Brushtail Possums in Care: Factors influencing post-release survival and the potential impacts of stress on release outcomes

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Introduction

Each year, tens of thousands of wild animals enter care through orphaning or other misadventure. These animals are looked after by dedicated teams of wildlife carer's and veterinarians, most of whom contribute their time and resources voluntarily. To date there is little knowledge of the fate of rehabilitated wildlife post-release and limited understanding of the factors that influence rehabilitation success. As such, scientists and wildlife carer's need to work together to develop an evidence-based framework for wildlife rehabilitation in Australia, which is where this project fits.

Our research team has two ongoing research interests in this field:

1. Determining the pre- and post-release factors that influence the fate of hand-reared orphaned animals after release into the wild through intensive tracking of released animals.
2. Understanding the extent to which captivity, related to care and treatment, can influence stress levels of wild animals brought into care, and the extent to which stress can compromise rehabilitation outcomes.

In this presentation I will summarise results from a recent study in which we assessed the factors correlated with short-term post-release survival for 20 hand-reared brushtail possums [1]. I will also highlight the importance of understanding the "stress" responses of orphaned, ill or injured animals that are taken into care for hand-rearing or rehabilitation, and how this could influence survival pre- or post-release. In particular, I will describe our ongoing research in this area and how interested carers could become involved.

Fate of hand-reared possums post-release

This presentation will highlight the results from a recent study which measured the survival of hand-reared brushtail possums post release [1]. Between August 2016 and February 2017, 20 juvenile brushtail possums were released in the Greater Sydney area and tracked for up to 40 days using VHF collars (Sirtrack, UG V5 VHF collar, New Zealand). This study found that the survival of brushtail possums was highly variable. After tracking for four weeks (or until collars fell off due to safety 'weak links'), 8 possums survived, 9 possums failed in the wild (i.e. killed by foxes or had to be returned into care due to injuries) and 3 possums had unknown fates (i.e. VHF signal lost). This study was specifically designed to test different approaches to possum release (i.e. hard vs soft release), neither of which had a significant effect on survival post release. However, other factors were influential, including behavioural phenotype (or personality) and the extent of humanisation (or habituation to humans). Most importantly, hand-raised possums that displayed highly humanised behaviours were less likely to be successful after release. Degree of "cautiousness" also affected post-release

success: animals that were possibly less cautious (i.e. animals that stayed with the handler longer before returning to their pen in the release test) were less likely to survive in the post-release phase. On the other hand, more exploratory individuals were more likely to survive once released. These results were compelling. The next step is to understand how fine-scaled movement (using GPS technology) affects post-release survival. We have begun such a study this year.

Stress responses to wildlife care and rehabilitation

Wild animals that enter captivity for rehabilitation or hand-rearing are exposed to a range of new stressors, such as living in abnormal social circumstances and a novel environment. When an animal is brought into care, their new habitat generally differs from the wild in terms of structure, appearance and smell, which can increase stress levels [2]. When in captivity, stress responses are commonly induced by cues such as crowding, and limited or overstimulation in housing e.g. artificial lighting, human contact and handling, unfamiliar diet, or excessive pain from injury [2]. However, there is very little information about the extent to which exposure to these stressors can elevate stress hormone concentrations and the potential flow-on effects for animal health and survival.

Stress physiology is a complex field in animal science, which is often misunderstood because of the way in which the term "stress" is used in modern life. "Stress" can most simply be described as "*a state in which homeostasis is lost*" [3; *p. 225*] or a state in which the internal balance of the body is disturbed physiologically. In many cases people think of "stress" as being a negative outcome of life events but mounting a stress response is an important component of day-to-day life.

Stress is beneficial in the short-term, and key to survival in the wild [2, 4]. The sympathetic nervous system (SNS) can be activated in response to a stressor causing a surge in adrenaline precursors through the bloodstream to allow for an immediate coping strategy to deal with the stressor [2], for example increasing blood flow and mobilising glucose stores. The stress response is the outcome of a highly organised neuroendocrine system, which involves a cascading hormonal reaction whereby the stressor triggers the production of hormones by specific regions of the brain, which in turn stimulate the adrenal glands to produce and secrete stress hormones into the bloodstream. The primary stress hormones include the glucocorticoid hormones cortisol and corticosterone [4, 5]. As such, these hormones are typically used as a proxy measure of stress levels in a variety of wild animal species.

While acute stress responses are important in the short-term, long-term exposure to stressors and chronically elevated glucocorticoid concentrations can be detrimental. A comprehensive review by Dickens et al. [2] describes the common impacts associated with chronic stress. These include impairment of the immune system, cardiovascular system, sympathetic nervous system and behavioural coping mechanisms.

High elevations of glucocorticoids can directly suppress the immune system, increasing vulnerability to disease and infection, and can also suppress reproductive function by reducing reproductive hormones. Chronic stress can harm the cardiovascular system as the SNS is constantly secreting catecholamines that can cause hypertension, myocardial infarction, higher cardiac output, and arrhythmias [6]. Such cardiovascular complications can cause further disease if left untreated. For example, irregular functioning of the cardiovascular system can disrupt other biological systems, such as the inflammatory response, that increases the risk of disease or infection.

