

Evaluating the utility of nitrogen stable isotope measurements of feces as an indicator of crop damaging sika deer

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Rapid increase in the number of sika deer (*Cervus nippon*) in the last decades aroused needs for quantitatively estimating crop herbivory by them, in order to evaluate how much damage do they cause to agriculture. Analysis of ingredients in deer feces using observation under microscopes or molecular techniques is useful to reveal whether they consumed crops. Nevertheless, these techniques in common tend to underestimate crop herbivory, because crops are usually digested more easily than wild plants and less likely to remain in the feces. Measurement of fecal nitrogen stable isotope ratio ($\delta^{15}\text{N}$) can be a complement to the molecular technique, considering that application of fertilizer leads to distinctively higher $\delta^{15}\text{N}$ values of crops than the adjacent wild plants. However, it remains unclear how much the dietary $\delta^{15}\text{N}$ affects the fecal $\delta^{15}\text{N}$, which is assumed to be a mixture comprised of the waste products from deer tissues and indigestible residues of diets. This study evaluated what extent the fecal $\delta^{15}\text{N}$ values were affected by dietary $\delta^{15}\text{N}$ values, by comparing $\delta^{15}\text{N}$ values of hair of sika deer, their diet, and the feces.

Sika deer hairs, fodders for rearing the deer, and their fresh feces were collected from 19 zoos and the nitrogen stable isotope ratio was measured using EA-IRMS. The composition of fodder was highly varied between zoos, which usually contained Leguminosae (ca. $\delta^{15}\text{N} = 0\text{‰}$) and graminoids (wide range of $\delta^{15}\text{N}$ values, although always higher than 0‰). Some zoos also fed vegetables, yeast, and wild plants, which resulted in a wide range of inter-zoo $\delta^{15}\text{N}$ variations of deer diet.

Our isotope measurement indicated that wild plants fed to the captive deer were less than 0‰ and nitrogen concentrations of wild plants were less than yeast, Leguminosae, and vegetables. Both clearly higher $\delta^{15}\text{N}$ values of crops than those of wild plants and higher nitrogen concentrations indicated that $\delta^{15}\text{N}$ is a good indicator of crop herbivory by deer. Also, the effects of individual deer profiles (sex and subspecies) slightly affected hair $\delta^{15}\text{N}$, based on data available from one particular zoo (Hirakawa zoological park, rearing 37 individuals of deer). Generalized linear mixed effect model resulted in male hair was 0.3‰ lower than female (selected model AICc = 31.6, full model AICc = 35.0). The results for hair indicated that deer individuals reared with same fodders have close $\delta^{15}\text{N}$ values, suggesting strong effects of dietary $\delta^{15}\text{N}$.

Pairwise comparisons between the fecal and hair $\delta^{15}\text{N}$ values based on intra-zoo averages revealed positive correlations between the fecal $\delta^{15}\text{N}$ and the hair $\delta^{15}\text{N}$ ($R^2 = 0.3$, $p = 0.01$) and the range of fecal $\delta^{15}\text{N}$ (2.1 to 7.4‰ ; from the lowest to the highest) was larger than those of hair (5.0 to 8.5‰). The contribution of the indigestible fraction of nitrogen in the diet, which hold shorter time-average of dietary $\delta^{15}\text{N}$ values, might affected the higher variation of fecal $\delta^{15}\text{N}$ values. Lastly, based on the measurements for zoos with feeding a single diet, or, mixed diet but coincidentally similar $\delta^{15}\text{N}$ values, we concluded that fecal $\delta^{15}\text{N}$ was intermediates of dietary $\delta^{15}\text{N}$ and deer $\delta^{15}\text{N}$. Fecal $\delta^{15}\text{N}$ values were ca. 2.0 to 3.5‰ higher than those of deer diets. Considering more than 3‰ difference of the averaged $\delta^{15}\text{N}$ values between wild plants and crops in this study, not only $\delta^{15}\text{N}$ measurement of deer tissues but also that of feces is a useful indicator of crop damaging deer. Future studies under more controlled rearing environment will reveal unknown determinants of fecal $\delta^{15}\text{N}$ values; the relative contribution of waste

products of deer tissues and directly diet-derived residuals in fecal nitrogen.

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