SD1 Attachment A1.1 – Appendices: Calculations and data tables

Appendix 1: Calculations

Estimation of the nutrient concentration in breast milk: Most studies and expert panels report a nutrient concentration in breast milk as mass (g, mg, or µg) per unit volume. Where breast milk concentration was reported as energy density (e.g. mass per kJ or kcal), concentration as mass per unit volume was calculated using 2720 kJ/L which is an average of reported energy content of breast milk of (Nommsen et al. 1991; EC Scientific Committee on Food 2003; Hester et al. 2012).

To compare the infant formula concentration to breast milk concentration, the nutrient amount (in units per 100 kJ) was converted to units/L using the midpoint of the Codex STAN 72-1981 energy range (2725 kJ/L). For simplicity, this midpoint was used to calculate both Standard 2.9.1 and Codex STAN 72-1981 amounts in units/L since it was determined that Codex STAN 72-1981 energy range was unlikely to pose a risk to infant health (see Section 3.2.3) and that the midpoint of the Standard 2.9.1 range (2825 kJ/L) was comparably close to the Codex STAN 72-1981 midpoint given uncertainties in measured breast milk concentrations and the determination of the AI or UL in infant populations. The midpoint was used since the actual energy content is likely to be an average amount within the specified range and the midpoint was assumed to be comparable to this average.

Estimation of daily volume of intake of breast milk or infant formula: The mean volume of breast milk intake for infants 0-<6 months is about 0.8 L/day (NHMRC and NZ MoH 2006, EFSA NDA Panel 2013, EFSA NDA Panel 2014). Since infant formula is based on breast milk nutrient composition, the average intake volume was assumed to be the same amount for fully formula fed infants. Since diets for infants aged 6-<12 months includes complementary foods as well as breast milk, the mean volume of breast milk intake for this age group is about 0.6 L/day (NHMRC and NZ MoH 2006).

Estimation of daily nutrient intake from infant formula: ANZ nutrient reference values (AI or EAR, UL) as reported by the NHMRC and NZ MoH (2006) were used to assess nutritional safety of minimum and maximum amounts of each nutrient, as applicable. Minimum and maximum nutrient amounts (in mass per 100 kJ) were multiplied by the midpoint of the energy content (in kJ/L) and the mean daily intake volume to obtain estimated daily nutrient intakes. This enabled comparison of the nutrient minimum to AIs or EARs, and the nutrient maximums to the ULs.

Section 2.2.3 provides further discussion about the comparison with ANZ NRVs.

The application of these calculations has been shown in the two examples provided below. Example (1) is the comparison of Codex STAN 72-1981 vitamin A amounts to breast milk and ANZ NRVs. The data for other nutrients presented in Appendix 2 was generated using the analogous calculations, as applicable. Example (2) showing calculations for fatty acids LA and ALA is presented due the complexities in expression of fatty acid amounts in standards and scientific reports.

Example (1) - comparison of vitamin A amount in Codex STAN 72-1981 to breast milk and NRVs

	Amount (*µg/100 kJ)*	
	Standard 2.9.1	Codex STAN 72-1981
Minimum	14	14
Maximum	43	43

1) Comparison with breast milk concentration:

$$\boldsymbol{Minimum\ vitamin\ A\ amount} = \frac{14\ \mu g}{100\ kJ} \times \frac{2725\ kJ}{L} = \frac{382\ \mu g}{L}$$

$$\boldsymbol{Maximum\ vitamin\ A\ amount} = \frac{43\ \mu g}{100\ kJ} \times \frac{2725\ kJ}{L} = \frac{1172\ \mu g}{L}$$

Breast milk nutrient concentrations are generally measured mass per unit volume (no conversion required) and for vitamin A, have been reported to be in the range of 150–1100 µg/L (Canfield et al. 2003) with Australian mothers at the lower end of this range (310 µg /L). Therefore, the vitamin A concentration based on the minimum and maximum amounts is consistent with breast milk concentrations.

2) Comparison to NRVs (AI or EAR, UL) for infants aged 0–<6 months:

$$\boldsymbol{Minimum\ intake\ of\ vitamin\ A} = \frac{14\ \mu g}{100\ kJ} \times \frac{2725\ kJ}{L} \times \frac{0.8\ L}{day} = \frac{305\ \mu g}{day}$$

$$\boldsymbol{Maximum\ intake\ of\ vitamin\ A} = \frac{43\ \mu g}{100\ kJ} \times \frac{2725\ kJ}{L} \times \frac{0.8\ L}{day} = \frac{937\ \mu g}{day}$$

The ANZ AI for vitamin A is 250 µg /day for infants 0–<6 months. If formula contained the minimum vitamin A amount, infants aged 0–<6 months would consume 305 µg/day and therefore would meet this requirement.

The ANZ UL for vitamin A is 600 µg /day for infants 0–<12 months. If formula contained the maximum vitamin A amount, infants aged 0–<6 months would consume 937 µg/day and would exceed the UL. Therefore, the maximum amount of vitamin A was determined to potentially exceed the UL and further analysis (including discussion of permitted forms of vitamin A) was undertaken, as explained in Section 3.6.1.

3) Comparison to NRVs (AI or EAR, UL) for infants aged 6–<12 months:

$$\boldsymbol{Minimum\ intake\ of\ vitamin\ A} = \frac{14\ \mu g}{100\ kJ} \times \frac{2725\ kJ}{L} \times \frac{0.6\ L}{day} = \frac{229\ \mu g}{day}$$

$$\boldsymbol{Maximum\ intake\ of\ vitamin\ A} = \frac{43\ \mu g}{100\ kJ} \times \frac{2725\ kJ}{L} \times \frac{0.8\ L}{day} = \frac{703\ \mu g}{day}$$

The ANZ AI for vitamin A is 430 µg /day for infants 6–<12 months. Assuming infants aged 6–<12 month receive 50% of nutrient intake from formula, and 50% from complementary foods (see Section 2.2.2), intakes from formula should meet 50% of the AI or 215 µg/day. If formula contained the minimum vitamin A amount, infants aged 6–<12 months would consume 229 µg/day and therefore would meet this requirement.

The ANZ UL for vitamin A is 600 µg /day for infants 0–12 months. Assuming infants aged 6–<12 month receive 50% of nutrient intake from formula, and 50% from complementary foods (see Section 2.2.2), intakes from formula should not exceed 50% of the UL or 300 µg/day. If formula contained the maximum vitamin A amount, infants aged 6–<12 months would consume 703 µg/day and would exceed the UL. Therefore, the maximum amount of vitamin A was determined to potentially exceed the UL and further analysis (including discussion of permitted forms of vitamin A) was undertaken, as explained in Section 3.6.1.

Example (2): **Comparison of LA and ALA amounts to breast milk and the EFSA AI (see Section 8.3.3)**

	Specified LA Amount		Specified ALA Amount	
	Standard 2.9.1 (% total FA)	**Codex STAN 72-1981 (mg/100 kJ)**	**Standard 2.9.1 (% total FA)**	**Codex STAN 72-1981 (mg/100 kJ)**
Minimum	**9**	**70**	**1.1**	**70**
Maximum	**26**	**330**	**4**	**330 (GUL)**

The following calculations assume 95% of fat is fatty acid (Greenfield and Southgate 2003*).*

1) Conversion of Standard 2.9.1 LA and ALA amounts to mg/100 kJ:

$$\boldsymbol{Amount\ FA} = \%\ \boldsymbol{total\ FA} \times \frac{\boldsymbol{g\ total\ fat}}{\boldsymbol{100\ kJ}} \times \frac{\boldsymbol{0.95\ g\ FA}}{\boldsymbol{g\ fat}} \times \frac{\boldsymbol{1000\ mg}}{\boldsymbol{g}} = \frac{\boldsymbol{mg\ FA}}{\boldsymbol{100\ kJ}}$$

$$\boldsymbol{Minimum\ LA} = 9\%\ LA \times \frac{1.05\ g\ total\ fat}{100\ kJ} \times \frac{0.95\ g\ FA}{g\ fat} \times \frac{1000\ mg}{g} = \frac{90\ mg\ LA}{100\ kJ}$$

$$\boldsymbol{Maximum\ LA} = 26\%\ LA \times \frac{1.5\ g\ total\ fat}{100\ kJ} \times \frac{0.95\ g\ FA}{g\ fat} \times \frac{1000\ mg}{g} = \frac{371\ mg\ LA}{100\ kJ}$$

$$\boldsymbol{Minimum\ ALA} = 1.1\%\ ALA \times \frac{1.05\ g\ total\ fat}{100\ kJ} \times \frac{0.95\ g\ FA}{g\ fat} \times \frac{1000\ mg}{g} = \frac{11\ mg\ ALA}{100\ kJ}$$

$$\boldsymbol{Maximum\ ALA} = 4\%\ ALA \times \frac{1.5\ g\ total\ fat}{100\ kJ} \times \frac{0.95\ g\ FA}{g\ fat} \times \frac{1000\ mg}{g} = \frac{57\ mg\ ALA}{100\ kJ}$$

