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Determination of vitamin D in foods:

Current knowledge and data gaps

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DETERMINATION OF VITAMIN D IN FOODS: CURRENT KNOWLEDGE AND DATA GAPS

Client Report FW 10011

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EXECUTIVE SUMMARY

As part of preliminary risk management activities, this report aims to collate information on the breadth and range of vitamin D fortified foods in New Zealand, technological issues associated with vitamin D fortification, and methodological issues associated with analyses of vitamin D in different food matrices. In addition, the quality and robustness of food composition information in the New Zealand Food Composition Database (NZFCD) was assessed for utility in contributing to a dietary intake assessment of vitamin D in the New Zealand food supply and New Zealand diet.

Based on voluntary notifications (172), New Zealand food products fortified with vitamin D included: baby foods, margarines, dairy desserts, food drinks and meal replacements, skim and reduced fat milks, protein beverages derived from legumes, yoghurts, and cereal bars. Dried milks, cheese and cheese products, analogues of cheese, and butter, which are approved for vitamin D fortification, were not identified in the Manufactured Food Database (MFD). Cereal and orange juice that may be fortified overseas, particularly in the United States (US), are not approved for vitamin D fortification in New Zealand.

Fortification can be achieved in multiple ways with varying efficiency. Most New Zealand companies stated no technological issues were experienced when fortifying with vitamin D. At least two companies have been fortifying for over 20 years, with a stable vitamin supplier and a prescribed level of fortification. In New Zealand, most companies contacted (7/9) add vitamin D in the form of vitamin D_3. Six companies undertake some analytical testing. Two companies commented on the variability of results and the lack of confidence in single results, leading one company to cease fortification of a product line with vitamin D.

Vitamin D is a complex, heat and light labile, fat soluble molecule. Analysis of the low concentrations expected in foods requires digestion of the food matrix, extraction from other fat soluble components in the food matrix, clean-up of the extract and a concentration step prior to detection. Instrumental methods for analysing vitamin D in foods include separation by high pressure liquid chromatography (HPLC) and detection by ultraviolet absorption (UV), a diode array detector (DAD) or mass spectrometry. Methods in current usage are all substantially similar and their fitness for purpose is best assessed in terms of demonstrated method performance, rather than definition of a standard method.

In an inter-laboratory collaborative study of five matrix-specific control materials, including canned salmon and vitamin D_3 fortified skim milk, processed cheese, cereal and orange juice, it was found that, with care, laboratories can obtain accurate results on vitamin D content using existing and various analytical methods. Experienced analysts can achieve relative standard deviations of 7% to 12%. Quality control data should be assessed to confirm robustness, accuracy and precision of results, with the availability of control materials of known vitamin D content being of particular importance.

Preliminary intake assessments using the 2009 New Zealand Total Diet Study (NZTDS) simulated diets, vitamin D concentrations from NZFCD (for unfortified foods) and MFD (for potentially fortified foods) were undertaken to identify key contributing foods. The NZTDS food list was refined to 89 foods for detailed enquiry regarding the origin and quality of reported vitamin D concentration data.

Of the 89 foods selected from the 2,717 foods in NZFCD, the majority of vitamin D values are British or British-derived values (38). Vitamin D concentrations of 11 foods are New Zealand analytical values, and a further 20 are derived, in part, from New Zealand data. For a further 3 foods, calculations used both New Zealand and British data. Lesser numbers of data are from the US (2), guessed or calculated from the MFD (8), or presumed to be zero (7).

Currently there is very limited New Zealand derived vitamin D concentration data in the NZFCD with single brands of bread, butter, cheese, yoghurt, milk, sausage roll and one fish species (salmon). Results for two egg and margarine samples are available. Additional validated data are available (but not included in the NZFCD) for a fortified infant formula, three fortified food drinks, a fortified fruit drink, eight fortified margarines, and five fortified milk products.

From an intake based approach, that is targeting those foods likely to have the greatest impact on vitamin D status, the priority foods to analyse to fill vitamin D concentration data gaps in New Zealand are: butter, milk, cheese, ice-cream, yoghurt, salad dressing, fish (fresh and canned), oysters, lambs' liver, meat pie, bread (wheatmeal and mixed grain), cake, biscuits (plain), pasta, pizza and brewed coffee.

1 INTRODUCTION

1.1 Background

There is a growing recognition that a significant proportion of New Zealanders have a less than optimal vitamin D status. For the general population, an optimal circulating 25-hydroxyvitamin D concentration is likely to be at least 50 nmol/L[1], but it may be higher in older groups (for example, 80 nmol/L) (Rockell *et al.*, 2008). In a nationally representative survey of New Zealanders aged 15 years and over (n=2,946), mean serum 25-hydroxyvitamin D concentrations were 47 and 52 nmol/L, in women and men, respectively (Rockell *et al.*, 2006). Two percent of males and 4% of females were vitamin D deficient (<17.5 nmol/L) and 45% of males and 52% of females had insufficient vitamin D (<50 nmol/L) (Rockell *et al.*, 2006). In the 2002 National Children's Nutrition Survey, 4% of 5-14 year olds were vitamin D deficient and 31% were vitamin D insufficient (Rockell *et al.*, 2005). In a study of newborns, 19% had 25-hydroxyvitamin D levels less than 25 nmol/L and 57% had levels less than 50 nmol/L (Camargo *et al.*, 2010). Ethnicity and season were determinants of vitamin D status of New Zealanders in each of the three New Zealand studies.

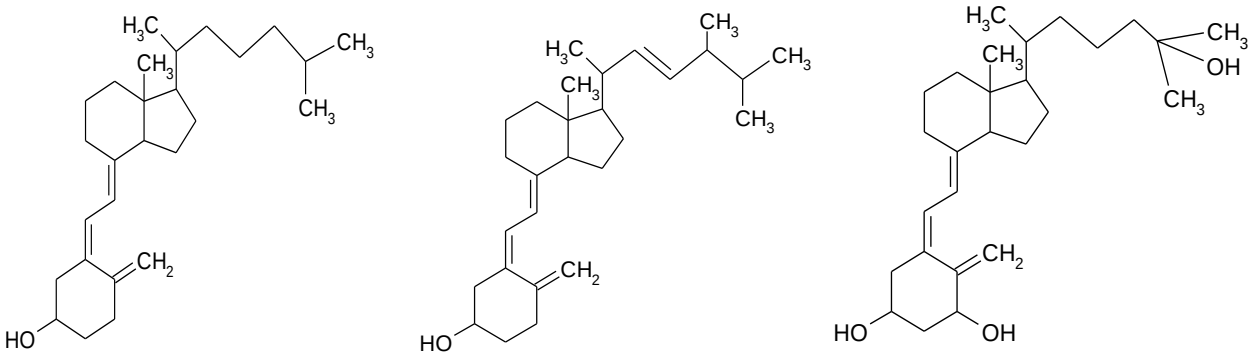
Lack of vitamin D concentration data precluded an assessment of dietary intake of vitamin D (Rockell *et al.*, 2006). The contribution of diet to serum 25-hydroxyvitamin D concentrations in New Zealand is not known.

In 2005, the New Zealand Food Safety Authority (NZFSA, now incorporated into the Ministry of Agriculture and Forestry; MAF) and the New Zealand Ministry of Health (MoH) commissioned a rapid review of vitamin D (Rockell *et al.*, 2008). The review aimed to clarify the role of vitamin D in the body, health implications of deficiency, the level required for optimum health, and current international strategies to improve vitamin D status. In 2010, NZFSA convened a Scientific Roundtable of Experts[2] to consider whether further preliminary risk management activities were required, with a specific focus on the success of food fortification as a means to improve vitamin D status.

1.2 Vitamin D Chemistry

Vitamin D exists in a number of forms, where the major physiologically relevant forms are vitamin D_2 (ergocalciferol) and vitamin D_3 (cholecalciferol) (Figure 1) (Truswell, 1998). Vitamin D_2 is produced by ultraviolet B (UVB) irradiation of the plant steroid ergosterol. Vitamin D_3 is synthesised in the skin of vertebrates through the action of UVB and 7-dehydrocholesterol. Vitamin D_2 is the less common form of vitamin D and has potentially lower bioavailability than Vitamin D_3 (Borradale and Kimlin, 2009). Vitamin D from sunlight or dietary sources is biologically inactive and is hydroxylated, via a two step process to 1,25 dihydroxyvitamin D (calcitriol), before it becomes metabolically active (Truswell, 1998).

[1] nmol/L = nanomoles per litre
[2] http://www.foodsafety.govt.nz/science/workshops-presentations/scientific-roundtable/index.htm



Vitamin D_2 (ergocalciferol)

Vitamin D_3 (cholecalciferol)

Activated vitamin D (calcitriol)

Figure 1: Chemical structures of vitamin D_2 (ergocalciferol), vitamin D_3 (cholecalciferol) and the activated form of vitamin D (calcitriol)

1.3 Sources of Vitamin D

1.3.1 Sunlight

Most vitamin D (usually between 50% and 90%) is produced via exposure of the skin to UVB through sunlight exposure (Borradale and Kimlin, 2009; Holick, 2008; Lips, 2010). In winter, however, oral intake of vitamin D may be the primary source as the UVB-related synthesis in the skin is limited. Similarly, oral intake of vitamin D is the primary source all the year round for people not exposed to sunlight, for example, due to wearing of clothing that prevents skin exposure (Jakobsen and Saxholt, 2009).

Vitamin D intake from sunlight depends on latitude, season, cloud cover, ozone level, surface reflection (for example, due to snow), altitude, outdoor practices, skin type, obesity, age and clothing (Engelsen, 2010; Lips, 2010; Rockell *et al.*, 2005; Webb *et al.*, 1988).

1.3.2 Naturally occurring food sources

Only a limited number of foods naturally contain vitamin D. Vitamin D_3 is present in animal foods such as eggs, fish species (North Sea salmon, herring and mackerel) and liver. Meat contains a small amount of vitamin D_3. Vitamin D_2 has been found in some mushrooms, where it appears to be formed from the action of UV on the provitamin, ergosterol (Teichmann *et al.*, 2007). Other plants sources may contain ergosterol that is not converted to vitamin D_2 (Lamberg-Allardt, 2006). Concentrations of vitamin D in unfortified foods reported in overseas studies are listed in Appendix 1.

1.3.3 Fortified foods

Few countries fortify foods with vitamin D. Canada and the United States (US) have previously fortified with vitamin D_2 (Lamberg-Allardt, 2006; Scientific Committee on Food,

2002). Vitamin D_3 is now the permitted form for addition to foods in Canada[3], while the USA[4] and European Union (EU)[5] permits use of either vitamin D_2 or vitamin D_3. In countries where it is permitted, foods fortified with vitamin D, such as milk products, margarines and breakfast cereals, are major contributors to vitamin D intake.

Vitamin D may be added to specific foods in New Zealand as regulated in Standard 1.3.2 Vitamins and Minerals, of the Australia New Zealand Food Standards Code (the Code), detailed in Table 1. In Australia, it is mandatory for edible oil spreads and margarines to contain no less than 55 µg/kg[6] of vitamin D. This mandatory requirement does not apply to these foods for sale in New Zealand (Standard 2.4.2, the Code) (Food Standards Australia New Zealand, 2002). Vitamin D may also be added to supplemented foods as regulated in the New Zealand Food (Supplemented Food) Standard (New Zealand Food Safety Authority, 2010). Vitamin D permissions are higher for supplemented foods compared to foods for sale under the Code.

