F. No. 3-9/Action on 13th Authority meeting minutes (Nutra Panel)/FSSA-14

Food Safety and Standards Authority of India

(A Statutory Authority under the Ministry of Health and Family Welfare, Govt. of India)

FDA Bhawan, Kotla Road, New Delhi-110002.

Dated, the 05th September, 2018

Note

1. The Food Safety and Standards (Health Supplements, Nutraceuticals, Food for Special Dietary Use, Food for Special Medical Purpose, Functional Food and Novel Food) Regulations, 2016 allows the use of vitamins/minerals only up to one RDA in Health supplements/Nutraceuticals.

2. In view of the growing concern towards unsupervised usage of Health supplements/Nutraceuticals and issues regarding 'Tolerable Upper Limits (TUL)' of vitamins/minerals, FSSAI approached ICMR for guidance on the subject matter. ICMR constituted a Committee to examine the matter, which has submitted its report to FSSAI.

3. A copy of report of the Expert Committee of ICMR, which specifies TUL of micronutrients for Indian population is placed at **Annexure** for the information of general public.

Dr. A. C. Mishra
Joint Director (Standards)
FSSAI, New Delhi.

Enclosure: as above

Copy To:

1. CITO, FSSAI for necessary action and uploading on FSSAI website.

Copy for information to:

1. PPS to Chairperson, FSSAI, New Delhi.
2. PS to CEO, FSSAI, New Delhi.
3. All Directors, FSSAI, New Delhi.

F. No. 3-9/Action on 13th Authority meeting minutes (Nutra Panel)/FSSA-14

भारतीय खाद्य सुरक्षा और मानक प्राधिकरण

(खाद्य सुरक्षा और मानक अधिनियम, 2006 के तहत स्थापित एक वैधानिक प्राधिकरण)

(मानक विभाग)

एफडीए भवन, कोटला रोड, नई दिल्ली- 110002

दिनांक: 05th सितंबर, 2018

नोट

खाद्य सुरक्षा और मानक (स्वास्थ्य अनुपूरक, न्यूट्रास्यूटिकल्स, विशेष आहार विषयक उपयोग के लिए खाद्य, विशेष चिकित्सीय प्रयोजन के लिए खाद्य, कृत्यकारी खाद्य और नूतन खाद्य) विनियमावली, 2016 के अनुसार स्वास्थ्य अनुपूरक/न्यूट्रास्यूटिकल्स में केवल एक आरडीए तक विटामिन / खनिजों के उपयोग का प्रावधान है।

2. स्वास्थ्य अनुपूरक/न्यूट्रास्यूटिकल्स के अनिरीक्षित उपयोग की बढ़ती चिंता को ध्यान में रखते हुए और विटामिन / खनिजों के 'सुरक्षित ऊपरी सीमाओं (टीयूएल)' से सम्बंधित मुद्दे के बारे में एफएसएसएआई ने मार्गदर्शन के लिए आईसीएमआर से संपर्क किया। आईसीएमआर ने इस विषय पर विचार विमर्श के लिए एक समिति का गठन किया, जिसने अपनी रिपोर्ट एफएसएसएआई को प्रस्तुत कर दी है।

3. भारतीय उपभोक्ताओं के लिए सूक्ष्म पोषक तत्वों के टीयूएल को निर्दिष्ट करने वाली आईसीएमआर समिति की रिपोर्ट की एक प्रति आम जनता की जानकारी के लिए अनुलग्नक में उपलब्ध की जा रही है।

अतिल मिश्रा

डॉ. ए.सी. मिश्रा
संयुक्त निदेशक (मानक)
एफएसएसएआई, नई दिल्ली।

संलग्नक: उपर्युक्त

प्रतिलिपि:

1. सीआईटीओ, एफएसएसएआई आवश्यक कार्रवाई और एफएसएसएआई वेबसाइट पर अपलोड करने के लिए।

प्रतिलिपि (जानकारी के लिए):

1. अध्यक्ष के लिए पीपीएस, एफएसएसएआई, नई दिल्ली।
2. मुख्य कार्यकारी अधिकारी के लिए पीएस, एफएसएसएआई, नई दिल्ली।
3. सभी निदेशक, एफएसएसएआई, नई दिल्ली।

ANNEXURE

Report of Expert Committee

To examine a) allowance of vitamins/minerals more than one RDA in health/dietary supplements and nutraceuticals and b) Safe Upper Limits

Submitted to

Director General
Indian Council of Medical Research
New Delhi

Date

Expert Committee to examine a) allowance of vitamins/minerals more than one RDA in health/dietary supplements and nutraceuticals and b) Safe Upper Limits (Copy of OM- see next page)

1a. Terms of Reference

- Define a framework for the definition of Safe Upper Limit, also called Tolerable Upper Limit (TUL), uncertainty factors and established risk assessment processes, keeping in mind all factors, including evidence for risk/hazard in humans, populations at different stages of the life cycle, causality, mechanisms of toxicity, and genetic susceptibility.
- Using this framework, define the TUL for vitamins and minerals, with due consideration of the RDA that has already been defined by the NIN/ICMR Committee 2010.
- To review if the allowance of vitamins and minerals in food fortification or in supplements should be up to 1 RDA, or whether this could exceed 1 RDA, with due consideration of safety factors.
- Provide a research framework to fill the existing gaps in information and knowledge for the future and to facilitate science based decisions on different levels of nutrient consumption with both efficacy and safety in mind.

1b. Members and Copy of OM

NATIONAL INSTITUTE OF NUTRITION
HYDERABAD

The Secretary DHR & Director General, ICMR has constituted the following committee consisting of scientists from the 'ICMR Expert Group on Nutrient Requirements and RDA' and some external experts to examine the matter raised by FSSAI on the below mentioned issues and to come up with appropriate recommendation (copy of letter enclosed).

(a) Allowance of vitamins/minerals more than one RDA in health/dietary supplements and Nutraceuticals and (b) Safe Upper Limits.

1. **Dr. A.V.Kurpad**, Professor of Physiology, St. John's Medical College, Bangalore. ***Chairperson***
2. **Dr. G.S.Toteja**, Scientist-G & Head (Nutrition) ICMR, New Delhi
3. **Dr. M.K.Bhan**, Former Secretary to Govt. of India, Ministry of Science & Technology, Dept. of Biotechnology, New Delhi.
4. **Dr. S.Radhakrishna,** Statistician, D-201, High Rise Apts, Lower Tank Bund Road, Gandhinagar, Hyderabad.
5. **Dr. Mahtab S Bamji**, INSA Honorary Scientist, Dangoria Charitable Trust, Hyderabad. Director-Grade-Scientist (Retd.), NIN, 211, Sri Dattasai Apartments, Charminar X Roads, Hyderabad.
6. **Dr. Kamala Krishnaswamy**, Former Director, NIN, H.No.2-98/2, Sriniketan Kakateeyanagar Colony, Habsiguda, Hyderabad.
7. **Dr. B. Sesikeran**, Former Director, NIN, Hyderabad.
8. **Ghafoorunissa**, Former Dy. Director (Sr. Gr.), NIN, Res. Flat No. 402, Maphar Comfortek Apt., 10-2-289/37/A, Shantinagar, Masab Tank, Hyderabad.
9. **Dr. K. Madhavan Nair**, Scientist F, Head, Micronutrient Research, NIN, Hyderabad.
10. **Prof. Ram Rajasekharan**, Director, CFTRI, Mysore.
11. **Dr. V. Prakash**, Former Director, CFTRI, Mysore.
12. **Dr. Prakash Shetty**, Prof. of Human Nutrition
13. **Dr. Prema Ramachandran**, Director, Nutrition Foundation of India, C-13, Qutab Institutional Area, New Delhi.
14. **Dr. A. Ramachandran**, Diabetes Research Centre, 4, Main Road, Royapuram, Chennai.
15. **Dr. Srinath Reddy K**, President, Public Health Foundation of India, PHD House, 2nd Floor, 4/2, Siri Fort Institutional Area, August Kranti Marg, New Delhi.

