



Original Research Article

Survey of vitamin D and 25-hydroxyvitamin D in traditional native Alaskan meats, fish, and oils

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ABSTRACT

Greater consumption of traditional foods has been associated with improved vitamin D status in Arctic and sub-Arctic populations, including Alaskan Native Americans. However, lack of vitamin D food composition data impairs epidemiological studies on health outcomes, and development of specific dietary recommendations. Vitamin D, including 25(OH)D₃ was quantified in samples of native fish, fish eggs, meats (caribou, goose, whale, seal) and traditionally prepared whale and seal oil collected from Alaskan tribes. Vitamin D₃, 25(OH)D₃, and vitamin D₂ were assayed in alkaline-saponified samples by UPLC-MS, after derivatization with 4-phenyl-1,2,4-triazole-3,5-dione, with in-house control materials and/or NIST SRM[®] 1546a Meat Homogenate included in each analytical batch. All but the land animals and bearded seal meat contained $\geq 2 \mu\text{g}$ vitamin D₃/100 g, with $> 10 \mu\text{g}/100 \text{ g}$ in steelhead trout; dried sheefish, whitefish, smelt; smoked/dried salmon; fermented sheefish eggs; whale and seal oils. Large between-sample differences in bearded seal oil suggested possible effects of season and/or maturity on vitamin D content. 25(OH)D₃ was $> 0.3 \mu\text{g}/100 \text{ g}$ in many foods, notably smoked salmon, beluga whale skin/fat and oil and spotted seal (but not other seal) oil, with the highest levels in dried beluga whale meat, skin/fat, and oil (up to 1.2). Vitamin D₂ was $< 0.2 \mu\text{g}/100 \text{ g}$ in all foods.

1. Introduction

Vitamin D is an essential nutrient for bone health and has been increasingly recognized as a factor in immune system modulation, musculoskeletal health, cancer, and other aspects of health and disease (Pludowski et al., 2013). Vitamin D₃ is readily formed in the epidermis with sun (ultraviolet light) exposure, from its precursor, 7-dehydrocholesterol (Wacker and Holick, 2015). However, many individuals and populations have suboptimal vitamin D levels, as measured by serum 25-hydroxyvitamin D₃ [25(OH)D₃] concentration (Holick, 2005; Ross et al., 2011), due to insufficient sun exposure or dietary intake. Populations at higher latitudes are particularly susceptible to deficiency (Barake et al., 2010; Chen et al., 2007; Webb et al., 1988), and in these groups, dietary sources of vitamin D become more significant. Studies have found higher serum 25(OH)D₃ in individuals than what would be predicted by ultraviolet light (UV) exposure and dietary intake determined by food intake surveys and food composition data, and part of the explanation might be underestimation of dietary intake due to a lack of reliable and complete data for vitamin D in foods, particularly for 25(OH)D₃ (Roseland et al., 2016; Taylor et al., 2016). Since 25(OH)D₃ has a higher biological activity than D₃ and is not routinely

measured in foods or included in food composition databases, it may be an overlooked component of vitamin D intake (Cashman et al., 2012; Norman, 2008; Ovesen et al., 2003).

Suboptimal vitamin D status has been documented in a variety of Arctic and sub-Arctic populations, including Alaskan and Canadian Northwest Native Americans (Frost and Hill, 2008; Lebrun et al., 1993; Sharma et al., 2011; Singleton et al., 2015; Weiler et al., 2007). There has been a trend toward westernization of the diet of Native populations (Ballew et al., 2006; Johnson et al., 2009; Murphy et al., 1995; Nobmann et al., 2005), although greater consumption of traditional foods in North American Arctic and sub-Arctic populations has been associated with better vitamin D status (Andersen et al., 2013; Bersamin et al., 2007; Luick et al., 2014; Mansuri et al., 2016; Mohatt et al., 2007; Ryman et al., 2015; Sheehy et al., 2014). “Traditional” foods are defined as those “composed of items from the local, natural environment that are culturally acceptable” (Kuhnlein and Receveur, 1996). However, a lack of food composition data for traditional Native foods impairs both epidemiological studies on vitamin D status and health and the development of specific dietary recommendations for increasing vitamin D intake in these populations (Johnson et al., 2009).

