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REVIEW

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A safe strategy for addition of vitamins and minerals to foods

Summary Addition of vitamins and minerals to foods must be done without health risk to any consumer group. International expert groups have aimed at establishing tolerable upper intake levels (ULs) for vitamins and minerals although lack of solid data on their safety is a major obstacle to this work. In this paper, we summarize the existing ULs and suggest the use of guidance levels (GLs) set by others and temporary guidance levels (TGLs) proposed here, whenever no consensus UL has been established for adults. We suggest the use of body surface area ratios to establish similar levels for younger age groups. The levels are applied in a model for calculation of safe fortification levels for all ages. We have estimated the upper 95th percentile intake of vitamins and minerals from food in various Danish age and gender groups and suggest that a daily multivitamin-mineral pill is included in the calculation of total dietary intake levels of all vitamins and minerals. By subtracting this dietary intake level from the UL, GL or TGL, we calculate the amount that can be safely used for fortification. Since safety must be assured for all age groups, the smallest difference relative to energy intake calculated for any age group is proposed as the maximal allowance (MA) for fortification with each nutrient. We suggest that the MA should be expressed in weight units per energy unit in order to distribute it equally between potentially fortifiable food groups according to their usual contribution to total energy intakes.

Key words vitamins – minerals – micronutrients – fortification – tolerable upper intake level – food

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Introduction

Regulation of fortification of foods with vitamins and minerals is necessary in order to ensure the safety of consumers in the European Union (EU). For several micronutrients there is a relatively low margin between the adequate nutritional level of intake and maximum safe intake. Therefore, a model is needed to predict the impact of fortified foods on the total intake of micronutrients in the general population in order to avoid the risk of excessive intakes in vulnerable sub-populations.

Flynn et al. have recently published a model for the safe addition of vitamins and minerals to foods [1]. This model is based on the tolerable upper intake level (UL) established by the EC Scientific Committee on Food (SCF) combined with average European 95th percentile intakes of micronutrients from non-fortified foods. Flynn's model is derived from adult intake data and ULs established for adults. However, the ULs established by the SCF are graduated into specific values for children and adolescents due to their increased intake of foods on a body weight basis as compared to adults. The knowledge of adverse effects by exaggerated micronutrient intake in these age groups is very limited and some nutrients are known not only to be highly essential for growth and healthy development, but also to be severely toxic when taken in too high doses. Examples are iron, calcium, vitamin D, vitamin A and vitamin B_6. Children and teenagers are often the target consumers for fortified food products, e. g., cereals, soft drinks, juices and candy, and their daily intakes of these food products can be

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substantial. This calls for special attention to this particular group to avoid the risk of adverse effects of micronutrients from fortified foods. Flynn et al. additionally assume that supplement users represent only an insignificant minority of the population [1]. However, recent Danish dietary surveys indicate that about 45 % of the adult Danish population and 64 % of the children regularly take vitamin and mineral supplements [2, 3]. The more prevalent use of supplements among children and adolescents increases their risk of exceeding the tolerable upper intake levels. The objective of the present paper is to further develop and extend the model previously proposed by Flynn et al. [1], by considering the common use of micronutrient supplements and by introducing age-differentiated ULs and thereby focusing in particular on the safety for children of all ages. Furthermore, we propose temporary guidance levels for a range of micronutrients for which no levels have been published by expert panels.

Elements of the model

The model used in the present paper is based on the model by Flynn et al. [1]. It employs three main elements, the ULs, the 95th percentile intake of micronutrients from non-fortified foods (CI_{95}) and supplements (SI), and the resulting maximal allowance (MA) for fortification. By using these factors, it is possible to estimate the level of each micronutrient that can be added to foods without any appreciable risk of adverse effects for any age group in the population, including individuals with high food intakes. The model is based on the following factors and mathematical formulae: (I) UL: the tolerable upper intake level established by the SCF or other expert committees. Where no UL has been established, guidance levels (GLs) suggested by the UK Expert Group on Vitamins and Minerals (EVM) or temporary guidance levels (TGLs) suggested here are used instead; (II) CI_{95}: the current 95th percentile dietary intakes of micronutrients from non-fortified foods; (III) SI: supplement intake; daily micronutrient intake from a normal vitamin/mineral supplement; (IV) MA: maximal allowance for intake of micronutrients from fortified foods: $MA = UL - (CI_{95} + SI)$; (V) EI_{95}: the 95th percentile energy intake; (VI) PFF_n: the fraction of foods in the market which is available for fortification; and (VII) ALA: Acceptable level of addition with each nutrient per 100 kcal portion of the food.

Each element in this model will be described in detail in the following sections.

$$ALA = \frac{MA}{EI_{95} \times PFF_n}$$

Tolerable upper intake level (UL)

The UL is defined as the maximum level of total chronic daily intake of a nutrient from all sources that is unlikely to create a risk of adverse health effects to humans [4]. The SCF has evaluated and published reports on ULs for 22 different vitamins and minerals. Recently, the Scientific Panel on Dietetic Products, Nutrition and Allergies (NDA Panel) under the European Food Safety Authorities (EFSA) has continued this work and has so far given four opinions on ULs. Other expert groups under the FAO/WHO Joint Expert Group on Food Additives (JECFA), the US Institute of Medicine (IOM), WHO/FAO, the EVM and the Nordic Council of Ministers have also published reports dealing with the upper safe levels or guidance levels for intake of vitamins and minerals. Our model for safe fortification of foods with micronutrients is primarily based on the ULs established by the SCF or the NDA Panel, and, where no opinions have yet been provided or where no UL was established for the EU, we use upper safe levels from other European expert committees or from the US IOM. These may be used until European consensus on ULs for these micronutrients has been achieved. In cases where no UL or GL has been proposed, we suggest using a temporary guidance level (TGL) until more knowledge about the safety of these micronutrients has been generated. Some vitamins and minerals that need special attention regarding their safety of use in fortified foods are briefly discussed below.