Contd../-

16. **Dr. Umesh Kapil**, Professor, Public Health Nutrition, AIIMS, New Delhi.
17. **Dr. D.Kanungo,** Former Additional DG, Ministry of Health and Family Welfare – **Expert Member**
18. **Director/Director I/c, NIN - Member Secretary**

The above committee under the chairmanship of Dr. A.Kurpad would be meeting soon to review the Terms of reference. I therefore request you to kindly give your consent to participate in the committee.

Date : 29th Feb. 2016

(T.Longvah)
Director I/c
निदेशक / DIRECTOR I/c
राष्ट्रीय पोषण संस्थान
NATIONAL INSTITUTE OF NUTRITION
भारतीय आयुर्विज्ञान अनुसंधान परिषद
Indian Council of Medical Research
जामे उस्मानिया पोस्ट, हैदराबाद-500007.
Jamai Osmania P.O., Hyderabad-500007.

2. Important Notice for Usage

1. *The Tolerable Upper Limit (TUL) for Nutrient Intake is NOT to be used for planning diets or fortification standards.*

2. *The TUL refers to the intake beyond which adverse effects could occur.*

3. *The current TUL is defined only for "unsupervised intake" of nutrients through entire life or required period of life for the general population*

4. *Special cases such as the supervised intake of antenatal supplements, and therapeutic foods, Foods for special medical purposes and any other short term intake of nutrients prescribed and supervised by medical professionals or qualified dieticians have not been considered in this report.*

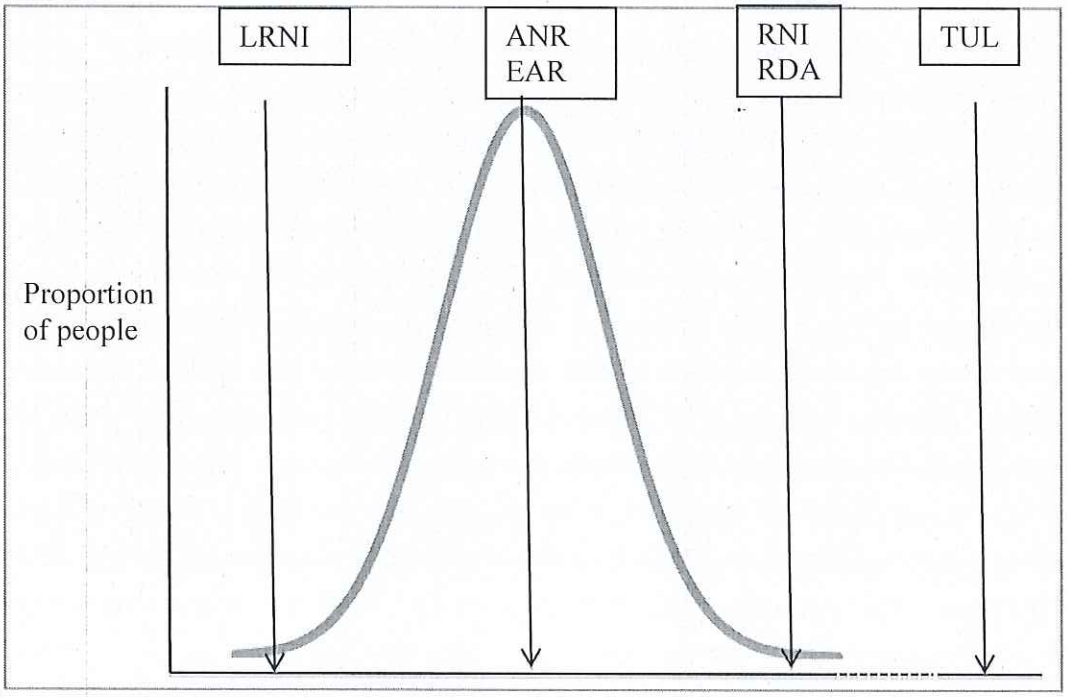
3. Introduction

The Food Safety Standards Authority of India Act 2006, section 22, restricts the use of minerals or vitamins or proteins or amino acids in amounts not exceeding the Recommended Daily Allowance (RDA) for Indians. On the other hand vitamins/minerals more than 1 RDA has been allowed in Foods for Special Dietary Use (FSDU) and Food for Special Medical Purpose (FSMP) to take care of special needs. The use of nutrient supplement in the country is growing in India and to keep a check on the misuse and its consequent adverse effects the FSSAI referred the matter to the Indian Council of Medical Research to examine the issue (Letter No 3-9/Action on 13th Auth. Meeting (Nutra panel)/FSSAI-2 dated 18th March 2015). Therefore the Director General (DG) of ICMR constituted a committee under the chairmanship of Dr.Anura Kurpad, vide email dated 28th January 2016 and OM dated 29th Feb. 2016 (Annexure) to examine the matter and give its comments pertaining to (a) Allowance of vitamins/minerals more than one RDA in health/dietary supplements and Nutraceuticals and (b) Safe upper limits. The committee in its first meeting at the National Institute of Nutrition (NIN) held on 28th April, 2016, assigned responsibilities to each member of the committee for preparing a draft Tolerable Upper Limit (TUL) for specific micronutrients according to the terms of reference. Thereafter, the draft TUL was circulated among all members for comments through emails. After a series of exchange of comments/deliberations vide emails, and a final consensus meeting at NIN on 10th June 2017, the "Report of Expert Committee to examine allowance of vitamins and minerals exceeding one RDA in health/dietary supplements and nutraceuticals and to determine their Safe Upper Limit" was finalized for submission to DG, ICMR. A lot of effort has been put in by the experts in evaluating published literature relevant to the ill effects of overconsumption of each specific nutrients. The recommended daily allowances (RDA) for Indians are now proposed to be supplemented with a new value, "Tolerable Upper Level," the largest daily intake which is unlikely to cause harm. It is important to note that the tolerable upper levels indicated for each nutrient are based on chronic exposure and supplements should not be treated casually.

4. Framework for Nutrient Requirements

Several countries recommend nutrient intakes for their populations. These are used to plan and evaluate the nutrient intakes of healthy people. Nutritional policies, food regulations and nutritional programs are based on these nutrient intake recommendations[2]. The recommended values differ from country to country and could range from a single value for a population group (as in the ICMR report for India[1]), to four different values that define a 'lower reference intake', an 'average requirement', a 'recommended intake' for individuals from a specific population, and an 'upper tolerable intake'[1,2].

Figure 1. Distribution of the requirements of a theoretical nutrient in a population, showing ANR/EAR and RNI/RDA. The TUL is also depicted as an intake in excess.



Note: The dashed line on the X-axis depicts a variable distance between the RNI/RDA and the TUL for different nutrients.