Research on the role of vitamin D intake and health requires reliable

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food composition data, including sampling that is relevant to the population being studied (Ahuja et al., 2013). Food sampling for the USDA National Food and Nutrient Analysis Program (NFNAP) (Haytowitz et al., 2008; Haytowitz and Pehrsson, 2018) has provided analytical food composition data for the USDA National Nutrient Database for Standard Reference (SR) (USDA, 2016), including foods for the What We Eat In America (WWEA) National Health and Nutrition Examination Survey (NHANES) (U.S. Department of Agriculture, Food Surveys Research Group, 2017). Sampling is based on statistical models that account for market share and sources of variability in the food supply, to obtain representative population-wide average food composition data (Perry et al., 2002). However, these models cannot be translated to Native populations, since traditionally harvested and prepared foods must be obtained from remote locations, directly from the tribes who may not have excess to spare. Thus, efforts to develop robust vitamin D data for traditional foods need to be focused on those that are significant sources of the nutrient. There is a lack of information on vitamin D in these foods to serve as a basis to narrow the number of foods for further investigation.

In general, foods naturally rich in vitamin D include fish and fish oils, and some meats and organ tissues. However, the level can vary widely among specific foods in these groups, and the content can vary based on location of sampling (Roseland et al., 2018). While some fish are high in vitamin D (e.g., salmon, trout), many others contain negligible vitamin D (Mattila et al., 1995; Roseland et al., 2018; Schmid and Walther, 2013). Thus, it is reasonable to expect that some Native Alaskan fish and meats might be high in vitamin D, including 25(OH)D₃. Kuhlein et al. (2006) reported on the vitamin D₃ content of several traditional native North American meats and fish sampled from Canadian tribes, but did not include 25(OH)D₃. Foods which are potentially good sources of 25(OH)D₃ include some fish and organ meats (Roseland et al., 2018), pork fat (Clausen et al., 2003), and eggs from vitamin D supplemented hens (Exler et al., 2010). Furthermore, the vitamin D content of a particular species of fish or mammal can be influenced by diet or environment (Liu et al., 2013; Mattila et al., 1997; Rao and Raghuramulu, 1996; Roseland et al., 2018). Processing prior to consumption also can influence vitamin D; variation has been observed based on removal of the skin from meats and fish is removed (Pierens and Fraser, 2015), sun drying (Barnkob et al., 2016), and differences in cooking methods (Jakobsen and Knuthsen, 2014; Mattila et al., 1999; Roseland et al., 2018). Specific environmental conditions and methods of preserving (e.g. smoking, drying), storing, and cooking foods likely vary among different indigenous populations, which have localized food supplies and often unique techniques for handling and preparation and that deserve consideration when evaluating dietary intake in specific populations.

An effort to obtain food composition data specifically for Alaskan and American Indian Native (AIAN) traditional foods was initiated by the United States Department of Agriculture (USDA) Agricultural Research Service (ARS) Nutrient Data Laboratory (NDL) (Beltsville, MD), with funding from the National Institutes of Health Office of Research on Minority Health (ORMH) and the Indian Health Service (IHS) (Rockville, MD). The main focus was to obtain food composition data for key nutrients, for inclusion in the American Indian/Alaskan Native database in SR, to support efforts on health studies of Native Americans (Amy and Pehrsson, 2003; Johnson et al., 2009; Pehrsson et al., 2005a, b; Perry et al., 2002). Meanwhile, efforts were being made in general to add data on vitamin D to SR (Holden et al., 2008), including studies on analytical methods and quality control (Byrdwell et al., 2008; Byrdwell, 2009; Phillips et al., 2008), vitamin D content of fish and shellfish (Byrdwell, 2009; Byrdwell et al., 2013), milk (Patterson et al., 2010), eggs (Exler et al., 2013), and mushrooms (Phillips et al., 2011). A key part of this work was validating methods for vitamin D and 25(OH)D. Until recently, methods for vitamin D were generally not standardized and often gave disparate results between laboratories using similar methods (Byrdwell et al., 2008; Phillips et al.,