2) Conversion of breast milk LA and ALA amounts to mg/100 kJ:

Breast milk from North American mothers was reported to contain LA at 8–17% of total fatty acids, and ALA at 0.5–1% of total fatty acids (LSRO 1998). The EFSA NDA Panel (2014) reported these amounts as 10–15% and 0.1–2.0 % of total fatty acids for LA and ALA, respectively, based on Greek and Finnish mothers. Since breast milk fat content and composition is highly influenced by maternal diet, the LSRO amounts were assumed to be more applicable to the ANZ population. LA and ALA amounts were converted to mg/100 kJ using the average total fat content (38 g/L) (Hester et al 2012) and the mean energy content of breast milk (2720 kJ/L) (EC SCF 2003, Hester et al 2012, EFSA 2014).

$$\textbf{Average total FA} = \frac{38\ g\ fat}{L} \times \frac{L}{2720\ kJ} \times \frac{0.95\ g\ FA}{g\ fat} = \frac{1.33\ g\ FA}{100\ kJ}$$

$$\textbf{Minimum LA} = 8\%\ LA \times \frac{1.33\ g\ total\ FA}{100\ kJ} \times \frac{1000\ mg}{g} = \frac{106\ mg\ LA}{100\ kJ}$$

$$\textbf{Maximum LA} = 17\%\ LA \times \frac{1.33\ g\ total\ FA}{100\ kJ} \times \frac{1000\ mg}{g} = \frac{226\ mg\ LA}{100\ kJ}$$

Applying this calculation to ALA in breast milk converts 0.5–1% of total fatty acids to a range of 0.67–13.3 mg ALA/100 kJ.

3) EFSA adequate intake

There are no ANZ AI amounts set for LA and ALA set by the NHMRC and NZ MoH (2006) The EFSA NDA Panel (2013) reported values for adequate intake for LA and ALA as 4% and 0.5% of daily energy intake, respectively, for infants aged 0–12 months. In order assess whether intakes based on the Codex STAN 72-1981 minimum amount would meet these values (as an indicative comparison), these were converted to g/day based on the assumptions:

- Mean EER for ages 0–<6 months is 2333 kJ/day; and for ages 6–<12 months is 3033 kJ/day or 1517 kJ/day from formula (see Table 5 in Section 3.2.2).
- 1 g fat is equivalent to 37 kJ (Standard 1.2.8) where a correction to allow for the fatty acid content of fat (95%) was applied (Greenfield and Southgate 2003).

Conversion of EFSA adequate intakes amounts to g/day:

$$\textbf{Adequate intake LA} = \frac{4\ kJ\ LA}{100\ kJ} \times \frac{2333\ kJ}{day} \times \frac{1\ g\ fat}{37\ kJ} \times \frac{0.95\ g\ FA}{g\ fat} = \frac{2.4\ g}{day}$$

$$\textbf{Adequate intakes ALA} = \frac{0.5\ kJ\ LA}{100\ kJ} \times \frac{2333\ kJ}{day} \times \frac{1\ g\ fat}{37\ kJ} \times \frac{0.95\ g\ FA}{g\ fat} = \frac{0.30\ g}{day}$$

Estimated intake for infants aged 0–<6months based on the Codex STAN 72-1981 minimum:

$$\textbf{Minimum intake of LA} = \frac{70\ mg}{100\ kJ} \times \frac{2725\ kJ}{L} \times \frac{0.8\ L}{day} \times \frac{1\ g}{1000\ mg} = \frac{1.5\ g}{day}$$

$$\textbf{Minimum intake of ALA} = \frac{12\ mg}{100\ kJ} \times \frac{2725\ kJ}{L} \times \frac{0.8\ L}{day} \times \frac{1\ g}{1000\ mg} = \frac{0.26\ g}{day}$$

For infants aged 0–<6 months, the estimated intake based on the Codex STAN 72-1981 minimum LA amount of 70 mg/100 kJ and the minimum ALA amount of 12 mg/100 kJ would be 1.5 g/day and 0.26 g/day, respectively (see Section 3.4.3). These amounts, as shown in the above conversion, are lower but essentially comparable (i.e. within conventional rounding rules) to the adequate intake amounts recommended by EFSA (2013).

Estimated intake for infants aged 6–<12 months based on the Codex STAN 72-1981 minimum:

$$\boldsymbol{Minimum\ intake\ of\ LA} = \frac{70\ mg}{100\ kJ} \times \frac{2725\ kJ}{L} \times \frac{0.6\ L}{day} \times \frac{1\ g}{1000\ mg} = \frac{1.1\ g}{day}$$

$$\boldsymbol{Minimum\ intake\ of\ ALA} = \frac{12\ mg}{100\ kJ} \times \frac{2725\ kJ}{L} \times \frac{0.6\ L}{day} \times \frac{1\ g}{1000\ mg} = \frac{0.20\ g}{day}$$

The estimated intake based on the Codex STAN 72-1981 minimum LA amount of 70 mg/100 kJ and the minimum ALA amount of 12 mg/100 kJ would be 1.1 g/day and 0.20 g/day, respectively. Assuming infants aged 6–<12 months receive 50% of their nutrient intake from formula, and 50% from complementary foods (see Section 2.2.2), intakes from formula should meet 50% of the adequate intakes amounts calculated above, or 1.2 g/day LA and 0.15 g/day ALA.

These amounts, as shown in the above conversion, are essentially comparable (i.e. within conventional rounding rules) to the adequate intake amounts recommended by EFSA (2013).

Appendix 2: Data tables for comparative analysis

Explanatory Notes:

A description of the calculations and conversions used to create data tables (Tables 20-22) has been shown in Appendix 1.

Table 21 shows: (1) amount range (i.e. minimum and maximum) as specified in Standard 2.9.1 and Codex STAN 72-1981; (2) converted nutrient amounts to units of mass per volume using the midpoint of the energy content as specified in Codex STAN 72-1981 (2725 kJ/L), and (3) reported mature breast milk concentrations in units of mass per volume to compare with Standard 2.9.1 and Codex STAN 72-1981 amounts. For energy and macronutrients (protein, fat, and carbohydrate), breast milk concentrations were sourced from several studies to assess the variability between published research on quantitating these nutrients in breast milk. Breast milk concentrations for all other nutrients were sourced from most recent available evidence, as cited.

Table 22 shows: (1) the minimum amounts from Standard 2.9.1 and Codex STAN 72-1981 (in mass per unit volume); (2) the estimated daily intake based on these minimum using the mean volume of intake of 0.8 and 0.6 L/day for ages 0–<6 months and 6–<12 months, respectively; and (3) the AI or EAR amounts derived by the NHMRC and NZ MoH (2006) to compare with Standard 2.9.1 and Codex STAN 72-1981 amounts.

Table 23 shows: (1) the maximum amounts from Standard 2.9.1 and Codex STAN 72-1981 (in mass per unit volume); (2) the estimated daily intake based on these maximums using the mean volume of intake of 0.8 and 0.6 L/day for ages 0-<6 months and 6-<12 months, respectively; and (3) the UL derived by the NHMRC and NZ MoH (2006) to compare with Standard 2.9.1 and Codex STAN 72-1981 amounts.

Abbreviations used in the Tables 21-25 : Std = Standard; conc. = concentration; min = minimum; max = maximum, NS = not specified by Standard 2.9.1 or Codex STAN 72-1981; (v) = voluntary maximum (this is a GUL in Codex STAN 72-1981); n/a = not applicable or not available; NA = no analysis; amount meets this assessment criteria and no further analysis has been described in the report; FA = fatty acid; Est. = estimated, mo = months. Other abbreviations are listed in the Abbreviations and Glossary of the Consultation Paper for Proposal P1028 (page 3–5).

