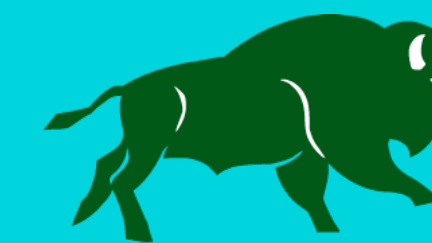




# Taxa- and species-specificity in calibration coefficients and their implications for quantitative fatty acid signature analysis (QFASA) in cetaceans

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Inspiring Conservation Leadership



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## Introduction

- Detailed, long-term diet estimates are key to assessing cetacean vulnerability to disturbances that impact prey (e.g., harmful algal blooms, fisheries)
- Quantitative fatty acid signature analysis (QFASA) provides a months-long average of prey species proportions<sup>1</sup>
- To estimate diet, QFASA compares a predator's fatty acid signature (FAS) to model mixes of prey FASs ("diets")
- Calibration coefficients (CCs) correct for changes in FA structure when they are deposited from prey into blubber
- CCs impede QFASA's broader use, however:
  - Calculating CCs requires blubber from professional care animals with known diets; numbers are limited<sup>2</sup>
  - Taxa- and species-specific differences in tissue structure, metabolism, and diet may cause CC variation, diminishing CC interchangeability<sup>2-4</sup>
- Just one set of cetacean-specific CCs exist, calculated in killer whales<sup>5</sup>; all other cetacean QFASA studies have applied mink-derived CCs with only partial success<sup>6,7</sup>

## Methods

- Derived the first set of bottlenose dolphin (*Tursiops truncatus*) CCs
  - Prey fish (n = 26, 4 species) and blubber from professional care dolphins (n = 2, D1 and D2) with known diets, from the US Navy Marine Mammal Program
  - To quantify FASs<sup>1</sup>:

Tissue processing → Lipid extraction via Soxtec apparatus → FAME derivatization and GC-FID