Table 1: New Zealand foods to which vitamin D may be added*

Food	Reference quantity	Maximum claim per reference quantity (µg) (proportion RDI)	Maximum permitted quantity per reference quantity (µg)
Dried milks	200 ml	2.5 (25%)	3.0
Modified milks and skim milk	200 ml	1.0 (10%)	1.6
Cheese and cheese products	25 g	1.0 (10%)	1.6
Yoghurts	150 g	1.0 (10%)	1.6
Dairy desserts	150 g	1.0 (10%)	1.6
Butter	10 g	1.0 (10%)	1.6
Edible oil spreads and margarine	10 g	1.0 (10%)	1.6
Beverages containing no less than 3% m/m protein derived from legumes	200 ml	1.0 (10%)	1.6
Analogues of yoghurt and dairy desserts containing no less than 3.1% m/m protein derived from legumes	150 g	1.0 (10%)	1.6

[3] http://www.hc-sc.gc.ca/fn-an/nutrition/vitamin/fortification_final_doc_1-eng.php

[4] http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?fr=184.1950&SearchTerm=vitamin%20d3

[5] http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2009:314:0036:0042:EN:PDF

[6] µg/kg = micrograms per kilogram

Food	Reference quantity	Maximum claim per reference quantity (µg) (proportion RDI)	Maximum permitted quantity per reference quantity (µg)
Analogues of cheese containing no less than 15% m/m protein derived from legumes	25 g	1.0 (10%)	1.6
Beverages containing no less than 0.3% m/m protein derived from cereals	200 ml	1.0 (10%)	1.6
Formulated Beverages	600 ml	2.5 (25%)	Not specified

* (Food Standards Australia New Zealand, 2002)
m/m = mass divided by mass or weight divided by weight
RDI = Recommended Daily Intake
ml = millilitres
g = grams

A summary of New Zealand manufactured fortified foods and their vitamin D concentrations is provided in Appendix 2. This summary includes only those foods for which information has been voluntarily submitted by food manufacturers to the Manufactured Food Database (MFD) as at December 2010. The voluntary nature of submissions to the MFD means the list in Appendix 2 is unlikely to be a complete list of all foods fortified with vitamin D in New Zealand.

New Zealand food products fortified with vitamin D (n=172) included: baby food, cereal bars, dairy desserts, food drinks, meal replacements, margarines, skim and modified milks, soy milks and yoghurt. Dried milks, cheese and cheese products, analogues of cheese, and butter, that are approved for vitamin D fortification under Standard 1.3.2 – Vitamins and Minerals of the Code and the New Zealand Food (Supplemented Food) Standard, were not identified in the MFD.

The number of vitamin D fortified foods in the MFD has increased from approximately 30 in 1999 to approximately160 in 2009 (Edmonds, 2010). The food type with the greatest increase was yoghurt, with lines fortified with vitamin D increasing from one in 2004 to 53 in 2009. Fortified skim milks and reduced fat milks increased from 17 to 32 during the same period, while fortified dairy desserts increased from none to 25.

1.3.4 Dietary supplements

Vitamin D may be sourced from dietary supplements (usually in pill, capsule, tablet or other controlled dosage form). A widely available source is cod liver oil. The vitamin D present in supplements can be in both vitamin D_2 and vitamin D_3. However, vitamin D_2 is rarely used as the fortificant in supplements (Rockell *et al.*, 2008). Dietary supplements are regulated under the Dietary Supplements Regulations 1985 (Parliamentary Counsel Office, 2010) and are administered by Medsafe.

1.4 Project Aim

As part of further risk management activities, ESR was commissioned to collate information on the breadth and range of vitamin D fortified foods in New Zealand, to identify technological issues associated with vitamin D fortification, and methodological issues associated with analyses of vitamin D in different food matrices. In addition, ESR was requested to critique the quality and robustness of food composition information in the New Zealand Food Composition Database (NZFCD). This critique would inform decision making around whether vitamin D concentrations of certain foods should be updated to ensure accurate vitamin D intake assessments.

2 TECHNOLOGICAL ISSUES ASSOCIATED WITH FORTIFICATION OF FOODS WITH VITAMIN D2 AND D3

2.1 Review of Technological Issues

Vitamin D is heat and light labile. In its native form, it is fat-soluble, but may be prepared in water-soluble forms, such as emulsions. Limited information regarding technological issues associated with fortification of foods with vitamin D was found in the scientific and open literature, despite the use of diverse search strategies.

2.1.1 Form of added vitamin D

Fortification can be achieved in multiple ways with varying efficiency. For example, vitamin D_3 was added to cheese by: addition of a commercial water-soluble emulsion, homogenisation of crystalline liposoluble vitamin D in a portion of cream used for cheesemilk standardisation, or addition of water-soluble vitamin D entrapped in multilamellar liposomes (Banville *et al.*, 2000). The recovery of vitamin D_3 in cheese was significantly higher when vitamin D_3 was entrapped in liposomes than for vitamin D_3 homogenised in cream or added as a commercially water-soluble emulsion. Commercial water- or fat-dispersible forms of vitamin D_3 were found to be equally effective in producing a uniformly distributed concentration of vitamin D_3 in processed cheese (Upreti *et al.*, 2002).

Vitamin D_3 was fortified into a Cheddar cheese-like matrix, yoghurt or ice cream in either a crystalline or emulsified form (Kazmi *et al.*, 2007). The emulsified form was more stable in cheese during a three month storage period at 4ºC, with approximately 6% of the crystalline vitamin D_3 lost under these conditions, while both forms of vitamin D_3 were stable in yoghurt and ice cream during storage for the expected shelf lives of the products.

2.1.2 Stability during processing and storage

Wagner *et al.* (2008) demonstrated that vitamin D_3 was stable during cheese processing and over one year of ripening at 3-8ºC. Approximately half (45%) of vitamin D_3 added to the milk used to make cheese was bound in the whey and unavailable for human biological processes. The vitamin D was uniformly distributed throughout the cheese (Wagner *et al.*, 2008). No loss of vitamin D_3 was found during manufacture of fortified processed cheese or storage for nine months at 21-29ºC or 4-6ºC (Upreti *et al.*, 2002). No off flavours were detected in the processed cheese due to fortification with vitamin D_3. Similar results were found for vitamin D_3 fortified Cheddar cheese, with vitamin D_3 stable over nine months of storage and fortified product being as acceptable in consumer tests as unfortified product (Ganesan *et al.*, 2011).

In an evaluation of increasing the level of vitamin D_3 fortification in high temperature processed reduced fat milks (high temperature-short time, pasteurised at 73ºC for 15 seconds, or ultra heat treated, pasteurised at 138ºC for 2 seconds) and low-fat yoghurt (pasteurised at 85ºC for 30 minutes) from 2.5 to 6.2 µg/serving, no loss of vitamin D_3 during processing was found (Hanson and Metzger, 2010). Vitamin D_3 was also found to be stable over the shelf life of each product (21, 60 and 42 days, respectively). No change in sensory characteristics was observed with the increase in fortification concentration.

In contrast, Banville *et al*. (2000) found that the vitamin D_3 concentration in fortified Cheddar cheese was stable during 3-5 months of ripening, but decreased after that time. The decrease was more marked when the vitamin D_3 had been added in the form of liposomes, rather than as an emulsion or in crystalline form.

Vitamin D_3 has also been shown to be stable in a non-fat food, with no change in the concentration of vitamin D_3 in fortified orange juice after storage for 30 days at 4ºC (Tangpricha *et al.*, 2003). Vitamin D_2 and D_3 were both found to be stable in fortified orange juice (Biancuzzo *et al.*, 2010).

2.1.3 Heat stability

Heating of processed cheese fortified with vitamin D_3 for 5 minutes at 232ºC resulted in approximately 25-30% loss of vitamin (Upreti *et al.*, 2002). However, under the same conditions (232ºC for 5 minutes) or 12 minutes at 100ºC, no loss of vitamin D_3 was reported in fortified Cheddar and low-fat cheeses (Wagner *et al.*, 2008).

2.1.4 Light stability

Ultra heat treated (UHT) low-fat milk was fortified with vitamin D_3 and stored in polyethylene terephthalate (PET) bottles with varying light transmittance for 12 weeks at 23ºC (Saffert *et al.*, 2009). In clear PET containers, 66% of vitamin D_3 was lost during this period, while increasing the pigmentation levels in the PET formulation decreased losses to 35%. The concentration of vitamin D_3 in samples stored in the dark in clear PET containers remained constant.

In model systems, riboflavin has been shown to increase the rate of light-induced oxidation of vitamin D_2 (Li and Min, 1998). Inclusion of microencapsulated lycopene in skim milk was shown to reduce the riboflavin-mediated photodegradation of vitamin D_3 by 45% (Montenegro *et al.*, 2006).

2.1.5 Small quantities added to foods and sample homogeneity

Only very small amounts of vitamin D are required to meet nutrient requirements (National Health and Medical Research Council/Ministry of Health, 2006). For example, an adequate intake of vitamin D for an adult female is 0.005 mg/day compared with 7 mg/day of vitamin E (National Health and Medical Research Council/Ministry of Health, 2006). This means that low concentrations are found or are added to foods and mixing problems become greater, meaning that sample homogeneity is difficult to ensure (John MacDonald, NP Analytical laboratories, USA, personal communication, Oct 2010).

2.2 Technological Issues in New Zealand

To gain information on technical vitamin D fortification issues in New Zealand, technical staff from nine food manufacturers that submitted vitamin D fortification data to the MFD were contacted during June 2010.

Seven of the nine food manufacturers contacted add vitamin D in the form of vitamin D_3; one adds vitamin D_2. Three manufacturers use a commercially available powdered pre-mix and two source vitamin D_3 as an oil from lanolin. One manufacturer uses a vitamin D fortified fruit preparation. Information relating to the form was not available from the remaining food manufacturers.

Six food manufacturers undertake some analytical testing. Information on testing was not available from three of the manufacturers. Two food manufacturers commented on the variability of results and the lack of confidence in single results. One manufacturer reported ceasing fortification of a product line with vitamin D because of poor confidence in the measured level and the associated commercial risk associated with the claimed vitamin D level.

Most food manufacturers stated no technological issues when fortifying with vitamin D. At least two manufacturers have been fortifying for over twenty years, with a reliable vitamin supplier and a prescribed level of fortification. One manufacturer reported challenges in achieving the desired level of fortification when fortifying low fat products; and one reported difficulties in fortifying heated foods, such as fruit preparations, with vitamin D_3.

3 METHODOLOGICAL ISSUES ASSOCIATED WITH THE MEASUREMENT OF VITAMIN D2 AND D3 IN FOODS

Vitamin D is a complex, light and heat labile, fat soluble molecule. Analysis of the low concentrations found in foods requires digestion of the food matrix, extraction from other fat soluble components in the food matrix, clean-up of the extract and a concentration step prior to detection. Instrumental methods for analysing vitamin D in foods include separation by high pressure liquid chromatography (HPLC) and detection by ultraviolet absorption (UV), diode array detection (DAD) or mass spectrometry (Byrdwell *et al.*, 2008; Greenfield and Southgate, 2003).