Vitamins and minerals with a UL specified by the SCF or the NDA

The SCF has currently established ULs for the following micronutrients: vitamin A, vitamin D, vitamin E, niacin, vitamin B_6, folic acid, calcium, magnesium, zinc, copper, iodine, and selenium. The NDA Panel has so far only established a UL for boron (see Table 1).

In general, the database on adverse effects of nutrients in children and adolescents is very limited. Therefore, in the absence of adequate data, the SCF in most cases has chosen to correct for differences in basal metabolic ratio by extrapolating the UL from adults to children on a body surface area basis (body weight$^{0.75}$). In a few instances, body weight (bw) alone was used in the reports by the SCF.

Some of the opinions on ULs published by the SCF were not available when Flynn et al. published their model [1], and several of the more recent SCF ULs are lower than those suggested by the US IOM [5, 6] that were used by Flynn et al. in their model [1]. Examples are the ULs for vitamin E, copper, iodine and zinc, where the ULs suggested by the US IOM were 2 to 3-fold higher than those later set by the SCF.

Table 1 Tolerable upper levels of vitamins and minerals for different age groups, established by the SCF

Micronutrient	UL[e]	Age 1–3 years	Age 4–6 years	Age 7–10 years	Age 11–14 years	Age 15–17 years	Adults	Ref.
Vitamin A (µg)	SCF	800	1100	1500	2000	2600	3000[g]	[60]
Vitamin D (µg)	SCF	25	25	25	50	50	50	[61]
Vitamin E (mg)	SCF	100	120	160	220	260	300[h]	[62]
Niacin (mg)[a]	SCF	150	220	350	500	700	900[f]	[63]
Vitamin B_6 (mg)	SCF	5	7	10	15	20	25	[7]
Folic acid (µg)[b]	SCF	200	300	400	600	800	1000	[64]
Calcium (mg)[c]	SCF	2500[c]	2500	2500	2500	2500	2500	[65]
Magnesium (mg)[d]	SCF	65[d]	250	250	250	250	250	[41, 66]
Zinc (mg)	SCF	7	10	13	18	22	25	[67]
Copper (mg)	SCF	1	2	3	4	4	5[f]	[68]
Iodine (µg)	SCF	200	250	300	450	500	600[i]	[69]
Selenium (µg)	SCF	60	90	130	200	250	300	[70]

[a] The ULs only apply for nicotinamide; [b] Does not include dietary folate from natural sources; [c] The SCF does not recommend an age-differentiation of the UL for calcium; [d] The UL only applies for magnesium supplements and for magnesium present in water. No UL was established by the SCF in 2001 for children under 4 years due to insufficient data. The UL for this age group was established by the US Institute of Medicine in 2003 [41]; [e] The EC SCF tolerable upper intake level; [f] The UL for adults does not apply during pregnancy or lactation; [g] The UL for adults does not apply for postmenopausal women, here the SCF recommends a maximum intake of 1500µg; [h] The UL does not apply for subjects with blood coagulation defects caused by vitamin K deficiency or by malabsorption or due to therapy with anticoagulants; [i] The UL established for iodine does not apply to populations with iodine deficiency disorders

Guidance levels (GLs) and temporary guidance levels (TGLs) for vitamins and minerals without a UL

For a number of micronutrients, there is a severe lack of adequate human toxicity data and the SCF and the NDA Panel therefore conclude that they are unable to set a UL for these nutrients. As an alternative, we have substituted with guidance levels (GLs) and temporary guidance levels (TGLs) in our model and suggest their use until sufficient data emerge to establish ULs (Table 2). A discussion of the basis for the GLs or TGLs used for these nutrients is given in the following sections. National expert committees in Europe or expert committees in the US have suggested ULs for some of these nutrients and, in most cases, we suggest using these until commonly agreed ULs have been established by the NDA Panel. For the remaining nutrients, where none of the expert committees have proposed a UL, there is ample human experience that the toxicity is low although there is no solid scientific evidence for it. To include these micronutrients in our model, we have established TGLs for their safe upper intake, based on a safety evaluation of the limited data available in humans. Furthermore, we suggest that the ULs, GLs and TGLs are extrapolated to children on a surface area basis ($bw^{0.75}$) due to differences in metabolic ratio in accordance with the practice of the SCF for their ULs (see Table 2) [7].

Temporary guidance level for upper safe intake of iron. Humans have no active excretion mechanism for iron and excessive iron intake can cause iron overload. Symptoms of acute iron overload are nausea, diarrhea, gastrointestinal bleeding and – with more severe intoxication – shock, coma or death. Hereditary hemochromatosis is due to a mutation in the hereditary hemochromatosis gene (HFE) and is the most common autosomal recessive genetic disease in Caucasians. This genetic disorder causes iron overload in homozygotes and originates from the Northern part of Europe [8]. In Denmark, 0.7 % are homozygotes and between 10 and 15 % of the Scandinavian populations are heterozygotes [9, 10]. Also the Netherlands, France and Ireland have a high prevalence of homozygosity (0.4–1.2 %) and heterozygosity (10–17.3 %) [9]. About 90 % of hemochromatosis cases are undiagnosed [10]. In heterozygous individuals, iron absorption and saturation of iron transfer is slightly increased compared to normal, but, with a normal diet without iron supplementation, this is unlikely to lead to iron overload [10]. Epidemiological studies indicate that iron overload is relatively common in Western countries, whereas the prevalence of iron deficiency has declined drastically in the developed countries during the past 15–20 years [9, 11]. Danish population-based studies have shown that 19 % of men (40–70 years) and 5.5 % of postmenopausal women have iron overload [11, 12]. Recent evidence pointing to iron-related cardiovascular hazards adds to the safety concern of increasing the iron intake for a large part of the population although data are somewhat conflicting [13–19].

