, heart rate, respiratory rate and packed cell volume (Eckert & Randall 1983; Radostits *et al* 1994; Schmidt-Nielsen 1997; Harris *et al* 1999; Zapata *et al* 2004). A less-immediate response (ie from min to h) can be observed in serum cortisol concentrations (Coles 1980; Kaneko *et al* 1997; Bateson & Wise 1998; Harris *et al* 1999; Zapata *et al* 2004; Bonacic *et al* 2006). The induced stress response is also characterised by an increase in circulating neutrophils and a decrease in lymphocytes and eosinophils (Harlow *et al* 1987; Ferre *et al* 1998). Although the neutrophil/lymphocyte ratio (N/L) and hormone levels are both stress indicators, there is evidence that they may not be correlated in wild animals, suggesting differential responses to stress (Müller *et al* 2011). Stress hormones can show changes relatively quickly (3–5 min), while leukocyte changes are detectable later (30 min) (Romero & Romero 2002; McLaren *et al* 2003; Lynn & Porter 2008). Therefore, it is generally recommended to record both metrics together, as complements, to have full assessment of the stress response (Davis *et al* 2008; Johnstone *et al* 2012). Also, the N/L ratio is positively related to the magnitude of the stressor (Davis *et al* 2008). Furthermore, the usefulness of these measures is enhanced when utilised in combination with behavioural observations (Goddard *et al* 1994, 1996).

Shearing has been shown to elicit responses consistent with acute unease in wild camelids (Bonacic *et al* 2003, 2006; Arzamendia *et al* 2010; Ovejero 2013), confirming the negative effects of capture observed in other wild ungulates (Cook *et al* 2000; Morgan & Trømborg 2007; Tarlow & Blumstein 2007). The guanaco (*Lama guanicoe*) is a large herbivore native to South America (Franklin 1983) and has a peculiar trait which allows the procurement of an extremely fine fibre via shearing, precluding the need for killing: thus providing a novel example of sustainable wildlife use.

Although preliminary investigations (Carmanchahi *et al* 2011; Taraborelli *et al* 2011) already alluded to physiological and behavioural changes occurring as a result of capture and shearing, there remains a lack of a comprehensive appraisal of the impact of handling operations on individuals with poor body condition scores and high neutrophil-lymphocyte ratio in blood. The objective of this study was to analyse both behavioural and physiological responses to provide a more accurate picture of the welfare of the guanaco. More detailed information is necessary to ascertain the appropriate protocols as regards guanaco handling during shearing (Marull & Carmanchahi 2008); typically this should integrate modern notions of animal welfare (Morton *et al* 1995).

In order to meet this aim, an investigation was carried out where different indicators of stress were investigated simultaneously during handling operations, including the frequency of different behavioural responses (hereafter stress behaviour), stress hormone levels (cortisol), neutrophil/lymphocyte ratios (hereafter N/L), and vital signs (heart-beat frequency, respiratory rate, body

temperature). As such, we predicted elevated levels in serum cortisol and N/L ratios with increased time spent in corrals and longer handling times, as well as poor body condition score. Furthermore, we expected that stress behavioural responses would be positively correlated with vital signs, N/L ratios, and serum cortisol levels during handling activities.

Materials and methods

Study area

This study was carried out in October 2009 (two capture-handling sessions within 15 days) and in October 2010 (three sessions within ten days) at La Payunia Provincial Reserve, Mendoza, central-west Argentina (between 36° 00' S 68° 34' W and 36° 36' S 69° 23' W, 1,300–2,000 m above sea-level; 450,000 ha). The climate is typically continental desert with an average temperature of 6°C in winter and 20°C in summer, and 255 mm of rainfall in a typical year (Candía *et al* 1993; Martínez Carretero 2004). The guanaco population in La Payunia has been estimated to have grown continuously by an average of 2% per year since 1982 until 2003 (Puig *et al* 2003). Population growth is due mainly to the prevention of poaching and management measures aimed both at species and habitat conservation. Nowadays, the area supports the largest migratory guanaco population in central Argentina, with more than 26,000 individuals in spring (Novaro *et al* 2005; Schroeder *et al* 2013). Guanacos operate within complex social systems structured into family, bachelor and mixed groups, as well as solitary adult males (Franklin 1983). Each year, prior to first period of capture, a temporary campsite is established in an abandoned oilfield at the base of Cerro El Loro (36° 06' S - 69° 09' W), approximately 140 km from the nearest human settlement. Each year the campsite is dismantled after the final capture period.