3.1 Official Methods of Analysis

The Association of Official Analytical Chemists (AOAC) International, the organisation responsible for establishing official, legally defensible analytic methods in the United States, has validated the following chemical methods for the analysis of vitamin D in foods and feeds:

- Method 980.26: Vitamin D in multivitamin preparations
- Method 981.17: Vitamin D in fortified milk and milk powder
- Method 982.29: Vitamin D in mixed feeds, premixes, and pet foods
- Method 992.26: Vitamin D_3 in ready-to-feed milk-based infant formula
- Method 995.05: Vitamin D in infant formulas and enteral products (for tube feeding)
- Method 2002.05: Cholecalciferol (Vitamin D_3) in selected foods (fortified milk, infant formula, gruel, margarine, cooking and fish oil)

The AOAC methods listed above are similar in principle. Each method involves four key steps:

1. A digestion step to break down the food matrix (alkaline saponification);
2. An extraction step to separate the vitamin D from the food matrix;
3. A clean-up step, to separate the vitamin D from other food components; and
4. Quantitative detection by HPLC with UV.

The methods vary in the choice of internal standards, extraction solvent (heptane, hexane, pentane or petroleum ether), the method of clean-up (alumina, cyano, reversed phase or silica solid phase), the choice of analytical column (Partisil, reverse-phase or silica) and the wavelength for UV detection (254 or 265 nm)[7] (Horwitz and Latimer, 2005). These methods are time consuming, labour intensive and require attention to detail by the analyst.

The methods have been validated for a limited range of foods (mostly dairy) that cover some foods currently fortified in New Zealand, but notably not fish, yoghurt, dairy foods and cheese. In addition, the methods may not be appropriate for low-fat fortified foods such as orange juice and cereals that are fortified in some countries (Appendix 1) (Byrdwell *et al.*, 2008). The AOAC methods target vitamin D_3, and not vitamin D_2, that may be present, either naturally or as a fortificant.

[7] nm = nanometres

More recently, mass spectrometry detection has been favoured over UV or DAD for its greater sensitivity, specificity, and reduced need for sample clean-up (Phillips *et al.*, 2008; Trenerry *et al.*, 2011).

3.2 Methods Utilised Internationally

A compilation of international vitamin D concentrations in foods is provided in Appendix 1. The methods utilised in the cited studies were reviewed. Fortified and naturally occurring vitamin D are analysed in essentially the same way, with differences in the selection of the internal standard used.

Finnish vitamin D concentrations (Appendix 1) for egg, fish, meat and yoghurt were obtained by Mattila and colleagues (Lamberg-Allardt, 2006). Whilst methodology for the cited concentration values was not provided, previous papers by these authors involved:

1. Homogenisation and addition of two internal standards (Vitamin D_2, 25-hydroxyvitamin D_2);
2. Digestion by alkali saponification;
3. Liquid-liquid extraction (petroleum ether:diethyl ether 1;1);
4. Purification with semi-preparative HPLC (µ-Porasil);
5. Further purification (Vydac 201 TP 54 and Spherisorb S5NH_2 + µ-Porasil or Zorbax ODS+Vydac 201 TP 54) if necessary; and
6. Detection by reverse-phase HPLC with DAD.

Recoveries of vitamin D_2, 25-hydroxyvitamin D_2, vitamin D_3 and 25-hydroxyvitamin D_3 from spiked meat and milk samples ranged from 53-69% and 48-61% respectively (Mattila *et al.*, 1995b), while spike recoveries from fish were in the range 76-81% (Mattila *et al.*, 1995a). The limits of determination (LOD) were 0.02-0.05 µg/100 g[8] for meat and dairy foods and 0.1-0.2 µg/100 g for fish samples. Repeatability, expressed as coefficient of variation (CV) was 3% for meat and dairy (n=14, mean = 0.22 µg/100 g) and 9% for fish (n = 39, mean = 11.1 µg/100 g) for vitamin D_3 and 13% for meat and dairy (n = 14, mean = 0.7 µg/100 g) and 17% for fish (n = 3, mean = 0.18 µg/100 g) for 25-hydroxyvitamin D_3. Unspiked blank samples were included to allow for any naturally present vitamin D_2 or D_3.

Vitamin D levels in most of the important fish species consumed in Germany were analysed by Ostermeyer and Schmidt (2006) by the following methodology:

1. Homogenisation and addition of two internal standards (vitamin D_2, ergosterol);
2. Digestion by alkali saponification overnight at room temperature;
3. Extraction using a solid phase cartridge (Chem Elut, (modified diatomaceous earth));
4. Purification with semi-preparative HPLC (LiChrosorb Si 60, normal phase); and
5. Detection by reverse-phase HPLC with electrochemical detection.

Average recoveries of vitamin D_2, vitamin D_3, ergosterol and 7-dehydrocholesterol ranged from 70 to 102% using an external standard method and from 84 to 106% using an internal standard method for quantification. CVs ranged from 1-17% and 1-14% for external and internal standard methods for quantification, respectively across different fish species.

[8] µg/100 g = micrograms per 100 grams

The following methodology was developed, validated and used to assess vitamin D_3 levels in Danish pork and dairy products (Jakobsen *et al.*, 2004; Jakobsen and Saxholt, 2009):

1. Homogenisation and addition of vitamin D_2 internal standard;
2. Digestion by alkali saponification for 45 minutes in a boiling water bath;
3. Extraction with petroleum ether:diethylether;
4. Clean-up with silica solid-phase extraction columns;
5. Purification with semi-preparative HPLC (amino + silica columns, normal phase); and
6. Detection by reverse-phase HPLC with DAD.

Samples were protected from UV radiation and oxidising agents with UV absorbing film on the windows, special covered lamps, and nitrogen to replace air before saponification and evaporation. The LOD for meat samples was 0.03 µg/100 g for a 50 g test sample. CVs were 9.1% and 8.9% for vitamin D_3 and 25-hydroxyvitamin D_3, respectively. Average recoveries of vitamin D_3 and 25-hydroxyvitamin D_3 from spiked samples were 95% and 99% respectively (Jakobsen *et al.*, 2004). Accuracy of the methodology for vitamin D_3 was assessed by analysis of a certified reference material (CRM) (Milk Powder, CRM 421, Institute for Reference Materials and Measurements, Geel, Belgium). All dairy analyses were made in duplicate (Jakobsen and Saxholt, 2009).

Details of the methods used to obtain the cited vitamin D concentration values for the United States were not available.

Key aspects of the methods outlined above are summarised in Table 2. The approach of Byrdwell *et al.* (2008) was adopted for this summary.

Table 2: Key aspects of vitamin D methods used internationally

Method reference	(Mattila *et al.*, 1995a)	(Mattila *et al.*, 1995b)	(Ostermeyer and Schmidt, 2006)	(Jakobsen *et al.*, 2004)	(Jakobsen and Saxholt, 2009)
Matrices tested	Fish and fish products	Milk products, raw meat and liver	Fish	Meat	Milk, butter
Internal standard	Vitamin D_2 25-hydroxyvitamin D_2	Vitamin D_2 25-hydroxyvitamin D_2	Vitamin D_2 Ergosterol (Provitamin D_2)	Vitamin D_2	Vitamin D_2 25-hydroxyvitamin D_2
Initial extraction solvent	Petroleum ether/diethyl ether (1:1)	Petroleum ether/diethyl ether (1:1)	Hexane	Petroleum ether/diethyl ether (1:1)	Petroleum ether/diethyl ether (1:1)
Cleanup steps	2	3	2	3	3
Quantification	HPLC-DAD or HPLV-UV	HPLC-DAD	HPLC-electrochemical detection	HPLC-DAD	HPLC-DAD

HPLC = high-pressure liquid chromatography
UV = ultraviolet detection
DAD = diode array detection

3.3 Assessment of Vitamin D Methods of Analysis

Two main approaches may be adopted in determining what method of analysis is appropriate for a given situation; standard methods or method performance criteria (Rose *et al.*, 2011). Both of these approaches have advantages. Standard methods are more likely to be the result of agreement between various stakeholders and may be considered to be the yardstick in event of disputes over analytical results. Method performance criteria allow for a more dynamic situation, where novel methods can be adopted, provided they meet the necessary performance criteria.

Byrdwell *et al.* (2008) reported on efforts by a United States Department of Agriculture (USDA) analytical methods committee "to determine the best analytical approach" for the analysis of vitamin D. The approach taken was to develop five control food materials to be analysed by laboratories using candidate methods. The five matrix-specific control materials included canned salmon and vitamin D_3 fortified skim milk, processed cheese, cereal and orange juice. Six US laboratories were able to obtain accurate results on vitamin D content using existing analytical methods and experienced analysts, with relative standard deviations of 7 - 12% (Phillips *et al.*, 2008). The level of agreement was found to be within the expected range for a collaboratively assessed single method. It was concluded that the most important step was the characterisation of the control material, as a means of assessing the performance of different analytical methods. This is consistent with a performance criteria approach to analytical methods.

The general similarity of analytical methods for vitamin D currently in international usage suggests that they are probably all potentially satisfactory and that the focus should be on the performance characteristics of the methods, rather than the details of methodological procedures. Control materials, such as CRMs are particularly important for this assessment.

3.4 Testing Services Available in New Zealand

Table 3 lists the New Zealand laboratories currently accredited by International Accreditation New Zealand (IANZ) for the analysis of vitamin D in foods (International Accreditation New Zealand, 2011). IANZ is the national authority for the accreditation of testing and calibration laboratories, inspection bodies and radiology services[9].

[9] http://www.ianz.govt.nz/

Table 3: New Zealand laboratories with IANZ accreditation for analysis of vitamin D in food

Company	Accreditation No.	Analyte	Matrix	Laboratory Method Reference[1]	Reference
AsureQuality Limited	175	Vitamin D_3	Foods Stockfoods	COST 91 (1986), JMA 1 (1985)	(Brubacher *et al.*, 1986; Indyk and Woollard, 1985)
	891	Vitamin D_3	Meat and Poultry	AOAC (18th Ed.)	(AOAC International, 2005)
	445	Vitamin D_3	Milk Powders	3.17.7 COST 91 JMA 1 p121-141 (1985)	(Brubacher *et al.*, 1986; Indyk and Woollard, 1985; Nordic Committee on Food Analysis, 2000)
Cawthron	107	Vitamin D	Oils, fats (spreads), fish	COST 91 (modified)	(Brubacher *et al.*, 1986)
Fonterra Ltd (Dairy Testing Laboratory)	504	Vitamins D_2 and D_3	Milk powders, casein and caseinates, soy powder, vitamin premixes	3.17.7	(Nordic Committee on Food Analysis, 2000)
	67	Vitamin D	Milk, milk powders	17.7 (modified)	(Nordic Committee on Food Analysis, 2000)
New Zealand Laboratory Services	10	Vitamin D Vitamin D_3	Foods (unspecified)	In-house methods AOAC:2003.05, JAOAC 86,400-406:2003	(Staffas and Nyman, 2003)
	426	Vitamin D_3	Milk powder	3.17.7	(Nordic Committee on Food Analysis, 2000)

[1] These references are how the methods are described in the laboratory's terms of registration. '3.17.7' or '17.7' refers to the section reference in NZTM3: New Zealand Dairy Industry Chemical Methods Manual

IANZ International Accreditation New Zealand
JMA Journal of Micronutrient Analysis
COST European Cooperation in the field of Scientific and Technical Research
AOAC Association of Official Analytical Chemists
JAOAC Journal of the AOAC International

A variety of food matrices, including multiple batches of fortified infant formula, food drinks, fruit drinks, margarines and milk products, were analysed for vitamin D by AsureQuality Ltd., using the Journal of Micronutrient Analysis (JMA) and European Cooperation in the field of Scientific and Technical Research (COST) approved methods (Thomson, 2006). Uncertainty (intra-sample variability) ranged from 5 - 12%. A comparison of the results for different batches of the same product showed that the variability between batches (inter-sample variability), expressed as the CV, ranged from 1- 46%. The highest variability was observed for a food drink product, where the measured concentration of vitamin D varied by a factor of three across five batches of the same product. The homogeneity of the food drink product and the consistency between replicates within a batch

suggested that most of the variability was a result of the manufacturing process (distribution between batches of the product) rather than a sub-sampling issue (Thomson, 2006).

