USDA Dietary Supplement Ingredient Database
Release 3.0 (DSID-3)

Omega-3 Fatty Acid Dietary Supplement Study

Research Summary

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March 2015

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1. Introduction

The Dietary Supplement Ingredient Database (DSID) evaluates levels of ingredients in dietary supplement products. The Nutrient Data Laboratory (NDL), Beltsville Human Nutrition Research Center, Agricultural Research Service (ARS), US Department of Agriculture, developed the DSID with the Office of Dietary Supplements (ODS) of the National Institutes of Health and other federal partners. The other partners are the National Center for Health Statistics of the Centers for Disease Control and Prevention, Food and Drug Administration, National Cancer Institute of the National Institutes of Health and National Institute of Standards and Technology (NIST) of the Department of Commerce. ODS is the primary funder of the DSID, which builds on the well-recognized strengths of the NDL in developing databases that support assessments of intakes of nutrients from foods.

For more detailed information on the history of this project and the DSID pilot studies, please read the “Background Information and Pilot Study Research Summary”, available on the DSID-3 website (http://dsid.usda.nih.gov).

2. Overview of the Omega-3 Fatty Acid Study

A study of dietary supplements containing omega-3 fatty acids (also known as n-3 fatty acids and n-3 long-chain polyunsaturated fatty acids [n-3 LCPUFA]) in dietary supplements estimated the relationship between the values on dietary supplement labels and analytical values for omega-3 fatty acids. The study focused on alpha-linolenic acid (ALA; C18:3n-3), eicosapentaenoic acid (EPA; C20:5n-3) and docosahexaenoic acid (DHA; C22:6n-3).

Omega-3 fatty acid dietary supplements were defined for this study as fish oil, plant oil and fish/plant oil blends sold for the primary purpose of increasing omega-3 fatty acids intake. Some but not all of the products analyzed had a label claim of the product’s amount of individual (ALA, EPA, DHA) or total omega-3 fatty acids.

Products identified as representative of the US market were purchased for this study. Samples of multiple lots of these products were sent to qualified laboratories for the analysis of fatty acids using validated methods and appropriate quality assurance measures. For the final analytical dataset, relationships between label values and analytical values for ALA, EPA and DHA were evaluated by regression analysis. In addition, the variability in the predicted ingredient contents was estimated. These statistical results and their National Health and Nutrition Examination Survey (NHANES) application tables were released for the first time in DSID-3 (http://dsid.usda.nih.gov) in 2015.

3. Sampling Plan

NDL develops sampling plans for food and beverages to select sample units representing the US market from multiple geographic areas of the United States

(Pehrsson et al., 2000). NDL has also consulted with statisticians to set up sampling plans for purchasing samples of dietary supplement products for the DSID studies. A sampling frame was developed for purchasing dietary supplements containing omega-3 fatty acids. US Census data was used to select purchase locations (six counties representative of the US population) in Alabama, California, Colorado, Michigan, Missouri and New York.

NDL identified omega-3 fatty acid products for purchase based on information from the NHANES dietary supplement files, *Nutrition Business Journal* (NBJ, 2006-2008) reports, national and local store surveys and internet searches. Products were purchased through three channels:

- Mass market (MM) channel: grocery stores, drug stores, club stores (e.g., Costco) and other retail outlets
- Natural/specialty (NS) channel: vitamin specialty shops and natural food stores
- Direct sales (D) channel: multilevel (network) marketers (e.g., Herbalife or Melaleuca) and internet vendors

The sampling plan combined both commonly reported omega-3 fatty acid dietary supplements (top market share [TMS] products) and representative products with a lower market share (LMS).

The primary factor in determining which omega-3 fatty acid supplements to include in the TMS category was the frequency with which they were reported in NHANES. Twenty (15 fish and 5 flax oil) supplements were identified as TMS based on NHANES 2003-06 data (the most recent data available at that time), market share information across sales channels, and the observed prevalence of fish vs. plant oil products. Multiple (3-7) lots of these products were purchased by contracted shoppers in the six states listed above.

After the TMS samples were sent for laboratory analysis, a sampling plan for LMS products was developed. Representative LMS retail products were selected based on results from local and regional store surveys conducted in seven US areas in 2008-09. Compared to products reported in NHANES 2003-06, a larger number of different omega-3 fatty acid supplement brands and products were observed in the store surveys. The NS stores had higher brand/product/form diversity than MM stores. Less omega-3 fatty acid product diversity was found in the MM channel because the same brands were sold in different types of stores.