Table 21: Comparison of breast milk nutrient concentration to the range (minimum-maximum) prescribed in Standard 2.9.1 and Codex STAN 72-1981

	Range specified (kJ/L)		Breast milk conc.[1] (kJ/L)	Main outcomes against assessment criteria comparing: • Standard 2.9.1 and Codex STAN 72-1981 amount • Codex STAN 72-1981 amount to breast milk	Section in Report
	Std.2.9.1	*Codex*			
Energy	2500–3150	2500–2950	(1) 2717 (2) 2725 (3) 2717 (4) n/a	Std 2.9.1 maximum is higher than Codex maximum. Mean energy content of breast milk is comparable to the midpoint of the energy Codex energy range (2725 kJ/L).	3.2

Macro-nutrients	Range specified (g/100 kJ)		Range calculated (g/L)		Breast milk conc.[1] (g/L)	Main outcomes against assessment criteria comparing: • Standard 2.9.1 and Codex STAN 72-1981 amount • Codex STAN 72-1981 amount to breast milk	Section in Report
	Std.2.9.1	*Codex*	*Std 2.9.1*	*Codex*			
Protein	0.45–0.7	0.45–0.7	12–19	12–19	(1) 8.5[2] (2) 13[3] (3) 13[3] (4) 12.7[3]	Specified protein range potentially allows protein content to be higher than mean breast milk concentration; numerous inconsistencies in minimum amino acid amounts	3.3
Fat	1.05–1.5	1.05–1.4	29–41	29–38	(1) NS (2) 38 (3) 24–59 (4) 40	Std 2.9.1 maximum different from Codex.	3.4
Carbohydrate	NS	2.2–3.3	n/a	60–89	(1) 65–83 (2) 67 (3) 60–85 (4) 74	Std 2.9.1 does not specify carbohydrate (amount is determined by difference from total energy minus energy from protein and fat) whereas it is a specified amount in Codex	3.5

Sources: (1) EC SCF (2003), (2) Hester et al (2012), (3) EFSA 2014 and (4) NHMRC and NZ MoH (2006). Hester et al (2012) is a systematic review of 21 studies that measured energy, protein, fat and carbohydrate content of mature breast milk.
This is true protein amount estimated by Raiha et al (1985) as the total crude protein content minus non-nutritional proteins and the NPN-fraction.
Crude protein (Total nitrogen x 6.25)

Table 21 Comparison to breast milk concentration fatty acids

Fatty acids	Range or amount specified			Range calculated		Breast milk conc.	Main outcomes against assessment criteria comparing: • **Standard 2.9.1 and Codex STAN 72-1981 amount** • **Codex STAN 72-1981 amount to breast milk**	Section in Report
	Unit	*Std.2.9.1*	*Codex*	*Std 2.9.1*	*Codex*			
				g/L				
Essential FA								
LA[1]	mg/100 kJ % total FA	(90–371) 9-26	70–330[2] NS	2.5–10.1	1.9–9.0	(106–226) 8-17	Expression of amounts of LA and ALA is different in Codex	3.4.3
ALA[2]	mg/100 kJ % total FA	(11–57) 1.1-4	12–NS NS	0.33–1.53	0.33–NS	(6.7–13.3) 0.5–1.0		3.4.3
LC-PUFA								
n-6 PUFA	% total FA	2 (max)	NS	n/a	n/a	n/a	Std 2.9.1 and Codex are not directly comparable.	3.4.4
AA	% total FA	1 (max)	NS	n/a	n/a	0.24–1.0		3.4.4
n-3 PUFA	% total FA	1 (max)	NS	n/a	n/a	n/a		3.4.4
DHA	% total FA	NS	0.5[v]	n/a	n/a	0.06–1.4		3.4.4
Trans FA	% total FA	4 (max)	3 (max)	n/a	n/a	n/a		3.4.5
Erucic acid	% total FA	1 (max)	1 (max)	n/a	n/a	n/a		3.4.5
Saturated FA								
Lauric acid + myristic acid	% total FA	NS	20 (max)	n/a	n/a	n/a		3.4.5
Ratios								
LA:ALA	n/a	5:1 (min) 15:1 (max)	5:1 (min) 15:1 (max)	n/a	n/a	n/a	Std 2.9.1 and Codex are not directly comparable.	3.4.3
LC-6 PUFA: LC-3 PUFA	n/a	≥ 1	NS	n/a	n/a	n/a		3.4.4
EPA:DHA	n/a	≤ 1	≤ 1	n/a	n/a	n/a		3.4.4
AA:DHA	n/a	NS	≥ 1	n/a	n/a	n/a		3.4.4

1 Sources: Essential FA (LA, ALA) from LSRO (1998) which cited studies of North American mothers. DHA and AA amount were taken from Brenna et al. (2007).
2 Values in parentheses are LA and ALA amounts calculated from the specified percentage of total fatty acids as shown in Appendix 1, Example (2).

Table 21 Comparison to breast milk concentration: vitamins

Vitamins (unit)	Range specified		Range calculated		Breast milk conc	Main outcome against assessment criteria: • **Compare Standard 2.9.1 and Codex STAN 72-1981 amount** • **Compare Codex STAN 72-1981 amount to breast milk**	Section in Report
	Std 2.9.1	*Codex*	*Std 2.9.1*	*Codex*	**unit/L**		
	Unit/100 kJ		unit/L				
Vitamin A (µg RE)	14–43	14–43	382–1172	382–1172	192–1120 retinol (450–600 RE)	Permitted forms differ; debate about bioavailability of β-carotene as a source	3.6.1
Vitamin D (µg)	0.25–0.63	0.25–0.6	6.8–17.2	6.8–16.4	0.1–1 (0.25–2.0)	Permitted forms differ; debate about bioavailability of vitamin D_2.	3.6.2
Vitamin E (mg)	0.11–1.1	0.12–1.2(V)	3.0–30	3.3–32.7	3-5.6 α-tocopherol (3.5 α-tocopherol)	Ratio of vitamin E min to PUFA content; maximum is guidance level in Codex	3.6.3
Vitamin K (µg)	1–5 (V)	1–6.5(V)	27–136	27–177	1.4–1.8 (0.85–9.2)	No issues. Breast milk has negligible amount; Vitamin K given prophylactically at birth	NA
Vitamin C (mg)	1.7–5.4(V)	2.5–17(V)	46–147	68–463	30–100 (35–90)	Codex max higher than Std 2.9.1. Codex min is marginally higher but history of safe use.	3.6.4
Niacin preformed (mg)	0.13–0.48(V)	0.07–0.36(V)	3.5–13.1	1.9–9.8	1.1–2.3 (1.8–2.2)	Codex min is different from Std 2.9.1; permitted forms are different.	3.6.5
Thiamin (µg)	10–48(V)	14–72(V)	273–1308	382–1962	154–238 (150–330)	No issues. Codex min and max are higher but history of safe use.	NA
Riboflavin (µg)	14–86(V)	19–119(V)	382–2344	518–3243	274–580 (350–600)	No issues. Codex min and max are higher but history of safe use.	NA
Vitamin B6 (µg)	9–36	8.5–45(V)	245–981	232–1226	70–310 (130)	Codex max is guidance level; vitamin B6 at 15 µg/g protein to support protein synthesis	3.6.6
Folate (µg as folic acid)	2–8(V)	2.5–12(V)	55–218	68–327	26–141 (as folate) (80 as folate)	Calculation of folic acid and comparison to breast milk, Codex max is different	3.6.7
Pantothenic acid (mg)	0.07–0.36(V)	0.096–0.48(V)	1.9–9.8	2.6–13.1	2–2.5 (2.5)	No issues. Codex min and max are higher but history of safe use.	NA
Vitamin B12 (µg)	0.025–0.17(V)	0.025–0.36(V)	0.71–4.6	0.71–9.8	0.16–0.64 (0.2–5)	No issues.	NA
Biotin (µg)	0.36–2.7(V)	0.4–2.4(V)	9.8–73.6	11–65	5–9 (5)	No issues.	NA

1 - Sources: All breast milk concentrations of vitamins as reported by the LSRO (1998) report with values in parentheses reported in EFSA 2014. Note that for most vitamins, concentration is highest early in lactation and decreases to the lower value in the range.
Codex defines units for vitamin A and vitamin E as µg of RE and mg of TE, respectively. See discussion in SD1 Section 7.2.3.