The NDA Panel recently evaluated the UL for iron [20]. The Panel could not establish a UL for iron intake due to inconsistent data on the different iron sources (haem and non-haem iron), and due to poor correlations between iron intake, biochemical indicators of iron

Table 2 Upper tolerable intake level (UL), Guidance levels (GLs) or temporary guidance levels (TGLs) for intake of vitamins and minerals, for which no UL has been established by the SCF[a]

Micronutrient	UL/GL/TGL[b]	Age 1–3 years	Age 4–6 years	Age 7–10 years	Age 11–14 years	Age 15–17 years	Adult men	Adult women	Ref.
β-carotene (mg)	TGL, Present paper[b]	5	5	5	5	5	5	5	Present paper[b]
Vitamin K (μg)	GL,EMV[c]	270	370	500	670	870	1000	1000	[21]
Thiamin (mg)	TGL, Present paper[b]	15	20	25	34	42	50	50	Present paper[b]
Riboflavin (mg)	GL, EMV[c]	12	16	22	29	37	43	43	[21]
Vitamin B_{12} (μg)	GL, EMV[c]	530	730	1000	1330	1730	2000	2000	[21]
Pantothenic acid (mg)	GL, EMV[c]	55	75	100	135	175	200	200	[21]
Biotin (μg)	GL, EMV[c]	270	370	500	670	870	1000	1000	[21]
Vitamin C (mg)	TGL, NDA[b,d]	270	370	500	670	870	1000	1000	[42], Present paper[b]
Phosphorus (mg)	UL, US[e]	3000	3000	3000	4000	4000	4000	3500[g]	[41]
Iron (mg)	TGL, JECFA[f]	10	14	20	30	40	50	50	[22]

[a] The EC SCF and other international expert committees may have evaluated these micronutrients, although no tolerable upper intake level (UL) could be established. GLs are obtained from other expert panels (see specific references for each micronutrient), and TGLs are suggested by the authors of this paper; [b] The basis for establishing the TGL is discussed in the text; [c] The GL established by EMV is used; [d] No UL was established by the NDA, but the TGL is based on the statement that 'supplemental daily doses of vitamin C up to about 1 g, in addition to normal dietary intakes, are not associated with adverse gastrointestinal effects' [42]; [e] The US Institute of Medicine established a UL for phosphorus in 2003; [f] The PMTDI at 50 mg/day established by JECFA is used to derive TGLs for children of all ages. For consistency, the bodyweight groups used by the SCF (see e. g., [7]) are also used for extrapolation for iron; [g] The UL for women is set for pregnant women [41]

status, and adverse effects, and, finally, due to uncertainty of the association between iron intake and chronic disease. The Panel pointed out that subjects at risk for iron overload 'should avoid iron-supplements and highly iron-fortified foods'. The US IOM has set a UL for iron intake at 45 mg/day (equivalent to 0.74 mg/kg bw/day) [5]. This value was based on the acute adverse effects observed in the gastrointestinal tract (GI) after intake of iron supplements [5]. Risk of chronic disease was not taken into account and it was stated that the proposed UL does not protect individuals with diseases resulting in iron-loading abnormalities. The US IOM also finds it 'prudent to recommend that men and postmenopausal women avoid iron supplements and highly fortified foods' [5]. The EVM also concluded that data are insufficient to establish a UL for iron intake, but suggest a GL of maximally 17 mg/day supplemental iron intake (equivalent to 0.28 mg/kg bw/day) [21]. They stress that this level does not apply to individuals with increased susceptibility to iron overload [21]. A provisional maximum tolerable daily intake (PMTDI) was established at 0.8 mg/kg bw by JECFA in 2003 based on findings indicating that the intake of supplements at 50 mg/day (ferrous iron) for long periods of time does not give rise to adverse effects in normal adult individuals [22]. Taking the JECFA evaluation as the basis, we suggest that a TGL is set at 50 mg/day for adults and is moderated for children and adolescents according to the PMTDI of 0.8 mg/kg bw as suggested by JECFA (see Table 2).

The Nordic Council of Ministers has recently reevaluated the Nordic Nutrition Recommendations (NNR) and now recommends a UL for iron at 25 mg/day for adults [23]. This UL is established as a maximum additional intake of 10 mg/day supplemental non-haeme iron to the habitual dietary iron giving a total intake of 25 mg/day for adults. This UL is based on the Framingham study, which has indicated that the homeostatic regulation of iron absorption is only able to prevent iron overload at intake levels up to 25 mg/day [23, 24]. Despite the more recently established Nordic UL at almost half of the level suggested by the US IOM and JECFA [5, 22], we suggest using the PMTDI at 50 mg/day as TGL for adults until a wider international consensus has been reached.

Food fortification with iron will render individuals with hemochromatosis unprotected and may lead to iron intakes in excess of the TGL for the youngest children. Along with other European experts [9, 10, 23, 25], we therefore recommend avoiding fortification of food with iron in order to protect all potential risk groups.

Temporary guidance level for upper safe intake of thiamin. Thiamin (vitamin B_1) is well absorbed in humans by an active, rate limited jejunal uptake mechanism [26]. At higher levels of intake, these vitamins are absorbed much less efficiently by passive diffusion [27]. The utilization of thiamin in the body depends on the carbohydrate metabolism; thus, increased physical activity, pregnancy and lactation increase thiamin re-

quirements because of greater energy needs. The recommended intake is set at 0.12 mg per MJ leading to average daily intake of around 1.1 to 1.5 mg [23, 28]. Thiamin has been utilized in large pharmacological doses for several decades and there have been no reports on adverse effects of oral thiamin, even at dosages of several hundred milligrams a day [29]. Adverse effects have only been reported in humans after parenteral administration of thiamin [29, 30]. Due to the lack of systematic oral dose-response studies as well as the low toxicity of oral thiamin in humans, the SCF concludes that no lowest-observed adverse effect level (LOAEL) and no-observed adverse effect level (NOAEL) can be established. The Dutch Nutrition Council reached the conclusion that 500 mg thiamin per person was a tolerable upper intake level [31]. The EVM also concludes that no UL can be established due to insufficient data for a dose-toxicity relationship, but suggests as a GL that a supplemental intake of 100 mg/day of water-soluble forms of thiamin is not suspected to cause any adverse effects. Flynn et al. have set the UL for thiamin intake at 50 mg/day and refer this value to the report from the US IOM [1]. However, IOM also concludes that no NOAEL or UL can be established, and note that 'supplements that contain up to 50 mg/day of thiamin are widely available without prescription, but effects of this level or more of intake do not appear to have been studied systematically' [6]. They additionally stress that since 'data on the adverse effects of thiamin intake are extremely limited, caution may be warranted.'