To identify representative LMS brands for purchase, a score was calculated for each brand based on the frequency with which this brand’s use was reported in store surveys, industry reports, NHANES and other national surveys. Brands were randomly assigned to a type (i.e., fish oil, flaxseed oil, fish oil/plant oil blend) if the brand sold more than 1 type. Brands with the highest scores comprised 75% of planned retail LMS products. Among the brands with lower scores, brand/type products were randomly selected to comprise the remaining 25% of planned retail LMS products. Products were purchased in the same six counties as the TMS products. NDL directed each shopper to buy 37 different LMS products from 20 different MM and NS stores in a specific sequence.

To identify LMS omega-3 fatty acid supplements sold via the D channel, a similar scoring approach was used. A list of more than 50 brand names was compiled using nonretail data from the sources previously mentioned. All of the top-scoring brands were designated for purchase and a random selection of the lower-scoring brands was also purchased.

For the entire study, multiple lots of 84 different omega-3 fatty acid products (TMS and LMS) were purchased and analyzed in 2008-2010. Supplements were purchased without consideration of the amount of label information about ingredient content. Of the 84 products, 59 contained fish oil, 17 had flaxseed oil and 8 consisted of fish oil/plant oil blends.

4. Laboratory Analysis and Quality Control

Products were sent to NDL for processing. Relevant information on each product purchased (e.g., ingredient names and levels, lot number, purchase location and date, and expiration date) was recorded in NDL's in-house database. Samples of 20 units (usually soft gels) were repackaged and placed in batches to be shipped to laboratories for the analysis of ALA, EPA, DHA, octadecatetraenoic acid, docosapentaenoic and eicosatrienoic acid. Results for octadecatetraenoic acid, docosapentaenoic and eicosatrienoic acid are not given in this report.

Laboratories were instructed to homogenize the material from all 20 units before sub-sampling for analysis (per US Pharmacopeia recommendations for analysis of dietary supplements). Two qualified analytical contract laboratories analyzed the sample sets using validated sample-handling protocols and appropriate methods to obtain analytical information about fatty acid levels. Gas chromatography methods were used by both laboratories and the data from both laboratories were combined. The method used by one of the laboratories included a combination of AOAC official method 991.39 (fatty acids in encapsulated fish oils methyl and ethyl esters) and AOAC official method 996.06 (fat total, saturated and unsaturated). The other laboratory used a method that combined AOAC official method 983.23 (fat in foods: chloroform-methanol extraction method) and AOAC official method 996.06.

Quality control (QC) materials were added to each batch of omega-3 fatty acid products for evaluation of laboratory precision and accuracy throughout the study. NIST Standard Reference Materials (SRMs), including NIST botanical oil SRM 1588c, NIST botanical oil SRM 3274 (flaxseed oil), NIST botanical oil SRM 3274 (borage oil) and NIST cod liver oil SRM 1588b were sent in each batch. Also, each batch included a set of product duplicates (2 sets of 20 soft gels of the same omega-3 fatty acid product but with different test sample identification numbers) and at least two in-house control materials that were analyzed for all ingredients in the study. A case of a single lot of an omega-3 fatty acid supplement with a matrix similar to that of the tested supplements was purchased to provide a sufficient amount for each in-house control material.

Analytical retests for ingredients in specific products were conducted to check unusually high or low results, high variability among product lots, or questionable data from batches where QC results showed a bias. For each sample analyzed, laboratory results reported in mg/g were compared to label levels and a percent difference from the label level was calculated.

5. Statistical Analysis

Initial statistical analysis focused on the analytical results for ALA, EPA and DHA in products that had a label level for at least one of these fatty acids (71 of 84 products, or 85%). The analytical data, which were reported in mg/g, were converted to mg/serving and mg/day and compared to label levels. The maximum recommended number of servings per day was used to calculate mg/day value. Percent differences from label levels were calculated using the following formula: ((analytical value – label value)/label value) × 100%.

To identify overly influential supplement observations, a jackknife technique was used to calculate Cook's distances and the restricted likelihood distances. Using a SAS mixed model procedure, weighted regression analysis was performed to estimate relationships between the label level and percent difference from label level for ALA, EPA and DHA. For each fatty acid, the label value was the independent variable and the percent difference from the label value was the dependent variable. Three models (mean, linear and quadratic) were used to fit the data for all three fatty acids. The most complex and statistically significant model was selected. Laboratory, supplement within label level and lot within supplement were modeled as random sources of variation. The accuracy of the models' predictions was assessed with validation techniques. Predicted analytical values were calculated from the predicted percent difference from the label level using the following formula: label value × (1 + predicted percent difference/100).

