

Short Communication

Total Daily Intakes from Commercial Chicken and Beef Infant Meals with Vegetable

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Abstract

The study considered a diet comprising of infant formula milk, lunch and dinner for eight months infants. Levels of four trace elements namely Cu, Fe, Se and Zn and minerals (Ca, K, Mg and Na) in the commercial infant food from the UK market that was used for lunch and dinner were determined using ICP-MS and ICP-AES respectively. This paper discusses total daily intakes from a diet of infant formula milk, chicken and beef meals with vegetable. Generally, finding showed that the levels of the trace elements and minerals were below the Recommended Nutrient Intakes (RNI). A feeding regime of chicken and vegetable based diet including infant formula milk suggested that the average daily consumptions of calcium, copper, iron, magnesium and sodium were below the Recommended Nutrient Intake (RNI). Values above the RNI were suggested in potassium, selenium and zinc by 19 mg/day, 1.4 µg/day and 0.1 mg/day respectively. However, the levels were within the Tolerable Upper Limits. In the beef feeding regimen, results suggest that Zn levels were equal to the RNI (5.00 mg/day). All the other trace elements and minerals were below the RNI except selenium whose values were higher by 0.4 µg/100g. In view of the fact that infants have rapid growth requiring more energy leading to a higher rate of food consumption relative to their body weight, they are more susceptible to consuming higher than recommended nutritional values. In light of these findings it is recommended that the manufacturers include information on the levels of the trace elements and minerals of the commercial foods on the labels.

INTRODUCTION

The World Health Organization (WHO) recommends that breastfeeding be done exclusively in the first 6 months of an infant, with subsequent introduction of complementary foods to accompany breastfeeding [1]. However, only a small number of European women practice breastfeeding of infants exclusively up to 3 months and only 33% in USA [2]. In light of this, the commercial foods and formula milk have been used to complement breast milk. This has not gone without challenges in the levels of the elements [3-5]. Research shows that complementary foods are widely introduced as an alternative at 4-6 months by at least 62% of parents [6].

Accordingly, a large assortment of commercial baby food products is seen in the market. In this regard, the requirement for commercial foods to have sufficient levels of trace elements and minerals cannot be over-emphasized.

The recommended levels for Cu, Fe, Se and Zn are 0.3mg/day (RNI); 7.8mg/day (RNI); 10µg/day (RNI) and 5mg/day (RNI) respectively [2,7,8]. In the same line, the mineral levels recommended are 500-800; 700; 75 and 320 mg/day (RNI) for Ca, K, Mg and Na respectively. The recommended levels were set by the European Food Safety Authority (EFSA); Joint Food and Agricultural Organisation/World Health Organisation Expert Committee on Food and Additives (JECFA); Scientific Committee for Food (SCF); Food and Nutrition Board (FNB) and the World Health Organisation (WHO). The aim was to ensure that the infants were fed with the right quantities of the essential elements and minerals.

In this respect, it is important to consider the total daily intake of nutrients when feeding an infant. Previous studies [7] provide the gastric capacity of 6-9 months infants as 249g per day. This was based on a 30g/kg body weight for 8 month infants with an average weight of 8.3kg. The total daily intake is calculated with the assumption that the child would have two main meals (lunch and dinner), the portion per meal becomes 124.5g as well as have 600ml of infant formula. These assumptions provide a good estimate of the total daily intake for comparison of different meals provided to infants.

The purpose of this paper was to determine the total daily intake of nutrients when feeding an eight months infant using two different regimens: a diet of infant formula milk, chicken and vegetable on one hand and another composed of infant formula milk, beef with vegetable, for breakfast, lunch and dinner. Levels of four trace elements namely Cu, Fe, Se and Zn and minerals (Ca, K, Mg and Na) were determined in the analysis of twelve brands of commercial infant food from the UK market using ICP-MS and ICP-AES respectively. Four brands for the 7+ age group that encompasses eight month old infants were used in this paper to compute the intakes for lunch and dinner.