2008). More recently, methodology for measurement of vitamin D and 25(OH)D by LC–MS has been validated (Byrdwell, 2009; Huang et al., 2009). The USDA Nutrient Data Laboratory (Beltsville, MD) has demonstrated that it is possible to obtain consistent results for vitamin D₃ and 25(OH)D₃ among laboratories (Roseland et al., 2016). Matrix-specific control materials and certified reference materials are, however, critical to include to account for the many factors that impact accuracy and precision of vitamin D quantification in different types of foods (Roseland et al., 2018).

The objective of this work was to measure vitamin D₃ and 25(OH)D₃ in samples of traditional meats and fish obtained from Native Alaskan tribes, to screen for foods that could be a rich dietary source of vitamin D, as a basis for focusing more detailed food composition studies as well as to add to the dearth of published data for traditional foods, using validated analytical methodology and including traceable quality control materials.

2. Materials and methods

2.1. Sample collection

The foods sampled included raw, preserved (dried, frozen, fermented, canned, smoked) and prepared (boiled or baked) fish, land and marine mammals, and marine mammal oils. Samples were collected from Alaskan Native tribes, according to the sampling framework described previously (Perry et al., 2002), from the locations shown in Fig. 1. The foods sampled were selected based on published research, informal surveys and interviews with tribal council, elders, and members (e.g., at clinical setting), collaboration (e.g., food frequency questionnaires from other government or academic studies and focus groups. Handling of Alaskan native endangered species samples was authorized under permit 782–1694 from the National Marine Mammal Laboratory of the Alaska Fisheries Science Center of the National Oceanic and Atmospheric Agency (Seattle, WA). All food collection was approved by the councils of each tribe, and where presentations and supportive documents were required they were submitted to the tribe.

Samples were obtained within and among villages and tribes, from festivals or markets, homes, senior centers (food parties) and schools, as summarized in Table 1. Multiple samples of each food were obtained when possible, although the number of samples was necessarily limited by the availability of extra food for this study. Prepared foods (smoked, boiled, dried, etc.) had been processed by traditional methods, but details of these procedures were not always available. The food samples were, as much as possible, weighed on site (as the amount collected was often considered a common portion), frozen, and then packed on dry ice or blue ice packs and shipped via overnight express to the Food Analysis Laboratory Control Center (FALCC) in the Biochemistry Department at Virginia Tech (Blacksburg, VA). Upon receipt at the FALCC, samples were inspected to confirm integrity and then stored frozen at $< -15 \pm 5^\circ\text{C}$ until prepared for analysis.

2.2. Sample preparation

Samples were thawed in the refrigerator ($4 \pm 2^\circ\text{C}$) prior to homogenization (meats and fish overnight, fish eggs 4–6 h, and oils until completely liquefied). Canned fish samples were homogenized using a Robot Coupe® industrial stainless steel food processor (RSI 6 V Blixer, Robot Coupe USA, Jackson, MS). Other fish, meat, and fish egg samples were homogenized in the same manner, except with the addition of liquid nitrogen to aid the homogenization process. Oils were thawed at $4 \pm 2^\circ\text{C}$ until liquefied, then stirred in a stainless steel bowl with a stainless steel spoon to homogenize. After homogenization, subsamples were dispensed in 15–30 g aliquots among 30-mL or 60-mL glass jars with Teflon™-lined lids (Qorpak®, Bridgeville, PA; #GLC-07098 and GLC-08640, respectively). The jars were sealed under nitrogen, surrounded with aluminum foil, and stored at $< -55^\circ\text{C}$ until

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