While IANZ accreditation provides general confidence in the laboratory, staff, methodology, procedures and documentation, it does not guarantee that each result being produced will be robust, accurate and precise. This needs to be confirmed using quality assurance control data assessed with each analytical batch, such as CRMs or spike recoveries (ideally of comparable concentration and matrix to the sample), a control sample, replicates, adequate LOD, and assessment of intra- and inter-batch variability. This would seem especially important for a challenging and labile analyte such as vitamin D. It is also consistent with a performance criteria approach to method selection.

4 ASSESSMENT OF DATA FOR VITAMIN D CONTENT OF NEW ZEALAND FOODS

4.1 Rationale for Identifying Key Vitamin D Concentration Data to Assess for Quality Assurance

The NZFCD contains in excess of 2,700 records of concentration values for vitamin D (Ministry of Health/Plant & Food Research, 2009). Most of these records are available in a database format and are marketed under the name FOODfiles[10]. A subset of 89 foods from this database was selected for detailed enquiry regarding the origin and quality of the reported vitamin D concentration data, based on an analysis to determine the main contributors to dietary vitamin D intake. Steps to identify the 89 foods are outlined below:

- Firstly, the 123 New Zealand Total Diet Survey (NZTDS) foods, that represent at least 70% of the most commonly consumed foods in New Zealand (Richard Vannoort, personal communication, August 2010), were assigned unfortified vitamin D concentrations from the NZFCD (Ministry of Health/Plant & Food Research, 2009). Average dietary vitamin D intakes for eight population groups were estimated using simulated diets as undertaken for the 2009 NZTDS. Key contributing food groups and individual foods to vitamin D intake were identified from these intake assessments (see section 4.2).
- The exercise was repeated to include vitamin D concentrations cited in the MFD (2009) for potentially fortified foods, namely for flavoured milk, yoghurt, dairy desserts and margarine (see section 4.3).
- The NZTDS food list was then refined by excluding foods that did not contribute to dietary intake of vitamin D. Specifically, fruits, vegetables and beverages (water, tea, beer, wine, carbonated drink), that make no contribution to vitamin D intake, based on New Zealand and international intake assessments, were deleted. Mushrooms were retained as there is some evidence they may contain vitamin D_2 (Lamberg-Allardt, 2006; Teichmann *et al.*, 2007).
- The edited list was augmented with foods not well covered in the NZTDS that are known sources of vitamin D. These additional foods included fish, salad dressing, chicken and lamb's kidney. Additional dairy products (spreadable butter, cream, Colby cheese, Edam cheese and processed cheese) were included since butter and cheese are likely contributors to intake. Some low fat, standard and premium dairy products (sour cream, ice cream, yoghurt) were included as vitamin D content appears to vary with fat content. Sausage roll and fish in coconut cream were also included as each recipe comprises two food sources of vitamin D and these composite foods are currently not in the NZTDS list of foods. According to the NZFCD, saveloy (0.9 µg/100 g) and Danish pastry (1.51 µg/100 g) have high vitamin D concentrations, and were therefore included. Margarine with a non-detected level of vitamin D and margarine at the maximum detected concentration of vitamin D, were included to represent an unfortified and a fortified product.
- Bread, egg, burger, meat pie and pizza were included as their vitamin D concentrations were variable in the 2009 NZFCD and because they are commonly

[10] http://www.crop.cri.nz/home/products-services/nutrition/foodcompdata/fcd-products/fcd-foodfiles/index.php

consumed foods. Toasted cheese sandwiches were included as variable results were reported for the vitamin D content of this food.

4.2 Preliminary Vitamin D Intake from Non-fortified Foods

Based on simulated diets and non-fortified food concentrations, the average vitamin D intake for various population sub-groups ranged from 1.5 to 4.2 µg/day. This is consistent with overseas intake estimates (Appendix 3). The two major contributing food groups, for seven of the eight population sub-groups, were 1) dairy products and 2) chicken, egg, fish and meats (CEFM). Lesser contributions came from grain foods, oils and takeaways (Table 4). Infant weaning foods were the major source of vitamin D for a six-month child.

Table 4: **Percentage contribution of food groups to vitamin D intake (unfortified foods)**

Food group	25+ yrs Male	25+ yrs Female	19-24 yrs Male	11-14 yrs Male	11-14 yrs Female	5-6 yrs M&F	1-3 yrs M&F	6 mnths M&F
Alcohol	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Beverage	0.3	0.3	0.1	0.0	0.0	0.0	0.0	0.0
Chicken, eggs, fish, meat (CEFM)	40.9	39.3	30.9	42.1	36.1	27.4	18.4	5.0
Dairy products	46.6	46.1	56.3	45.4	47.1	57.2	66.0	9.7
Fruit	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Grain foods	8.4	9.7	7.9	8.1	10.8	10.8	6.4	1.1
Infant weaning foods	0.0	0.0	0.0	0.0	0.0	0.0	8.6	84.1
Nuts	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Oils	2.9	3.9	2.9	2.9	4.1	3.1	0.0	0.0
Sweets & spreads	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.0
Takeaways	0.9	0.6	1.8	1.3	1.8	1.3	0.6	0.1
Vegetables	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

yrs = years mnths = months M&F = males and females

Within the food groups, individual foods making the greatest contribution to vitamin D intake from non-fortified foods were fresh fish (6.4-24.6%), butter (3.6-29.3%), standard milk (5.3-46.3%), egg (1.8-13.8%), low-fat milk (2.8-10.8%), canned fish (1.8-7.2%), mixed grain bread (0.6-4.8%), salad dressing (2.9-3.9%), cake (0.1-3.0%), wheatmeal bread (0.3-2.4%), cheese (0.5-2.6%), oysters (0.5-0.9%), flavoured milk (0.6-3.7%), plain biscuit (0.4-2.7%), meat pie (0.1-1.1%), ice cream (0.3-1.8%), pasta (0.1-0.6%), brewed coffee (0.3%, adults only), pizza (0.1-0.6%) and lamb's liver (0.3-0.4%). The percentage ranges shown here refer to the percentage contribution to dietary vitamin D intake across the various population sub-groups. Since these foods account for the greatest contribution to vitamin D intakes, they were identified as key foods to query with respect to concentration data.

4.3 Preliminary Vitamin D Intake from Fortified Foods

When fortified vitamin D concentration values, according to label information (Manufactured Food Database, 2009), for flavoured milk, yoghurt, dairy dessert and margarine were included in the intake estimates, average estimated vitamin D intakes ranged from 2.7 to 5.8 µg/day.

Three food groups accounted for most vitamin D intake; dairy products, oils, and CEFM (Table 5). Infant weaning foods were the major source of vitamin D for a six-month infant (Table 5). Major individual food sources of vitamin D in descending order for an adult male were margarine (28.2%), fresh fish (17.1%), butter (14.8%), standard milk (9.7%), egg (6.6%), low-fat milk (6.2%), canned fish (3.7%), mixed grain bread (2.1%), salad dressing (2.0%), yoghurt (2.0%), cake (1.9%), wheatmeal bread (1.2%), cheese (1.0%) and flavoured milk (0.6%). Dairy dessert and ice cream feature as major vitamin D sources for other population sub-groups. The foods identified above account for the greatest contribution to vitamin D intake, if fortified foods are included, and are considered to be key foods to query with respect to concentration data.

Table 5: Percentage contribution of food groups to vitamin D intake (fortified foods)

Food group	25+ yrs Male	25+ yrs Female	19-24 yrs Male	11-14 yrs Male	11-14 yrs Female	5-6 yrs M&F	1-3 yrs M&F	6 mnths M&F
Alcohol	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Beverage	0.2	0.2	0.1	0.0	0.0	0.0	0.0	0.0
Chicken, eggs, fish, meat (CEFM)	28.5	28.5	23.0	26.7	21.3	15.2	10.4	4.1
Dairy products	34.6	36.9	45.0	34.6	38.1	46.8	69.7	20.4
Fruit	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Grain foods	5.8	7.1	5.9	5.1	6.4	6.0	3.6	0.9
Infant weaning food	0.0	0.0	0.0	0.0	0.0	0.0	4.9	68.9
Nuts	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Oils	30.2	26.9	24.7	32.7	33.2	31.3	11.1	5.6
Sweets and spreads	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.0
Takeaways	0.6	0.4	1.3	0.8	1.0	0.7	0.3	0.1
Vegetables	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

yrs = years mnths = months M&F = males and females

4.4 Data Assessment of Targeted Foods

For each of the 89 foods selected from the 2,717 foods in FOODfiles 2010 (the database version of NZFCD), reported vitamin D concentrations, date of analysis, data origin, sample number, methods of analysis and lab accreditation status, were provided by Plant and Food Research (Sivakumaran and Huffman, 2011) (Appendix 4).

Of the 89 foods, the majority of vitamin D concentrations (38 foods) are British or British-derived values. These include concentrations taken directly from the British Food Composition Tables 1991 (17 foods) and concentrations imputed from the British Food

Composition Tables through recipe calulations or values derived from similar foods (21 foods). Vitamin D concentrations for 3 foods were derived from a combination of British and New Zealand data. Vitamin D concentrations of 11 foods are New Zealand analytical values, and a further 20 are derived, in part, from New Zealand data. Lesser numbers of data are from USDA (2 foods), guessed or calculated from the MFD (8 foods), or presumed to be zero (7 foods).

The British or British-derived values were sourced in or prior to 2002, with only very brief general information supplied regarding analytical methodology, for example, 'reverse-phase HPLC, biological assay and spectrometry GLC (gas liquid chromatography)' (Sivakumaran and Huffman, 2011). It should be noted that all methods currently in common use in New Zealand and internationally, and summarised in section 3, are HPLC-based. The biological assay and GLC are no longer commonly used for vitamin D analysis. The lack of more detailed methodology precludes assessment of the robustness of these results.

The New Zealand analytical vitamin D concentrations, reported as the sum of vitamin D forms (Table 6), were analysed by the Institute of Food, Nutrition, and Human Health (IFNHH), Massey University, Palmerston North during 2008-2009. The vitamin D concentrations were derived from four different methods (AOAC 2002.05; AOAC 982.29; Nielson, 1994; and the Nordic Committee on Food Analysis, 2000). The four methods were similar, involving saponification, extraction, normal phase HPLC clean-up and reverse phase HPLC analysis with UV and DAD to determine vitamin D_2 and D_3, which are reported as the sum (vitamin D). Variations in methodology included differences in extraction solvents and internal standards, as previously described (section 3). At the time of analysis, IFNHH was IANZ accredited for the analysis of vitamin D in food, but it is not currently accredited for this analysis (International Accreditation New Zealand, 2011).

Accuracy of the vitamin D results was substantiated by results from an international proficiency testing programme conducted by the Swedish National Food Administration in 2009. IFNHH and a second New Zealand analytical laboratory were two of six laboratories that achieved a satisfactory rating for vitamin D_3 in milk within that international proficiency round (Sivakumaran and Huffman, 2011).