Two hundred and ten guanacos were captured during the two events in 2009, while 103 were captured in three capture events in 2010. A high percentage of captured guanacos were males (2009: 71.6%, 2010: 81.7%); and most were adults (2009: 78.3%, 2010: 82%). When the number of guanacos captured was high, the shearing continued into the following morning.

Capture, handling and shearing were performed in compliance with a government-agreed protocol 'Protocol of good practices of management in wild guanacos (*Lama guanicoe*)' (Marull & Carmanchahi 2008), 'Guidelines for the use of animals in research' and to rulings 14346 of the Argentina National Law, and Province Park Permit No 795/10 of the Direction of Renewable Natural Resources, Secretary of the Environment, Mendoza Province.

Stages of management

Operations consisted of chasing and grouping guanacos via horseback into a huge corral trap (chasing distance 8 km; see Carmanchahi *et al* 2011; Taraborelli *et al* 2011; <http://www.payunmatru.com/>). The corral trap consisted of two arms forming a 'V' shape, with a distal opening 1,500 m

wide. These 2-m high arms consisted of black netting in the form of 2-cm knotless nylon mesh netting and were 2,000 and 3,000 m in length (north and south arm, respectively). These funnelled into a 300-m wide opening while another entrance, 50-m wide, separated by a sliding door made from green netting to prevent the escape. The funnel led to a series of inter-connecting square holding corrals, separated by wooden doors. These decreased in length and size: corral 1) 70 × 3 m (length × width) (210 m²); corral 2) 20 × 3 m (60 m²); and corral 3) 10 × 3 m (30 m²). Corral 3 led to a 0.6 × 0.4 m timber-walled chute (see Carmanchahi *et al* 2011). The corral walls had light brown rug material and fishing net to reduce potential stress from external visual stimuli. To avoid wounds due to escape attempts, corral 3 also had nylon mesh netting as ceiling. All the corrals had native forage available for the animals. In the chute, each guanaco was handled, blindfolded, hobbled, and placed onto a wooden stretcher by two or three individuals.

Measures

Enclosed time was defined as the period animals entered the corral trap until the immobilising stage (range 3–30 h). Handling time referred to the period between immobilisation and release. Total handling time ranged between 5–22 min. During veterinary procedures, sex, age category (juveniles < 2 years old; adults ≥ 2 years old; from dentition development), and vital signs (heart beat frequency per min; respiratory rate per min; body temperature, measured with a digital rectal probe) were registered and blood samples taken for physiological analyses (serum cortisol levels and N/L ratios). Body condition was investigated by palpating the degree of sharpness of spinous processes, muscle mass and fat cover adjacent to the lumbar vertebrae (Audige *et al* 1998); palpation was always carried out by the same person. Scores ranged from 1 to 5: Score 1 = Very poor condition (cachexia), lumbar spinous processes are very sharp and there is little muscle and no fat cover; Score < 2 = Poor condition (skinny or leanness), spinous processes are slightly enveloped, not prominent and can be easily felt by palpation with slight finger pressure; Score 2–<3 = Moderate/normal condition (slim), spinous processes are slightly enveloped and not prominent and are felt by palpation with moderate finger pressure; Score 3–<4 = Good condition (normal), spinous processes are enveloped and are felt by palpation only with firm finger pressure; and Score > 4–5 = Very good condition (fat), lumbar spinous processes are well enveloped and unable to be palpated (Audige *et al* 1998). The guanacos captured in Payunia ranged from 2–3.5 body condition. Male guanacos were marked via application of a blue collar and females a red one. Each collar was numbered, thus enabling identification of recaptured animals.