Despite the deficiencies in the database on adverse effects of thiamin intakes in humans, especially during development, we suggest using the level of 50 mg/day as TGL until more data have emerged, since no adverse effects have been reported at this level of supplementation. This value should be extrapolated to children based on the differences in metabolic ratio ($bw^{0.75}$) as seen in Table 2.

Guidance level for upper safe intake of riboflavin, vitamin B_{12}, pantothenic acid, and biotin. These four water-soluble vitamins all have in common that their toxicity is low. There are no published data from studies in experimental animals or humans that connect oral intake of riboflavin, vitamin B_{12}, pantothenic acid or biotin with genotoxic, carcinogenic, teratogenic, or reproductive effects or with toxic effects in humans [32, 33]. At physiological concentrations, the uptake of these vitamins occurs by active, saturable transport systems. At higher levels of intake, these vitamins are absorbed much less efficiently by passive diffusion [32, 33].

Biotin

The animal and human toxicity studies on biotin are very limited, and systematic oral intake dose-response studies of biotin in humans are totally lacking [33]. Therefore, the SCF has concluded that it is not possible to derive a numerical UL for biotin [33]. Although Flynn et al. refer the UL of 2.5 mg/day for biotin to the US IOM, they also conclude in their report that 'there are not sufficient data on which to base a tolerable upper intake level (UL) for biotin' [6]. The EVM has established a GL for supplemental intake of biotin [21]. Based on a study where 9 mg biotin were given daily to 20 non-insulin dependent diabetes patients for up to 4 years, the EVM suggested an uncertainty factor of 10, due to the low number of subjects in the study. In addition to the maximum intake from food of 0.07 mg, this gives an overall GL of 0.97 mg/day [21]. We therefore suggest using this GL of 1 mg/day until more studies on effects of biotin intakes are provided. The GL should be extrapolated to children based on $bw^{0.75}$ (Table 2).

Vitamin B_{12}

Vitamin B_{12} has a long history of safe use as a therapeutic agent given in high dosages for treatment of disorders associated with vitamin B_{12} malabsorption, for example, pernicious anemia [34]. In vitamin B_{12} replacement therapy, oral or intramuscular dosages of between 1000 and 5000 µg vitamin B_{12} have been used for many years, with no evidence of adverse effects. No systematic toxicological studies have been reported for vitamin B_{12}.

The SCF concluded that there are no clearly defined adverse effects produced by vitamin B_{12} and, thus, no basis for deriving a UL [34]. The EVM suggested an upper intake guidance level of 2000 µg/day, based on the clinical experience with this vitamin [21]. We therefore suggest using this GL for vitamin B_{12} for adults. The GLs for children are derived by extrapolating on a $bw^{0.75}$ basis (Table 2).

Pantothenic acid

Pantothenic acid has low toxicity in humans. There are no reports on toxicity of pantothenic acid or panthenol in humans. In therapeutic trials, pantothenic acid has been given to patients in gram doses without any reported side-effects [35]. Owing to the lack of systematic oral dose-response intake studies and the very low toxicity of pantothenic acid (calcium pantothenate or panthenol), the SCF concludes that no LOAEL and NOAEL can be established and no numerical UL can be derived [35].

The EVM has suggested the use of 200 mg/day as GL for the upper intake level of pantothenic acid [21]. This was based on experience from a randomized double-blind, placebo-controlled study (n = 47) where a dose of 2000 mg/day was used [36]. The EVM applied an uncertainty factor of 10 to the dose to allow for inter-individual variability due to the small number of subjects, and to the general incomplete data on hazardous effects of riboflavin in humans [21]. We suggest using this GL un-

til more data are provided and extrapolating this value to children based on $bw^{0.75}$ basis (Table 2).

Riboflavin

In a study with 55 migraine patients treated with 400 mg/day of riboflavin (vitamin B_2) (or placebo) for 3 months, mild, self-reported, adverse effects were described by Schönen et al. [37]. No adverse effects were reported by Zempleni et al. in a study with oral or i. v. doses up to 60 mg in healthy adults [38]. In a 13-week oral feeding study with riboflavin, a 6 % growth retardation was found in female rats given 200 mg/kg bw/day [39]. Based on this study, the SCF has set the NOAEL for riboflavin at 50 mg/kg bw [32]. However, as a result of insufficient data on adverse effects in humans, the SCF was unable to set a UL for riboflavin intake. They stress that, although no studies have reported significant adverse effects in humans, 'this does not mean that there is no potential for adverse effects from high intakes.' Based on the study by Schönen et al. [37], the EVM established a GL for supplemental intake of riboflavin [21]. To allow for inter-human variability and for general incomplete data on hazardous effects in humans, an uncertainty factor of 10 was applied [21]. In addition to the maximum intake from the diet (Table 3), this gives a total intake of 43.3 mg/day. Until more data on adverse effects in humans have emerged, we suggest using this GL and extrapolating it to children based on body surface area (Table 2).

Guidance level for upper safe intake of vitamin K. Vitamin K is a lipid soluble vitamin. The current consideration of a tolerable upper level for vitamin K is focused on phylloquinone, the predominant dietary source. The SCF concludes in their opinion on vitamin K that, in the limited numbers of human studies, 'there is no evidence of adverse effects associated with supplementary intakes of vitamin K in the form of phylloquinone of up to 10 mg/day (more than two orders of magnitude higher than the recommended dietary intake of vitamin K) for limited periods of time.' The SCF is, therefore, not able to establish a UL for vitamin K intake. The same conclusion was reached by the US IOM and by the EVM [5, 21]. The EVM has, however, established a GL for safe upper intake by applying an uncertainty factor of 10 to the dose of 10 mg/day that has been investigated in a small number of subjects (n = 8) [40] resulting in a GL of 1 mg/day that is unlikely to result in adverse effects [21]. We suggest using this GL until further knowledge on potential adverse effects of vitamin K is gathered. We also suggest differentiating this GL to other age groups on a $bw^{0.75}$ basis (see Table 2).