Table 6: Foods with 2008-2009 vitamin D analytical values from New Zealand*

FoodID	Food Name	No. of samples	Vitamin D value (µg/100g)	Method
A1007	Bread, white, sliced, prepacked	1	0.27	AOAC 2002.05, 982.29
F1016	Milk, high calcium, 0.1% fat, May, Anchor and Meadow Fresh	1	0.66	AOAC 2002.05, 982.29
F1051	Butter, spreadable, 'Fernleaf-semisoft'	1	4.45	AOAC 2002.05, 982.29
F1056	Cottage Cheese, Light, 1% fat	1	4.00	(Nielsen, 1994)
F1052	Yoghurt, Greek Style, Fresh 'n' Fruity	1	7.30	(Nielsen, 1994)
G1001	Egg, chicken, boiled	2	1.75	(Nordic Committee on Food Analysis, 2000)
G1008	Egg, whole, raw	2	1.50	(Nordic Committee on Food Analysis, 2000)
H1043	Sausage roll, individual size, microwaved	2	0.29	(Nordic Committee on Food Analysis, 2000)

FoodID	Food Name	No. of samples	Vitamin D value (µg/100g)	Method
J1006	Margarine, Poly, 70% fat, reduced salt	2	17.19	AOAC 2002.05, 982.29
J1008	Margarine, Poly, 50% fat, 'Flora Light'	1	16.25	AOAC 2002.05, 982.29
K1001	Salmon, King, New Zealand, raw	NA	20.14	NA

* (Sivakumaran and Huffman, 2011)
NA=not available

The New Zealand analysed results for naturally occurring vitamin D in egg (1.50-1.75 µg/100 g) and salmon (20.14 µg/100 g) are consistent with overseas data for these two foods (1.8-2.8 µg/100 g and 4.2-29.8 µg/100 g, respectively; see Appendix 1), providing confidence in the New Zealand data relative to other studies.

The New Zealand analysed results for margarine, butter and yoghurt (16.25 and 17.19, 4.45, and 7.30 µg/100 g, respectively) are up to 20 times higher than very limited comparative data from overseas (0.3-10.7, 0.13-1.4, and <0.02 µg/100 g, respectively). These New Zealand foods are most likely fortified foods and the difference may reflect different fortification practices between countries. New Zealand-made cottage cheese (4.0 µg/100 g) contains higher vitamin D concentrations than US-made cottage cheese (0.0-0.01 µg/100 g) (United States Department of Agriculture, 2010), but consistent with concentrations found in US processed cheese (5.7-8.3 µg/100 g) (Phillips *et al.*, 2008). The New Zealand result for vitamin D in white bread is notably higher than the zero value reported for three white breads in the US (United States Department of Agriculture, 2010).

The NZFCD results reported for vitamin D concentration in margarine (16.25 and 17.19 µg/100 g) are consistent with values for New Zealand fortified margarines reported by Thomson (2006) (6.7-14.2 µg/100 g).

Values for vitamin D in the NZFCD are based on one or two samples per food. With such limited samples, it is not possible to be sure of how representative these results are of the New Zealand food supply. Based on the consistency of the New Zealand data for naturally occurring vitamin D_3 with concentration data from overseas, and results from the proficiency round (based on vitamin D in milk), there is little reason to doubt the accuracy of the New Zealand data.

5 DATA GAPS

Currently there are very limited New Zealand-derived vitamin D concentration data in the NZFCD with single brands of bread, butter, cheese, yoghurt, milk, sausage roll and one fish species (salmon). Results for two egg and margarine samples are available (Table 6).

Additional validated data is available for a fortified infant formula, three fortified food drinks, a fortified fruit drink, eight fortified margarines, and five fortified milk products (Thomson, 2006).

The individual foods, and associated New Zealand concentration data, estimated to make the greatest contributions to vitamin D intake (sections 4.2 and 4.3) are provided in Table 7. Where no FoodID is provided, but vitamin D concentrations are provided, the data were sourced from Thomson (2006). Where no FoodID or vitamin D concentration values are provided, the food is considered to be a potential contributor to vitamin D intake for New Zealanders, but no New Zealand-specific concentration information is available.

Table 7: New Zealand data for key fortified and non-fortified foods contributing to vitamin D intake

Food	FoodID	Food name	Vitamin D (µg/100g)
Margarine*		Gold 'n Canola Lite spread; Gold 'n Canola Standard; Flora Light Spread; Flora Canola; Flora Proactive Light; Flora Proactive; Olivio-Bertolli Light; Olivio-Bertolli (n= 40)	6.7-14.2
Butter	F1051	Butter, spreadable, "Fernleaf-semisoft" (n=1)	4.45
Milk (3.25% fat)			NA
Milk (0.5% fat)	F1016	Milk, high calcium, 0.1% fat, May, Anchor and Meadow Fresh (n=1)	0.66
Milk (flavoured)*		Calci Kids ChocoZoom, Calci Kids Chocozoom UHT, Primo Chocolate (n=15)	0.16-0.52
Cheese	F1056	Cottage cheese, light, 1% fat (n=1)	4.00
Ice-cream			NA
Yoghurt*			NA
Dairy dessert*		Calci Kids Dairy Food (n=5)	1.12
Egg	G1001 G1008	Egg, chicken, boiled (n=2) Egg, whole, raw (n=2)	1.75 1.50
Salad dressing			NA
Fish (canned)			NA
Fish (fresh)	K1001	Salmon, King, fillet, New Zealand, raw (n=1)	20.14
Oysters			NA
Lambs liver			NA

Food	FoodID	Food name	Vitamin D (µg/100g)
Meat pie			NA
Bread (wheatmeal)	A1007	Bread, white, sliced, prepacked (n=1)	0.27
Bread (mixed grain)	A1007	Bread, white, sliced, prepacked (n=1)	0.27
Cake			NA
Biscuits (plain)			NA
Pasta			NA
Pizza			NA
Coffee beans			NA

* These foods were identified as potentially fortified (Manufactured Food Database, 2010)
NA = no New Zealand data is available for these foods

By estimating which foods have the greatest impact on vitamin D intake, the priority foods identified for vitamin D analysis are butter, milk, cheese, ice-cream, yoghurt, salad dressing, fish (fresh and canned), oysters, lambs liver, meat pie, bread (wheatmeal and mixed grain), cake, biscuits (plain), pasta, pizza and brewed coffee. The contributions of these foods to dietary vitamin D intakes of New Zealand subpopulations are summarised in sections 4.2 and 4.3.

6 PRIORITISED PLAN TO UPDATE VITAMIN D DATA

1 Consider including the vitamin D results reported by Thomson (2006) in the NZFCD, given the methods used and associated method performance information.

2 Analyse samples of butter, milk, cheese, ice-cream, yoghurt, salad dressing, fish (fresh and canned), oysters, lambs liver, meat pie, bread (mixed grain and wheatmeal), cake, biscuits (plain), pasta, pizza, and brewed coffee as priority foods for vitamin D concentration.

3 Since fortification with vitamin D is voluntary in New Zealand, intake will depend on brand choices. Attention may be given to market share of fortified and non fortified foods within the foods identified in 2 above.

4 Attention be given to adequate validation of the analytical methodology. Detection by mass spectrometry is recommended and, if available, analysis of a CRM, preferably of the same matrix and concentration, per batch. In the absence of a suitable CRM, spike recoveries and at least one duplicate be analysed per analytical batch (for repeatability) and one quality control sample per analytical run, for determination of uncertainty between analytical runs. It would be desirable to have the analytical laboratory have two to three samples of different matrices, validated by a second accredited laboratory, as a cross-check.

5 Sample numbers should be adequate to reflect intra- and inter- batch variability. If necessary, sample numbers per food should reflect the relative importance of the food to dietary intake, that is, more samples of those foods likely to make the greatest contribution to vitamin D dietary intake.

7 REFERENCES

AOAC International. (2005) Official Methods of Analysis of AOAC International (OMA), 18th Edition. Accessed at: http://www.eoma.aoac.org/. Accessed: 3 February 2009.

Bailey RL, Dodd KW, Goldman JA, Gahche JJ, Dwyer JT, Moshfegh AJ, Sempos CT, Picciano MF. (2010) Estimation of total usual calcium and vitamin D intakes in the United States. Journal of Nutrition; 140(4): 817-822.

Banville C, Vuillemard JC, Lacroix C. (2000) Comparison of different methods for fortifying Cheddar cheese with vitamin D. International Dairy Journal; 10(5-6): 375-382.

Biancuzzo RM, Young A, Bibuld D, Cai MH, Winter MR, Klein EK, Ameri A, Reitz R, Salameh W, Chen TC, Holick MF. (2010) Fortification of orange juice with vitamin D_2 or vitamin D_3 is as effective as an oral supplement in maintaining vitamin D status in adults. American Journal of Clinical Nutrition; 91(6): 1621-1626.

Borradale D, Kimlin M. (2009) Vitamin D in health and disease: an insight into traditional functions and new roles for the 'sunshine vitamin'. Nutrition Research Reviews; 22: 118-136.

Brubacher G, Müller-Mulot W, Southgate DAT. (1986) Methods for the determination of vitamins in food – recommended by COST 91. London: Elsevier Science Publishers.

Byrdwell WC, DeVries J, Exler J, Harnly JM, Holden JM, Holick MF, Hollis BW, Horst RL, Lada M, Lemar LE, Patterson KY, Philips KM, Tarrago-Trani MT, Wolf WR. (2008) Analyzing vitamin D in foods and supplements: Methodologic challenges. American Journal of Clinical Nutrition; 88(Suppl): 554S-557S.

Calvo MS, Whiting SJ, Barton CN. (2005) Vitamin D intake: A global perspective of current status. Journal of Nutrition; 135(2): 310-316.

Camargo CA, Ingham T, Wickens K, Thadhani RI, Silvers KM, Epton MJ, Town GI, Espinola JA, Crane J. (2010) Vitamin D status of newborns in New Zealand. British Journal of Nutrition; 104(7): 1051-1057.

Clausen I, Jakobsen J, Leth T, Ovesen L. (2003) Vitamin D_3 and 25-hydroxyvitamin D_3 in raw and cooked pork cuts. Journal of Food Composition and Analysis; 16(5): 575-585.

Delgado-Zamarreno MM, Sanchez-Perez A, Gomez-Perez MC, Hernandez-Mendez J. (1995) Automatic determination of liposoluble vitamins in butter and margarine using Triton X-100 aqueous micellar solution by liquid chromatography with electrochemical detection. Analytica Chimica Acta; 315(1-2): 201-208.

Edmonds JC. (2010) Considerations for increasing vitamin D in the food supply. In: Proceedings of UV Radiation and its Effects - an Update 2010, NIWA UV Workshop, Queenstown, 7-9 May 2010. Accessed at: http://webcat.niwa.co.nz/documents/NIWAis77.pdf. Accessed: 5 October 2011.

Elmadfa I, Freisling H. (2009) Nutritional status in Europe: methods and results. Nutrition Reviews; 67 (Suppl 1): S130-S134.

Engelsen O. (2010) The relationship between ultraviolet radiation exposure and vitamin D status. Nutrients; 2(5): 482-495.

Food Standards Australia New Zealand. (2002) Australia New Zealand Food Standards Code. Accessed at: http://www.foodstandards.gov.au/foodstandards/ Accessed: August 2011.

Ganesan B, Brothersen C, McMahon DJ. (2011) Fortification of Cheddar cheese with vitamin D does not alter cheese flavor perception. Journal of Dairy Science; 94(7): 3708-3714.

Greenfield H, Southgate DAT. (2003) Food composition data. Production, management and use. Second Edition. Rome: Food and Agriculture Organization of the United Nations.