MATERIALS AND METHODS

The materials used to determine the levels of trace elements and minerals include pestle and mortars (12), beakers, volumetric flasks (20, 50, 100ml), analytical balance, microwave vessels, weighing boats, falcon tubes, pipettes (1, 5, 10, 20, 25ml), micropipettes. The equipment used were CEM MARS 6®

microwave; freeze drier (Labconco FreeZone⁶, Fischer Scientific, UK); ICP-AES ACTIVA system (Horiba JobinYvon, UK) and ICP-MS (Nexlon 350X, Perkin Elmer, US)

Collection of samples

Thirteen samples of a popular brand of baby food were obtained from the supermarkets in UK in June 2015. The age range for the foods, weights and sample identification are shown in Table 1. The samples comprised of vegetable, chicken and beef based diets for three age ranges 4-6 months, 7+ and 10+ months. However, this paper focuses on the foods for the 7+ months in looking at the feeding regimen for an 8 month old infant. The foods were placed under normal conditions in the fridge (at 4°C).

Reagents

Ultrapure water was prepared in the laboratory (18.2 MΩ/cm at 25°C) using Milli pak 2Φ Millipore (Direct-Q[®] 8UV, 0.2μm) equipment. The ultrapure water was used to make solutions in the experiment. Supra pure quality of nitric acid (70% concentrated, Sigma Aldrich, Poole, UK) was used in the sample digestion. Standard solutions of the analytes in this paper (trace elements Cu, Fe, Se and Zn and minerals Ca, K, Mg and Na) were made from seven point dilutions of 1000mg/L respective stock solutions. The standard solutions were supplied by Sigma Aldrich (Poole, UK).

All the glassware and plastic apparatus used in the experiment were cleaned in an acid bath made up of 5% Nitric acid (laboratory grade, Fischer Scientific, UK), for 24 hours to remove impurities before putting any samples. The apparatus were rinsed using deionised water and dried before use.

Freeze drying procedures

The food samples were put in 50ml falcon tubes and weighed on an analytical balance to determine the fresh weights. Subsequently, para film was used to seal the falcon tubes with holes made to allow evaporation of water during the freeze drying process. The food samples were put in a freeze drier (Labconco FreeZone⁶, Fischer Scientific, and UK) at -50°C and a pressure of 0.100mbar for 36 hours. The samples were freeze dried to a constant weight to ensure they were fully dry. The samples were removed from the freeze drier, weighed to confirm the dry masses then stored at room temperature, away from direct sunlight in the falcon tubes.

Microwave digestion

The dry food samples were each homogenised until powdered in individual pestles and mortars that had been cleaned in the acid bath for 24 hours. About 0.5g of each sample was weighed in five replicates and put in clean XP-1500 microwave vessels (CEM, UK). This was followed by the addition of 4ml of supra pure quality concentrated nitric acid (70% analytical grade, Sigma Aldrich, Poole, UK) to each replicate in the fume hood. Blank samples were made using 4ml of nitric acid (70% analytical grade, Sigma Aldrich, Poole, UK). The samples were left open in the fume hood for one hour to reduce pressure build-up in the microwave vessels. Digestion was done in a CEM MARS 6[®] microwave using the food program (Table 2).

The microwave digested samples were removed, taken to the

fume hood and the vessels opened slowly to release pressure. The samples were cleaned off the stoppers and the sides of the vessels using ultrapure water (18.2 Ω/cm at 25°C) then transferred into a 50ml volumetric flasks and filled up to the mark. An aliquot of 5ml of this sample was transferred to 20ml volumetric flask. Indium internal standard was added at a concentration of 1.25μg/ml (25μl aliquot of 1000mg/L stock solution in 20ml volumetric flask) to each replicate sample and sample blank. The volumetric flasks were then filled up to the mark with ultrapure water. The section that follows explains the preparation of mixed standards for ICP-MS.

ICP-MS

Cadmium, copper, iron, selenium and zinc were analysed by ICP-MS. Preparation of the mixed standard was done as indicated in the next section.

