A TARGETED METABOLOMICS ASSAY TO MEASURE PURINES IN THE DIET OF MANAGED AND FREE-RANGING COMMON BOTTLENOSE DOLPHINS (*TURSIOPS TRUNCATES*)

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Abstract

Ammonium urate nephrolithiasis occurs in common bottlenose dolphins (*Tursiops truncatus*) managed under human care but not in free-ranging dolphins (Smith et al., 2013). In mammals, purine-rich diets, such as the piscivorous diet of dolphins, can predispose to urate urolith formation (Osborne et al., 1995). The total purine content of food is measured commercially by summing the concentrations of four purine metabolites: adenine, guanine, hypoxanthine, and xanthine. Nevertheless, several other dietary purine metabolites can be converted into uric acid, and individual metabolite concentrations can vary with fish species, cold storage methods, and storage time (Clifford et al., 1976). A method using high-performance liquid chromatography with tandem mass spectrometry was developed to quantify the purine metabolites in frozen stored whole fish species [striped mullet (*Mugil cephalus*) and ladyfish (*Elops saurus*)] fed to collection dolphins and fresh frozen species [Atlantic herring (*Clupea harengus*) and west coast Loligo squid (*Loligo opalescens*)] consumed by free-ranging dolphins. The method accurately quantifies three additional metabolites: adenosine monophosphate, inosine monophosphate, inosine, and uric acid. The mean total purine content (n=5) measured using this assay, including these additional purine metabolites, was approximately twice that which would be quantified by the commercial assay. Additionally, mean inosine monophosphate concentrations were much greater on an energy basis in ladyfish when compared with the other species (656 vs. 2-18 μmol/Mcal ME; $p < 0.05$). Thus, this assay provides a more accurate determination of the total purine content of whole fish and reveals the extent to which individual purine metabolite concentrations vary among fish species.

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