6. Results

Based on regression analysis, predicted mean percent differences from label levels for the three major omega-3 fatty acids (ALA, EPA and DHA) are reported per serving (Table 1) and per day (Table 2).

Table 1. Predicted Mean Percent Differences from Label Levels Per Serving Amount

Omega-3 Fatty Acid	Range of Predicted Mean % Difference	Most Common Label Level	Mean %Difference at Most Common Label Level
ALA	-14.1 to 6.3%	450 mg	3.6%
EPA	-5.4%	180 mg	-5.4%
DHA	-1.7%*	120 mg	-1.7%*

* Not statistically significantly different from label level

Table 2. Predicted Mean Percent Differences from Label Levels Per Day Amount

Omega-3 Fatty Acid	Range of Predicted Mean % Difference	Most Common Label Level	Mean % Difference at Most Common Label Level
ALA	0.25%*	1350 mg	0.25%*
EPA	-5.5%	360 mg	-5.5%
DHA	-1.7%*	240 mg	-1.7%*

* Not statistically significantly different from label level

Regression estimates for the mean predicted percent differences from label amounts varied by fatty acid. For the per-serving label amounts, the mean percent differences from the most common label level were 3.6% for ALA, -5.4% for EPA and -1.7% for DHA. For the per-day label amounts, the mean predicted percent differences from the most common label levels were 0.3% for ALA, -5.5% for EPA and -1.7% for DHA.

The per-serving results have been applied to NHANES dietary supplement data and the results have been released in DSID-3. The per-day results are the best data for comparing ingredient levels among products because many product labels (including 65% of products analyzed in this study) recommend multiple servings per day.

7. Use of Regression Equations

The regression equations for omega-3 fatty acids released in DSID-3 predict the mean percent differences from label levels for ALA, EPA and DHA in dietary supplements consumed in the United States. The predicted amounts are linked to labeled levels for three omega-3 fatty acids and are not specific to any brand or supplement. These predictions (predicted mean values) are included in DSID-3 for research purposes and are not meant to provide analytical estimates for omega-3 fatty acid levels in individual products.

The predicted standard error of the mean (SEM) is the standard error (SE) for this mean prediction. The regression equations also estimate the SE for an individual product at specific label levels. This SE is much larger than the SEM because it represents the error of prediction for an individual product vs. the error of prediction of a mean value for many products. Results (for per-serving amounts) predicted by regression for the mean percent difference from label level and the SEs were linked to NHANES products at the labeled levels reported for those omega-3 fatty acids. The predicted results from the DSID can be used instead of information from labels to more accurately assess ingredient intakes from dietary supplements in large population surveys.

Detailed information about the DSID-3 data files and instructions for appropriate use of the files are described in a separate report, “DSID-3 Data Files and Description” which is available on the DSID-3 website. Please refer to this report for further details about how to interpret and use each data file.

8. Future Research

Supplements for the DSID omega-3 fatty acid supplement study were purchased without regard to label claim information about fatty acid content. Some products had only a total omega-3 fatty acid content claim; some had only individual omega-3 fatty acid content claims; and some products claimed content levels for both. Approximately 85% of the products in the study listed a label amount for at least one of the major omega-3 fatty acids (ALA, EPA and DHA). The data in the DSID-3 is based on the statistical evaluation of the analytical results for these products.

Additional statistical evaluation of the omega-3 fatty acid data is not completed. NDL scientists plan to compare the analytical results for products with labeled levels of individual omega-3 fatty acids to results for products not labeled. This may provide additional information for a future omega-3 fatty acid ingredient calculator. Also being investigated is how best to evaluate total omega-3 fatty acid analytical results and label claims. Of the 57 fish oil products with a label claim for total omega-3 fatty acids, 1/3 of the products defined this term by summing EPA + DHA; another 1/3 defined this term by summing EPA + DHA + other omega-3 fatty acids; and the rest of the products did not define this term. An assortment of ingredient content claims and definitions were also noted for products with plant oils (label amounts for ALA and/or total omega-3 fatty acids). The DSID will publish an omega-3 fatty acid ingredient calculator when statistical evaluations of the data are complete. A possible consensus definition of 'total omega-3 fatty acids' is being investigated.

Additional DSID studies are underway to evaluate ingredient quantities in prescription prenatal multivitamin/minerals (MVMs) and to analyze botanical dietary supplements containing green tea and flavonoids. Future data releases will include results from these studies and a study that is monitoring adult MVMs over time for changes in both label and analytical levels. Estimates of vitamin D, vitamin A and chromium in adult MVMs, previously not reported, will be released in the future.

9. References

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