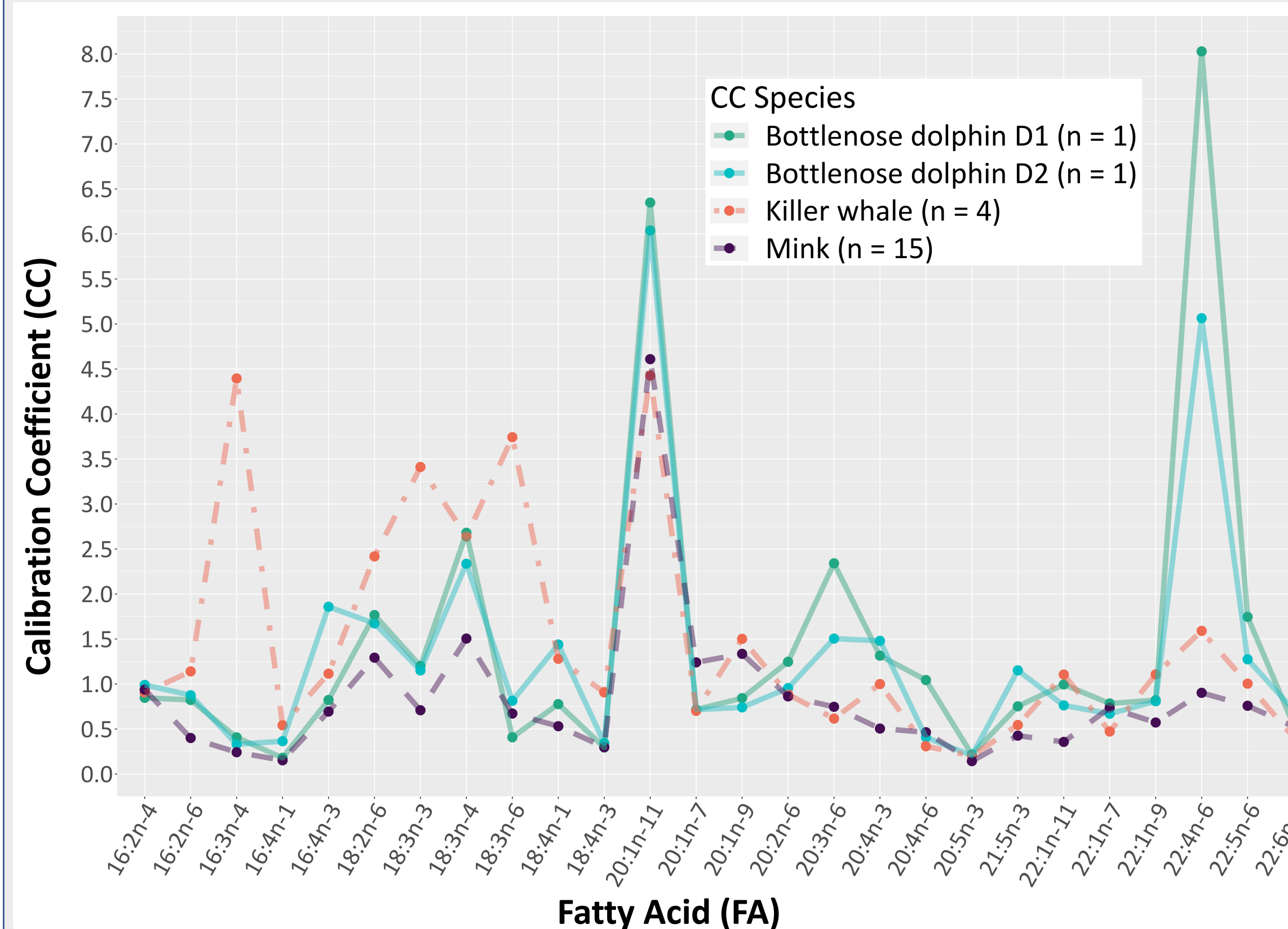


- Used dolphin and prey FASs to calculate CCs for each FA (k) and each blubber layer (inner, middle, outer) (Fig 1)

$$CC_k = \frac{\text{measured proportion of } FA_k \text{ in predator}}{\text{expected proportion of } FA_k \text{ based on } FA_k \text{ values in the prey and the known proportions of prey in predator diet}}$$

- Used R package qfasar<sup>8</sup>, dietary FAs > 0.1% of the total FASs (k = 23 – 51, based on CC set), and the Aitchison distance to estimate dolphin diet, applying CCs from the other dolphin, killer whales (n = 4)<sup>5</sup>, and herring-fed mink (n = 15)<sup>4</sup>
- Assessed diet estimate accuracy and CC interchangeability by calculating percent error of QFASA diet estimates: absolute value (prey's % in dolphin's known diet — % in estimated diet) / % in known diet \* 100

Bottlenose dolphin calibration coefficients (CCs) generate more accurate QFASA diet estimates than killer whale or mink CCs, supporting the importance of species-specific values



**Figure 1.** Calibration coefficients (CCs; ratio of the measured proportion of each FA in a predator to the expected proportion based on its diet) used in the QFASA diet estimates for bottlenose dolphins. Dolphin CCs were calculated from two professional care dolphins with different known diets (D1 ate capelin and herring, while D2 ate capelin, herring, finger mullet, and squid). Killer whale and mink CCs were reported previously<sup>4,5</sup>. Only CCs from dolphin and killer whale inner blubber layers are shown, as those provided the most accurate diet estimates (see **Results and Implications**). Only FAs used to estimate dolphin diet are shown (see **Methods**).