In 2007, the United Nations University's Food and Nutrition Program, in collaboration with the Food and Agriculture Organization (FAO), the World Health Organization (WHO), and UNICEF, attempted to harmonize the recommendations used across several countries, and coined the term Nutrient Intake Values (NIV), using primary data from several countries. These were primarily, Dietary Reference Values (DRV, UK), Nutrient Reference Values (NRV, Australia, New Zealand), Reference Values for nutrient supply (Germany, Austria, Switzerland), and Dietary Reference Intakes (DRI, USA, Canada)[2,3]. The relationship between average requirement and the recommended intake are shown in Figure 1.

Two of the NIV's were recommended for comparability across all countries for specific life stages and genders: average nutrient requirement (ANR) which is equivalent

to the Estimated Average Requirement (EAR[4]) and Upper Nutrient Level (UNL) equivalent of the Tolerable Upper Limit (TUL[4]). The other values, like the Reference Nutrient Intake (RNI)/ Recommended Dietary Allowance (RDA)/ Recommended Dietary Intake (RDI), which are very similar to each other, and the Lower Reference Nutrient Intake (LRNI), were derived values from the two recommended NIVs (Fig 1). The IOM suggested that, although their recommendations were based on dietary intakes in the United States and Canada, their values could be adapted to other populations by adjusting for nutrient bioavailability[4].

Each of these terms is defined in Table 1.

Table 1. Definition of terms used in the framework of nutrient requirements.

• *Average Nutrient Requirement (ANR)* • *Estimated Average Requirement (EAR)*	Refers to the average daily nutrient intake level estimated to meet the requirements of half of the healthy individuals in a particular life stage and gender group. It is used primarily to evaluate populations or groups.
• *Recommended Nutrient Intake (RNI)* • *Recommended Dietary Allowance (RDA)*	Refers to the daily dietary nutrient intake level that is sufficient to meet the nutrient requirements of nearly all (97–98 percent) healthy individuals in a particular life stage and gender group. This is derived from the ANR/EAR as the mean plus 2 standard deviations (SD) of the distribution of requirements. The term is used to primarily evaluate individual diets. *The RDA is inappropriate for dietary assessment of groups as it is the intake level that exceeds the requirement of a large proportion of individuals within the group.*
• *Upper Nutrient Level (UNL)* • *Tolerable Upper Level (TUL)*	Refers to the highest average daily nutrient intake level that is likely to pose no risk of adverse health effects to almost all individuals in the general population. As intake increases above the UL, the risk of adverse effects will increase.
• *Adequate Intake (AI)* • *Safe Intake*	These values are used when ANR or RDA cannot be determined. The Safe intake or AI is the recommended average daily intake level based on observed or experimentally determined approximations or estimates of nutrient intake by a group of apparently healthy people that are assumed to be adequate.
• *Lower reference nutrient intake (LRNI)* • *Lower threshold intake (LTI)*	Refers to a value derived from the ANR/EAR and is calculated as the ANR/EAR minus 2 SD of the distribution of requirements. This value is sufficient to meet the needs of the bottom 2% of individuals. However, countries have used a different cut off such as 5% or 10% to evaluate nutrient insufficiency, although the concern is that these values would set a very low expectation of the individual nutrient intake adequacy level.

References provided in text

An additional term that is used is the Acceptable Macronutrient Distribution Ranges (AMDR). The AMDR is a range of macronutrient intakes that is associated with a reduced risk of chronic diseases but at the same time provides adequate intakes of essential nutrients. It is usually expressed as a percentage of energy, with lower and upper limits. In the US and Canada, the AMDRs refer to appropriate ranges of usual intakes of *individuals*, whereas the WHO standards are mean intake goals for the *population*. Based on the latter, the mean intake goal for total fat intake is 15% to 30%

of the energy intake, and implies that it is acceptable for half of the individuals in a population to have intakes below 15%[2,5].

Tolerable Upper Limit (TUL)

The TUL is the maximum level of habitual intake from all sources of a nutrient or related substance judged to be unlikely to lead to adverse health effects in humans[10]. An adverse effect is a change in morphology, physiology, growth, development, reproduction or lifespan of an organism, system, or (sub) population that results in an impairment of functional capacity, an impairment of capacity to compensate for additional stress, or an increase in susceptibility to other diseases. The potential risk of adverse effects increases after the intake increases above the TUL. The TUL applies to chronic daily use and it is important to first assess the characteristics of the individual or group, the source of the nutrient, the physiological state of the individual and the duration of sustained high intakes. The bioavailability of a nutrient, which is its accessibility to normal metabolic and physiological processes, also plays a role in the nature and severity of adverse effects at excessive intakes. However, in some cases, the unabsorbed nutrient may also have effects on the lower parts of the intestine. This is particularly relevant for iron, in which the unabsorbed iron may have effects on the intestinal microbiome. The most appropriate approach is to establish the TUL for age/gender/life stage sub-populations, since adverse effects of nutrients are influenced by growth and physiological stages.

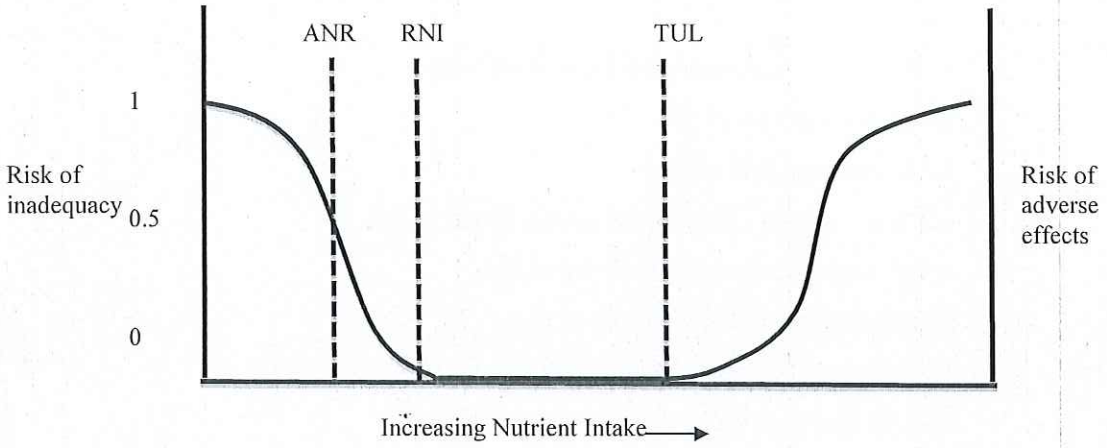
The increased availability and consumption of fortified (many with multiple micronutrients) foods and food supplements has sparked concerns about excessive intake of nutrients. It is important to assess the safety of fortification by comparing eventual micronutrient intakes with the TUL. The safety of fortification by comparing the total micronutrient intake from all sources (natural and fortified food; supplements), with the TUL. In principle, high levels of micronutrient additions should be avoided even if a micronutrient has no recommended TUL, particularly if there is no evidence of derived benefit from levels of intake in excess of the RNI.

The conceptual basis for calculating the risk of inadequacy or excess

An individual has inadequate intake when the intake does not meet the requirement of the nutrient, and an excessive intake if the intake is more than the TUL. Since the RNI is the sum of the ANR plus 2 SD, recommending this intake for an individual will mean that his/her chances of being at risk of an inadequate intake, when eating this intake, is <2.5%. In a population, the risk of inadequacy or excess translates to the proportion of people whose usual intake of a nutrient does *not meet* their

requirement, or *exceeds* the TUL. These can be easily calculated from normalized population level data on usual intakes and the requirement/TUL(Figure 2). The safety of fortification could also be assessed by this method, when for example, the population intakes of micronutrients from all sources (natural and fortified food, food supplements) are evaluated against the TUL.