Chronic stress also affects behavioural coping mechanisms and the SNS of an individual. When homeostasis is overloaded, behaviours such as tonic immobility, aggression, anxiety, and fear could arise and alter body weight e.g. obesity or malnutrition can eventuate [7]. Also, during chronic stress, the SNS is continually active causing the body to relentlessly mobilise energy stores [7]. Depleted energy stores have a sub-lethal effect on the individual and can cause starvation, which will ultimately affect overall survival.

From the descriptions above, it is evident that chronic stress compromises the survival of animals in care, but there is limited information on the stress-levels in wildlife in care.

How to measure stress in animals in care

Traditionally behaviour has often been assessed as an indicator of stress. However, studies of wildlife in care found that while regular handling may decrease behavioural signs of stress, physiological stress parameters remain high [8]. This has led to the hypothesis that these animals suffer from 'learned helplessness' [8]. As such, there is a need to determine whether there are more meaningful ways to assess the stress response of animals in care to determine whether stress in care and rehabilitation outcomes are correlated.

The common stress hormones, cortisol and corticosterone, are secreted into the bloodstream and subsequently metabolised and excreted in the faeces. The metabolites can be measured in the faeces and they calibrate well with stress hormones in blood approximately 1-2 days prior (roughly the time taken for the metabolites to transit through the gut). The advantage of using faecal samples to determine stress hormone concentrations is that collection is non-invasive and nonstressful compared with blood sampling, which induces stress and confounds results [5].

Research Program

Our new study aims to determine whether there is a correlation between various measures of stress (determined by measuring faecal glucocorticoid metabolite concentrations) in care and outcomes for brushtail possums throughout rehabilitation and hand-rearing. The stress hormone profile for each possum will be calculated and we will compare both the temporal profile and the average stress level (following [9] and [10]) to factors such as: time in care, survival, and type of illness/injury. Knowing key timeframes, treatment interventions or transitional stages when animals are under greater stress means that carers can be aware of when animals may be more prone to stress-related disorders, and possibly when anxiolytic medications may be beneficial. Also, by characterising the mean fluctuations in cortisol over time and establishing a range of "normal" values, we can use this method to identify chronically stressed animals that might need additional intervention.

Acknowledgements

This research was conducted in collaboration with WIRES and depended on the help of a number of extremely dedicated wildlife carers. We are extremely grateful for the involvement of these carers and their possums. The new research projects described here are funded by the NSW Industry and Community Engagement Fund (as part of the NSW Government Boosting Business Innovation Program) and the University of Sydney.

References

[1] Gillies, C. 2017. Factors influencing the success of hand-reared juvenile brushtail possums (*Trichosurus vulpecula*) released into the wild. Unpublished BSc(Honours) Thesis. School of Life and Environmental Sciences. The University of Sydney

[2] Dickens, MJ, Delehanty, DJ & Michael Romero, L 2010, 'Stress: An inevitable component of animal translocation', *Biological Conservation*, vol. 143, no. 6, pp. 1329-1341.

[3] Reeder, DM & Kramer, KM 2005, 'Stress in free-ranging mammals: Integrating physiology, ecology, and natural history', *Journal of Mammalogy*, vol. 86, no. 2, pp. 225-235.

[4] Sapolsky, RM, Romero, LM & Munck, AU 2000, 'How do glucocorticoids influence stress responses? Integrating permissive, suppressive, stimulatory, and preparative actions', *Endocrine Reviews*, vol. 21, no. 1, pp. 55-89.

[5] Sheriff, MJ, Dantzer, B, Delehanty, B, Palme, R & Boonstra, R 2011, 'Measuring stress in wildlife: Techniques for quantifying glucocorticoids', *Oecologia*, vol. 166, no. 4, pp. 869-887.

[6] Heinz, R 1999, 'Excess catecholamine syndrome: Pathophysiology and therapy', *Annals of the New York Academy of Sciences*, vol. 881, no. 1, pp. 430-444.

[7] Romero, LM, Dickens, MJ & Cyr, NE 2009, 'The reactive scope model — a new model integrating homeostasis, allostasis, and stress', *Hormones and Behavior*, vol. 55, no. 3, pp. 375-389.

[8] Hogan, LA, Johnston, SD, Lisle, AT, Keeley, T, Wong, P, Nicolson, V, Horsup, AB, Janssen, T & Phillips, CJC 2011, 'Behavioural and physiological responses of captive wombats (*Lasiorhinus latifrons*) to regular handling by humans', *Applied Animal Behaviour Science*, vol. 134, no. 3, pp. 217-228.

[9] Fanson KV *et al.* (2017) One size does not fit all: Monitoring faecal glucocorticoid metabolites in marsupials. *General and Comparative Endocrinology* **244**: 146-156.

[10] Raña, A. 2017. The influences of physiological stress, personality, and behavioural mechanisms on problem solving by common brushtail possums. Unpublished BSc(Honours) Thesis. School of Life and Environmental Sciences. The University of Sydney