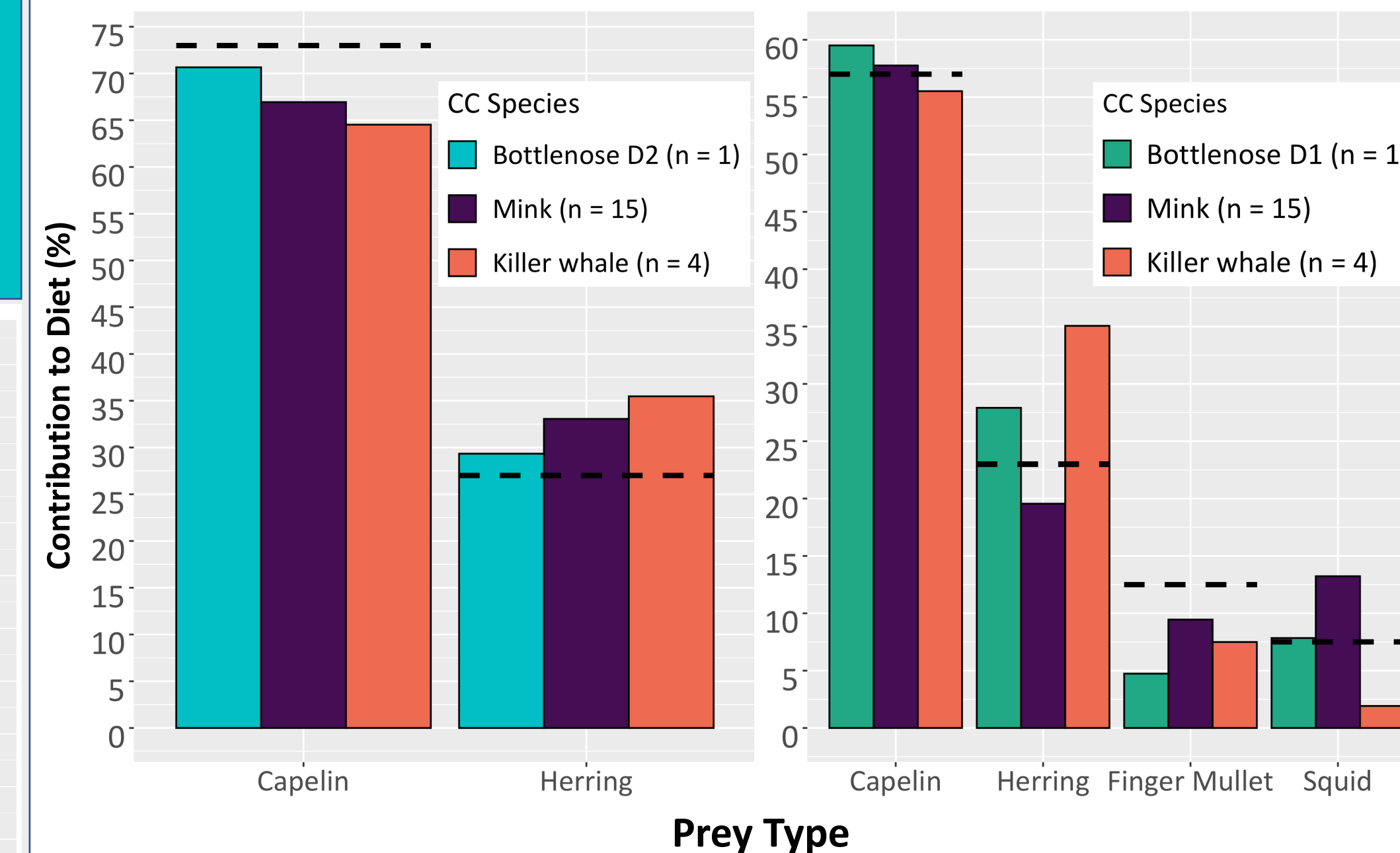
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**References:** (1) Budge et al., 2006. (2) Rosen and Tollit, 2012. (3) Meynier et al., 2010. (4) Thiemann et al., 2008. (5) Remili et al., 2022. (6) Choy et al., 2019. (7) Xie et al., 2022. (8) Bromaghin, 2017.

All samples were collected during routine animal care and under the authority codified in U.S. Code, Title 10, Section 7524.

## Results and Implications

- Layer-specific dolphin CCs yielded the most accurate diet estimates (e.g., inner blubber CCs used to estimate diet based on inner blubber FASs) (Fig 2)
  - D2-derived CCs estimated D1 diet with 5.9% total error (TE); D1-derived CCs estimated D2 diet with 23.1% TE
- Mink CCs yielded lower-error estimates (15.3% and 29.2% TE for D1 and D2, respectively) than inner blubber layer killer whale (KW) CCs (21.5% and 43.4%)



**Figure 2.** QFASA diet estimates for bottlenose dolphins D1 (left) and D2 (right) using dolphin, mink, and KW CCs. Dashed lines indicate known diet: 73% capelin and 27% herring (D1) and 57% capelin, 23% herring, 12.5% finger mullet, and 7.5% squid (D2).

- Findings support previous QFASA studies: mink-derived CCs estimated higher-percent prey with relative accuracy but under- or over-estimated lower-percent prey<sup>5-7</sup>
- Diet complexity may influence CC interchangeability more than taxa-specific metabolic or structural differences<sup>2</sup>
  - Mink and KW CCs estimated D1 diet with relatively similar TEs; for D2 (with more complex diet) all TEs were larger, and mink and KW TEs differed
  - While mink ate only herring, KWs ate capelin, herring, mackerel, and salmon
  - ↑ diet complexity generates CCs that represent more prey and FASs, yielding more accurate diet estimates
  - OR ↑ diet complexity increases the possibility of prey FAS overlap, causing over- or under-estimation of some prey
- If predator-specific CCs are not available, assessing prey species FAS overlap and comparing QFASA diet estimates from multiple sets of CCs is encouraged
- Bottlenose dolphin-specific CCs make QFASA available for detailed, long-term diet estimates in cetaceans worldwide