Upper tolerable intake level for phosphorus. The tolerable upper intake level for phosphorus has not been evaluated by the SCF or NDA, but the US IOM has established a UL for intake of phosphorus [41]. Since information is lacking about potential adverse effects of phosphorus up to the intake level associated with ectopic mineralization, the UL was based on the level where the upper boundary of normal serum values of phosphorus is reached. The UL was set to 4000 mg/day for adults. As seen in Table 2, the US IOM recommends a somewhat lower UL for the youngest children to account for potentially increased susceptibility due to the smaller body size. During pregnancy, absorption efficiency for phosphorus rises by about 15 % and, thus, the UL is set to a 15 % lower value for pregnant women, giving a UL of 3500 mg/day [41]. We suggest using these levels proposed by the US IOM as ULs until the NDA panel has reached a conclusion for a UL for phosphorus.

Temporary guidance level for upper safe intake of vitamin C. In the opinion issued by the NDA Panel, no conclusion regarding a UL for vitamin C is reached due to insufficient data [42]. However, it is stated that 'supplemental daily doses of vitamin C up to about 1 g, in addition to normal dietary intakes, are not associated with adverse gastrointestinal effects.' The NNR has set the UL for vitamin C to 1 g/day for adults, since higher doses may be associated with diarrhea, other gastrointestinal disturbances, increased oxalate formation and kidney stones in susceptible individuals [23]. A guidance level was not estimated by EMV since all data on adverse effects of vitamin C are related to bolus doses as opposed to doses from food, but they state that 1 g/day appears to be safe [21]. The US IOM has set the UL to 2 g/day for adults based on a LOAEL of 3 g/day for osmotic diarrhea in adults [43]. Based on relative body weights, they moderate this level to children down to 1 year [43]. Based on these evaluations, we suggest using 1 g/day as TGL for adults, and relatively less for children based on their higher metabolic ratio (see Table 2). We urge that further research should be done to evaluate the safety of chronic intakes of vitamin C at this dose level in potentially vulnerable groups, especially in combination with high iron intakes.

Temporary guidance level for upper safe intake of β-carotene. In their opinion, the SCF states that 'existing evidence from human trials indicates that supplemental β-carotene (20 mg/day or more) is contraindicated for use in current, heavy smokers.' However, they find the scientific basis insufficient to set a UL for β-carotene [44]. Although the adverse effects of β-carotene on human lung cancer risk have only been observed in groups at elevated risk for lung cancer [45, 46], the studies available with cohorts at lower risk have insufficient power to exclude that adverse effects of high doses of β-carotene may also affect others [47]. Animal studies in ferrets indicate that β-carotene *per se* affects cell differentiation and proliferation in the bronchus and that sub-

Table 3 95th percentile energy intake and intake of vitamins and minerals from food intake and supplements in Denmark

EI_{95}[a]	Age 1–3 years		Age 4–6 years		Age 7–10 years		Age 11–14 years		Age 15–17 years		Adult men		Adult women	
n	278		232		340		251		146[b]		272		315	
Kcal/day	2200		2500		2800		3300		3200		3800		2900	
MJ/day	9.2		10.5		11.6		13.6		13.4		15.7		12.1	
Micronutrient	CI_{95}[a]	Suppl.	CI_{95}[a]	Suppl.	CI_{95}[a]	Suppl.	CI_{95}[a]	Suppl.	CI_{95}[a]	Suppl.	CI_{95}[a]	Suppl.	CI_{95}[a]	Suppl.
Vitamin A (µg)	1100	400	1360	400	1190	400	1170	800	1100	800	1780	800	1220	800
β-carotene (µg)	4900	–	9000	–	10400	–	9200	–	7900	–	9300	–	11000	–
Vitamin D (µg)	3.0	10	3.8	10	5.2	10	4.9	5	6.4	5	7.2	5	7.2	5
Vitamin E (mg)	8.3	5	10	5	11	5	12	10	10	10	12	10	10	10
Vitamin K (µg)[c]	80	30	90	30	95	30	110	90	115	90	145	90	140	90
Thiamin (mg)	1.3	0.7	1.6	0.7	1.7	0.7	1.9	1.4	1.9	1.4	2.2	1.4	1.7	1.4
Riboflavin (mg)	2.3	0.8	2.3	0.8	2.7	0.8	2.8	1.6	2.9	1.6	2.9	1.6	2.5	1.6
Niacin (mg)[d]	10	9	12	9.0	15	9	16	18	17	18	26	18	19	18
Vitamin B_6 (mg)	1.5	0.8	1.6	0.8	1.8	0.8	2.0	2	2.0	2	2.4	2	1.9	2
Folic acid (µg)[e]	270	75	370	75	440	75	450	200	450	200	500	200	460	400[b]
Vitamin B_{12} (µg)	6.6	1	8	1	8	1	8	1	8	1	10	1	8	1
Pantothenic acid (mg)	5	2	6	2	7	2	7	6	7	6	8	6	6	6
Biotin (µg)	35	8	40	8	45	8	45	150	45	150	50	150	40	150
Vitamin C (mg)	105	40	160	40	200	40	220	60	230	60	200	60	210	60
Calcium (mg)	1400	200	1440	200	1730	200	1940	320	2010	320	1800	320	1770	320
Phosphorus (mg)	1570	–	1690	–	1970	–	2180	–	2240	–	2360	–	1940	–
Magnesium (mg)[f]	20	85	20	85	30	85	30	300	40	300	40	300	40	300
Iron (mg)	8.9	8	11	8	12	8	14	14	14	14	17	14	13	14
Zinc (mg)	10	5	12	5	14	5	16	15	17	15	18	15	14	15
Copper (mg)[c]	3	0.34	3.5	0.34	4.6	0.34	5.4	0.9	6.1	0.9	9	0.9	9	0.9
Iodine (µg)[g]	91	70	260	70	290	70	330	150	340	150	340	150	280	150
Selenium (µg)	35	20	46	20	53	20	57	40	57	40	70	40	50	40