Behavioural responses to handling

We recorded the behaviour of guanacos during handling time in the chute until release using continuous focal sampling (Lehner 1996). All observations were recorded on a voice recorder. Stress behaviours were recorded as the number of occurrences of spitting, kicking, escape attempts and vocali-

sations (snorts and sharp shouts; Arzamendia *et al* 2010; Taraborelli *et al* 2011). We calculated rates as the number of stress behaviours/time of handling in min per individual. Increases in rates of these behaviours have been described as acute signs of stress in other ungulate species (Cook *et al* 2000; Tarlow & Blumstein 2007; Arzamendia *et al* 2010).

Data from 62 guanacos were obtained, 24 in 2009 (14 adults and three juvenile males, five adults and two juvenile females), and 38 in 2010 (23 adults and six juvenile males, six adults and two juvenile females).

Stress hormone response

The shorn individual was moved, immediately, to another sector for veterinary procedures. Blood samples were collected by venipuncture of the femoral vein via sterile syringe using 18 G needles. One blood sample was taken per individual and divided into two sub-samples, one for stress hormones analysis and the other for blood cell analyses. Blood samples were obtained from 62 guanacos (eleven adults and four juvenile females, 23 adults and six juvenile males).

For stress hormone analyses, the blood sample was centrifuged for 15 min at 3,000 rpm in a portable 12 V centrifuge (Mobilespin, Vulcan Technologies, Grandview, Missouri, USA). The serum was fractionated into cryovial tubes, labelled, and frozen in liquid nitrogen until transportation to the laboratory (Institute of Medicine and Experimental Biology of Cuyo, IMBECU, CCT-Mendoza, Argentina), where samples were preserved at –20°C. Serum cortisol concentration (ng ml⁻¹) was determined using Immunotech Cortisol Radioimmunoassay Kit 1 M 1841 (Immunotech, Beckman Coulter, Czech Republic) (Carmanchahi *et al* 2011; Ovejero 2013). Sensitivity of the assay was 3.62 ng ml⁻¹. Intra-assay coefficient of variation was < 5.8% (n = 10 replicates per sample). Zapata *et al* (2004) found concentrations of 7.9 (± 1) ng ml⁻¹ baseline plasma cortisol in semi-domesticated male guanacos in farms in Chile.

