



ELSEVIER

Contents lists available at ScienceDirect

MethodsX

journal homepage: www.elsevier.com/locate/mex

Method Article

Comparison of methanol and isopropanol as wash solvents for determination of hair cortisol concentration in grizzly bears and polar bears



Thomas Kroshko^a, Luciene Kapronczai^a, Marc R.L. Cattet^{b,c},
Bryan J. Macbeth^d, Gordon B. Stenhouse^e, Martyn E. Obbard^f,
David M. Janz^{g,*}

^a Toxicology Centre, University of Saskatchewan, Saskatoon, SK, S7N 5B3, Canada

^b RGL Recovery Wildlife Health & Veterinary Services, Saskatoon, SK, S7H 4A6, Canada

^c Department of Veterinary Pathology, University of Saskatchewan, Saskatoon, SK, S7N 5B4, Canada

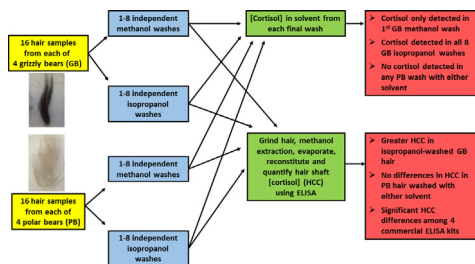
^d Department of Ecosystem and Public Health, University of Calgary, Calgary, AB, T2N 4Z6, Canada

^e Foothills Research Institute, Hinton, AB, T7V 1X6, Canada

^f Wildlife Research and Monitoring Section, Ontario Ministry of Natural Resources and Forestry, Trent University, Peterborough, ON, K9J 7B8, Canada

^g Department of Veterinary Biomedical Sciences, University of Saskatchewan, 52 Campus Drive, Saskatoon, SK, S7N 5B4, Canada

GRAPHICAL ABSTRACT



ABSTRACT

Methodological differences among laboratories are recognized as significant sources of variation in quantification of hair cortisol concentration (HCC). An important step in processing hair, particularly when collected from wildlife, is the choice of solvent used to remove or “wash” external hair shaft cortisol prior to quantification of HCC. The present study systematically compared methanol and isopropanol as wash solvents for their efficiency at removing external cortisol without extracting internal hair shaft cortisol in samples

* Corresponding author.

E-mail address: david.janz@usask.ca (D.M. Janz).

collected from free-ranging grizzly bears and polar bears. Cortisol concentrations in solvents and hair were determined in each of one to eight washes of hair with each solvent independently.

- There were no significant decreases in internal hair shaft cortisol among all eight washes for either solvent, although methanol removed detectable hair surface cortisol after one wash in grizzly bear hair whereas hair surface cortisol was detected in all eight isopropanol washes.
- There were no significant differences in polar bear HCC washed one to eight times with either solvent, but grizzly bear HCC was significantly greater in hair washed with isopropanol compared to methanol.
- There were significant differences in HCC quantified using different commercial ELISA kits commonly used for HCC determinations.

© 2017 The Author(s). Published by Elsevier B.V. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

ARTICLE INFO

Method name: Hair cortisol concentration

Keywords: Hair cortisol, HCC, Hair analysis, Long-term stress, Wash solvent, Wildlife, Bear

Article history: Received 14 September 2016; Accepted 25 January 2017; Available online 30 January 2017

Method details

Background

Quantification of hair cortisol concentration (HCC) has emerged as a promising tool to investigate long-term stress in mammals. Since the first report of hair cortisol in wildlife [1], there are now many publications that have used this approach in wildlife, veterinary, zoo, and human research [2,3]. However, methodologies used for sample collection, storage, preparation and cortisol quantification vary considerably, and recent papers have identified concerns about the reproducibility of HCC determinations [2–6]. One of these concerns is the presence of cortisol on the exterior of the hair shaft and how best to remove it [3,7].

Given the certainty that cortisol concentrations in blood and sweat are greater than that found within the hair shaft, it is clear that not removing it will cause artefactual increases in HCC [3]. This is especially critical when measuring HCC in free-ranging wildlife such as bears, because hair is commonly contaminated with blood, feces, urine and lipids [8–10]. Intact hair is typically washed with isopropanol [7,9,10] or methanol [8,11,12] three to five times in brief (3 min) intervals, depending on the extent of external contamination [8]. Multiple short washes are required to prevent the solvent from penetrating into the hair and removing internal hair shaft (medulla or hair matrix) cortisol, which also complicates the solvent choice because methanol is more effective at removing external cortisol than isopropanol, but also penetrates the hair shaft more readily [13,14]. Another methodological consideration is the technique used to quantify HCC. Most researchers have adapted enzyme-linked immunosorbent assays (ELISA) designed for saliva (e.g., [7,9]) or serum (e.g., [8]), although there has been some use of radioimmunoassay and liquid chromatography–tandem mass spectrometry [11,15,16]. The main objective of the present study was to compare isopropanol and methanol as wash solvents to remove external hair shaft cortisol contamination, without lowering internal hair shaft cortisol, in grizzly bear (*Ursus arctos*) and polar bear (*Ursus maritimus*) hair. A secondary objective was to compare the HCC results obtained from commonly-used commercial cortisol ELISA kits.

Hair sample preparation and analysis – solvent wash dynamics

Guard hair samples from four grizzly bears and four polar bears were collected during previous studies and stored in paper envelopes in the dark at room temperature [8,17]. Three of the grizzly bear hair samples were collected from bears captured using leg snares in 2007 or 2008, following an approved animal use protocol (AUP 20010016) by the University of Saskatchewan Committee on Animal Care and Supply. One of the grizzly bear hair samples was collected opportunistically in 2007 from a bear killed in self-defence. All grizzly bear samples were collected from animals

Download English Version:

<https://daneshyari.com/en/article/5518449>

Download Persian Version:

<https://daneshyari.com/article/5518449>

[Daneshyari.com](https://daneshyari.com)