PREPARATION OF THE MIXED STANDARD FOR ICP-MS

Mixed standards for ICP-MS (Nexlon 350X, Perkin Elmer, US) were prepared by taking 1ml of 1000mg/L stock solutions of each element (cadmium, copper, selenium and zinc.) obtained from Sigma Aldrich (Poole, UK) and making up to the mark with ultrapure water in a 50ml volumetric flask. This gave an intermediate standard of 20μg/L. A seven point dilution was then made into 20ml volumetric flasks as shown in Table 3 that follows.

In each standard, the same amount of the Indium internal standard was added as used in the samples (25μl) and the solution made up to 20ml using ultra-pure water (18.2MΩ/cm, 25°C).

Analysis by ICP-MS

Analysis was done using a Nexlon 350X, Perkin Elmer (US). The elements, Cu, Fe, Se and Zn had atomic weights as 62.929, 55.934, 81.9 and 65.926 respectively. The samples were introduced using a peristaltic pump that was connected to Meinhard nebulizer by a cyclon spray chamber. The analysis parameters were as shown in Table 4 below.

Kinetic Energy Discrimination (KED) mode was used in the analysis. In this mode, helium is used instead of argon in the collision cell to avoid interference by ArO which has equal mass with Fe (56). This implies that the data becomes more reliable due to the removal of interferences enabling only the quantification of the selected isotope. Calibration curves were obtained and used in the quantification of the elements. The standards for ICP-AES were prepared as explained in the next section. Cu, Fe, Se and Zn and minerals (Ca, K, Mg and Na)

ICP-AES

Calcium, magnesium, potassium, sodium were analysed by ICP-AES. The mixed standards were made as explained in the next section (Table 5).

PREPARATION OF MIXED STANDARDS FOR ICP-AES

Mixed standards for ICP-AES ACTIVA system (Horiba JobinYvon, UK) were prepared by taking 10ml aliquot of

Table 1: Baby foods, age of infant and sample identification code.

Food brand	Age in months	Weight (g)	Sample ID
Baby cauliflower cheese	4-6	125	W1
Grandpa's Sunday lunch	4-6	125	W2
Orchard Chicken	4-6	125	W3
My first Bolognese	4-6	125	W4
Creamy cauliflower cheese	7+	200	X1
Grandma's Sunday lunch	7+	200	X2
Yummy harvest chicken	7+	200	X3
Scrummy spaghetti Bolognese	7+	200	X4
Broccoli cheese	10+	250	Y1
Chicken Sunday lunch	10+	250	Y2
Autumn orchard chicken	10+	250	Y3
My favourite spaghetti Bolognese	10+	250	Y4

shows the age range for the foods, weights and sample identification

Table 2: Conditions for microwave digestion.

	Conditions
Sample weight	0.5 g
Vessel type	HP-500
Nitric acid (HNO ₃)	4 ml
Pressure	Max 350 psi
Power	1500 W
Temperature	Step 1: Ramp to 210°C for 20 minutes Step 2: Hold at 210°C for 15 minutes Step 3: Allow cooling for 30 minutes to room temperature.

Shows the experimental conditions of the microwave digestion

Table 3: Calibration standards concentration for ICP-MS.

Standard	1	2	3	4	5	6	7
Concentration (µg/L)	0	10	20	40	60	80	100
volume (µl) of aliquot	0	10	20	40	60	80	100

Shows the seven point calibration range used in ICP-MS.

Table 4: Analysis Parameters for ICP-MS.

Parameter	Condition
Nebulizer gas flow (ml/min)	0.94
Auxillary gas flow (ml/min)	1.20
Plasma gas flow (ml/min)	18.00
ICP RF power (W)	1600
Sample flush	55seconds (24rpm)
Read delay	35 seconds (20rpm)
Analysis	-20rpm
Wash	60seconds (rpm)
Number of sweeps	25
Number of readings	1
Replicates	3
KED gas used	Helium

Shows the parameter of ICP-MS that were used in the analysis concentrations below:

1000mg/L stock solution of each standard (Ca, Mg, K, and Na) into 100ml volumetric flask. Ultrapure water was added to make up to the mark. Seven point dilutions were made into 20ml volumetric flasks following the

Indium internal standard (25µl) was put and the solution made up to the mark.