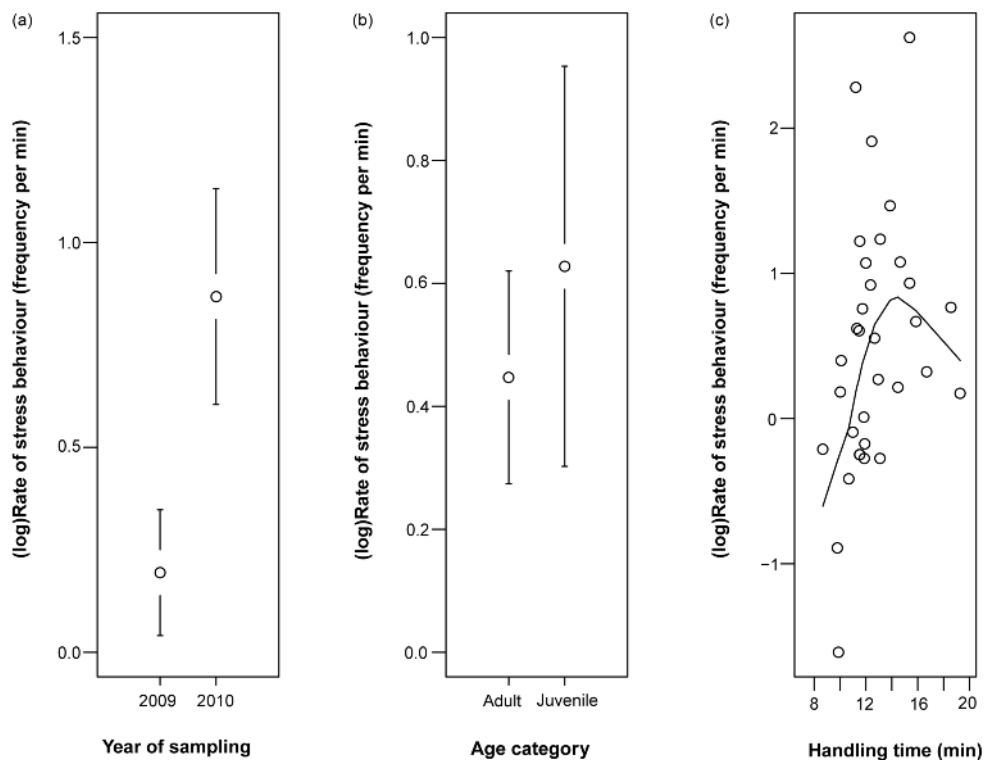
Neutrophil/lymphocyte ratio

Sub-samples for blood cell analysis were immediately homogenised with heparin following venipuncture. Blood smears were fixed in 99% methanol stained with Giemsa to perform WBC differentials. Before performing cell counts, white and red blood cell visibility was enhanced by applying a pre-packaged dilution (Ery-tic®, Leukotic® from Bioanalytic, Umkirch, Freiburg, Germany). N/L ratios were assessed for 24 guanacos (nine adults and three juvenile females, ten adults and two juvenile males). The small sample size was due to considerable logistical constraints in the field; analyses were required to be carried out the same day as blood extraction, meaning a significantly reduced period remained for counting cells in the field.

Statistical analysis

Three generalised linear models (GLM; McCullagh & Nelder 1983) were designated to test for the effects of handling operations on guanacos. The response variables were stress behaviour rate, serum cortisol levels and N/L ratio; whilst explanatory variables were study year, sex, age, time of day, handling time, period of time enclosed, body

Figure 1



The rate of stress behaviour ratio (frequency per min) (a) and the years of sampling (2009 and 2010). Data are expressed as means (\pm SEM). The rate of stress behaviour ratio (frequency per min) (b) and age category of guanacos (adult and juvenile). Data are expressed as mean (\pm SEM). The rate of stress behaviour ratio (frequency per min) (c) and handling time (min).

temperature and body condition score. Stress behaviour rate variable was \log_{10} -transformed to meet model assumptions of normal distribution and homogeneity of variances. A *posteriori* graphic visualisation confirmed a normal distribution of the error structure.

GLMs were performed using R statistical software version 3.1.1 (R Development Core Team 2013). Model selection was based on the Akaike information criterion (AIC; Akaike 1974) and model selection was obtained using the 'stepAIC' function from MASS package. This function performs a step-wise model selection, starting with full models (models including a given response variable and all potential explanatory variables), and successively removing the variables/factors that least contribute to model explanation, selecting the best one (with the lowest AIC value). We considered models differing in AIC value by more than $\Delta AIC > 1$ to be significantly different from each other (Ims & Yoccoz 1997). Model selection was carried out using the 'AICtab' function from the bbmle package (Bolker 2012). We also performed a correlation test to evaluate the relationship between serum cortisol levels and N/L ratio. In this case, we only included data from 15 individuals with complete data of serum cortisol levels and N/L ratio.