Table 21 Comparison to breast milk concentration

Minerals or Electrolytes (unit)	Range specified		Range calculated		Breast milk conc.[1]	Main outcome against assessment criteria: • Compare Standard 2.9.1 and Codex STAN 72-1981 amount • Compare Codex STAN 72-1981 amount to breast milk	Section in Report
	Std 2.9.1	*Codex*	*Std 2.9.1*	*Codex*			
	unit/100 kJ		unit/L				
Chloride (mg)	12–35	12–38	327–953	327–1036	411–453 (400)	No issues.	NA
Sodium (mg)	5–15	5–14	136–409	136–382	124–207 (140-160)	No issues.	NA
Potassium (mg)	20–50	14–43	545–1363	382–1172	430–543 (500)	Codex min is higher than Std 2.9.1	3.7.1
Calcium (mg)	12–33(V)	12–35(V)	327–899	327–954	194–268 (200–300)	Codex and Std. 2.9.1 are different for calcium:phosphorus ratio	3.7.3
Phosphorus (mg)	6–25	6–24(V)	164–600	164–654	107–164 (107–164)	Difference in specified Ca:P ratio. Codex max is GUL but no evidence to indicate this should be mandatory maximum	3.7.2
Magnesium (mg)	1.2–4.0	1.2–3.6(V)	33–109	33–98	26–49 (15–64)	No issues. Codex max is GUL but no evidence to indicate this should be mandatory maximum	NA
Iron (mg)	0.2–0.5	0.1–0.3	5.5–13.6	2.7–8.2	0.2–0.8 (0.2–0.4)	Codex min and max are substantially different from Std 2.9.1; bioavailability different from breast milk	3.7.4
Iodine (µg)	1.2–10	2.5–14(V)	33–273	68–382	59–178 (50–100)	Codex min and max are higher than Std 2.9.1; Breast milk concentration is region-specific (difficult to compare)	3.7.6
Copper (µg)	14–43	8.5–29(V)	382–1172	232–790	200–700 (329–390)	Codex min and max are substantially different from Std 2.9.1;	3.7.7
Zinc (mg)	0.12–0.43	0.12–0.36(V)	3.3–12	3.3–9.8	1–2 (1.91–0.77)	Codex max is substantially different from Std 2.9.1; ratio to other nutrients	3.7.5
Manganese (µg)	0.24–24	0.25–24(V)	6.5–654	6.8–654	2.0–6.6 (3–30)	Codex max is a GUL.	3.7.8
Selenium (µg)	0.25–1.19	0.24–2.2	6.8–32	6.5–60	5–22 (3–84)	Codex max is higher than Std 2.9.1; Breast milk concentration is region-specific (difficult to compare)	3.7.9
Chromium (µg)	NS–2.0(V)	NS	n/a–55	n/a	0.18–0.39 (0.19–10.8)	No maximum in Codex	3.7.10
Molybdenum (µg)	NS–3(V)	NS	n/a–82	n/a	1.5–2.6 (0.72–4)	No maximum in Codex	3.7.10

1 - Sources: Breast milk concentrations of minerals and electrolytes taken from LSRO (1998) or, in parentheses, from EFSA (2014). Note that for most minerals, concentration is highest early in lactation and decreases over the period of lactation to the lower value in the reported range.

Table 21 Comparison to breast milk concentration cont'd

Optional substances (unit)	Range specified		Range calculated		Breast milk conc.[1]	Main outcome against assessment criteria comparing: • Standard 2.9.1 and Codex STAN 72-1981 amount • Codex STAN 72-1981 amount to breast milk	Section in Report
	Std.2.9.1	*Codex*	*Std 2.9.1*	*Codex*			
	unit/100 kJ		unit/L				
Choline (mg)	1.7–7.1	1.7–12(V)	46–194	46–327	81.9 (160–210)	Estimate of breast milk concentration complex due to different forms. Codex prescribes mandatory addition.	3.8.1
L-carnitine (mg)	0.21–0.8	0.3–NS	5.7–22	8.2–n/a	7.2–12.9 (5.9–10.4)	Codex min is higher than Std 2.9.1. Codex prescribes mandatory addition with no max specified.	3.8.2
Inositol (mg)	1.0–9.5	1–9.5(V)	27–259	27–259	149–312 (130–325)	Codex prescribes mandatory addition.	3.8.3

1 Sources: All breast milk concentrations of nutritive substances as reported by the LSRO (1998) report with values in parentheses reported in EFSA 2014.

Table 22: Comparison of estimated intake from minimum amounts to the AI amount: energy

	Energy midpoint		Intake for 0–<6 months			Intake for 6–<12 months			Outcome against assessment criteria comparing: • Codex STAN 72-1981 minimum to AI	Section in Report
			Est. from minimum		AI	Est. from minimum		AI[1]		
	Std 2.9.1	*Codex*	*Std 2.9.1*	*Codex*		*Std 2.9.1*	*Codex*			
Energy	kJ/L		kJ/day							
	2825	2725	2260	2180	2292	1695	1635	1517	Est. mean energy intake is marginally lower than average EER for 0–<6 mo.	3.2

Table 22: Comparison of estimated intake from minimum amounts to the AI amount: macronutrients

Macro-nutrients	Minimum amount		Unit	Intake for 0–<6 months			Intake for 6–<12 months			Outcome against assessment criteria comparing: • Codex STAN 72-1981 minimum to AI	Section in Report
	Std 2.9.1	*Codex*		Est. from minimum		AI	Est. from minimum		AI[1]		
	g/L			*Std 2.9.1*	*Codex*		*Std 2.9.1*	*Codex*			
Protein	13	12	g/day	10.2	9.8	10.0	7.6	7.4	7.0	Codex min (aligned with Std.2.9.1) meets ANZ AI	3.3
Fat	3.0	2.9	g/day	23.7	22.9	31.0	17.8	17.4	15	Codex min does not meet AI for infants 0–<6 mo.	3.4
Carbohydrate	NS	60	g/day	NS	48	60	n/a	45	48	Codex min does not AI for infants 6–<12 mo	3.5

1 - The AI for 6–<12 month age group, as listed here, is 50% of the AI amount set by NHMRC and MoH (2006) since it is assumed that infants in this age group were assumed to consume 50% of their nutrient intake from infant formula and 50% from complementary foods.

Table 22: Comparison of estimated intake from minimum amounts to the AI amount: fatty acids

Fatty acids	Minimum amount			Unit	Intake for 0–<6 months			Intake for 6–<12 months			Outcome against assessment criteria comparing: • Codex STAN 72-1981 minimum to AI	Section in Report
					Est. from minimum		AI	Est. from minimum		AI[1]		
	Unit	*Std 2.9.1*	*Codex*		*Std 2.9.1*	*Codex*		*Std 2.9.1*	*Codex*			
LA	g/L	2.5	1.9	mg/day	2.0	1.5	n/a (2.3)	1.5	1.1	n/a (1.4)	No ANZ AI; Codex min does not meet AI set by EFSA (indicated in parentheses)	3.4.3
ALA		0.33	0.33		0.26	0.26	n/a (0.29)	0.20	0.20	n/a (0.22)	No ANZ AI; Codex min does not meet AI set by EFSA (indicated in parentheses)	3.4.3
n-6 PUFA	% total FA	2 (max)	NS	g/day	n/a	n/a	4.4	n/a	n/a	4.6	No ANZ AI for n-6 PUFA; see Section 8.4.2 for discussion of AA.	3.4.4
n-3 PUFA		1 (max)	NS		n/a	n/a	0.5	n/a	n/a	0.5	No ANZ AI for n- 3 PUFA; see Section 8.4.1 for discussion of DHA.	3.4.4

1 - The AI for 6–<12 month age group, as listed here, is 50% of the AI amount set by NHMRC and MoH (2006) since it is assumed that infants in this age group were assumed to consume 50% of their nutrient intake from infant formula and 50% from complementary foods.

Table 22: Comparison of estimated intake from minimum amounts to the AI amount: vitamins

Vitamins (unit)	Minimum amount		Intake for 0–<6 months			Intake for 6–<12 months			Outcome against assessment criteria comparing: • Codex STAN 72-1981 minimum to AI	Section in Report
	Std. 2.9.1	*Codex*	Est. from minimum		AI	Est. from minimum		AI		
			Std 2.9.1	*Codex*		*Std 2.9.1*	*Codex*			
	unit/L		unit/day							
Vitamin A (µg RE	382	382	305	305	250	229	229	186[2]	No issues. Codex min meets AI for 0–<12 mo.	3.6.1
Vitamin D (µg)	6.8	6.8	5.5	5.5	5	4.1	4.1	2.5	Debate about bioavailability of vitamin D_2 and increased recommended intakes.	3.6.2
Vitamin E (mg TE)	3.0	3.3	2.4	2.6	4	1.8	2.0	2.5	Codex min does not meet AI for 0–<12 mo.	3.6.3
Vitamin K (µg)	27	27	22	22	2	16	16	1.25	No issues. AI is low because assumes prophylactic vitamin K administered at birth.	NA
Vitamin C (mg)	46	68	37	55	25	28	41	15	No issues. Codex min meets AI for 0–<12 mo	NA
Thiamin (µg)	273	382	218	305	200	164	229	150	No issues. Codex min meets AI for 0–<12 mo	3.6.5
Riboflavin (µg)	382	518	305	414	300	229	311	200	No issues. Codex min meets AI for 0–<12 mo.	NA
Niacin preformed (µg)	3.7	1.9	3.0	1.5	2	2.2	1.1	2	Codex min does not meet AI for 0–<12 mo	3.6.5
Vitamin B6 (µg)	245	232	196	185	100	147	139	150	Codex min meets AI for 0–<12 mo; prescribed ratio of B6 to protein	3.6.6
Folate (µg folic acid)	55	68	44	55	39	33	41	48	Codex min meets AI for 0–<12 mo; bioavailability of folate versus folic acid	3.6.7
Pantothenic acid (mg)	1.9	2.6	1.5	2.1	1.7	1.1	1.6	1.1	No issues. Codex min meets AI for 0–<12 mo	NA
Vitamin B12 (µg)	0.7	0.7	0.50	0.55	0.4	0.4	0.4	0.25	No issues. Codex min meets AI for 0–<12 mo	NA
Biotin (µg)	9.8	11	7.8	8.7	5	5.9	6.6	3	No issues. Codex min meets AI for 0-<12 mo	NA

1 - The AI for 6–<12 month age group, as listed here, is 50% of the AI amount set by NHMRC and MoH (2006) since it is assumed that infants in this age group were assumed to consume 50% of their nutrient intake from infant formula and 50% from complementary foods.