Hanson AL, Metzger LE. (2010) Evaluation of increased vitamin D fortification in high-temperature, short-time-processed 2% milk, UHT-processed 2% fat chocolate milk, and low-fat strawberry yogurt. Journal of Dairy Science; 93(2): 801-807.

Henderson L, Irving K, Gregory J. (2003) Adults aged 19 to 64 years. Vitamin and mineral intake and urinary analytes. Accessed at: http://www.food.gov.uk/multimedia/pdfs/ndnsv3.pdf. Accessed: August 2011.

Heudi O, Trisconi MJ, Blake CJ. (2004) Simultaneous quantification of Vitamins A, D3 and E in fortified infant formulae by liquid chromatography-mass spectrometry. Journal of Chromatography A; 1022(1-2): 115-123.

Holick MF. (2008) Sunlight, UV-radiation, vitamin D and skin cancer: How much sunlight do we need? Advances in Experimental Medicine and Biology; 624: 1-15.

Horwitz W, Latimer G. (2005) Official methods of analysis of AOAC International, 18th Edition. Maryland, USA: AOAC International.

Indyk H, Woollard DC. (1985) The determination of vitamin D in fortified milk powders and infant formulas by HPLC. Journal of Micronutrient Analysis; 1(2): 121-141.

International Accreditation New Zealand. (2011) Programs-International Acceditation New Zealand. Accessed at: http://cabis.ianz.govt.nz/ianzwebportal. Accessed: August 2011.

Jakobsen J, Clausen I, Leth T, Ovesen L. (2004) A new method for the determination of vitamin D_3 and 25-hydroxyvitamin D_3 in meat. Journal of Food Composition and Analysis; 17(6): 777-787.

Jakobsen J, Saxholt E. (2009) Vitamin D metabolites in bovine milk and butter. Journal of Food Composition and Analysis; 22(5): 472-478.

Jenab M, Salvini S, van Gils CH, Brustad M, Shakya-Shrestha S, Buijsse B, Verhagen H, Touvier M, Biessy C, Wallström P, Bouckaert K, Lund E, Waaseth M, Roswall N, Joensen

AM, Linseisen J, Boeing H, Vasilopoulou E, Dilis V, Sieri S, Sacerdote C, Ferrari P, Manjer J, Nilsson S, Welch AA, Travis R, Boutron-Ruault MC, Niravong M, Bueno-de-Mesquita HB, van der Schouw YT, Tormo MJ, Barricarte A, Riboli E, Bingham S, Slimani N. (2009) Dietary intakes of retinol, β-carotene, vitamin D and vitamin E in the European Prospective Investigation into Cancer and Nutrition cohort. European Journal of Clinical Nutrition; 63(Suppl. 4): S150-S178.

Kazmi SA, Vieth R, Rousseau D. (2007) Vitamin D_3 fortification and quantification in processed dairy products. International Dairy Journal; 17(7): 753-759.

Lamberg-Allardt C. (2006) Vitamin D in foods and as supplements. Progress in Biophysics and Molecular Biology; 92(1): 33-38.

Li TL, Min DB. (1998) Stability and photochemistry of vitamin D_2 in model system. Journal of Food Science; 63(3): 413-417.

Lips P. (2007) Vitamin D status and nutrition in Europe and Asia. Journal of Steroid Biochemistry and Molecular Biology; 103(3-5): 620-625.

Lips P. (2010) Worldwide status of vitamin D nutrition. Journal of Steroid Biochemistry and Molecular Biology; 121(1-2): 297-300.

Manufactured Food Database. (2009) Fortified foods available in New Zealand, December 2009. Wellington: New Zealand Food Safety Authority.

Manufactured Food Database. (2010) Fortified foods available in New Zealand, December 2010. Wellington: New Zealand Food Safety Authority.

Mattila P, Piironen V, Uusi-Rauva E, Koivistoinen P. (1995a) Cholecalciferol and 25-hydroxycholecalciferol contents in fish and fish products. Journal of Food Composition and Analysis; 8(3): 232-243.

Mattila PH, Piironen VI, Uusi-Rauva EJ, Koivistoinen PE. (1995b) Contents of cholecalciferol, ergocalciferol, and their 25-hydroxylated metabolites in milk products and raw meat and liver as determined by HPLC. Journal of Agricultural and Food Chemistry; 43(9): 2394-2399.

Ministry of Health/Plant & Food Research. (2009) The Concise New Zealand Food Composition Tables. Wellington: Ministry of Health.

Montenegro MA, Nunes IL, Mercadante AZ, Borsarelli CD. (2006) Photoprotection of vitamins in skimmed milk by an aqueous soluble lycopene−gum Arabic microcapsule. Journal of Agricultural and Food Chemistry; 55(2): 323-329.

Moore C, Murphy MM, Keast DR, Holick MF. (2004) Vitamin D intake in the United States. Journal of the American Dietetic Association; 104(6): 980-983.

National Health and Medical Research Council/Ministry of Health. (2006) Nutrient Reference Values for Australia and New Zealand including Recommended Dietary Intakes. Canberra/Wellington: National Health and Medical Research Council/Ministry of Health.

New Zealand Food Safety Authority. (2010) New Zealand Food (Supplemented Food) Standard 2010. Accessed at: http://www.foodsafety.govt.nz/elibrary/industry/Zealand_Food-Regulations_Regulations.pdf. Accessed: 20 September 2011.

Nielsen SS. (1994) Introduction to the chemical analysis of foods. Boston, MA: Jones and Bartlett Publishers, Inc.

Nordic Committee on Food Analysis. (2000) Cholecalciferol (vitamin D_3) and ergocalciferol (vitamin D_2). Determination by HPLC in foodstuff. Standard No.167. Oslo: Nordic Committee on Food Analysis (NMKL).

Nowson CA, Margerison C. (2002) Vitamin D intake and vitamin D status of Australians. Medical Journal of Australia; 177(3): 149-152.

Ostermeyer U, Schmidt T. (2006) Vitamin D and provitamin D in fish : Determination by HPLC with electrochemical detection. European Food Research and Technology; 222(3-4): 403-413.

Parliamentary Counsel Office. (2010) Dietary Supplements Regulations 1985, SR 1985/208 Accessed at: http://www.legislation.govt.nz/regulation/public/1985/0208/latest/DLM102109.html. Accessed: 20 September.

Perales S, Delgado MM, Alegría A, Barberá R, Farré R. (2005) Liquid chromatographic determination of Vitamin D_3 in infant formulas and fortified milk. Analytica Chimica Acta; 543(1-2): 58-63.

Phillips KM, Craig Byrdwell W, Exler J, Harnly JM, Holden JM, Holick MF, Hollis BW, Horst RL, Lemar LE, Patterson KY, Tarrago-Trani MT, Wolf WR. (2008) Development and validation of control materials for the measurement of vitamin D_3 in selected US foods. Journal of Food Composition and Analysis; 21(7): 527-534.

Rockell J, Skeaff CM, Logan V, Green T. Vitamin D - A review prepared for the New Zealand Food Safety Authority and the Ministery of Health, in press.

Rockell JE, Green TJ, Skeaff CM, Whiting SJ, Taylor RW, Williams SM, Parnell WR, Scragg R, Wilson N, Schaaf D, Fitzgerald ED, Wohlers MW. (2005) Season and ethnicity are determinants of serum 25-hydroxyvitamin D concentrations in New Zealand children aged 5-14 y. Journal of Nutrition; 135(11): 2602-2608.

Rockell JEP, Skeaff CM, Williams SM, Green TJ. (2006) Serum 25-hydroxyvitamin D concentrations of New Zealanders aged 15 years and older. Osteoporosis International; 17(9): 1382-1389.

Rose M, Poms R, Macarthur R, Popping B, Ulbreth F. (2011) What is the best way to ensure that valid analytical methods are used for food control? Quality Assurance and Safety of Crops & Foods; 3: 123-134.

Saffert A, Pieper G, Jetten J. (2009) Effect of package light transmittance on the vitamin content of milk, part 3: Fortified UHT low-fat milk. Packaging Technology and Science; 22(1): 31-37.

Scientific Committee on Food. (2002) Opinion on the tolerable upper intake level of Vitamin D. European Commission, SCF/YCS/NUT/UPPLEV/38 Final. Accessed at: http://ec.europa.eu/food/fs/sc/scf/out157_en.pdf. Accessed: August 2011.

Sivakumaran S, Huffman L. (2011) Vitamin D concentration in selected New Zealand foods-a review. Client report for MAF. Palmerston North: Plant & Food Research.

Staffas A, Nyman A. (2003) Determination of cholecalciferol (vitamin D_3) in selected foods by liquid chromatography: NMKL collaborative study. Journal of AOAC International; 86(2): 400-406.

Tangpricha V, Koutkia P, Rieke SM, Chen TC, Perez AA, Holick MF. (2003) Fortification of orange juice with vitamin D: A novel approach for enhancing vitamin D nutritional health. American Journal of Clinical Nutrition; 77(6): 1478-1483.

Teichmann A, Dutta PC, Staffas A, Jagerstad M. (2007) Sterol and vitamin D_2 concentrations in cultivated and wild grown mushrooms: Effects of UV irradiation. LWT - Food Science and Technology; 40(5): 815-822.

Thomson B. (2006) Fortification overages of the food supply: Vitamin A, Vitamin D and calcium. ESR Client Report FW0637. Christchurch: ESR.

Trenerry VC, Plozza T, Caridi D, Murphy S. (2011) The determination of vitamin D_3 in bovine milk by liquid chromatography mass spectrometry. Food Chemistry; 125(4): 1314-1319.

Truswell AS. (1998) Essentials of human nutrition. Oxford: Oxford University Press.

United States Department of Agriculture. (2010) USDA National Nutrient Database for Standard Reference, Release 23. Accessed at: http://www.ars.usda.gov/ba/bhnrc/ndl Accessed: August 2011.

Upreti P, Mistry VV, Warthesen JJ. (2002) Estimation and fortification of vitamin D_3 in pasteurized process cheese. Journal of Dairy Science; 85(12): 3173-3181.

Wagner D, Rousseau D, Sidhom G, Pouliot M, Audet P, Vieth R. (2008) Vitamin D_3 fortification, quantification, and long-term stability in Cheddar and low-fat cheeses. Journal of Agricultural and Food Chemistry; 56(17): 7964-7969.

Webb AR, Kline L, Holick MF. (1988) Influence of season and latitude on the cutaneous synthesis of vitamin D_3: exposure to winter sunlight in Boston and Edmonton will not promote vitamin D_3 synthesis in human skin. Journal of Clinical Endocrinology and Metabolism; 67(2): 373-378.