Analysis by ICP-AES

The analysis was carried out using ICP-AES ACTIVA system (Horiba JobinYvon, UK). The elements were analysed at the wavelength in nanometres as follows: Ca: 370.603; K: 766.490; Mg: 279.079 and 279.806; Na: 589.592. The analysis parameters for ICP-AES are given in Table 6 that follows.

Spiking

The spiking solution was made by placing 20µl of Se ; 100 µl of Cu and 1ml of Fe and Zn stock standards (1000mg/L) in 100ml volumetric flask and filling to the mark with ultrapure water. The samples were prepared by adding 1ml of the spiking solution into the microwave vessels containing each replicate. 4ml of 70% concentrated nitric acid were added and the samples digested using one touch methods, food program.

Data analysis and statistics

Data analysis in this paper was carried out using Microsoft excel to determine the total daily intake of nutrients when feeding an 8 months old infant. Previous studies [7] showed the gastric capacity of 6-9 months infants as 249g per day. The total daily intake was calculated with the assumption that the child would have two main meals (lunch and dinner), giving the portion per meal as 124.5g, as well as 600ml of infant formula. These assumptions provide a good estimate of the total daily intake for comparison of different meals provided to infants.

FINDINGS

Appendix 1 gives results on the concentration of elements in fresh foods obtained from the samples replicates. The findings suggest that the foods majorly contained potassium, iron and zinc. Grandma's Sunday lunch (X2) had the highest amounts of potassium (185.8-200.2 µg/100g) and iron (137.9-144.4 µg/100g). Zinc was highest in creamy cauliflower cheese (X1) with levels of 133.2-137.7 µg/100g. Quantities of calcium (36.9-37.9 µg/100g) were also highest in creamy cauliflower cheese (X1). The levels of potassium (45.9-47.3) and zinc (48.4-49.7) in sample X4 were relatively alike. Scrummy spaghetti Bolognese

(X4) had the lowest levels of all the elements except copper. The amounts of Se were below 2.00 µg/100g in the samples.

The means of the fresh sample replicates were computed giving results in Table 7 that are presented with the respective standard deviations. Table 7 suggests that levels of copper were highest in Grandma's Sunday lunch (X2) at $19.0 \pm 1.2 \mu\text{g}/100\text{g}$. Iron, potassium and Zinc were the major trace elements and minerals in the foods going as far as $141.7 \pm 2.6 \mu\text{g}/100\text{g}$ and $193 \pm 5.2 \mu\text{g}/100\text{g}$ in Grandma's Sunday lunch (X2) and, $134.8 \pm 2 \mu\text{g}/100\text{g}$ in Creamy cauliflower cheese (X1) respectively. Generally, all the foods had levels of selenium below 2.00 µg/100g. The low standard deviation values indicate a high level of precision in the measurement of the nutrient content of sample replicates.

Total daily intakes from the feeding regimen

Table 8 below gives the total daily intakes from the computation for a chicken and vegetable based commercial diet from the UK market. The foods for 7+ months old were used in the computation considering the gastric capacity for infants of 8 months. The intake of 600ml of milk was used based on the Scientific Advisory Committee recommendation on the average amount of milk intake for infants up to 12 months. However, the menu has a limitation in that it does not represent the exact quantities taken by infants as it is the maximum portion size. The regime also only considers infant formula milk for breakfast and ready meals for lunches and dinners in calculating the total daily intake while in some instances cereals or different menus may be used. The menu does not also account for food wastage. Further the nutrient intake from breast milk, other homemade foods and snacks are not put into consideration.

Table 5: Calibration standards concentration for ICP-AES.

Standard	1	2	3	4	5	6	7
Concentration (µg/ml)	0	7.5	15	30	45	60	75
volume (ml) of aliquot	0	1.5	3	6	9	12	15

Shows the seven point calibration used in ICP-AES

Table 6: Analysis parameters for ICP-AES.