Results

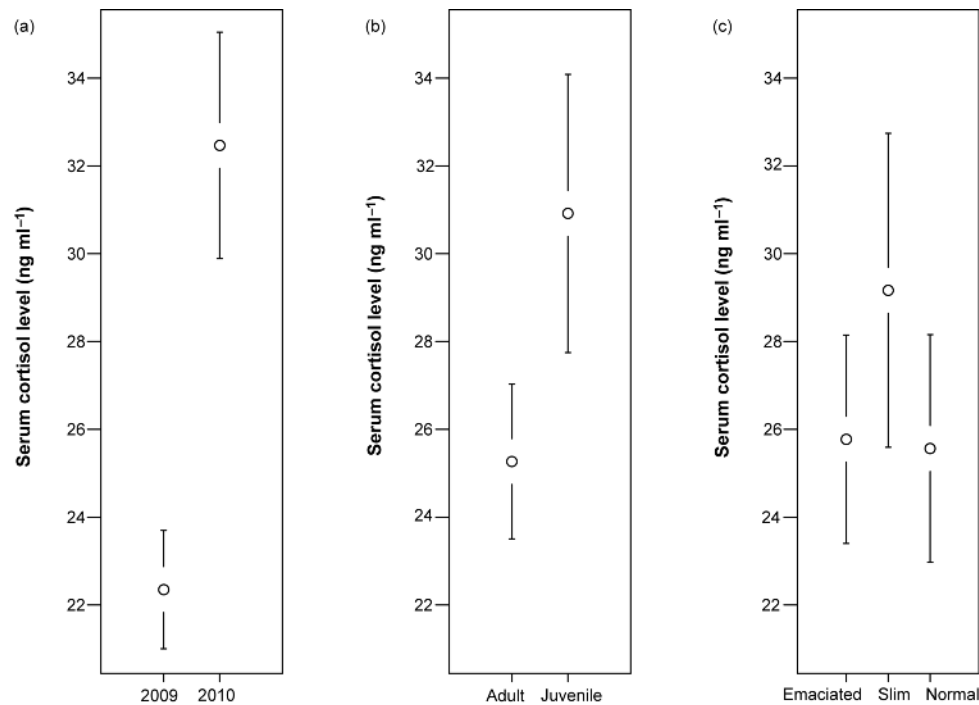
Stress behavioural responses

Two models of stress behavioural responses received almost 83% of the model-selection weight (Appendix A; [see supplementary material to papers published in *Animal Welfare* on the UFAW website: <http://www.ufaw.org.uk/t-ufaw-journal/supplementary-material>]). Stress response rates were affected by year of sampling, age category and handling time. During 2010, guanacos exhibited higher stress levels compared to 2009, as indicated by heart rate (3.7 ± 0.7 and 2.3 ± 0.4 frequency per min, respectively; Figure 1[a]) and juveniles showed more stress than adult guanacos (3.4 ± 0.9 and 2.7 ± 0.4 frequency per min, respectively; Figure 1[b]). Stress behaviour rates increased as handling time enhanced. However, after 15 min of being manipulated, stress behaviour rates began to decline (Figure 1[c]).

Stress hormone response

Serum cortisol level in shorn guanacos was affected by the year of management, age category and body condition of the guanacos (Figure 2). Time of day did not have an effect on cortisol levels. Two models of serum cortisol level received almost 82% of the model-selection weight (Appendix A; [<http://www.ufaw.org.uk/t-ufaw-journal/supplementary-material>]).

Figure 2



Showing (a) serum cortisol level (ng ml⁻¹) and the years of sampling (2009 and 2010), (b) serum cortisol level (ng ml⁻¹) and age category of guanacos (adult and juvenile) and (c) serum cortisol level (ng ml⁻¹) and the body condition of guanacos (emaciated, slim and normal). Data are expressed as means (\pm SEM).

Serum cortisol levels were higher during 2010's handling activities (34.9 [\pm 1.9] ng ml⁻¹) compared to 2009 (21.8 [\pm 1.2] ng ml⁻¹; Figure 2[a]), and were also higher in juveniles (32.1 [\pm 1.8] ng ml⁻¹) compared to adults (28.9 [\pm 2] ng ml⁻¹; Figure 2[b]). Guanacos with the lowest (emaciated) and highest (normal) body scores presented lower levels of serum cortisol (29.9 and 27.6 ng ml⁻¹, respectively) than guanacos with mean body condition scores (slim; 31.7 ng ml⁻¹).

Correlations between serum cortisol levels and heart frequency, and serum cortisol levels and respiratory rates did not show significant correlations ($R = 0.32$; P -value = 0.08 and $R = 0.13$; P -value = 0.48, respectively).

Neutrophil/lymphocyte ratio

The best-supported model of neutrophil/lymphocyte (N/L) ratios received 68% of the model-selection weight, with an AIC score 1.8 units better than the second best supported model (27% of weight; Appendix A [<http://www.ufaw.org.uk/t-ufaw-journal/supplementary-material>]). The N/L ratio was heavily influenced by year of sampling and body condition of individuals (Figure 3).

Higher N/L ratios were seen in 2010 than 2009 (7.8 [\pm 2.4] and 5.7 [\pm 1.6], respectively; Figure 3[a]). Guanacos with low (emaciated) and medium (slim) body condition scores showed higher N/L ratios (8.7 [\pm 5.9] and 8.9 [\pm 3], respectively) compared to individuals with a better body condition (5.4 [\pm 0.9]; Figure 3[b]). Further, the N/L ratio showed a positive correlation with serum cortisol levels ($R = 0.80$; $P < 0.001$; Figure 4).

