

## ABSTRACT

The development of non-invasive methods, particularly fecal determination, has made possible the assessment of hormone concentrations in wild animal populations. However, measuring fecal metabolites needs careful validation for each species and for each sex. We investigated whether radioimmunoassays (RIAs) previously used to measure fecal testosterone (fT) in male baboons and fecal estrogens (fE) in female baboons were well suited to measure these hormones in the opposite sex. We compared fE and fT concentrations determined by RIA to those measured by liquid chromatography combined with triple quadrupole mass spectrometry (LC/MS/MS), a highly specific method. Additionally, we conducted a biological validation to assure that the measurements of fecal concentrations reflected physiological levels of the hormone of interest. Several tests produced expected results that led us to conclude that our RIAs can reliably measure fT and fE in both sexes, and that within-sex comparisons of these measures are valid: (i)  $fT_{\text{RIA}}$  were significantly correlated to  $fT_{\text{LC/MS/MS}}$  for both sexes; (ii)  $fT_{\text{RIA}}$  were higher in adult than in immature males; (iii)  $fT_{\text{RIA}}$  were higher in pregnant than non-pregnant females; (iv)  $fE_{\text{RIA}}$  were correlated with  $17\beta$ -estradiol ( $fE_2$ ) and with estrone ( $fE_1$ ) determined by LC/MS/MS in pregnant females; (v)  $fE_{\text{RIA}}$  were significantly correlated with  $fE_2$  in non-pregnant females and nearly significantly correlated in males; (vi)  $fE_{\text{RIA}}$  were higher in adult males than in immature males.  $fE_{\text{RIA}}$  were higher in females than in males, as predicted, but unexpectedly,  $fT_{\text{RIA}}$  were higher in females than in males, suggesting a difference in steroid metabolism in the two sexes; consequently, we conclude that while within-sex comparisons are valid,  $fT_{\text{RIA}}$  should not be used for intersexual comparisons. Our results should open the field to important additional studies, as to date the roles of testosterone in females and estrogens in males have been little investigated.