[a] *EI_{95}* 95 % energy intake based on Danish dietary surveys [49, 50]; *CI_{95}* the 95th percentile dietary intakes of micronutrients from non-fortified foods; *SI* Supplement, the daily intake of micronutrients from a normal vitamin/mineral supplement; [b] 60 % of the sample were girls, which may explain the relatively low energy intake; [c] Intake estimates are less precise due to inadequate food database; [d] The data apply only for nicotinamide; [e] Dietary folate from natural sources are not included in the calculation of MA (see Table 4). The recommendation of 400µg/day of folic acid for women of child-bearing age is used for women: [f] Only intake from magnesium supplements and water are included in the calculation of MA (see Table 4); [g] Intake data include mandatory iodine fortification of salt

stances in cigarette smoke enhance this effect [48]. Sources of β-carotene in the diet include its natural abundance in many foods, the use of beta-carotene as a colorant and as a pro-vitamin A in supplements. The difference in dose between the 95 % confidence interval for dietary levels (5–9 mg/day) and the adverse intake levels is quite narrow (see Table 3), although differences in bioavailability of β-carotene from foods and from supplements may cause a larger difference in plasma levels. Moreover, the mechanisms behind the adverse effects are not fully understood and may affect population groups and target organs other than those presently known. Based on these uncertainties, we suggest avoiding the use of β-carotene for food fortification in Europe, i. e., setting the TGL for β-carotene equal to the average dietary level of 5 mg/day for all age groups leading to a zero allowance for fortification of food with β-carotene.

Energy and nutrient intakes from diet and supplements

Diet

Estimated dietary intakes of energy, vitamins and minerals for selected groups of the population used in the model are presented in Table 3. The intake estimates are based on Danish nation-wide dietary surveys mainly carried out in the period 2000–2002 [49]. Data were collected across the years in order to cover for seasonal variation in dietary habits. A representative sample of the population (2169 females and 1995 males) aged 4–75 years provided information on their food consumption habits. Since the youngest children were not part of the sample, intake estimates for this group (1–3 years) are from a Danish dietary survey in 1995 (n = 278) [50]. Seven-day prospective food records were used in both surveys as described in Andersen et al. [49, 50]. The result of the food registration was expressed as daily mean intakes for each participant in the survey. Using data at the individual level, it was possible to describe the intake distribution of both foods and nutrients in the population. Calculation of vitamin and mineral intakes included correction for losses during food preparation and cooking. The Danish intake data for iodine include contributions from mandatory iodized salt (13 mg/kg) used in households and for bread production.

Supplements

Since approximately half of the adult population and about two-thirds of the children in Denmark take supplements regularly [2, 3], this nutrient source has to be included in estimates of total vitamin and mineral intakes. About 28 % of children and 26 % of adults in Denmark daily consume a vitamin pill all year around and another 28 % and 12 %, respectively, take vitamin pills daily in the winter season according to the most recent dietary survey covering the years 2000–2002 [2, 3]. The frequency of regular intake has only decreased about 10 % from 1995 to 2002 [2, 3] indicating that a fairly large and stable fraction of the population use multi-vitamin supplements. Also, a recent American survey shows that 34 % of children < 6 years take micronutrient supplements daily or almost daily and that an additional 12 % take supplements regularly [51]. The most common supplement used in Denmark is a combined multi-vitamin-mineral tablet usually containing 100 % of reference values for recommended daily intakes of vitamins and minerals. Two sets of reference values are used in the Danish legislation on dietary supplements; one valid for children aged 1–10 years and one for older children and adults. In this model, the contribution from supplement intake (SI) to the total intake of micronutrients is chosen equal to 100 % of the reference values (Table 3).

Establishment of a safe model for all age groups

Based on the ULs (Table 1), the surrogate GLs and TGLs (Table 2), we can establish a model for safe addition of vitamins and minerals to foods. However, not all foods can or will be fortified. In agreement with Flynn et al. (2003), we estimate that about half of the Danish diet consists of foods which are potentially fortifiable. Among these, not all foods will be fortified. Even consumers who prefer fortified foods will probably not be able to get more than 25 % of their total energy intake from fortified foods. This fraction (PFF_n) should be adjusted according to the future development in market shares from fortified foods. Assuming that 25 % of the energy intake comes from fortified foods, the acceptable levels of addition (ALAs) can be distributed in energy portions: $ALA_{25} = MA/(EI_{95} \times 0.25)$. Table 4 presents the results for the most exposed age group in portions of 100 kcal. This portion size is chosen similar to Flynn et al. (2003).

As an example, the most vulnerable group for vitamin B_6 intake are the youngest children between 1 and 3 years. For this group, the 95th percentile intake (CI_{95}) of vitamin B_6 is 1.5 mg/day, and intake from supplements, 0.8 mg, giving a total intake of 2.3 mg vitamin B_6. The difference up to the UL at 5 mg/day gives the MA = 2.7 mg. EI_{95} is 2200 kcal/day for children aged 1–3 years (Table 3). MA is then divided with $EI_{95} \times 0.25$, giving an ALA_{25} of 0.5 mg vitamin B_6 per 100 kcal. We suggest that for light products an energy density similar to the non-light analog should be applied. In this way, addition to light products will be accepted at the same level as their high-energy analogs.