2 - For infants aged 6–<12 months, the AI used for vitamin A was based on reported intake from breast milk (NHMRC and MoH 2006).

Table 22: Comparison of estimated intake from minimum amounts to the AI amount: minerals

Minerals or Electrolytes (unit)	Minimum amount		Intake for 0–<6 months			Intake for 6–<12 months			Outcome against assessment criterion comparing • Codex STAN 72-1981 minimum to AI or EAR	Section in Report
	Std. 2.9.1	*Codex*	Est. from minimum		AI	Est. from minimum		AI[1]		
			Std 2.9.1	*Codex*		*Std 2.9.1*	*Codex*			
	unit/L		unit/day							
Chloride (mg)	327	327	262	262	n/a	196	196	n/a	No issues. No ANZ AI.	NA
Sodium (mg)	136	136	109	109	120	82	82	65	No issues; Codex min meets AI for 0–<12	NA
Potassium (mg)	545	382	436	305	400	327	229	350	Codex min does not meet AI for 0–<12 mo.	3.7.1
Calcium (mg)	327	327	262	262	210	196	196	135	No issue. Codex min meets AI for 0–<12 mo	NA
Phosphorus (mg)	164	164	131	131	100	98	98	138	Codex min does not meet AI for 6–<12 mo	3.7.2
Magnesium (mg)	33	33	26	26	30	20	20	38	No issue. Codex min meets AI for 0–<12 mo	NA
Iron (mg)	5.5	2.7	4.4	2.2	0.2	3.3	1.6	3.5 (EAR)	Codex min does not meet AI for 0–<6 mo or EAR for 6–<12 mo.	3.7.4
Iodine (µg)	33	68	26	55	90	20	41	55	Codex min does not meet AI for 0–<12 mo, evidence of iodine deficiency in ANZ	3.7.6
Copper (µg)	382	232	305	186	200	229	139	110	Codex min does not meet AI for 0–<6 mo	3.7.7
Zinc (mg)	3.3	3.3	2.6	2.6	2	2.0	2.0	1.25 (EAR)	No issues. Codex min meets AI for 0–<12 mo.	NA
Manganese (µg)	654	681	5.2	5.5	3	3.9	4.1	600[2]	No issues Codex min meets AI for 0–<12 mo.	NA
Selenium (µg)	6.8	6.5	5.5	5.2	12	4.1	3.9	7.5	Codex min does not meet AI for 0–<12 mo	3.7.9
Chromium (µg)	NS	NS	n/a	n/a	0.2	n/a	n/a	5.5	No min amount, cannot compare to AI.	3.7.10
Molybdenum (µg)	NS	NS	n/a	n/a	2	n/a	n/a	3	No min amount, compare to AI.	3.7.10

1 - The AI for 6–<12 month age group is 50% of the AI amount set by NHMRC and MoH (2006) since it is assumed that infants in this age group will consume 50% of their nutrient intake from infant formula and 50% from complementary foods. **2** - The AI for infants 6–<12 months is greater than the AI for infants 0-<6 months because the concentration of manganese in food is much greater that in breast milk.

Table 22: Comparison of estimated intake from minimum amounts to the AI amount: optional substances

Nutritive substances (unit)	Minimum amount		Intake for 0–<6 months			Intake for 6–<12 months			Outcome against assessment criterion, comparing • Codex STAN 72-1981 minimum to AI	Section in Report
			Est. from minimum		AI	Est. from minimum		AI[1]		
	Std. 2.9.1	*Codex*	*Std 2.9.1*	*Codex*		*Std 2.9.1*	*Codex*			
	unit/L		unit/day							
Choline (mg)	46.3	46.3(V)	37	37	125	28	28	75	Codex min does not meet AI for 0–<12 months.	3.8.1
L-carnitine (mg)	5.7	8.2	4.6	6.5	n/a	3.5	4.9	n/a	No issues (no AI)	3.8.2
Inositol (mg)	27.3	27.3(V)	21.8	21.8	n/a	16.4	16.4	n/a	No issues (no AI)	3.8.3

1 - The AI for 6-<12 month age group is 50% of the AI amount set by NHMRC and MoH (2006) since it is assumed that infants in this age group will consume 50% of their nutrient intake from infant formula and 50% from complementary foods.

Table 23: Comparison of estimated intake from maximum amounts to NHMRC and MoH (2006) upper level of intake (UL)

Vitamins (unit)	Maximum amount		Intake for 0–<6 months			Intake for 6–<12 months			Outcome against assessment criterion comparing • Codex STAN 72-1981 maximum to UL	Section In Report
			Est. from maximum		UL	Est. from maximum		UL[1]		
	Std 2.9.1	*Codex*	*Std 2.9.1*	*Codex*		*Std 2.9.1*	*Codex*			
	unit/L		unit/day							
Vitamin A (µg RE)	1172	1172	937	937	600	703	703	300	Std 2.9.1 and Codex max exceed UL	3.6.1
Vitamin D (µg)	17.2	16.4	14	14	25	10	10	12.5	No issues. Codex max does not exceed UL	NA
Vitamin E (mg TE)	31	33	25	26	n/a	19	20	n/a	No issues (no UL to compare)	NA
Vitamin K (µg)	136	177	109	142	n/a	82	106	n/a	No issues (no UL to compare)	NA
Vitamin C (mg)	147	463	118	370	n/a	88	278	n/a	No issues (no UL to compare)	NA
Thiamin (µg)	1308	1962	1046	1570	n/a	785	1177	n/a	No issues (no UL to compare)	NA
Riboflavin (µg)	2344	3243	1875	2594	n/a	1406	1946	n/a	No issues (no UL to compare)	NA

Vitamins (unit)	Maximum amount		Intake for 0–<6 months			Intake for 6–<12 months			Outcome against assessment criterion comparing • Codex STAN 72-1981 maximum to UL	Section In Report
			Est. from maximum		UL	Est. from maximum		UL[1]		
	Std 2.9.1	*Codex*	*Std 2.9.1*	*Codex*		*Std 2.9.1*	*Codex*			
Niacin preformed (µg)	13.1	9.8	10.5	7.8	n/a	7.9	5.9	n/a	No issues (no UL to compare)	NA
Vitamin B6 (µg)	981	1226	785	981	n/a	589	736	n/a	No issues (no UL to compare)	NA
Folate (µg folic acid)	218	327	174	262	n/a	131	196	n/a	No issues (no UL to compare)	NA
Pantothenic acid (mg)	9.8	13.1	7.84	10.5	n/a	5.9	7.9	n/a	No issues (no UL to compare)	NA
Vitamin B12 (µg)	4.6	9.8	3.7	7.8	n/a	2.8	5.9	n/a	No issues (no UL to compare)	NA
Biotin (µg)	74	65	59	52	n/a	44	39	n/a	No issues (no UL to compare)	NA

1 The UL for 6–<12 month age group is 50% of the UL amount set by NHMRC and MoH (2006) since it is assumed that infants in this age group will consume 50% of their nutrient intake from infant formula and 50% from complementary foods.

Table 23: Comparison of estimated intake from maximum amounts to NHMRC and MoH (2006) upper level of intake (UL)

Minerals or Electrolytes (unit)	Maximum amount		Intake for 0–<6 months			Intake for 6–<12 months			Outcome against assessment criterion, comparing: • Codex STAN 72-1981 maximum to UL	Section in Report
			Est. from maximum		UL	Est. from maximum		UL[1]		
	Std 2.9.1	*Codex*	*Std 2.9.1*	*Codex*		*Std 2.9.1*	*Codex*			
	unit/L		unit/day							
Chloride (mg)	953	1036	762	829	n/a	572	622	n/a	No issues (no UL to compare)	NA
Sodium (mg)	409	382	327	306	n/a	245	229	n/a	No issues (no UL to compare)	NA
Potassium (mg)	1363	1172	1090	938	n/a	818	703	n/a	No issues (no UL to compare)	NA
Calcium (mg)	899(V)	954(V)	719	828	n/a	539	621	n/a	No issues (no UL to compare)	NA
Phosphorus (mg)	600(V)	654(V)	480	523	n/a	360	392	n/a	No issues (no UL to compare).	NA
Magnesium (mg)	109	98 (V)	87	78	n/a	65.4	59	n/a	No issues	NA

Iron (mg)	13.6	8.2	10.9	6.6	n/a	8.2	4.9	n/a	No issues. (no UL to compare)	NA
Iodine (µg)	273	382(V)	218	306	n/a	164	229	n/a	Codex is a GUL	3.7.6
Copper (µg)	1172	790(V)	938	632	n/a	703	474	n/a	Codex and Std 2.9.1 are substantially different; Codex is a GUL	3.7.7
Zinc (mg)	12	9.8 (V)	9.6	7.8	20	7.2	5.9	10	Codex is a GUL; Est. intake exceeds UL	3.7.5
Manganese (µg)	654	654(V)	523	523	n/a	392	392	n/a	Codex is a GUL; research reports of high exposure in infants	3.7.8
Selenium (µg)	32	60(V)	26	48	n/a	19.2	36	n/a	Codex is a GUL; max exceeds UL; new FDA proposed rule	3.7.9
Chromium (µg)	55(V)	n/a	44	n/a	4	33	n/a	2	Guidance limit in Std 2.9.1 and no max in Codex.	3.7.10
Molybdenum (µg)	82(V)	n/a	66	n/a	n/a	49.2	n/a	n/a	Guidance limit in Std 2.9.1 and no max in Codex.	3.7.10

1 The UL for 6–<12 month age group is 50% of the UL amount set by NHMRC and MoH (2006) since it is assumed that infants in this age group will consume 50% of their nutrient intake from infant formula and 50% from complementary foods.