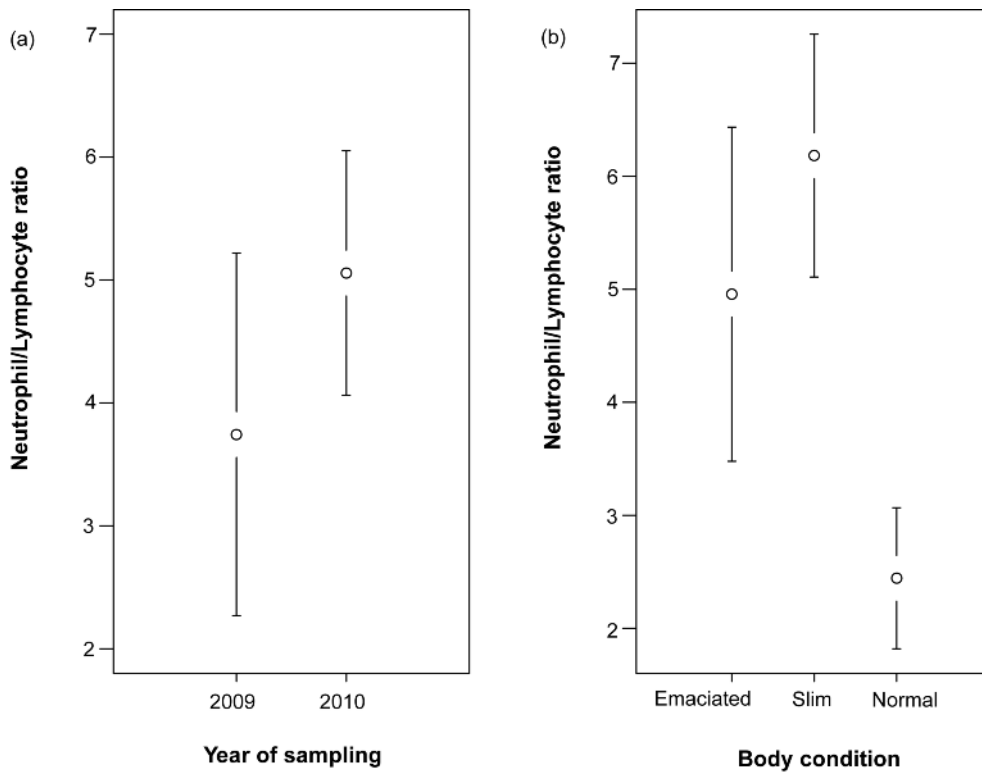
Discussion

This paper describes the attempt to quantify the impact of handling operations on guanacos through the simultaneous measurement of behavioural responses, serum cortisol concentration, and neutrophil/lymphocyte ratios. The results show that handling activities promote behavioural and physiological changes indicative of stress. It is clear that handling time is an important stressor during shearing activities. Guanacos that were under manipulation for between 12 and 15 min displayed high rates of stress behaviours. Handlers sought to shear and take blood samples as quickly as possible, whilst also ensuring every precaution was taken not to injure the animal. There is, therefore, a trade-off between optimising handling time and minimising damage to the individual.

Another factor to consider is the age of the individuals. Juvenile guanacos showed higher stress behaviour rates than adults and higher levels of serum cortisol, suggesting that special care may be needed when manipulating young individuals.