Table 4 Safe maximum levels of vitamins and minerals that can be added to foods based on the most susceptible risk group

Micronutrient	Most sensitive risk group (years)	UL/GL/TGL[a]	CI_{95}[b]	SI[c]	MA[d]	% Foods fortified (of total available)[e]				
						100 %	50 %	25 %	10 %	5 %
						Acceptable levels of addition (ALAs) per 100 kcal portion (units as in first column)[d]				
Vitamin A (µg)	1–10[j]	–[j]	1360[l]	400	–	0	*0*	0	0	0
β-carotene (mg)	All ages[k]	5[k]	11.0[l]	–	–	0	*0*	0	0	0
Vitamin D (µg)	7–10	25	5.2	10	9.8	1	*1*	3	7	14
Vitamin E (mg)	1–3	100	8.3	5.0	87	8	*16*	32	79	158
Vitamin K (µg)	1–3	270	80	30	160	15	*29*	58	145	291
Thiamin (mg)	1–3	15	1.3	0.7	13	1	*2*	5	12	24
Riboflavin (mg)	1–3	12	2.3	0.8	8.9	1	*2*	3	8	16
Niacin[f] (mg)	1–3	150	10	9	131	12	*24*	48	119	238
Vitamin B_6 (mg)	1–3	5	1.5	0.8	2.7	0.2	*0.5*	1.0	2.0	5.0
Folic acid[g] (µg)	1–3	200	270	75	125	11	*23*	45	114	227
Vitamin B_{12} (µg)	1–3	530	6.6	1	522	47	*95*	190	475	950
Pantothenic acid (mg)	1–3	55	5	2	48	4	*9*	17	44	87
Biotin (µg)	1–3	270	35	8	227	21	*41*	83	206	413
Vitamin C[h] (mg)	1–3	270	105	40	230	21	*42*	84	209	418
Calcium (mg)	15–17	2500	2010	320	170	11	*21*	43	106	213
Phosphorus (mg)	7–10	3000	1970	–	1030	74	*147*	294	736	1471
Magnesium[i] (mg)	1–3 and > 10[j]	–[j]	40[m]	300	–	0	*0*	0	0	0
Iron (mg)	1–10	–[j]	12[l]	8	–	0	*0*	0	0	0
Zinc (mg)	All ages[j]	–[j]	18[l]	15	–	0	*0*	0	0	0
Copper (mg)	All ages[j]	–[j]	9[l]	0.9	–	0	*0*	0	0	0
Iodine (µg)	1–17	–[j]	340[l]	150	–	0	*0*	0	0	0
Selenium (µg)	1–3	60	35	20	5	0	*1*	2	5	9

[a] The UL, GL or TGL for the most sensitive age group is shown (see Tables 1 and 2); [b] *CI_{95}* the current 95th percentile dietary intake of micronutrients from non-fortified foods in Denmark (see Table 3); [c] *SI* Supplement intake; the content of micronutrients in a normal vitamin/mineral supplement used in Denmark (see Table 3); [d] Where no amount is given (–), the MA and the amount available for fortification is either zero or negative; [e] By total available is meant the 50 % of all foods that are assumed to be fortifiable. The highlighted column corresponds to fortification of 25 % of all foods, and is the level suggested for regulating food fortification; [f] The data apply only for nicotinamide; [g] Dietary folate from natural sources is not included in the calculation of MA; [h] The TGL applies only for supplemental vitamin C; [i] For magnesium, only supplements and intake from water are included in the calculation of MA; [j] The 95 % intake from the diet and daily supplement exceeds the UL in several or all age groups, see Tables 1 and 3 for UL and intake data; [k] Fortification with β-carotene is not recommended; [l] The 95th percentile for the age group with the highest intake is given; [m] Estimated intake from water

Discussion

Comparison to the model proposed by Flynn et al.

Our model is essentially similar in design to that previously suggested by Flynn and co-workers [1]. One major difference is the incorporation of the daily intake of vitamin and mineral supplements. This is particularly important for children, who are given supplements more often than adults. Furthermore, most of the marketed fortified food products are targeted at the younger consumers. As examples, 56 % of all fortified foods marketed between 1985 and 2001 in Germany contained added sugar [52] and a food survey conducted in the period 1989 and 1991 in the US showed that children (< 10 years) consumed fortified foods twice as often as adults between 25 and 50 years of age [53].

Another modification in our model is the general use of age-differentiated ULs, GLs and TGLs based on the differences in basal metabolic ratio between children and adults. Most of the ULs established by the SCF are extrapolated to younger age groups (see Table 1). Since there is generally very little knowledge about the safety of elevated intakes of vitamins and minerals in children, it seems prudent to ensure a dose modification based on a general pharmacodynamic principle.

A third difference from the model proposed by Flynn et al. [1] is that we estimate that 50 % of fortifiable foods will be fortified and, hence, that the MA can be distributed among only 25 % of the energy intake. Only a few published studies are concerned with intake of fortified

foods, and most are confined to single food groups or intake of specific nutrients. Godfrey et al. recently estimated the impact of fortified foods in France, Germany, Italy, Spain and the UK [54]. Based on trade statistics and market research data, the average intake of fortified foods per capita was estimated to be around 3 %, except for the UK where mandatory fortification of white flour increases the average intake to 5.5–7 % per capita. However, in this study, food survey data for adults from the UK and France showed that for frequent consumers of fortified beverages (carbonated beverages, juices, fruit drinks and milk), these alone may contribute up to 17 % of the total diet [54]. The authors argue, however, that this is an unrealistically high level, and that consumers are unlikely to obtain more than 4 % of their diet from fortified foods [54]. On the contrary, other recent studies show that the intake of fortified foods is considerably higher than 4 %. These studies additionally demonstrate that children and adolescents are more frequent consumers of fortified foods than adults, and that the intake is generally increasing over time. In a study on zinc intake in 7474 US preschool children (1994–1998), 78 % of the cereals consumed by these children were fortified, and the intake of zinc from fortified foods doubled from 14 % in 1994 to 28 % in 1998. In a study from the period 1989–1991 (n = 11710), Berner et al. have showed that between 70 and 80 % of children < 10 years consumed one or more fortified food during a 3-day dietary survey, whereas only 31–38 % of the adults (25–50 years) consumed fortified foods [53]. In the German DONALD study from the years 1985–2001 the intake of fortified foods was highest in the youngest age groups and the intake of fortified foods in 2 to 3-year-olds contributed with 9.02 ± 7.80 E% (mean ± SD) of the total energy intake [52]. This demonstrates a large range in the amount of fortified foods consumed by children and that some children have a very high intake of fortified foods. Although the number of studies on intake of fortified foods is very limited, a prudent estimate is that no more than 25 % of the total energy intake will be fortified giving room for an expanding market also including high level consumers and children. There is a need for newer consumption data covering all age groups in order to provide a more detailed and updated picture of the trends in the intake of fortified foods.