	Condition
Mode of analysis (s)	Normal
Rinsing time (s)	20
Pump	High speed
Transfer time (s)	15
Stabilisation (s)	10 seconds
Pump speed	High speed

Shows the parameter of ICP-AES used in the analysis

Table 7: Mean concentrations and standard deviations per 100g of fresh food samples.

	Cu	Fe	Zn	Se	Ca	K	Mg	Na
X1	4.4 ± 0.1	74.5 ± 5.4	134.8 ± 2	1.2 ± 0.1	37 ± 0.4	47 ± 0.7	4 ± 0	31 ± 0.1
X2	19.0 ± 1.2	141.7 ± 2.6	111.7 ± 2.9	1.2 ± 0.3	16 ± 0.3	193 ± 5.2	10 ± 0.2	18 ± 0.5
X3	17.1 ± 1.7	106.9 ± 7	64.2 ± 1.6	0.7 ± 0.2	8 ± 0.1	84 ± 2.3	5 ± 0.1	8 ± 0.3
X4	6.7 ± 0.3	58.2 ± 0.6	49.0 ± 0.6	0.4 ± 0.2	4 ± 0.1	47 ± 0.5	3 ± 0	6 ± 0.2

Suggests that levels of copper were highest in Grandma's Sunday lunch (X2) at $19.0 \pm 1.2 \mu\text{g}/100\text{g}$. Low standard deviations values show a high precision in the measurement of the nutrient content.

Table 8: Total daily intake of trace elements and minerals based on the gastric capacity of eight month old^d for a chicken and vegetable diet.

Meal	Infant formula		Lunch (Chicken based, X2)			Dinner (vegetable based X1)			Total daily intake	RNI mg/day
Amount	100ml	600ml ^a	100g	SD	124.5g ^b	100g	SD	124.5g ^c	a+b+c	
Fe	1.2	7.2	0.1	0.02	0.18	0.07	0.04	0.09	7.5	7.8
Zn	0.8	4.8	0.1	0.02	0.14	0.13	0.01	0.2	5.1	5
Ca	50	300	16	1.7	20	37	2.7	46.6	366	525
K	70	420	193	32.7	241	47	4.9	58.5	719	700
Mg	6.4	38.4	10	1.1	13	4	0.2	4.4	56	75
Na	16	96	18	2.9	22	31	0.7	38.8	157	320
Cu	0.03	0.18	0.019	0.008	0.024	0.004	0.001	0.01	0.2	0.3
Se*	1.4	8.4	1.2	2.0	1.5	1.20	2.0	1.5	11.4	10

Reference Nutrient Intake values (RNI) and formula milk concentrations adopted from (Zand, et al. (2012)

^aVolume of milk recommended for 6-9 months infant

^bDaily consumption for lunch

^cDaily Consumption for supper

^dGastric capacity of eight month old: Average weight of about 8.3kg for a 6-9 month infant (30g/kg body weight that was divided by two for dinner and lunch (30x8.3)/2 giving 124.5g)

Se* concentration is in µg/100g

Table 9: Total daily intake of trace elements and minerals based on the gastric capacity of eight month old^d for a beef and vegetable diet.

Meal	Infant formula		Lunch (Beef based, X4)			Dinner (Vegetable based X1)			Total daily intake	RNI (mg/day)
Amount	100ml	600ml ^a	100g	SD	124.5g	100g	SD	124.5g		
Fe	1.2	7.2	0.1	0.02	0.07	0.07	0.04	0.09	7.4	7.8
Zn	0.8	4.8	0.05	0.02	0.06	0.13	0.01	0.2	5.0	5.0
Ca	50	300	4.0	1.7	5.0	37.0	2.7	46.6	352	525
K	70	420	47.0	32.7	58	47.0	4.9	58.5	536	700
Mg	6.4	38.4	3.0	1.1	4.0	4.0	0.2	4.4	46.0	75.0
Na	16	96.0	6.0	2.9	8.0	31.0	0.7	38.8	143	320
Cu	0.03	0.18	0.01	0.008	0.008	0.004	0.001	0.01	0.2	0.3
Se*	1.4	8.4	0.4	2.000	0.498	1.20	2.000	1.5	10.4	10.0