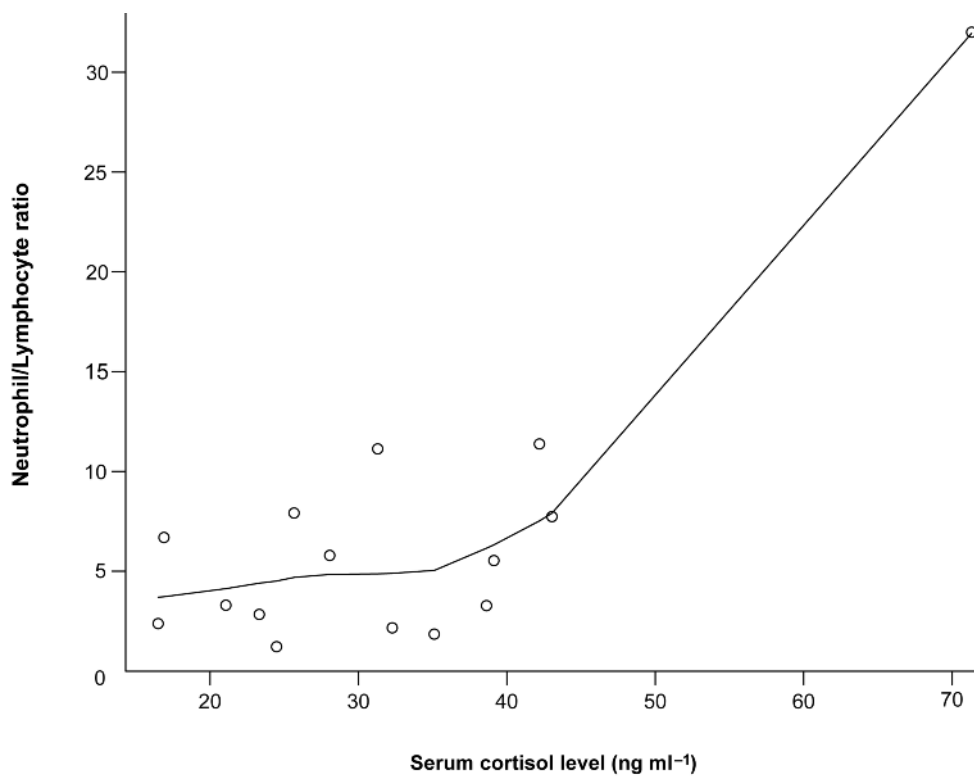
Body condition score was a novel variable that we consider important to record at the time of shearing. Guanacos with poor body scores showed higher levels of serum cortisol and higher N/L ratios. These results were more pronounced when we observed the levels of N/L ratio in guanacos with emaciated and slim scores, compared with the low levels by guanacos with normal scores. The high N/L ratios in guanacos with lower body condition scores could be because animals in poor condition are less capable of avoiding injury. Therefore, the influx of neutrophils into the blood from bone marrow

Figure 3



Showing (a) neutrophil/lymphocyte ratios (N/L) and the years of sampling (2009 and 2010) and (b) neutrophil/lymphocyte ratios (N/L) and the body condition of guanacos (emaciated, slim and normal). Data are expressed as means (± SEM).

Figure 4



The relationship between neutrophil/lymphocyte ratios (N/L) and the serum cortisol level (ng ml⁻¹).

and the adherence of circulating lymphocytes to the endothelial cells and transmigration to lymph nodes, spleen, bone marrow and skin, ensure that the different types of cells will be present in the stress response (Dhabhar 2002; Davis *et al* 2008; Johnstone *et al* 2012). Although cortisol levels in guanacos do not seem to be associated with body condition, as reported for other vertebrate species (Müller *et al* 2011), our results show a positive correlation between N/L and serum cortisol levels, in accordance with results of Bonacic *et al* (2003) in vicuñas (*Vicugna vicugna*). Therefore, we consider that the body condition score together with age should be considered important *in situ* markers of the potential stress response to handling activities.

Our results show that shearing activities induced behavioural and physiological changes. It is important to minimise handling time to reduce the stress as indicated by the high frequency of stress behaviours. Therefore, human contact must be kept to a minimum. Unease is a very difficult parameter to measure in animals and particularly in South American camelids. If we assume that wild camelids undergoing close contact with humans are always anxious to be released, then handling protocols should be as quick as possible to avoid stressing the animals (Bonacic & Gimpel 2006).