Table 24: Summary of results comparing vitamin and mineral intake estimated from the minimum amount to the ANZ AI (NHMRC and NZ MOH 2006)

Nutrient	Conclusion from this assessment : Does intake estimated from minimum amount meet the ANZ AI/[1]			
	Standard 2.9.1		Codex STAN 72-1981	
	Age 0–<6 mo	Age 6–<12 mo[2]	Age 0–<6 mo	Age 6–<12 mo[2]
Vitamin A	✓	✓	✓	✓
Vitamin D	✓	✓	✓	✓
Vitamin E	✗	✗	✗	✗
Vitamin K	✓	✓	✓	✓
Vitamin C	✓	✓	✓	✓
Thiamin	✓	✓	✓	✓
Riboflavin	✓	✓	✓	✓
Niacin	✓	✓	✗	✗
Vitamin B6	✓	✗	(✗)	(✗)
Folate	✗	✗	✗	✗
Pantothenic acid	(✗)	✓	✓	✓
Vitamin B12	✓	✓	✓	✓
Biotin	✓	✓	✓	✓
Sodium	(✗)	✓	(✗)	✓
Chloride	n/a	n/a	n/a	n/a
Potassium	✓	(✗)	✗	✗
Calcium	✓	✓	✓	✓
Phosphorus	✓	✗	✓	✗
Magnesium	(✗)	✓	(✗)	✓
Iron	✓	(✗)	✓	✗
Zinc	✓	✓	✓	✓
Iodine	✗	✗	✗	✗
Copper	✓	✓	(✗)	✓
Manganese	✓	✗	✓	✗
Selenium	✗	✗	✗	✗
Chromium	n/a	n/a	n/a	n/a
Molybdenum	n/a	n/a	n/a	n/a

[1] ✓= Minimum amount meets the ANZ AI; ✗ = Minimum amount does not meet the ANZ AI; n/a means cannot be determined because no ANZ AI has been set (chloride) or minimum amounts are not set in Standard 2.9.1 or Codex (chromium and molybdenum). Values in parentheses mean estimated intake is only marginally less than AI.

[2] For older infants (aged 6-<12 months) the estimated intake was compared to 50% of the AI since it was assumed that these infants receive 50% of their nutrient intake form infant formula and 50% of from complementary foods.

Appendix 3: Protein content of infant formula - Table of reviewed evidence

Table 25: Summary of studies (2000-2015) on the relationship between protein content of infant formula and increased obesity risk[1]

1st Author (year)	**Study description:** Type, infant subgroup, N (total at baseline) Feeding groups and/or protein content (g/100 kJ) Feeding duration Measured outcomes	**Key results**	**Comments**
Scaglioni (2000)	Prospective cohort, Term infants, N=147 BF versus bottle-fed Duration = n/a Anthropometric at 0, 1 and 5 yrs	Protein intake at 1yr associated with overweight at 5yrs. High BMI mothers showed higher prevalence of overweight at 5y in bottle-fed compared to BF.	Study supports hypothesis that in children of overweight mothers, bottle feeding increases risk of obesity compared to BF
Hoppe (2004)	Prospective cohort, Term infants, N=251		Study supports hypothesis that high protein intake increases growth and assertion that BF protects against overweight (see Casazza 2013)
Koletzko - ECOT[2] (2009)	RCT, healthy, term infants, N=1678 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF), 1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Anthropometric at 24 mo	HP associated with higher weight (weight-for-length z score) at 24 mo.	HP has highest possible protein content for duration of trial. FoF for 6 mo+ is very high protein content compared to IF and breastmilk.
Ohlund (2010)	Prospective cohort, N=127 Anthropometric at 18 mo		Study supports hypothesis that parental BMI is predictor of child overweight
Escribano - ECOT (2011)	RCT, healthy, term infants, N=805 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF), 1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Anthropometric, kidney volume at 6 mo	LP and BF had lower weight gain and BMI z-score compared to HP. Kidney volume greater in HP.	Short term growth outcomes measured. Study supports hypothesis that growth effects of HP is mediated by effects on kidney.
Mennella (2011)	RCT, healthy, term infants, N=64 LP=0.49 (cow's milk), HP=0.67 (hydrolysed) Feeding duration: 0.5 – 7.5 mo Anthropometric measured monthly to 7.5 mo	LP (cow's milk protein) had higher weight gain than HP (hydrolysed protein).	Study based on hydrolysed protein IF (HP) and standard cow's milk IF (LP) which are not comparable; suggests intact cow's milk protein (not amount) increase weight gain.

[1] Abbreviations: BF = breastfed or breastfeeding, IF = infant formula, FoF = Follow-on formula, LP = low protein content, HP = high protein content, E= energy, MFGM= milk fat globule membrane, BMI = body mass index, IGF =insulin growth factor, yr(s) = year(s), mo= months, wks = weeks.
All other abbreviations as defined in the Consultation Paper for this assessment (p. 3–5).
[2] ECOT is the European Childhood Obesity Trial, also known as the European Childhood Obesity Project Group or CHOP.

1st Author (year)	Study description: Type, infant subgroup, N (total at baseline) Feeding groups and/or protein content (g/100 kJ) Feeding duration Measured outcomes	Key results	Comments
Trabulsi (2011)	RCT, healthy, term infants, N=336 BF, LP=0.46, HP=0.51 Feeding duration 5-120days Anthropometric measured 0-120 days	Growth outcomes for LP and HP not significantly different. HP had higher weight gain/day, weight-for-age z-score at 120 days compared to BF.	LP enriched with lactalbumin to meet minimum amino acids so cannot compare directly to HP.
Socha - ECOT (2011)	RCT, healthy, term infants, N=1678 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF), 1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Anthropometric at 6,12,24 mo; serum measures at 6mo	HP associated with high weight-for-length and BMI at 2 yrs, Serum branched chain amino acids and IGF1 were increased in the HP group.	Study supports the hypothesis that the effects of HP on growth is mediated by metabolic-endocrine response related to insulin.
Escribano -ECOT (2012)	RCT, healthy, term infants, N=80 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF), 1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Anthropometric at 6,12,24 mo; body fat mass at 6 mo	HP had significantly higher weight gain/mo in first 6 mo and tended to have higher fat mass at 6 mo (not significant) which was linked to high BMI at 12 and 24 mo.	Study supports HP in first 6 months linked to higher weight gain but low power study so fat mass measure was not significant different between groups.
Hornell (2013)	Systematic review of 37studies: 8 RCT, 19 cohort, 10 cross-sectional	HP intake in infancy was associated with increased growth and higher BMI in childhood. Protein intake between 15 E% and 20 E% associated with increased risk of overweight.	Current protein range (0.45-0.70 g/100 kJ) expressed as %E (using 1 g protein = 17 kJ) is 7.6-12% so this would not confer increased risk of overweight.
Weber -ECOT (2013)	RCT, healthy, term infants, N=1678 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF), 1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Anthropometric at 6 mo to 6 yrs	HP had small but sig. higher BMI at 6 yrs compared to LP. BMI at 95th percentile (not 50th, 85th, 90th) sig. higher in HP compared to LP but not after adjusting for confounding.	Long term follow up study to Koletzko (2009). Unknown clinical significance of small differences measured at 6 yrs.
Luque -ECOT (2013)	RCT, healthy, term infants, N=1678 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF), 1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Anthropometric at 6mo; kidney volume and serum IGF1	HP showed higher plasma IGF1 which was correlated with higher BMI z-score and kidney volume.	Secondary analysis from ECOT. Study supports hypothesis that effects of HP on growth is mediated by kidney growth however may not be clinically relevant in long term.
Rzehak -ECOT (2013)	RCT, healthy, term infants, N=1090 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF),1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Serum IGF variants at 6 mo	HP had greater serum concentrations of total IGF1, free IGF1, and molar ratio of IGF1/IGF3 compared to LP	Study supports the hypothesis that effects of HP on growth is mediated by metabolic-endocrine responses related to insulin
Inostroza (2014)	RCT, infants of high BMI mothers, N=248 LP=0.39 + probiotics; HP=0.65. Feeding duration: 3–6 mo Anthropometric, plasma markers at 3–6 mo	HP greater weight gain compared to LP. LP weight gain compared to 2009 WHO growth standards.	Study supports other findings that infants of high BMI mothers at risk for greater weight gain.