Reference Nutrient Intake values (RNI) and formula milk concentrations adopted from (Zand, et al. (2012)

^aVolume of milk recommended for 6-9 months infant

^bDaily consumption for lunch

^cDaily Consumption for supper

^dGastric capacity of eight month old: Average weight of about 8.3kg for a 6-9 month infant (30g/kg body weight that was divided by two for dinner and lunch (30x8.3)/2 giving 124.5g)

Se* concentration is in µg/100g

Generally, based on the results of the hypothesised infant feeding regime, the average daily consumption of calcium from the ready to eat foods was 159 mg/day lower than the RNI (525 mg/day). These results correspond with those obtained in the study by [7] where a meat and vegetable regime were compared and calcium levels were lower. Similarly, the total daily intake of copper was lower by 0.1 mg/day, iron by 0.3 mg/day; magnesium by 19 mg/day and sodium by 163mg/day. The rest of the elements suggested excessive daily intakes of 19 mg/day, 1.4µg/day and 0.1 mg/day in potassium, selenium and zinc respectively.

Given that iron and zinc are known to have poor bioavailability

as a result of some inhibitors such as calcium, phytate and tannin [7], then the levels of iron are considerably low. If a moderate bioavailability of 10% of iron is put into account, the dietary reference intakes recommended are 11.0 mg/day [8] as opposed to 7.8 mg/day. Phytates are the main stores of phosphorous in plants. The phytates bind to iron, manganese, zinc, and to a small extent, calcium slowing down their absorption. The main sources of phytates are whole grains such as rice and wheat [9].

In a similar manner, considering that the upper limits for zinc intake are 5.00 mg/day while the adequate intakes are suggested as 3.00 mg/day [8], the quantities of zinc are slightly higher. Nevertheless, the tolerable upper limit is placed at 7.00 mg/day

for 1-3 year olds (Scientific Committee for Food, (2006) adopted from Pandelova, et al. (2012) [2] implying that the quantities are within this level. Nonetheless, it worth noting that excessive intake of the zinc may diminish the intake of other elements such as [10] and this can have adverse effects such as anaemia in infants since copper is essential in the formation of haemoglobin.

Table 9 below gives the diet using a beef and vegetable based food regime. The amount of zinc in the food regime consisting of beef and vegetables suggests that the levels were equal to the RNI (5.00 mg/day). All the other trace elements and minerals were below the RNI except selenium whose values were higher by 0.4 µg/100g. Selenium levels were higher, with more contribution from the infant formula milk and the vegetable based at 8.4 and 1.5µg/day respectively.

CONCLUSION

Based on the hypothesised infant feeding regime of a chicken and vegetable based diet that includes formula milk, lunch and dinner for 8 months infants, the average daily consumption of calcium from the ready to eat foods was 159 mg/day lower than the RNI (525 mg/day). Similarly, the total daily intake of copper was lower by 0.1 mg/day, iron by 0.3 mg/day; magnesium by 19 mg/day and sodium by 163mg/day. The other elements suggested excessive daily intakes of 19 mg/day, 1.4µg/day and 0.1 mg/day in potassium, selenium and zinc respectively. Nevertheless potassium and selenium were within the Tolerable upper Limits of 800 mg/day and 60µg/day for 6-12 months old infants. Considering that the RNI for zinc intake is 5.00 mg/day while the adequate intake is suggested as 3.00 mg/day [8] the quantity of zinc in foods was slightly higher. Nevertheless, the Tolerable Upper Limit is placed at 7.00 mg/day for 1-3 year olds.

Given the rapid growth of infants early in life and requirement for more energy leading to a higher rate of food consumption relative to their body weight, infants are more susceptible to consuming higher than recommended nutritional values. In light of these findings it is recommended that the manufacturers include information on the levels of the trace elements and minerals on the labels. This will help in determining the portions of foods to give the infants depending on the diet. A further analysis to determine the total daily intake considering a diet using breast milk instead of formula milk is recommended.

APPRECIATION

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