Figure 2: Showing risk of inadequacy or excess (Y-axis), for a range of population intakes



References

1. Indian Council of Medical Research. Nutrient requirements and recommended dietary allowances for Indians. A Report of the expert group of the Indian Council of Medical Research. National Institute of Nutrition, Hyderabad, 2010.
2. King JC, Vorster HH, Tome DG. Nutrient intake values (NIVs): A recommended terminology and framework for the derivation of values. Am J ClinNutr 2007; 81: S16-S26.
3. King JC, Garza C. Harmonization of nutrient intake values. Am J ClinNutr 2007; 81: S3-S12.
4. Institute of Medicine. Dietary reference intakes: The essential guide to nutrient requirements. Otten JJ, Hellwig JP, Meyers LD, Editors. National Academy of Sciences. The National Academies Press, Washington DC. 2006.
5. World Health Organization. Diet, nutrition and the prevention of chronic diseases. Geneva: World Health Organization, 2003.

5. Abbreviations used:

- AI: Adequate Intake
- AMDR: Acceptable Macronutrient Distribution Ranges
- ANR: Average Nutrient Requirement
- Cu: Copper
- DRI:Dietary Reference Intakes
- EAR: Estimated Average Requirement
- Fe: Iron
- ICMR: Indian Council of Medical Research
- IFA: Iron and Folic Acid
- IOM: Institute of Medicine
- LOAEL: Lowest Observed Adverse Effect Level
- LRNI: Lower Reference Nutrient Intake
- NFHS: National family Health Survey
- NIN: National Institute of Nutrition
- NNMB: National Nutrition Monitoring Bureau
- NOAEL: No Observed Adverse Effect Level
- RCT: Randomized Control Trial
- RDA: Recommended Daily Allowance
- RNI: Recommended Nutrient Intake
- TUL: Tolerable Upper Limit
- UF:Uncertainty Factor
- UL: Upper Limit
- UNL: Upper Nutrient Limit
- Zn: Zinc

6. A framework for arriving at Tolerable Upper Limits (TUL)

The Tolerable Upper Intake Level (TUL) is not a recommended intake amount. Instead, it indicates the level above which the risk for harm begins to increase. It is defined as the highest average daily intake of a nutrient that is likely to pose no risk of adverse health effects for nearly all persons in the general population. TUL is a reference value meant to guide policy-makers and scientists responsible for ensuring food safety and thus good health among population groups.

Arriving at a value for the TUL of a nutrient requires the definition of a framework for uncertainty factors and established risk assessment processes, keeping in mind all factors, including evidence for risk/hazard in humans, populations at different stages of the life cycle, causality, mechanisms of toxicity, and genetic susceptibility. This framework is used to define TUL for vitamins and minerals, with due consideration of the RDA that has already been defined by the NIN/ICMR Committee 2010.

It is important to state that TUL is defined only for "unsupervised intake" of nutrients through entire life or required period of life for the general population. Special cases such as the supervised intake of antenatal supplements, and therapeutic foods, Foods for special medical purposes (FSMP) and any other short term intake of nutrients prescribed and supervised by medical professionals or qualified dieticians have not been considered in this report.

Examples of Adverse effects occurring due to both extremes of intakes are shown below:

Nutrient	Intakes below RDA	Intakes if >TUL
Calcium	Osteoporosis	Hypercalcemia/renal stones
Iron	Anemia	GI side effects
Zn	Growth Failure	Impairs Cu status
Vitamin A	Ocular lesions, Morbidity and Mortality	Liver damage, teratogenecity
Vitamin C	Scurvy	GI side effects
Vitamin D	Skeletal Deformities	Hypercalcemia
Folic Acid	Megaloblastic Anemia	Masking of Vitamin B12 deficiency

The risk associated with nutrient intakes has a U-shaped curve. When intakes are below the RDA, the risk of deficiency disorders increases and if the intake exceeds upper limits, there is a possibility of adverse effects, at least in case of some of the nutrients (Fig 1 in Section 4). The RDA is the minimum level, since by definition, if intakes get below RDA, deficiency risk increases. The RDA is a measure of nutrient adequacy and not a risk based safety limit. Several naturally occurring foods, or combinations of foods with enhanced bioavailability are found to have higher amounts of nutrients that surpass RDA.

The TUL is the limit beyond which toxicity may become a possibility. The levels at which the earliest evidence of toxicity reported is the Lowest Observed Adverse effect level (LOAEL) and this may be very close to the TUL or several-fold higher or may even be indeterminable. This indicates the margin of safety i.e. narrow to a very wide margin respectively. Hence, there is a need to define these limits. The range between RDA and TUL is considered to be a "safe range" of nutrient intakes. The limits, both lower (RDA) and upper (TUL), apply to the total per day consumption of a nutrient through all sources both food as well as nutrient supplements.

Steps followed to arrive at TUL's:

Step 1: Hazard Identification

This was done by evaluation of all published information from India relative to the nutrient's potential to cause harm in humans. Global data regarding TUL initially came from supplementation studies but more recently there has been data from long term use of fortified food (for example, mandatory folic acid fortification of wheat flour in USA). Most of the Indian data are from side effects reported from supervised supplementation (for example, IFA supplementation in adolescents and pregnant women). These data were analyzed for the nature of adverse effects, severity, dose response and its persistence or chronicity. If the excess nutrient intake resulted in multiple adverse effects of a varied nature, the most critical, i.e. in severity and persistence at Lowest Intake was considered. Clinical case studies were also considered to understand the nature of the adverse events but the intake levels were not considered since most of them were of very short duration and often much higher than long term intake levels.

Step 2- Dose Response Assessment

A quantitative evaluation of relationship between the level of intake and any adverse effect is done to fix the "no observed adverse effect level" (NOAEL). The highest level of intake with no adverse effect, is the "no observed adverse effect level" (NOAEL); the "lowest observed adverse effect level" (LOAEL) is the lowest intake level above which, adverse effects have been reported. Uncertainty factor (UF) is a factor used as a denominator applied to keep the TUL at a level lower than the actual level where adverse effects were observed; UF provides the allowances for possible uncertainties due to a variety of reasons like robustness of the research and data, number of studies, sample size, human versus animal data and other inherent variations and limitations of the published evidence. If the data is robust then the UF is low and vice versa. The Institute of Medicine USA used a range of UF from 1 to 5. This committee did not consider data from animal experiments

Step- 3 Derivation of UL

a) UL = NOAEL ÷ UF or sometimes UL = LOAEL ÷ UF (from Indian data). When NOAEL is not available and LOAEL is used for derivation of UL, a higher uncertainty factor may be used by the experts.

b) If data from Indian population was not available international reference TUL values from sources like Institute of Medicine (IOM- USA), Council for Responsible Nutrition (CRN) European Commission -Scientific Committee on Food (EC SCF 2003) or WHO have been considered. It was also found that in most instances TUL arrived at from published data on Indian population was similar to the international TUL.

Specific points to be considered

No TUL have been recommended for infants 0 to 6 months' age, since no other foods or supplements other than breast milk are recommended, as per the law in India.

When no Indian evidence for TUL for any nutrient was available, the international values for TUL from other countries were used, and the lowest TUL value was chosen.

However, as mentioned earlier this does not apply to the use of nutrients as supplements in any age group for clinical indications and under supervision of qualified health care professionals.