Studies by Bonacic and Macdonald (2003), Bonacic *et al* (2006) and Arzamendia *et al* (2010) on vicuñas, and by Carmanchahi *et al* (2011) and Taraborelli *et al* (2011) on guanacos, reported high values of serum cortisol as a consequence of chasing and handling. Our results are similar but, additionally, we obtained high N/L ratio values in guanacos with poor body condition, which strengthen and complement the results of serum cortisol. Glucocorticoid production enables rapid physiological and behavioural adjustments to unpredictable changes in the environment, allowing animals to react more efficiently to adverse circumstances, so minimising their exposure to stress and increasing their chances of survival and a progressive return to 'normal' life (Hofer & East 1998; Arlettaz *et al* 2007).

The high serum cortisol and N/L ratio levels in 2010 could be a consequence of environmental factors, since both years differed considerably in terms of the weather conditions. The winter of 2010 had 20% more days with temperatures below 0°C compared to 2009 (Payunia weather station data). Various authors have shown environmental stress due to extreme temperatures to affect both herbivores and the pastures upon which they feed (Romero *et al* 2009; Sakai & Larcher 2012; Arias & Velapatiño 2015). Additionally, spring and winter 2009 saw 23 mm rainfall, compared to 47 mm for the same period in 2010. Another indicator of environmental stress is the temporal primary productivity variation estimated via the reflectance vegetation index. In the management area, the enhanced vegetation index (EVI) was 0.088 (\pm 0.0006) in 2010 and 0.106 (\pm 0.0006) in 2009 (mean maximum EVI), and mean EVI was 0.069 (\pm 0.0004) in 2010 and 0.079 (\pm 0.0004) in 2009 (Schroeder unpublished data). Finally, migrant guanacos arrived later at La Payunia Reserve, October as opposed to September (P Taraborelli, personal

observation 2010). Thus, it is possible that the guanacos' basal serum cortisol and N/L ratio were higher in 2010 as a result of adverse and less productive conditions.

Changes in behaviour allow animals to either escape or counter challenges, while the autonomic and neuroendocrine response provides the animal with the resources needed to meet the demands of the altered behaviour, as well as maintaining homeostasis during the aversive situation (Moberg 1985). The major challenge in disturbance ecology is to link such behavioural and physiological reactions to long-term detrimental impacts. This is because disturbance *per se* is not necessarily detrimental for the organism (Arlettaz *et al* 2007). It is, therefore, important to monitor shorn individuals for long periods. Our research group has assessed the effects of capture and shearing on survival, reproduction, population density and structure, daily movements, ranging behaviour and spatial distribution in La Payunia's guanaco population. Capture and shearing did not reduce breeding success and early survival of young guanacos and maximum post-shearing survival rate was estimated to be 0.997 (Carmanchahi *et al* 2014). And, furthermore, we found that capture and shearing do not have a significant effect on the spatial distribution pattern of this population (Carmanchahi *et al* 2014). Also, density declined and population structure changed significantly after assembly of corrals, returning to pre-assembly levels one month later. Average daily distance moved by radio-collared guanacos during the first two days after shearing was three times further than daily distances moved during the subsequent 30 days. However, we consider there to be some important considerations to take into account to preserve the welfare of the shorn individuals, most notably the body condition, age and handling time.

Animal welfare implications

Based on our results, we make the following recommendations to reduce unease responses in guanacos: i) reduce handling time; ii) guanacos with poor body condition score (low fat deposition) should not be sheared; iii) greater care should be taken with juvenile guanacos as these are more prone to stress, in terms of both behavioural reaction and cortisol secretion; and iv) environmental conditions of the study area should be considered throughout the entire year and shearing juveniles and individuals with poor body condition score should be avoided in times of adverse conditions. Incorporating these considerations into animal welfare practices would reduce stress conditions during handling of wild guanacos. We also consider that this protocol should be followed to assess any changes in the animal's behavioural and/or physiological expression as an indirect measure of potential unease and suffering due to aversion, fear or anxiety during capture or captivity in camelids.

These simple management measures are likely to improve animal welfare, facilitating the move towards sustainable management of a wild species, emblematic of the desert biomes of South America. In addition, they could be used by those who make policy decisions regarding management practices of wildlife species and form part of animal welfare

protocols. We believe that the sustainable use of guanacos would play an important role in the future of this species and its habitat, since it would lead to persistence of long-term functional populations, and reduce processes of habitat degradation due to overgrazing of sheep.

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