1st Author (year)	Study description: Type, infant subgroup, N (total at baseline) Feeding groups and/or protein content (g/100 kJ) Feeding duration Measured outcomes	Key results	Comments
Fleddermann (2014)	RCT, N=550 LP=0.45, HP=0.52 Duration of feeding 28–120 days Anthropometric at 30–120 days	No difference in weight gain between LP and HP. Growth per energy intake higher in LP.	LP was higher fat content, enriched with lactalbumin, and added tryptophan and phenylalanine. Cannot compare directly to HP.
Kirchberg -ECOT (2014)	RCT, healthy, term infants, N=691 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF),1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Plasma amino acids and acyl carnitine at 6mo	HP had higher plasma amino acid concentrations compared to LP and BF. Plasma acylcarnitine elevated in HP reflects degradation of excess branched chain amino acids.	Study supports the hypothesis that effects of HP on growth are related to insulin-mediated responses through insulinogenic branched chain amino acids.
Timby (2014)	RCT, healthy, term infants, N=160 LP=0.48 (E=60 kcal/L)+MFGM; HP=0.46 (E+ 66 kcal/L) Feeding duration 0-6mo Anthropometric and cognitive function at 6 mo	Low P supplemented with MFGM associated with positive outcomes in cognitive function.	Study tested LP/low E vs HP/high E but protein in g/100 kJ was similar amount in both groups. LP supplemented with MFGM so cannot compare to HP.
Martin (2014)	RCT, N=120 BF=0.57; LP=0.39 + probiotics; HP=0.65 Feeding duration 3–12 mo Anthropometric, plasma measures at 6 &12 mo	LP had lower weight-for-age and Z-scores compared to BF or HP groups. Higher infant growth in HP also associated with high maternal weight	Study supports findings that infants of high BMI mothers have higher weight gain. LP had added probiotics so the interventions cannot be compared directly.
Ziegler (2015)	RCT, healthy, term infants, N=194 LP=0.39 g/100 kJ (modified); HP = 0.51 g/100 kJ Feeding duration 3–12 mo. Anthropometric, plasma biochemistry at 3–12 mo	Weight gain (g/day) similar in both LP and HP at 3-12 months but both LP and HP show higher weight for age z-score compared to BF.	LP modified to remove of caseino-macropeptide (a patented process) and give higher tryptophan and lower threonine content. Intervention IF not comparable. However, unlike ECOT, the HP in this trial is more comparable to typical IF protein content.
Gruszfeld -ECOT (2015)	RCT, healthy, term infants, N=691 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF),1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Carotid intima-media thickness (cIMT) at 5 years	HP versus LP intake in infancy does not influence cIMT at 5 yrs.	cIMT is a marker for subclinical arteriosclerotic disease and related to obesity. This is a secondary analysis of the ECOT study.
Abrams (2015)	Systematic review 4 RCT studies (6 papers) met inclusion criteria (of 29 studies reviewed)	HP has a small effect on growth compared to control formula (2 studies). LP shows similar growth to BF infants (4 studies)	Study indicates issues with RCTs to date: Test formulas (HP, LP) very different in protein content; HP much higher than usually consumed; and may contain other nutrient components to explain effects.
Escribano -ECOT (2016)	RCT, healthy, term infants, N= 1678 BF=0.41, LP=0.43(IF), 0.53(FoF); HP= 0.70(IF),1.1(FoF) Feeding duration: IF=8wks-6 mo: FoF=6 mo-1yr Neuropsychological testing at 8 yrs	No differences between feeding groups in any of the 23 psychological and behavioural outcomes measured.	High attrition rate (537 assessed in neuropsychological testing). Children who withdrew differed from participants in numerous factors that could influence neurodevelopment.

Appendix 4: Comparison of vitamin E amounts

As discussed in Section 3.6.3, Standard 2.9.1 differs from Codex in the specifications used to calculate the minimum vitamin E amount needed to allow for vitamin E that may be consumed in PUFA oxidation. Standard 2.9.1 has a vitamin E factor which is 0.5 mg vitamin E per g PUFA for all types of PUFA whereas the factor in Codex differs depending on the number of double bonds present in each PUFA.

Using the different factors, Tables 26.1 and 26.2 show the amount of vitamin E (as mg alpha-tocopherol equivalents) calculated for theoretical infant formulas containing (1) the PUFA fat source only and no added DHA; and (2) the PUFA fat source and added DHA to the maximal permitted amount (0.5% of FA) as well as AA (which in Codex must be present in at least the same amount as DHA) and EPA (which should not be present in amounts greater than DHA).The fat sources shown are typical vegetable oils used in infant formula which are blended to provide a fatty acid profile that is similar to breast milk (McSweeney et al. 2013). Tables show minimum amounts of vitamin E that should be in formula containing the minimum or maximum fat content in Codex (1.05g/100 kJ and 1.4 g/100 kJ, respectively) to allow for vitamin E (α-TE) to be expressed in g/100 kJ. Because the purpose of this calculation is to determine the difference between calculating vitamin E using the different specifications in the two Standards, the calculation assumes that each fat source was the sole PUFA source (i.e. does not include PUFA present in bovine milk fat (which is mostly removed during processing).

Table 26.3 shows the minimum quantity of vitamin E calculated for a theoretical formula containing the minimum amounts of LA and ALA in Codex with (1) no added DHA and (2) with added DHA to the maximal permitted amount (0.5% of FA) and the same assumptions for EPA and AA. As above, values were calculated for infant formula containing the minimum and maximum fat content specified in Codex.

Table 26.4 shows vitamin E calculated from a commercial brand of infant formula currently in the marketplace that included sufficient label information to calculate the difference using the specifications in the two standards.

Table 26.1: Minimum vitamin E required in a theoretical infant formula corresponding to the polyunsaturated fat content of various fat sources assuming no added DHA

Fat source	FA composition[1]		PUFA amount[2]			Using Standard 2.9.1 specification		Using Codex specification			Difference = Total vitamin E (Codex) − Total vitamin E (Std. 2.9.1)
						Vitamin E from PUFA amount[3]	Total vitamin E[4]	Vitamin E from PUFA amount[5]		Total vitamin E[4]	
	g/100 g total FA		g PUFA/100 kJ			mg/100 kJ	mg/100 kJ	mg/100 kJ		mg/100 kJ	
	LA	ALA	LA	ALA	Total			LA	ALA		
Minimum fat amount = 1.05 g/100 kJ											
Coconut oil	1.5	-	0.015	0	0.015	0.008	0.128	0.0075	0	0.128	0
Palm oil	9.4	0.3	0.094	0.003	0.097	0.049	0.169	0.047	0.0023	0.169	0
Palm kernel oil	2.0	-	0.02	0	0.02	0.010	0.130	0.01	0	0.130	0
Safflower oil	77.7	-	0.777	0	0.777	0.389	0.509	0.389	0	0.509	0
Safflower oil (high oleic)	12.2	0.2	0.122	0.002	0.124	0.062	0.182	0.061	0.0015	0.183	0.001
Soybean oil	53.2	7.8	0.532	0.078	0.61	0.305	0.425	0.266	0.059	0.445	0.020
Sunflower oil	68.2	0.5	0.682	0.005	0.687	0.344	0.464	0.341	0.004	0.465	0.001
Maximum fat amount = 1.4 g/100 kJ											
Coconut oil	1.5	-	0.020	0	0.020	0.010	0.130	0.0100	0	0.130	0
Palm oil	9.4	0.3	0.125	0.004	0.129	0.065	0.185	0.0625	0.003	0.186	0.001
Palm kernel oil	2.0	-	0.027	0	0.027	0.014	0.134	0.0135	0	0.134	0
Safflower oil	77.7	-	1.033	0	1.033	0.517	0.637	0.517	0	0.637	0
Safflower oil (high oleic)	12.2	0.2	0.162	0.003	0.165	0.083	0.203	0.081	0.002	0.203	0
Soybean oil	53.2	7.8	0.708	0.104	0.812	0.406	0.526	0.354	0.078	0.552	0.026
Sunflower oil	68.2	0.5	0.907	0.007	0.914	0.457	0.577	0.454	0.005	0.579	0.002

[1] Source for LA and ALA amounts: (McSweeney et al. 2013, p.466).