APPENDIX 1: CONCENTRATIONS OF VITAMIN D IN FOODS

Table A1. 1: International vitamin D_3 concentration data of naturally occurring in foods

Product	Average (range) vitamin D_3 concentration (µg/100 ml or µg/100 g)*	Country	Reference
Bread			
Bread, white	0.0	USA	(United States Department of Agriculture, 2010)
Cheese			
Cottage cheese	0.0-0.1	USA	(United States Department of Agriculture, 2010)
Eggs			
Egg	2.8 2.0 (1.8-2.2)	Finland USA	(Lamberg-Allardt 2006) (United States Department of Agriculture, 2010)
Egg Yolk	7.8 5.4	Finland USA	(Lamberg-Allardt 2006) (United States Department of Agriculture, 2010)
Fish			
Anchovies	5.7	Finland	(Mattila *et al.*, 1995a)
Eel	25.6	Finland	(Lamberg-Allardt 2006)
Cod	7.0 0.5 0.9 (0.6-1.2)	Finland Germany USA	(Mattila *et al.*, 1995a) (Ostermeyer and Schmidt, 2006) (United States Department of Agriculture, 2010)
Herring	15.4 12.1 2.8	Finland Germany USA	(Lamberg-Allardt, 2006) (Ostermeyer and Schmidt, 2006) (United States Department of Agriculture, 2010)
Mackerel	3.2 5.0 (4.0-8.4)	Finland Germany	(Mattila *et al.*, 1995) (Ostermeyer and Schmidt, 2006)
Perch	1.4	USA	(United States Department of Agriculture, 2010)
Pollack	1.3 1.3	Germany USA	(Ostermeyer and Schmidt, 2006) (United States Department of Agriculture, 2010)
Salmon	24.8 (21.3-29.8) 12.4 14.2 (13.1-17.0) 7.5 (4.2-10.7)	USA Finland USA Germany	(Phillips *et al.*, 2008) (Lamberg-Allardt 2006) (United States Department of Agriculture, 2010) (Ostermeyer and Schmidt, 2006)
Rainbow trout	7.6 8.1 19.1	Finland Germany USA	(Mattila *et al.*, 1995a) (Ostermeyer and Schmidt, 2006) (United States Department of Agriculture, 2010)
Tuna	1.7 7.2 3.8 (2.0-6.7)	Finland Finland USA	(Mattila *et al.*, 1995a) (Lamberg-Allardt 2006) (United States Department of Agriculture, 2010)
Meat			

Product	**Average (range) vitamin D_3 concentration (µg/100 ml or µg/100 g)***	**Country**	**Reference**
Beef	0.22 (0.12-0.35)	USA	(United States Department of Agriculture, 2010)
Chicken	0.29 0.00-0.19	Finland USA	(Mattila *et al.*, 1995b) (United States Department of Agriculture, 2010)
Pork	0.05-0.21 0.11 1.1 (0.5-2.6)	Denmark Finland USA	(Clausen *et al.*, 2003) (Mattila *et al.*, 1995b) (United States Department of Agriculture, 2010)
Ham	0.8 (0.5-0.9)	USA	(United States Department of Agriculture, 2010)
Liver, beef	0.8 <0.05 1.2	Finland Finland USA	(Lamberg-Allardt 2006) (Mattila *et al.*, 1995b) (United States Department of Agriculture, 2010)
Liver, pork	0.40	Finland	(Mattila *et al.*, 1995b)
Milk products			
Yoghurt, plain	<0.02 0.0-0.1	Finland USA	(Mattila *et al.*, 1995b) (United States Department of Agriculture, 2010)

* Results in the United States Department of Agriculture National Nutrient Database are expressed in terms of 'µg per serving' , with serving sizes given in grams. Figures in this table have been recalculated to a 'µg/100 g' basis

Table A1. 2: International vitamin D_3 concentrations of potentially fortified foods

Product	Average (range) vitamin D_3 concentration (µg/100ml or µg/100g)*	Country	Reference
Butter	0.2 (0.13-0.34) 0.3 0.2 1.4	Denmark Finland Finland USA	(Jakobsen and Saxholt, 2009) (Lamberg-Allardt, 2006) (Mattila *et al.*, 1995b) (United States Department of Agriculture, 2010)
Cheese	0.11 6.95 (5.66-8.26) 0.00-0.70	Finland USA USA	(Mattila *et al.*, 1995b) (Phillips *et al*, 2008) (United States Department of Agriculture, 2010)
Cereal	3.31 (2.42-4.30)	USA	(Phillips *et al*, 2008)
Margarine	4.5 (3.2-9.0) 10.7	Spain USA	(Delgado-Zamarreno *et al.*, 1995) (United States Department of Agriculture, 2010)
Cream	0.09 (0.02-0.15) 0.07	Denmark Finland	(Jakobsen and Saxholt, 2009) (Mattila *et al.*, 1995b)
Infant formula	6-15 6.8 (6.7-7.0)	Switzerland Spain	(Heudi *et al.*, 2004) (Perales *et al.*, 2005)
Milk			
Milk (skimmed)	0.005** (0.003-0.007) 1.0 (0.78-1.09) 1.2	Denmark USA USA	(Jakobsen and Saxholt, 2009) (Phillips *et al*, 2008) (United States Department of Agriculture, 2010)
Milk (whole)	0.009** (0.008-0.013) 0.7 <0.02 1.3	Denmark Spain Finland USA	(Jakobsen and Saxholt, 2009) (Perales *et al*, 2005) (Mattila *et al.*, 1995b) (United States Department of Agriculture, 2010)
Milk (whole, organic)	0.008** (0.003-0.019)	Denmark	(Jakobsen and Saxholt, 2009)
Orange juice	1.5 (0.94-2.84)	USA	(Phillips *et al*, 2008)

* Results in the United States Department of Agriculture National Nutrient Database are expressed in terms of 'µg per serving' , with serving sizes given in grams. Figures in this table have been recalculated to a 'µg/100 g' basis

** Data from Denmark (Jakobsen and Saxholt, 2009) = apparently, no fortified products were assessed

APPENDIX 2: NEW ZEALAND FOODS FORTIFIED WITH VITAMIN D

Table A2. 1: New Zealand foods fortified with vitamin D*

Category	Number of brands	Number of distinct product lines	Vitamin D concentrations (µg/100 g)
Baby foods	1	2	0.8
Butters and margarines	4	20	10.0 (n = 16) 18.0 (n =2)
Dairy desserts	2	22	0.7 (n = 5) 1.0 (n = 17)
Food drinks and meal replacements	5	17	0.5 (n = 1) 1.0 (n = 4) 4.1 (n = 1) 4.6 (n = 3) 8.3 (n = 4) 9.2 (n = 3) 11.5 (n = 1)
Skim milks and reduced fat milks	5	28	0.5 (n = 27) 0.6 (n = 1)
Protein beverages derived from legumes	2	4	0.5 (n = 3) 2.0 (n = 1)
Yoghurts	5	70	0.7 (n = 17) 0.76 (n = 1) 1.0 (n = 52)
Miscellaneous	2	9	1.9 (n = 1) 2.0 (n = 1) 10.0 (n = 2) 12.8 (n = 5)

* (Manufactured Food Database, 2010)

APPENDIX 3: INTERNATIONAL INTAKE ASSESSMENTS OF VITAMIN D

Table A3. 1: Summary of international intake assessments of vitamin D

Country	Major Food Sources	Vitamin D Intake (µg/day)			Reference
Australia	Margarine, canned fish, eggs		male	female	(Nowson and Margerison, 2002)
		Mean	2.6-3.0	2.0-2.2	
Canada and US	Supplements, milk, Cereals		male	female	(Calvo *et al.*, 2005)
		Mean	8.1	7.3	
		Mean[1]	5.1	3.1	
		Median	NR	NR	
Europe	NA		male	female	(Elmadfa and Freisling, 2009) In (Lips, 2007)
		Mean	2.3-6.2	2.0-5.1	
			4.0	2.8	
Europe (Greece, Spain, Italy, France, The Netherlands, Germany, United Kingdom, Denmark, Sweden, Norway)	Fish/shellfish, added fats, meat and meat products, dairy products		male	female	(Jenab *et al.*, 2009)
		Mean	5.5	3.6	
Lebanon			male	female	In (Lips, 2007)
		Mean	NA	2.5	
New Zealand	NA		male	female	(Nowson and Margerison, 2002) (Food Standards Australia New Zealand, 2002)
		Mean	2.0-2.4	2.0-2.4	
Northern India	NA		male	female	In (Lips, 2007)
		Mean	NA	0.4	
Tunisia	NA		male	female	In (Lips, 2007)
		Mean	NA	1.9	
United Kingdom	Fish and fish products, meat and meat products, cereals and cereal products, fat spreads		male	female	(Henderson *et al.*, 2003) (Calvo *et al.*, 2005)
		Mean	4.2	3.7	
		Mean[11]	1.4	1.1	
		Median	3.4	2.7	
United States	Dairy products		male	female	(Moore *et al.*, 2004)
		Mean	5.5-5.9	3.9-4.5	
United States	NA		male	female	(Bailey *et al.*, 2010)
		Mean	5.1-5.6	3.6-4.5	

NA=not available

[11] Contributed by fortified food

APPENDIX 4: CONCENTRATION AND ANALYTICAL DETAILS OF VITAMIN D IN SELECTED NEW ZEALAND FOODS

Table A4. 1: Concentration and analytical details of vitamin D in selected New Zealand foods from the New Zealand FOODfiles 2010 (Sivakumaran and Huffman, 2011)

	FoodID	Food Name	Vit D VALUE µg/100g	Date	Source Code	Source description	No of samples	Method of analysis	Service provider	Laboratory accreditation
1	A1007	Bread, white, sliced, prepacked	0.27	2008	z	NZ analytical data	1	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
2	A1008	Bread, wheatmeal, sliced, prepacked	0.27	2008	grz	Imputed from A1007	n/a	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
3	A1009	Bread, multi-grain,light, sliced, prepacked	0.46	2008	rz	Imputed from NZ analytical data	n/a	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
4	A117*	Cracker, wholemeal, reduced fat	0	before 2002	b	Borrowed from British Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4]: Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	unknown
5	A108	Cake, chocolate, standard	0.4	before 2002	bcrz	Derived from recipe calculation	n/a	n/a	n/a	n/a
6	A113	Muffin,fruit,toasted	0.42	before 2002	br	Imputed from British Food composition Tables 1991[3]	n/a	Food Standard Agency (2002)[4]: Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	unknown
7	A27	Cake,Madeira	1.65	1998	ru	Imputed from related food from USDA[5]	n/a	unknown	n/a	unknown
8	A29	Cake,fancy,iced	0.37	before 2002	bcrz	Derived from recipe calculation	n/a	n/a	n/a	n/a
9	A53	Bread roll,mixed grain,spmkt fresh	0		bcr	Derived from recipe calculation	n/a	n/a	n/a	n/a
10	A4	Biscuit,plain,`Digestive'	0	2002	b	Data borrowed from Co FIDS[4] 2002 (11-506)	n/a	Food Standard Agency (2002)[4]: Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	n/a
11	A72	Biscuit,chocolate coated,`Toffee Pop'	0.08	2002	g	Value was guessed based on the ingredients present in the food	n/a	n/a	n/a	n/a
12	A91	Biscuit,basic,NZ recipe	1.7	before 2002	bcrwz	Derived from recipe calculation	n/a	n/a	n/a	n/a
13	C1018	'Milo',powder,fortified09	16.4	2002	rz	Borrowed from the old record C71	n/a	unknown	n/a	n/a
14	C29	Soy drink,`So Good'	0	2004	p	Ingredients in the food are not good source for Vitamin D. Presume zero	n/a	n/a	n/a	n/a
15	C30	Soy drink,`So Good Lite'	0	2004	p	Ingredients in the food are not good source for Vitamin D. Presume zero	n/a	n/a	n/a	n/a

