An Evaluation of the Vitamin D₃ Content in Fish: Is the Vitamin D Content Adequate to Satisfy the Dietary Requirement for Vitamin D?


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Abstract

It has been suggested that the major source of vitamin D should come from dietary sources and not sun exposure. However, the major fortified dietary source of vitamin D is milk which often doesn’t contain at least 80% of what is stated on the label. Fish has been touted as an excellent source of vitamin D especially oily fish including salmon and mackerel. Little is known about the effect of various cooking conditions on the vitamin D content in fish. We initiated a study and evaluated the vitamin D content in several species of fish and also evaluated the effect of baking and frying on the vitamin D content. Surprisingly, farmed salmon had approximately 25% of the vitamin D content as wild salmon had. The vitamin D content in fish varied widely even within species. These data suggest that the tables that list the vitamin D content are out-of-date and need to be re-evaluated.

Keywords

vitamin D; fish; HPLC; sunlight; diet; salmon

Introduction

Vitamin D deficiency is now recognized as a world-wide problem for both children and adults (1–3). Because of concern about sun exposure and skin cancer, both children and adults either avoid sun exposure or use sun protection which puts them at high risk for vitamin D deficiency. Thus, their only source of vitamin D is from the diet or supplements. The major fortified foods in the United States that contain vitamin D include milk (100 IU/8 oz), some orange juices (100 IU vitamin D₂/8 oz), some breads, yogurts and cheeses. Irradiated mushrooms contain vitamin D₂ in varied amounts. Naturally vitamin D is found in the flesh of oily fish. Based on the dietary tables, it is suggested that salmon contains approximately 400 IU of vitamin D₃/3.5 oz.
Methods

A vitamin D assay was developed to determine the vitamin D content in foods. The details are shown in Figure 1. Typically 1 gram of fish flesh was saponified and then extracted for its lipid content. The lipid extract was chromatographed on a preparative C-18 reverse phase column. The vitamin D fraction was collected and chromatographed on a straight phase high performance liquid chromatography system (HPLC). The vitamin D fraction was collected and chromatographed on a reverse phase HPLC to quantify the vitamin D$_2$ and vitamin D$_3$ content based on the UV absorption and area under the curve (Figure 2).

For confirmation, a subset samples were recovered from the reverse phase HPLC and applied to a liquid chromatography tandem mass spectroscopy system as previously described (4) (Figure 3).

Results

The average content of vitamin D$_3$ found in wild caught salmon was 988 ± 524 (mean ± SEM) IU of vitamin D$_3$/3.5 oz which is a typical amount that is served for dinner (Table). In contrast, farmed salmon had approximately 25% of the vitamin D content present in the flesh of wild salmon. The mean concentration in the flesh of farmed salmon was 240 ± 108 IU of vitamin D$_3$. Blue fish is a very oily fish and thus thought to be an excellent source of dietary vitamin D$_3$. However, our analysis revealed in four different samples of blue fish, a mean concentration of 280 ± 68 IU of vitamin D$_3$/3.5 oz. White fish that typically has very little fat in its flesh including cod and gray sole were found to have 104 ± 24 IU and 56 ± 36 of vitamin D$_3$/3.5 oz respectively. In addition, farmed trout contained 388 ± 212 IU of vitamin D$_3$ and tuna Ahi-yt had 404 ± 440 IU of vitamin D$_3$.

Little is known about the effect of cooking on the vitamin D content in fish. When farm salmon was baked, almost all of the vitamin D content, i.e. 240 IU of vitamin D$_3$ was recovered from 3.5 oz. of salmon. The initial concentration in the uncooked salmon was 245 IU of vitamin D$_3$. However, when the salmon was fried in vegetable oil, approximately 50% (123 IU of vitamin D$_3$ was recovered.)

We also evaluated the vitamin D content in mackerel which is traditionally considered to be an excellent source of vitamin D$_3$ because of its oily content. However, in the one sample that we tested, we only observed 24 IU of vitamin D$_3$ in 3.5 oz.

Conclusion

It has been suggested by those who have little knowledge of human nutrition and little expertise in the field of vitamin D that everyone can obtain enough of their vitamin D requirement from their diet and that any unprotected sun exposure should be avoided (5). However, most experts agree that 1,000 IU of vitamin D$_3$ is required if there is no exposure to sunlight (1–3,6,7). It has been assumed that fish, especially oily fish such as salmon, mackerel and blue fish are excellent sources of vitamin D$_3$. However, our analysis of the vitamin D content in a variety of fish species that were thought to contain an adequate amount of vitamin D did not have an amount of vitamin D that is listed in food charts. There needs to be a reevaluation of the vitamin D content in foods that have been traditionally recommended as good sources of naturally occurring vitamin D.

Acknowledgments

This work was supported in part by the UV Foundation
References


Procedure for Fish Vitamin D Assay

Homogenization

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Saponification According To AOAC Method

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Extraction with Hexane, Wash with ddH₂O

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Dried Down Extract, Redissolved in MeOH

↓

Applied to C18 Cartridge

↓

1st NP-Alltech Column Silica 0.5% IPA in Hexane

↓

2nd-Zorbax Column Silica 0.5% IPA in Hexane

↓

RP-Vydac 201 TP 54, 25% MeOH in Acetonitrile

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Quantification HPLC

Figure 1.
Figure 2.
High performance liquid chromatography (HPLC) tracings and the ultraviolet absorption spectrum of the vitamin D₃ that was recovered from the final reverse phase (RP) HPLC. The top panel is the standard vitamin D₃ and the bottom panel is the HPLC chromatograms and UV absorption spectrum of a sample of salmon flesh that had been processed according to the procedure outlined in Figure 1. NP1 represents the first normal phase chromatography profile and NP2 represents the second HPLC profile of the vitamin D region recovered from the first NP1 chromatography.
Figure 3.
Top panel is the liquid chromatography tandem mass spectroscopy profile of the vitamin D$_3$ recovered from a representative salmon fish sample. The bottom panel represents the vitamin D$_2$ internal standard.
### Table

**Vitamin D Content in Fish**

<table>
<thead>
<tr>
<th>Fish Type</th>
<th>Vitamin D (IU/OZ)</th>
<th>N</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blue fish</td>
<td>280 ±68</td>
<td>4</td>
</tr>
<tr>
<td>Cod</td>
<td>104 ±24</td>
<td>3</td>
</tr>
<tr>
<td>Grey sole</td>
<td>56 ± 36</td>
<td>9</td>
</tr>
<tr>
<td>Salmon Farm</td>
<td>240 ±108</td>
<td>9</td>
</tr>
<tr>
<td>Salmon Wild</td>
<td>988 ±524</td>
<td>9</td>
</tr>
<tr>
<td>Trout Farm</td>
<td>388 ±212</td>
<td>4</td>
</tr>
<tr>
<td>Tuna Ahi-YT</td>
<td>404 ±440</td>
<td>4</td>
</tr>
</tbody>
</table>