6. Framework of fixing nutrient levels for fortification of staple foods, processed foods and dietary supplements.

The level of nutrients recommended for fortification of staple foods is country-specific. The fortification strategies in India have been based on available evidence and international guiding principles. Fortification is the addition of one or more essential nutrients to a food whether or not it is normally contained in the food, for the purpose of preventing or correcting a demonstrated deficient dietary intake of one or more nutrients in the population or specific population groups[1]. The goal of any national food fortification policy is to prevent nutritional deficiencies; by bridging the gap between the requirement of the nutrient, the current intake and in exceptional cases of very wide spread and severe deficiencies, the fortification can be up to 100% of the RDA. If the nutrients are not part of a food matrix, it shall be per serving, or if multiple servings per day, then the total intake per day should not exceed 1 RDA. In processed and packaged foods the level of fortification is about 15 to 30% of RDA (computed per 600 calories of the processed food). However, it is preferable not to embark on multiple micronutrient fortification of multiple food stuffs.

When these precautions are followed, food fortification carries a minimal risk of chronic toxicity.

This depends mostly on the quantity of micronutrients recommended for fortification and the daily consumption of the fortified food which serves as the vehicle for the nutrient(s). The following formed the basis of fixing the level of nutrient for fortification:

1. RDA's have been determined as being more appropriate for fixing fortification levels, especially in India which is still at a nascent stage in its food fortification program and would benefit by having more diversity in fortified products.
2. Habitual Indian diets are predominantly cereal-pulse-vegetarian ones with low amounts of flesh foods. This has been used in computation of RDA and therefore no extra allowance has been made while computing the fortification levels.
3. While ensuring adequate intake of the nutrient to vulnerable segments, it is also necessary to ensure that those who are already getting adequate intake should not be at risk of any adverse effects of excess intake. In the Indian context, the group which could probably fall into the latter category is that of adult men because they consume the greatest quantity of the identified vehicles, especially cereals.
4. Why the requirements of adult man have been chosen while fixing the fortification limits of micronutrients? For a majority of the micronutrients, the highest recommended intakes apply to adult males[2]. The exception is iron (17 mg/d). Adult males face the lowest risk of micronutrient deficiencies because their overall food

intake is high enough to supply the relatively small micronutrient requirements per unit body weight. An intake of 400g of cereal is calculated to meet the requirement of calories and certain micronutrients for a reference man which is close to the actual intake reported from rural NNMB surveys across the country (Tables 2 and 3 and Figure 3).

Table 2. Consumption of Cereals by different Age Groups

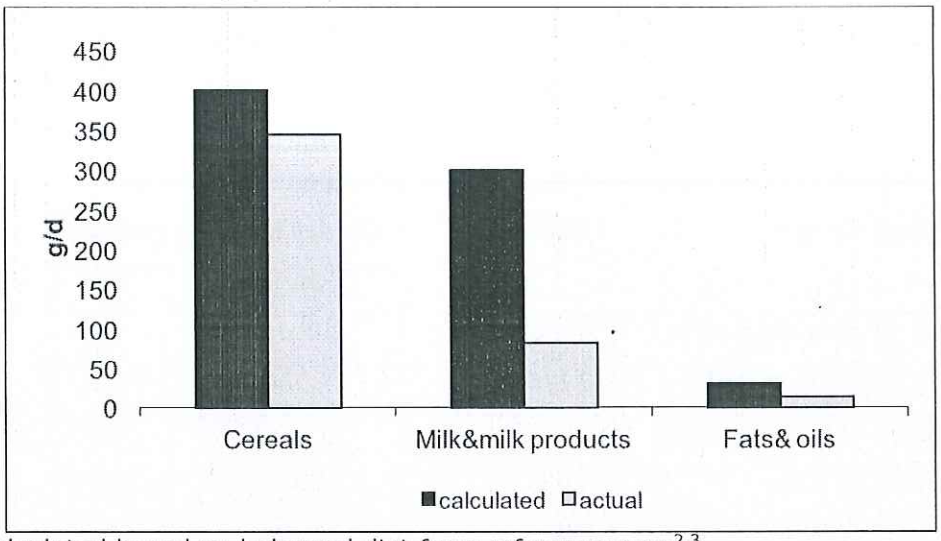
Age Group	Number	Cereal intake (g/day)	
		Mean	SD
1-3y (Boys & Girls)	2895	131	82
4-6y (Boys & Girls)	2915	209	97
7-9y (Boys & Girls)	2963	262	110
10-12y Boys	1654	301	124
10-12y Girls	1577	289	124
13-15y Boys	1529	347	133
13-15y Girls	1538	324	131
16-17y Men	898	386	148
16-17 y Women	991	346	144
Adult Men	11274	444	166
Adult Women	6118	391	141
Pregnant Women	322	354	138
Lactating Women	693	395	152

Table 3. Consumption of Cereals in different States – Adult Men, Moderate Activity

States	N	Mean (g/day)	SD
Kerala	756	325	133
Tamil Nadu	982	424	146
Karnataka*	1302	481	191
Andhra Pradesh	1119	494	190
Maharashtra*	1343	378	160
Gujarat*	1322	431	145
Madhya Pradesh*	1407	480	120
Orissa	1044	501	81
West Bengal	968	363	84
Pooled	11274	444	166

***Habitual diet consists of millet as staple[3]**

Figure 3. Cereal, milk and visible fats and oils in adult male



Note: Calculated based on balanced diet for a reference man[2,3]

Though the cereal consumption is region-specific, it is unlikely to exceed the limits even in the context of consumption of rice and wheat together. Cereal intakes appeared to be adequate and had been considered as the major vehicle for fortification of iron, folic acid and vitamin B12[4,5].

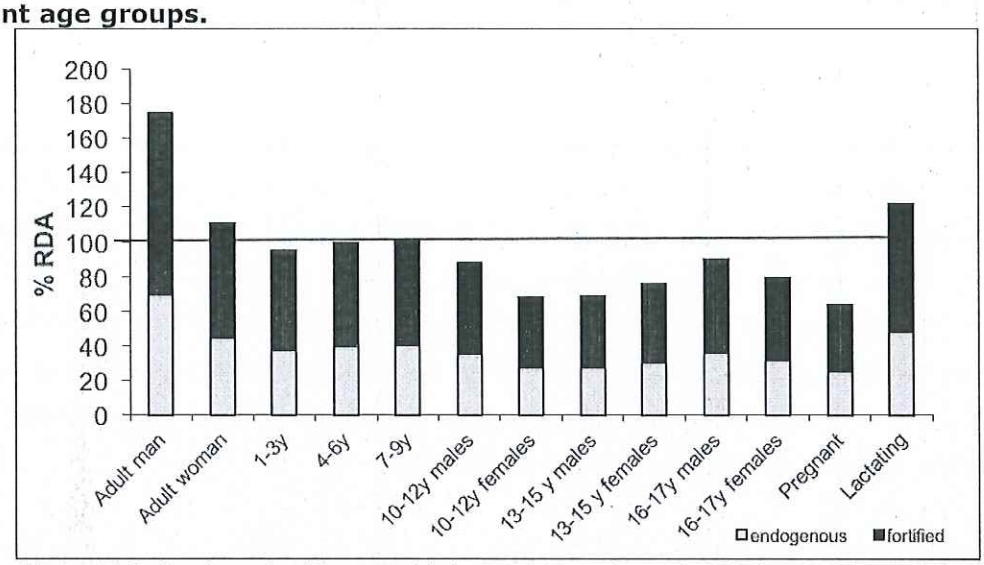
5. Milk and oil, were consumed daily across population groups but the intakes were insufficient (Figure 3, only 80 mL/d of milk as against 300mL suggested and visible fat intake was only 13 g/d as against 30g recommended). Considering the universal usage of these two food stuffs, the deliberations also included consideration of fortification with vitamin D and vitamin A.
6. In addition to the above vehicles, salt has been a universal vehicle for iodine for decades; has been considered for fortification with iron also.
7. Fortified processed foods: Processed foods are those in which food raw materials have been treated industrially (e.g. bread) so as to preserve them and formulated by mixing several different ingredients. Though not consumed by large segments of the population in India, being a market driven strategy it has the great potential to contribute to daily requirements of nutrients.

