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The effect of stress and stress hormones on dynamic colour-change in a sexually dichromatic Australian frog

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Running headline: Colour change in *Litoria wilcoxii*

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Abstract

Rapid colour changes in vertebrates have fascinated biologists for centuries, herein we demonstrate dynamic colour change in an anuran amphibian, the stony creek frog (*Litoria wilcoxii*), which turns from brown to bright (lemon) yellow during amplexus. We show this by comparing the colour of baseline (unpaired males) and amplecting (paired) males. We also investigate the possible role of stress and stress hormones on this colour change. Frogs were subjected to four different levels of stressors (handling, toe-clipping, saline injection and adrenocorticotropic hormone [ACTH] injection) and the colour change was measured using digital photography. A comparison of baseline colour and stress hormone (corticosterone) levels was also conducted to give further insight to this topic. From the images, the Red Blue Green (RGB) colour values were calculated, and a principal components analysis (PCA) was used to create a single colour metric (the major axis) as an index of colour in the visible spectrum. A moderate stressor (toe-clipping) led to a significant change in colour (within 10 minutes) similar to that of amplecting males. Surprisingly, neither a mild stressor (handling and saline injection)