[2] PUFA amounts were converted to g/100 kJ from FA composition data (in g/100 g total FA) using Codex minimum fat amount (1.05 g fat/100 kJ = 1.00 g FA/100 kJ) or Codex maximum fat amount (1.4 g fat/100 kJ = 1.33 g FA/100 kJ), as indicated in table.

[3] Where amount of vitamin E relevant to FA double bonds was calculated from the Standard 2.9.1 conversion of 0.5 mg vitamin E per g PUFA.

[4] Total vitamin E is the vitamin E amount calculated from FA double bonds plus Codex minimum vitamin E amount (0.12 mg/100 kJ).

[5] Where amount of vitamin E relevant to FA double bonds was calculated from the Codex STAN 72-1981 conversion factors: 0.5 mg vitamin E/g LA; 0.75 mg vitamin E/g ALA; 1.0 mg vitamin E/g AA; 1.25 mg vitamin E/g EPA; 1.5 mg vitamin E/g DHA.

Table 26.2: Minimum vitamin E required in a theoretical infant formula corresponding to the polyunsaturated fat content of various fat sources assuming added DHA is also present at maximum permitted amount

Fat source	PUFA amount[1,2]						Using Standard 2.9.1 specification		Using Codex specification					
							Vitamin E from PUFA amount[3]	Total vitamin E[4]	Vitamin E from PUFA amount[5]					Total vitamin E[4]
	g PUFA/100 kJ						mg/100 kJ	mg/100 kJ	mg/100 kJ					mg/100 kJ
	LA	ALA	DHA	AA	EPA	Total			LA	ALA	DHA	AA	EPA	
Minimum fat amount = 1.05 g/100 kJ														
Coconut oil	0.015	0	0.005	0.005	0.005	0.03	0.015	0.135	0.008	0	0.008	0.005	0.006	0.146
Palm oil	0.094	0.003				0.112	0.056	0.176	0.047	0.002				0.188
Palm kernel oil	0.020	0				0.035	0.018	0.138	0.010	0				0.149
Safflower oil	0.777	0				0.792	0.396	0.516	0.389	0				0.527
Safflower oil (high oleic)	0.122	0.002				0.139	0.070	0.190	0.061	0.002				0.201
Soybean oil	0.532	0.078				0.625	0.313	0.433	0.266	0.059				0.463
Sunflower oil	0.682	0.005				0.702	0.351	0.471	0.341	0.004				0.484
Maximum fat amount = 1.4 g/100 kJ														
Coconut oil	0.020	0	0.0067	0.0067	0.0067	0.040	0.020	0.140	0.010	0	0.010	0.007	0.008	0.155
Palm oil	0.125	0.004				0.149	0.075	0.195	0.063	0.003				0.210
Palm kernel oil	0.027	0				0.047	0.024	0.144	0.014	0				0.159
Safflower oil	1.033	0				1.053	0.527	0.647	0.517	0				0.661
Safflower oil (high oleic)	0.162	0.003				0.185	0.093	0.213	0.081	0.002				0.228
Soybean oil	0.708	0.104				0.832	0.416	0.536	0.354	0.078				0.577
Sunflower oil	0.907	0.007				0.934	0.467	0.587	0.454	0.005				0.604

[1] Source for LA and ALA amounts: (McSweeney et al. 2013, p.466). The maximum permitted DHA = 0.5% of FA (or 0.5 g DHA/100 g of FA) as specified in Codex was used and therefore AA and EPA included to same concentration.

[2] LA and ALA amounts in each fat source were converted from g/100 g total FA to g/100 kJ using Codex minimum fat amount (1.05 g fat/100 kJ = 1.00 g FA/100 kJ) or Codex maximum fat amount (1.4 g fat/100 kJ = 1.33 g FA/100 kJ), as indicated in table.

[3] Where amount of vitamin E relevant to FA double bonds was calculated from the Standard 2.9.1 conversion of 0.5 mg vitamin E per g PUFA.

[4] Total vitamin E is the vitamin E amount calculated from FA double bonds plus Codex minimum vitamin E amount (0.12 mg/100 kJ).

[5] Where amount of vitamin E relevant to FA double bonds was calculated from the Codex STAN 72-1981 conversion factors: 0.5 mg vitamin E/g LA; 0.75 mg vitamin E/g ALA; 1.0 mg vitamin E/g AA; 1.25 mg vitamin E/g EPA; 1.5 mg vitamin E/g DHA.

Table 26.3: Vitamin E in a theoretical infant formula using minimum specified LA and ALA amounts plus added DHA[1]

PUFA	PUFA amount[2]	Using Standard 2.9.1 specification		Using Codex specification		Difference = Total vitamin E (Codex) – Total vitamin E (Std. 2.9.1)
		Vitamin E from PUFA amount[3]	Total vitamin E[4]	Vitamin E from PUFA amount[5]	Total vitamin E[4]	
	g PUFA/100 kJ	mg/100 kJ	mg/100 kJ	mg/100 kJ	mg/100 kJ	
Minimum fat amount = 1.05 g/100 kJ						
LA	0.070	0.035	0.169	0.035	0.183	0.014
ALA	0.012	0.006		0.009		
DHA	0.005	0.0025		0.0075		
AA	0.005	0.0025		0.005		
EPA	0.005	0.0025		0.0063		
Maximum fat amount = 1.4 g/100 kJ						
LA	0.070	0.035	0.171	0.035	0.189	0.018
ALA	0.012	0.006		0.009		
DHA	0.0067	0.0034		0.010		
AA	0.0067	0.0034		0.0067		
EPA	0.0067	0.0034		0.0084		

[1] Minimum LA and ALA amounts in Codex were used since maximum amounts are a GUL (for LA) or not specified (for ALA). Therefore a midpoint amount could not be derived.

[2] Assumes DHA added to the maximum permitted amount (0.5% of FA) and therefore AA and EPA included at same concentration to meet Codex restrictions for these PUFA. Values were converted from % of FA (or g PUFA/100 g FA) to g PUFA/100 kJ using Codex minimum fat amount (1.05 g fat/100 kJ = 1.00 g FA/100 kJ) or Codex maximum fat amount (1.4 g fat/100 kJ = 1.33 g FA/100 kJ).

[3] Where amount of vitamin E relevant to FA double bonds was calculated from the Standard 2.9.1 conversion of 0.5 mg vitamin E per g PUFA.

[4] Total vitamin E is the vitamin E amount calculated from FA double bonds plus Codex minimum vitamin E amount (0.12 mg/100 kJ).

[5] Where amount of vitamin E relevant to FA double bonds was calculated from the Codex STAN 72-1981 conversion factors: 0.5 mg vitamin E/g LA; 0.75 mg vitamin E/g ALA; 1.0 mg vitamin E/g AA; 1.25 mg vitamin E/g EPA; 1.5 mg vitamin E/g DHA

Table 26.4: Vitamin E amount based on label information of a commercial infant formula containing PUFA and added DHA

Vitamin E content (as labelled) = 0.34 mg TE/100 kJ

PUFA	PUFA amount[1]	Using Standard 2.9.1 specification		Using Codex specification		Difference = Total vitamin E (Codex) – Total vitamin E (Std. 2.9.1)
		Vitamin E from PUFA amount[2]	Total vitamin E[3]	Vitamin E from PUFA amount[4]	Total vitamin E[3]	
	g PUFA/100 kJ	mg/100 kJ	mg/100 kJ	mg/100 kJ	mg/100 kJ	
LA	0.220	0.110	0.250	0.110	0.262	0.012
ALA	0.035	0.0175		0.0263		
DHA	0.00217	0.0011		0.0033		
AA	0.00227	0.0011		0.0023		
EPA	0.0	0		0		
EPA = maximum permitted						
LA	0.220	0.110	0.251	0.11	0.264	0.014
ALA	0.035	0.0175		0.0263		
DHA	0.00217	0.0011		0.0033		
AA	0.00227	0.0011		0.0023		
EPA	0.00217	0.0011		0.0027		

[1] Amounts are as labelled except for EPA amount which was not listed. Codex restricts EPA content to be no greater than DHA content so calculation was carried assuming a minimum EPA amount of zero or maximum amount of 0.00217 g/100 kJ.

[2] Where amount of vitamin E relevant to FA double bonds was calculated from the Standard 2.9.1 conversion of 0.5 mg vitamin E per g PUFA.

[3] Total vitamin E is the vitamin E amount calculated from FA double bonds plus Codex minimum vitamin E amount (0.12 mg/100 kJ).

[4] Where amount of vitamin E relevant to FA double bonds was calculated from the Codex STAN 72-1981 conversion factors: 0.5 mg vitamin E/g LA; 0.75 mg vitamin E/g ALA; 1.0 mg vitamin E/g AA; 1.25 mg vitamin E/g EPA; 1.5 mg vitamin E/g DHA