Fortification level of iron in staple foods

Considering the RDA of iron of 17mg/d and an average consumption of staple foods rice and or wheat of 400 g/day, the fortification of iron per kg of cereal would be 42 mg/kg. At this level of fortification, the different groups will receive between 39% (pregnant women) and 100% (adult man) of their RDAs. Considering the presence of endogenous iron in cereals, the overall intake of iron from cereals is presented in Figure

4, showing that all the age groups and pregnant women would meet above 60% of the RDA.

Figure 4. Contribution of iron from endogenous and fortified sources in different age groups.



Note *TUL for iron is 45 mg which corresponds to 2.65 times RDA. None of the groups exceeded this level.*

However, due allowances were made for the variable intake of cereals in different population groups and the fact that iron requirements are lowest for adult men. On this basis, a higher level **of 60 mg/kg** which is in concordance with the FAO/WHO guidelines was adopted[1,7]. This translates to an intake of 200% of RDA by an adult man (34 mg/day), which is still well below the TUL level of iron (45 mg/day).

Fortification level in iodized salt (double fortified salt)

RDA is also the basis for fixing the levels of iodine (150 µg) and iron (10 mg which is 50% RDA) in double fortified salt and is based on approximate consumption of 10 g of salt by an adult male.

Fortification level of Vitamins A and D in oil and milk

On account of the fact that more than one vehicle lends itself to fortification with vitamins A and D (milk and vegetable oil), and also given the tropical climate for vitamin D, 1/3rd of the daily requirement has been fixed as the fortification level.

Fortification of processed foods

It is assumed that about 30% (600 kCal) of an individual's daily energy intake is derived from fortified processed foods and therefore its contribution would also be limited to a maximum of 1/3rd of RDA[1]. All processed packaged foods should carry

nutrient labels, indicating total nutrient content and this should be within 15-30% of the RDA.

Levels of vitamins and minerals in food supplements:

According to The Food Safety and Standards, Health Supplements, Nutraceuticals, Food for Special Dietary Use, Food for Special Medical Purpose, Functional Food and Novel Food) Regulations, 2016 the quantity of nutrients added to the articles of food shall not exceed the RDA as specified by the Indian Council of Medical Research and in case such standards are not specified, the standards laid down by international food standards body, namely, Codex Alimentarius Commission, shall apply.

In case of food products falling under health supplements of vitamins and minerals, the individual nutrient content added shall not be less than 15% RDA, where a nutrient content claim is being made: provided that, if claim of higher nutrient content is made, the nutrient content shall not be less than 30% RDA.

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8. Individual Nutrients

The following section contains the recommendations for individual nutrients, based on the frameworks laid out above, and from evidence available for adverse effects.

8a. Iron

A search was carried out using PubMed, Cochrane Review and Google Scholar. The search terms included iron, supplementation, side effects, adverse effects, GI disorders, adolescents, pregnant women, children, RCT, meta-analysis, programme evaluation, India.

Six studies (RCT, programme evaluation, tolerance test) among children (2-5y), adolescent girls (10-19y) and pregnant women (16-28 weeks of gestation) were included. No studies in other age group/gender with side effects were found (Appendix 1; Table-1). Different doses and forms of iron were supplemented along with folic acid and vitamin B12, with different periodicity (daily and weekly). Iron supplementation was shown to significantly improve iron biomarker levels in these studies. There was no GI side effects with supplementation of 20 to 30 mg iron in children, while 60 to 180 mg iron in pregnant women, 60 to 300 mg iron in adolescent girls was found to be associated with GI side effects such as abdominal pain, dyspepsia, nausea, vomiting, constipation and diarrhoea. These results support the IOM recommended TUL for iron supplementation of 40 mg/d (for 6mo-13 y) and 45 mg/d (14->70 y) (Appendix 1).

An exception to TUL in the case of iron: It is estimated that an additional requirement of about 900 - 1000 mg of iron is needed during pregnancy, which cannot be met from diet. Considering this and the severity of anemia, iron supplementation much above the TUL (60-200 mg) is recommended by WHO and Ministry of Health and Family Welfare, GoI. However, these are given for a short period and monitored by health workers.

Recommendation:

In the absence of wide spread use of iron fortified foods in the country, the expert group considered studies that were conducted within the frame work of National Nutrition Anemia Prophylaxis program. The dosage of iron supplements varied from 20 mg for under 5 anemic children, 45 mg for 5-10 years, 60-100 mg for 10-19 years and women of reproductive age which were advised to be taken with the meals to reduce the adverse gastrointestinal effects.

Among children of 2-5 year age group no adverse gastrointestinal effects have been reported after oral dosage at 20-30 mg daily supplemental iron preparations for 7-

12 months. This suggests that the risk of adverse effects from supplement including food sources (8.5 mg/1000kcal), is considered to be low for this age group. Since these are within the tolerable upper intake level of iron of 40 mg suggested by Institute of Medicine, the same is adopted.

Iron supplementations in the dose range of 60-180 mg have been reported to produce adverse gastrointestinal disturbances among 10-19 year adolescents, and pregnant women. The side effects of oral iron preparations increased with increase in dosage. However, these studies clearly demonstrated benefit from supplemental iron intake. In absence of studies with iron doses less than 60 mg/day, it was recommended to consider the upper intake level of 45 mg, suggested by Institute of Medicine.

Table of evidence and Bibliography provided in Appendix 1.

8b. Zinc

Literature survey was carried out on PubMed, Cochrane Review and Google Scholar. The search included 'Zinc supplementation Human India' and 'Effect of zinc supplementation on Copper status'. The abstracts were further filtered for data on blood copper level and on studies conducted in India.

Eight zinc supplementation trials were found in literature. All the studies (except one which targeted those between 6 months and 15 years of age)were seen to be carried out on pre-school aged children(<59 months) as a part of clinical trials. These studies investigated the effect of zinc supplementation on plasma/serum copper status. One study assessed the effect on iron status. There were no studies on the effect of zinc supplementation on blood copper level among adults. The dose of zinc supplements tested ranged from 5mg-40mg. None of the doses tested resulted in any significant reduction in blood copper levels. Since these results do not contradict the IOM recommended TUL for zinc, it may be considered to be the same for Indians (Appendix 2).

Recommendation:

In the absence of robust data on dietary zinc intake and copper status and since most of the studies have been carried out with zinc supplements of dosage ranging from 5-40 mg in under 5 year old children with diarrhoea or in malnourished children, tolerable upper limits, as recommended by IOM, is recommended for Indians.

Table of evidence and Bibliography provided in Appendix 2.