Underestimations of micronutrient intakes

In order to declare a certain content of a micronutrient in foods, it is often necessary to add excessive amounts to ensure the content throughout shelf-life. In Denmark, it is a currently accepted practice to add up to 150 % of the declared amount. This 'over-fortification' is well known from control analyses of fortified foods and is not a problem as long as no part of the population is at risk of exceeding the ULs. However, in the new situation in Europe, where fortification may become common for many foods, over-fortification may become a problem, since it may lead to underestimation of the actual exposure. An example of the many difficulties in predicting the impact of food fortification is the mandatory folic acid fortification of cereals and grain products in the US. The aim of the fortification program was to increase the intake of folate in the North American population by about 100 μg/day (70–130 μg/day) to limit the occurrence of neural tube defect in newborn babies [55]. Two studies have recently estimated that the actual folic acid consumption is about twice the level intended to be the outcome of the fortification, namely, 190 μg/day and 215–240 μg/day, respectively [56, 57]. This illustrates the difficulties in predicting the impact of food fortification and raises concerns for voluntary food fortification where variations in amounts, types of micronutrients, food brands and consumer patterns will add to the complications in predicting the actual intake of each micronutrient in the population.

The safety of ULs

The ULs reported by the SCF were established in order to evaluate the safety of current intake levels of different micronutrients in Europe. The opinions were not specifically aimed at evaluating the safety of future food fortification and most of the reports only conclude that the current high (97.5th percentile) intake of the individual micronutrients is below the established UL and can, therefore be regarded as safe. The safety of an excessive long-term (life-long) intake of micronutrients at levels close to the UL is, thus, in general not included in the risk assessment due to lack of data. However, by implementing the ULs in the present model or in the model by Flynn et al. [1], they are used as upper safe levels to which the intake can be elevated without any appreciable risk to the population. This may actually be an overinterpretation of the ULs. The safety of long-term daily exposure to elevated doses of multiple vitamins and minerals is still essentially unknown. More knowledge on the effects of long-term exposure at intake levels close to the UL of all micronutrients is, therefore, urgently needed in order to assure the safety of daily consumption of fortified foods. In their opinions, the SCF often emphasizes that knowledge on adverse effects during development is inadequate and that care should be taken. Other population groups may also be at a higher risk in specific cases. The SCF attempts to identify these sensitive risk groups in their opinions and has specifically included safety considerations for pregnant and lactating women in all their reports. In the case of copper and niacin, for example, they conclude that the established ULs for adults do not account for pregnant and

lactating women due to inadequate data relating to this critical life stage (see Table 1). For vitamin B_6, the SCF highlights that 'a major deficit in the database for this vitamin is the absence of information from adequate developmental neurotoxicity studies' since there are 'no data on the neuronal toxicity of excessive vitamin B_6 intake during development of the nervous system.' The SCF also identifies other sensitive sub-groups in the population in their 'characterization of risk.' As seen in Table 1, the established ULs for iodine, vitamin E and A do not apply for specific subgroups, e. g., populations with iodine deficiency, subjects with blood coagulation defects or postmenopausal women at risk of bone fracture (see footnotes in Table 1).

Also, other yet unidentified sub-groups of the population may be at higher risk when large doses of micronutrients are consumed over a long period of time. As an example, there was unambiguous trust among all nutritionists and toxicologists that beta-carotene was safe at high doses and it was even subscribed to smokers by some physicians. This situation is now completely reversed because two large-scale human trials have indicated that this pro-vitamin may not be safe for population groups with an elevated risk for lung cancer. Another example is the French SUVIMAX study, where 12741 subjects were randomized to either placebo or daily low doses of vitamin C (120 mg), vitamin E (30 mg), β-carotene (6 mg), selenium (100 µg) and zinc (20 mg) for 8 years [58]. In this study, the incidence of all types of skin cancer was increased in women receiving supplements compared to women in the placebo group. On the contrary, the risk of all cancers for men receiving supplements decreased by 31 %. Consequently, the long-term intake of low doses of some micronutrients may have adverse effects in particular population groups and beneficial effects in others. Risk-benefit and risk-risk analyses may, therefore, be the way ahead for most micronutrients, although lack of detailed knowledge about risks is a major obstacle [59]. With the present limited knowledge about the safety of most of the micronutrients, we suggest applying the GLs and TGLs listed in Table 2 for those micronutrients, where no UL has yet been established. Research should be initiated to substantiate the safety of these provisional limits, and ULs, GLs and the proposed TGLs should be re-evaluated whenever new knowledge about the safety of these vitamins and minerals becomes available.

Conclusion

The proposed model for safe addition of vitamins and minerals to foods is designed to protect the whole population including children of all ages against possible adverse effects of excessive intakes of vitamins and minerals from fortified foods. The present model takes into account the regular use of a vitamin and mineral supplement and differentiates ULs for children of all ages. Although the model is designed to ensure the safety of 95 % of the population including children of all ages, it should be emphasized that knowledge on the safety of long-term exposure to elevated vitamin and mineral intakes is still lacking especially in potentially sensitive sub-populations like infants, children, adolescents, pregnant or postmenopausal women, and the elderly. More research-based knowledge on the safety of long-term exposure to elevated intakes of micronutrients is, thus, greatly warranted to guarantee the safety of long-term regular use of fortified foods.

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