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DIETARY ISOTOPIC ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) VALUES FROM CAYO SANTIAGO  
MACAQUES SAMPLED BEFORE AND AFTER HURRICANE MARIA

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**Abstract**

**Objectives:** Dietary isotopic analysis of primate tissues is a powerful tool for understanding responses to environmental change and the utilization of fallback foods by primate populations. This study aims to assess dietary change in the Cayo Santiago macaques following Hurricane Maria. Because pre-Maria samples were biobanked for genetic studies, I also investigate the influence of preservative agents commonly used for archived tissues. The effects of these preservation methods on stable isotope values is not well understood, and so the first objective of this study is to **(1)** identify any biogenic dietary isotopic signal changes in skeletal muscle tissue from the Cayo Santiago rhesus macaque population that were preserved in *RNALater* relative to those that were frozen fresh. Then using previously biobanked skeletal muscle tissue from  $n=29$  macaques of the Cayo Santiago population from 2016 (prior to hurricane Maria), I ran statistical analyses to **(2)** compare differences in the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  dietary composition between group HH (2016) and KK (2018). The last objective of this study is to **(3)** assess dietary variability within group KK, by modeling isotope values as a function of age, sex, and mother-infant dependent status (MIDS).

**Methods:** Macaque muscle tissues included; *RNALater* treated muscle tissues of HH group ( $n=29$ ) sampled from 2016, and both fresh and *RNALater* preserved samples of KK group ( $n=67$ ) sampled from 2018. **(1)** This study first uses t-tests to compare  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values between the 2018 fresh and RNA-later preserved muscle replicates to understand effects on the biogenic dietary isotopic signal. **(2)** I then used linear models to analyze the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values in skeletal muscle tissue between group HH (2016) and group KK (2018), as well as food sources collected on the island in 2018 (species=10), to determine the dietary consequences of the deforestation on the island. **(3)** To identify any dietary variance in age, sex, and MIDS, I used

linear models to analyze biobanked fecal (n=65), hair (n=60), and fresh muscle (n=67) samples within group KK (post-hurricane) for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values.

**Results: (1)** These results show a significant ( $p < 0.001$ ) decrease in  $\delta^{15}\text{N}$  values (3.9‰) and a slight but significant ( $p < 0.01$ ) increase in  $\delta^{13}\text{C}$  values for RNALater preserved tissues (0.20‰). **(2)** Comparisons between the HH and KK group  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  values from muscle tissue preserved in RNALater revealed a slight (0.34‰) but significant ( $p < 0.001$ ) increase in the average  $\delta^{13}\text{C}$  value from 2016 to 2018, and positive relationship with age in HH and KK (coefficient 0.03;  $p < 0.01$ ). No significant difference in the average  $\delta^{15}\text{N}$  value was identified. **(3)** Fecal, fresh muscle, and hair showed males having slight (0.84‰) but significant ( $p < 0.05$ ) lower  $\delta^{15}\text{N}$  values than females.  $\delta^{13}\text{C}$  in hair also showed: males having lower values than females (mean difference 0.16‰;  $p = 0.037$ ), and a positive relationship with age (coefficient = 0.04;  $p = 0.002$ ). For both  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  in hair, mothers with infants also show higher values than that of females without and males (mean difference = 0.29‰,  $p = 0.01$ ; mean difference = 0.39‰;  $p = 0.05$ , respectively).

**Discussion: (1)** In this study, I determine that comparative dietary studies of  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  values in RNALater preserved muscle tissue and fresh muscle tissues will yield erroneous results. The use of a correction factor is needed to determine original biogenic  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  values. This may vary by species consuming different diets, as well as length of time the sample was retained in the preservative agent. **(2)** I interpret these results to indicate that the vegetative loss from Hurricane Maria led to a modest increase in  $\text{C}_4$  resource consumption. Based on behavioral observations and isotopic data, I interpret that the macaques consumed relatively more seaweed,  $\text{C}_4$  grasses and commercial monkey chow due to substantial loss of tree foliage. **(3)** Furthermore, I identified subtle differences between dietary protein intake in males and females; males showed lower values than females in all tissue types and in both  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ . Mothers with infants showed  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  enrichment in hair values indicating that mothers with dependent infants modestly exploit more  $\text{C}_4$  vegetation than females without. These results have broad implications for post-weaning behaviors of the rhesus macaque females on the island. Lastly, results show a slight increase in  $\text{C}_4$  consumption with age, which I suggest is representative of subtle changes in foraged vegetation as the macaques age, from more  $\text{C}_3$  arboreal vegetation to more  $\text{C}_4$  grasses.