8c. Calcium

There are different criteria used to define adverse events due to excessive intake of calcium. These are:

- Hypercalcemia: It is defined as serum calcium level >10.5 mg/dl (2.63mmol/l). Hypercalcemia can result due to excessive intake of calcium or vitamin D but it is more commonly observed in conditions such as malignancy and primary hyperparathyroidism. Clinical signs and symptoms of hypercalcemia include anorexia, polyuria, weight loss, arrhythmia, fatigue, soft tissue calcifications.
- Hypercalciuria:It is defined as urinary calcium excretion > 250 mg/day in women and >275-300 mg/day in men or urinary calcium: creatinine ratio > 0.3 mg/mg creatinine. It usually occurs when serum calcium levels are > 12 mg/dl.
- Vascular and soft tissue calcification – This occurs as a result of long standing hypercalcemia or increased serum phosphate levels. It is usually associated with metabolic disorders such as hyperparathyroidism, sarcoidosis or connective tissue disease such as scleroderma. Some evidence indicates increased cardiovascular disease (CVD) risk with calcium supplements. For example, a meta-analysis involving 11 randomized controlled trials of calcium supplements in 12,000 older patients had found that there was a 30 percent increased risk of heart attack independent of age, gender, and type of supplement (Bolland, Barber et al. 2008). But this finding was not consistent with other studies where most of the observational studies revealed inverse (Iso, Stampfer et al. 1999)or null associations (Van der Vijver, van der Waal et al. 1992).

A few studies found enhanced CVD risk in men but not in women. For example,in a study which included a total of 132,823 participants in the Cancer Prevention Study II Nutrition Cohort, and which was followed from baseline (1992 or 1993) through 2012 for mortality outcomes, it was found that supplemental calcium intake of ≥1000 mg/day in men was associated with significant higher all cause and CVD-specific mortality but not in women (Yang, Campbell et al. 2016). Another study, the National Institutes of Health-AARP Diet and Health Study from the US, which included a total of 388 229 men and women aged 50 to 71 years also found that high intake of supplemental calcium was associated with an excess risk of CVD death in men but not in women (Xiao, Murphy et al. 2013).

Although calcium is present in approximately 80 % of kidney stones (Coe et al., 1992), the role of calcium and other nutrients, acting alone or in concert as risk factors, is not completely understood and may be a function of physiological context. Various dietary and non-dietary factors are associated with stone formation, making data difficult to interpret. Evidence from calcium supplementation studies on the risk of nephrolithiasis

is inconsistent. In healthy postmenopausal women participants from the Women's Health Initiative study done in U.S.A, total calcium intake of 2,100 mg per day (average of 1,100 mg daily from dietary calcium plus the additional 1,000 mg of supplemental calcium) was found to be associated with increased kidney stones (17% higher risk in the supplemented group compared to placebo) (Wallace, Wactawski-Wende et al. 2011). However, in a study which prospectively examined data again from the NHS for an 8-year period relative to dietary factors and the risk for kidney stones in women 27 to 44 years of age, the inverse relationship between calcium intake from foods and the risk of kidney stone formation remained, but there was no apparent relationship between supplement use and risk (Curhan, Willett et al. 2004). The suggested discrepancy between the risks from food sources of calcium and from calcium supplements may in part be due to the timing of the supplement intake (Curhan et al., 2007). Calcium present in the food binds oxalate and prevents its absorption. If taken between meals, the calcium would have less opportunity to bind oxalate, and so oxalate absorption would be increased. These observations suggest that the relationship between calcium supplementation and a meal may be important.

Overall, the data indicates that the calcium content of foods does not cause stone formation and on the contrary it may be protective against it. On the other hand, calcium supplements are emerging as a cause of concern based on observational data, at least for some groups under certain circumstances.

Studies of associations between calcium supplement use and risk for incident prostate cancer provide mixed results. A prospective study on men (n=65,321; age 50-74 years) from the Cancer Prevention Study II, which examined associations between calcium and dairy product intake and risk for incident prostate cancer, found a small increase in overall prostate cancer risk for calcium intakes of 2,000 mg/day and higher compared with intakes less than 700 mg/day (Rodriguez et al., 2003). On the other hand, a randomized controlled trial (n= 672 men; mean age 61.8 years) from the United States which examined risk for prostate cancer from supplemental calcium (3 g/day of calcium carbonate for 4 years) over a follow up period of 12 years, did not find a higher risk of prostate cancer in the calcium-supplemented group as compared with controls (relative risk [RR] = 0.83; 95% CI: 0.52–1.32) (Baron et al., 2005). Overall, the final verdict in this area is inconclusive. Although observational studies suggest that total calcium intake of 2,000 mg/day or higher may be associated with increased risk for prostate cancer, this data may not be sufficiently robust to serve as an indicator for a UL.

There is a concern about high calcium intake interfering with the absorption of other nutrients such as zinc and iron. Studies in adolescent girls have shown that

calcium intakes of 1,500 to 1,700 mg/day do not interfere with iron or zinc absorption (McKenna, Ilich et al. 1997, Yin, Zhang et al. 2007).

Recommendation:

Based on the above considerations, the TUL defined by the IOM (2011) are presented in Table 4. The European Food Safety Authority (2012), on the other hand, has not defined TUL for infants, children and adolescents stating that the data are insufficient (Table 5).

Table 4: TUL of calcium intake by IOM 2011

	Men	Women
9–13 y	3,000 mg	3,000 mg
14–18 y	3,000 mg	3,000 mg
19–30 y	2,500 mg	2,500 mg
31–50 y	2,500 mg	2,500 mg
51–70 y	2,000 mg	2,000 mg
> 70 y	2,000 mg	2,000 mg
Pregnancy		
14-18 y		3000 mg
>18 y		2500 mg
Lactation		
14-18 y		3000 mg
>18 y		2500 mg

Table 5: TUL of calcium intake by European Food Safety Authority (2012)

Age	Male	Female	Pregnant	Lactating
Infants	Data insufficient			
Children & adolescents	Data insufficient			
Adults	2500	2500	2500	2500

Table of evidence and Bibliography provided in Appendix 3.

8d. Sodium:

The importance of salt reduction is certainly fixing the lower limit rather than the upper limit. The focus of toxicity may not arise but it is self-limiting salt and even the Vitamins and Mineral Study 3rd Edition from the Council for Responsible Nutrition who have published one of the latest studies have also eliminated Sodium from the upper limit list and hence I only concentrated on Potassium as one can understand. Therefore, the main message that needs to go in is reduction of salt to the WHO level of 5 g per

day at the most inclusive of invisible salt. That does not mean that is the upper limit because in India we have data for upto at 9-10 g and a large amount of which comes from salted snacks and pickles.

The global level surveys (many of them) clearly indicate the various socio-demographics, self-rated and calculated level of salt intake, perceived and calculated sources of salt intake, interest groups in reducing salt intake, responsibility for salt reduction and communication protocols in the various traditional Foods and salt.

Recommendation:

Thus, as far as Sodium Chloride is concerned there is a requirement to bring in awareness by propelling behavioral change, understanding perceptions on salt and exercise that is worth for salt and a way forward with our commitments with a focus *"can we change our Food habits and Dietary intake either per day or per week by adopting a lifestyle which is good to us and our family by lowering our salt intake if one is taking more than 5 gm per day. The lower you eat added salt perhaps better it is in the long run".* It is an integrated approach from all angles and a habit forming one and being careful in not taking excess salt can make a lot of difference in the NCD issue that India is burning with. Certainly, on an average, India's salt consumption is high and it is time as a general principle if it is halved perhaps we will be more proactive with the societal health.

One will also get into some controversy or conflict of interest with the iodine recommendation, when a lesser sodium intake is recommended. Therefore, at this point of time perhaps to"reduce salt intake as much as possible, but at any cost not to exceed 5 g per day inclusive of invisible salt", may be good for better health.