	FoodID	Food Name	Vit D VALUE µg/100g	Date	Source Code	Source description	No of samples	Method of analysis	Service provider	Laboratory accreditation
16	C4	Coffee, brewed	0	before 2002	b	Borrowed from British Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4]: Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	Unknown
17	D20*	Whole wheat biscuits,`Weet-Bix'	0	1999	br	Imputed from British Food Composition Table 1991[3]	n/a	n/a	n/a	n/a
18	D32	Corn flakes, Kellogg's	0	before 2002	p	Ingredients in the food are not good source for Vitamin D. Presume zero	n/a	n/a	n/a	n/a
19	E25	Oats,rolled,raw	0	1995	b	Borrowed from British Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4]: Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	Unknown
20	E27	Rice, white, polished ,boiled	0	1998	b	Borrowed from British Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4]: Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	Unknown
21	E55	Pasta, fresh, cooked, assorted type	0	1997	br	Borrowed from British Food Composition Table 1991[3]	n/a	n/a	n/a	n/a
22	F1001	Cheese, Edam	0.2	2008	br	NZ analytical data	n/a	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
23	F1006	Cheese, Colby	0.28	2008	cz	NZ analytical data	n/a	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
24	F1015	Cheese, Cheddar, Mild	0.3	2008	cz	NZ analytical data	n/a	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
25	F1014	Cheese, Cheddar, Tasty	0.3	2008	crz	Imputed from NZ analytical food F1015	n/a	AOAC 2002.05 and 982.29[1]	n/a	n/a
26	F1016	Milk, High Calcium, 0.1% fat, May, Anchor & Meadowfresh	0.66	2008	z	NZ analytical data	1	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
27	F1028	Milk, whole, 3.3% fat, Composite	0.48	2008	cz	NZ analytical data. Average of the two season	2	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
28	F1035	Milk, trim, 0.5% fat, Composite	0.33	2008	cz	NZ analytical data	2	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
29	F1042	Milk, lite, 1.5% fat, Composite	0.33	2008	cz	NZ analytical data	2	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
30	F42	Milk, UHT, chocolate flavour	0.01	1986	br	Imputed from British Food Composition Table 1991[3]	n/a	n/a	n/a	n/a

	FoodID	Food Name	Vit D VALUE µg/100g	Date	Source Code	Source description	No of samples	Method of analysis	Service provider	Laboratory accreditation
31	F1046	Butter, salted	5.19	2008	cz	NZ analytical data (average of two brands one with two season)	3	AOAC 2002.05[1] and 982.29[2]	IFNHH[3]	Yes
32	F1050	Butter, unsalted,' Mainland'	5.2	2008	rz	Imputed from NZ analytical data F1046	n/a	AOAC 2002.05[1] and 982.29[2]	IFNHH[3]	Yes
33	F1051	Butter, spreadable,' Fernleaf-semisoft'	4.45	2008	z	NZ analytical data	1	AOAC 2002.05[1] and 982.29[2]	IFNHH[3]	Yes
34	F1056	Cottage Cheese, Light,1% fat	4	2009	z	NZ analytical data	1	Nielsen (1994)[6]6	IFNHH[3]	NO
35	F1059	Milk,1% fat,' Anlene',fortified	2.5	2009	fm	NZMFD 2009[7]	n/a	n/a	n/a	n/a
36	F12	Cheese, cream	0.24	1987	b	Borrowed from British Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	Co FIDS[4]	Unknown
37	F122	Ice cream, vanilla, low fat	0.1	2003	br	Borrowed from Co FIDS related food	n/a	n/a	n/a	n/a
38	F19	Cheese, processed	0.22	1985	b	Borrowed from British Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	Co FIDS[4]	Unknown
39	F23	Cream, sour, cultured	0.16	1985	b	Borrowed from British Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	Co FIDS[4]	Unknown
40	F24	Cream, standard	0.15	1986	b	Borrowed from British Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	Co FIDS[4]	Unknown
41	F29	Ice cream, vanilla, premium	0.1	2003	b	Borrowed from Co FIDS 2002[4]	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	Co FIDS[4]	Unknown
42	F1052	Yoghurt, Greek Style, Fresh'n'Fruity	7.3	2009	z	NZ analytical data	1	Nielsen (1994)[6]	IFNHH[3]	NO
43	F56	Yoghurt, asst fruits, low fat, sweetened	0.01	2002	br	Borrowed from Co FIDS[4] related food	n/a	n/a	n/a	n/a
44	F57	Yoghurt,plain,unsweetened	0	2001	lz	NZ analytical data, lower than detection limit	unknown	unknown	unknown	unknown
45	F58	Cheese,feta	0.5	2001	b	Borrowed from British Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	Co FIDS[4]	unknown

	FoodID	Food Name	Vit D VALUE µg/100g	Date	Source Code	Source description	No of samples	Method of analysis	Service provider	Laboratory accreditation
46	F59	Cheese, ricotta	0.3	2001	rz	Imputed from NZ related food	n/a	n/a	n/a	n/a
47	F70	Dessert, dairy food, chocolate flavour	0	2001	rz	Imputed from NZ related food	n/a	n/a	n/a	n/a
48	F71	Yoghurt, berry, low fat, art sweetened	0	2001	rz	Imputed from NZ related food	n/a	n/a	n/a	n/a
49	F80	Cream, sour, reduced fat	0	2001	rz	Imputed from NZ related food	n/a	n/a	n/a	n/a
50	G1001	Egg, chicken, boiled	1.75	2007	z	New Zealand analytical data	2	NMKL (2000)[7]	IFNHH[2]	Yes
51	G1002	Egg, fried in vegetable oil	1.75	2007	rz	Imputed from G1001	n/a	NMKL (2000)[7]	IFNHH[2]	Yes
52	G1008	Egg, whole, raw	1.5	2007	z	New Zealand analytical data	2	NMKL (2000)[7]	IFNHH[2]	Yes
53	G1009	Egg, whole, scrambled	1.75	2007	rz	Imputed from G1001	n/a	NMKL (2000)[7]	IFNHH[2]	Yes
54	H1039	Potato, fries, independent shops, straight cut (2009)	0	2007	b	Data borrowed from Co FIDS[4]	n/a	Food Standard Agency (2002)[4]: Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	n/a
55	H1043	Sausage roll, individual size, microwaved	0.29	2007	z	NZ analytical data	2	NMKL (2000)[7]	IFNHH[2]	Yes
56	H179	Pizza, frozen, individual size, Hawaiian baked	0.06	2002	br	Imputed from Co FIDS[4]	n/a	n/a	n/a	n/a
57	H190	Pizza, BBQ chicken, large, baked, common, thick crust	0.1	2004	br	Imputed from Co FIDS[4]	n/a	n/a	n/a	n/a
58	H194	Fish, fillet, crumbed, frozen, baked	0	2004	bc	Imputed from Co FIDS[4]	n/a	n/a	n/a	n/a
59	H22	Fish, battered, deep fried	0	1990	g	Value was assumed zero	n/a	n/a	n/a	n/a
60	H31	Pie, mince, individual size, supermarket, ready to eat	0	2002	br	Imputed from Co FIDS[4]	n/a	n/a	n/a	n/a
61	H61	Fish,fingers,baked	0	1990	b	Borrowed from Food Composition Table 1991[3]	n/a			
62	H8	Burger, 'Big Mac', McDonald's	0.37	1980	g	Value was guessed based on the ingredients present in the food	n/a	n/a	n/a	n/a
63	J1006	Margarine,Poly,70% fat, reduced salt	17.19	2008	z	NZ analytical data (fortified food)	2	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes
64	J1008	Margarine,Poly,50% fat,'Flora Light'	16.25	2008	z	NZ analytical data (fortified food)	1	AOAC 2002.05 and 982.29[1]	IFNHH[2]	Yes

	FoodID	Food Name	Vit D VALUE µg/100g	Date	Source Code	Source description	No of samples	Method of analysis	Service provider	Laboratory accreditation
65	J1009	Margarine, Mono olive bld,75% fat,' Olivani'	0	2008	p	Presumed to zero	n/a	n/a	n/a	n/a
66	J13	Oil, canola	0	before 2002	b	Borrowed from Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	Unknown
67	K1001	Salmon, King, fillet, NZ, raw	20.14	2008	z	Data obtained from Massey University[7]	unknown	unknown	unknown	unknown
68	K101	Tuna, canned in brine, drained-	5.8	1990	gz	Value obtained by guessing	n/a	n/a	n/a	n/a
69	K167	Salmon, flesh, smoked, export quality	17	before 2002	br	Imputed from British Food composition Tables 1991[3]	n/a	n/a	n/a	n/a
70	K185	Salmon, Sockeye, canned, drained	9.8	1997	ru	Borrowed from USDA 1997[5] (15087)	n/a	n/a	n/a	n/a
71	K187	Tuna, in oil, canned, drained	3	2003	b	Borrowed from Co FIDS 2002[4] (16-230)	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	Unknown
72	K42	Sardines, drained solids, canned	11	before 2002	br	Imputed from British Food composition Tables 1991[3]	n/a	n/a	n/a	n/a
73	K60	Hoki, flesh, baked	5	1990	g	Value was guessed based on the ingredients present in the food	n/a	n/a	n/a	n/a
74	K79	Orange Roughy, flesh, deep fried	4	1990	g	Value was guessed based on the ingredients present in the food	n/a	n/a	n/a	n/a
75	K89	Snapper, flesh, baked	5	1990	g	Value was guessed based on the ingredients present in the food	n/a	n/a	n/a	n/a
76	L1014	Apple, assorted variety, flesh & skin, fresh	0	2009	p	Not good source, presumed zero	n/a	n/a	n/a	n/a
77	M232	Chicken, breast, flesh, grilled	0	1988	br	Imputed from British Food composition Tables 1991[3]	n/a	n/a	n/a	n/a
78	Q1005	Peanuts all types, dry-roasted, without salt	0	2010	p	Not good source, presumed zero	n/a	n/a	n/a	n/a
79	S49	Dressing, Home style, Eta	0.6	1996	br	Imputed from British Food composition Tables 1991[3]	n/a	n/a	n/a	n/a
80	S7	Dressing, 'Vinaigrette'	0.19	before 2002	br	Imputed from British Food composition Tables 1991[3]	n/a	n/a	n/a	n/a
81	T4	Mussel, Green, steamed	0	before 2002	bcr	Derived form recipe calculation	n/a	n/a	n/a	n/a

	FoodID	Food Name	Vit D VALUE µg/100g	Date	Source Code	Source description	No of samples	Method of analysis	Service provider	Laboratory accreditation
82	T6	Oyster, Dredge,r aw	1	before 2002	br	Imputed from British Food composition Tables 1991[3]	n/a	n/a	n/a	n/a
83	W11	Honey	0	before 2002	b	Borrowed from Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	Unknown
84	X1008	Potato, assorted variety, flesh & skin, boiled	0	2009	p	Not a food source presumed to zero	n/a	n/a	n/a	n/a
85	X60	Mushrooms, flesh and stem, raw	0	before 2003	b	Borrowed from Food Composition Table 1991[3]	n/a	Food Standard Agency (2002)[4] Reverse phase HPLC, Biological assay and spectrophotometry GLC	n/a	Unknown
86	Z1001*	Infant Formula Prepared	1	2008	cm	Averaged from the different brands (NIP)	n/a	n/a	n/a	n/a
87	Z519*	Infant custard/fruit dish	0.03	before 2006	bgrc	Calculated from Food Composition Table 1991[3]	n/a	n/a	n/a	n/a
88	Z522*	Infant, cereal	0	before 2006	br	Borrowed from Food Composition Table 1991[3]	n/a	n/a	n/a	n/a
89	Z533*	Infant lambs fry & bacon	0.02	2003	bcr	Calculated from Co FIDS 2002[4]	n/a	n/a	n/a	n/a

b = British data,
c = computer calculated after data entry
f = fortified
g = guessed by estimation or calculation
l = analytical value less than limit of detection/quantification
m = NIP (nutrient information panel) data from MFD
p = presumed
r = value derived from a related food
u = USDA data
w = value derived from sample with unknown water content
z = New Zealand analytical data