Bibliography provided in Appendix 4.

10. Potassium

When it comes to Potassium of course there are large number of studies which have been well documented and the take home message on Potassium is summarized in the attachment file which are page no. 106-109 from the Vitamin and Mineral Safety, 3rd Edition from the Council for Responsible Nutrition, IADSA, 2014.

In brief the report proposes the following advisory: Potassium is an essential element obviously required for a large number of physiological functions as an electrolyte and also for osmolar regulations. The cation is widely distributed in Food and manifestation of Potassium deficiency is well documented but when it comes to fixing the upper limit for Potassium both EFSA and the EVM group came to the conclusion that the available data is insufficient to establish upper limit but came with the rider that Potassium intake from Foods in healthy individuals should be of the order of 3-4 g per

day in adults and at any cost should not exceed 6 g per day and for the supplemental Potassium as KCI of about 3 g per day. This also comes with the rider that certain groups which are sensitive to increases in Potassium intakes in particular cases with impaired renal excretion of Potassium must go by medical advice and take the supplemental quantities under medical supervision. Most of the Potassium does come from Fruits and Vegetables and there is no discernible Scientific justification for the FDA threshold of 100 milligram Potassium for regulation of products such as drugs that require a prescription caution statement.

Recommendation:

Summary for Potassium TUL:

CRN UL, Supplemental intake	1,500 mg/day (500 mg, 3 times a day)
IOM UL, total intake	Not determined
EFSA UL, total intake	Not determined
EC Supplement maximum	Not determined
EVM, guidance level, supplemental intake	3,700 mg/day

Bibliography provided in Appendix 4.

8f. Iodine

A search was carried out using Pubmed, Cochrane Review and Google Scholar. The search terms included Iodine supplementation, side effects, adverse effects, adults, pregnant women.

Adults

Two hundred and twenty-five healthy women with underlying thyroid abnormalities were given 750µg iodine daily for 28 days. It was found that iodide intake of 750µg/day adversely affect thyroid function, especially in individuals with borderline hypothyroidism **(1).**

Pregnant women

A cross sectional study conducted on 1844 pregnant women who received 200µg/d and 100µg/day iodine supplementation during pregnancy. There was an increased risk of TSH above µU/mL in pregnant women who consumed 200µg or more of iodine supplements daily **(2).**

Recommendation:

In line with international recommendations, the TUL for iodine in adults is 900ug/day (0.9 mg).

Bibliography and Tables provided in Appendix 5.

8g. Vitamin A

A search was carried out using Pubmed, Cochrane Review and Google Scholar. The search terms included Vitamin A supplementation, side effects, adverse effects, Liver abnormalities, infants, children adults, and pregnant women, elderly.

Infants

One hundred sixty seven infants under 6 months of age who received 3 doses of 25000 IU of vitamin A at 6.5, 11.8 and 17.0 weeks of age had the higher incidence of bulging of the fontanelle **(1) .**

Children

A single dose of 500 000 IU of Vitamin A showed adverse effects like vomiting and more than 30 children died **(2).**

Studies conducted in Indonesia, Brazil, India, Ghana, Congo, Mexico, USA and Canada on 33,179 children upto 7 years of age who were given mega dose of 100,000 t0 200,000 IU Vitamin A concluded that the megadose had the higher incidence of LRTI in normal weight children **(3).**

Adults

A study which included 41 subjects who were given 25,000 IU for 6 years and 100,000 IU for 2 years and 6 months found cirrhosis in 17 subjects, mild chronic hepatitis in 10, noncirrhotic portal hypertension in 5 and increased storage in 9 subjects **(6).**

Pregnant Women

During pregnancy (n=22,748) 15,000 IU of preformed Vitamin A from food and supplements and 10,000 IU from supplements showed the higher cranial-neural-crest-defects among the babies born to women who consumed high levels of Vitamin A **(9)**.

Elderly

Administration of 25000 IU daily for 4 years had significant difference in alkaline phosphatase, triacylglycerol and after 49 months of follow-up, alkaline phosphatase was 7% higher, triacylglycerol was 11% higher, cholesterol was 3% higher, and HDL was 1% lower **(10).**

Recommendation:

In line with international recommendations, the TUL for Vitamin A in adults is 3000 ug/dy.

Bibliography and Tables provided in Appendix 6.

8h. Vitamin D

When the ULs for vitamin D were originally established in 1997, it was noted that the available data were limited relative to adverse outcomes and dose–response relationships. The adverse effects were considered primarily in terms of acute toxicity, which was defined as the condition of hypercalcemia or, in some cases, hypercalciuria with or without hypercalcemia.

Although information on chronic excess intake is limited, data have emerged recently that caution us about increased levels of vitamin D. Diet, supplements and sun exposure are known to impact Vitamin D status. The long-term effects of high intakes that are less than the toxic levels but may result in an increase in serum 25-hydroxyvitamin D (25OHD) levels into upper ranges previously considered to be physiologically safe are yet to be studied. It is also required to compare the effects at these high physiological levels of 25OHD achieved through supplementation versus sun exposure, and further research is needed to clarify the relative adverse effects of different sources of vitamin D (1).

Indicators of adverse outcome ofexcess intake of Vitamin D

- Intoxication and related hypercalcemia and hypercalciuria
- Serum calcium
- Measures in infants include retarded growth, hypercalcemia
- Emerging evidence for all-cause mortality, cancer, cardiovascular risk, falls and fractures

Vitamin D toxicity generally presents with non-specific symptoms that may vary and it often includes anorexia, weight loss, polyuria, and heart arrhythmia. The condition eventually leads to vascular and tissue calcification with subsequent renal and cardiovascular damage. Although research data supports the viewpoint that the biomarker plasma 25(OH)D concentration must rise above 750 nmol/L to produce vitamin D toxicity, the more prudent upper limit of 250 nmol/L might be retained to ensure a bigger safety margin (2).

Increased serum 25OHD levels and resulting hypercalcemia are the hallmarks of vitamin D toxicity. Although intakes of either vitamin D_2 or vitamin D_3 can cause toxicity, there is evidence that higher levels of vitamin D_2 can be tolerated in humans. Similarly,

in laboratory animal experiments, vitamin D_3 has been reported to be more toxic (Roborgh and de Man, 1960).

Recommendation:

The TUL for Vitamin D for adults is 100 ug/day.

Evidence and bibliography provided in Appendix 7.

8i. Vitamin E

Vitamin E comprises four tocopherols (α, β, γ, δ) and four tocotrienols (also α, β, γ, δ). α-tocopherol constitutes 90% of them and shows the strongest biological activity. Vitamin E (α-tocopherol) is recognised as the key, essential lipophilic antioxidant in humans protecting lipoproteins, PUFA, cellular and intra-cellular membranes from damage (FAO 2001; Raederstorff *et al* 2015). Many clinical trials with vitamin E involving subjects with various diseases have not shown any consistent pattern of adverse effects at any intake level of vitamin E.

Epidemiological Studies on Vitamin E supplementation in Humans

Vitamin E Clinical trials: The larger clinical trials that have tested the effect of α-tocopherol on Cardiovascular events in different populations

ATBC Study: The α-Tocopherol-Carotene Cancer Prevention Study (1994). Among 29,133 male smokers in Finland, aged 50 – 69 y, supplementation with vitamin E at 50 mg/d (all-rac--tocopheryl acetate, and therefore 50 IU/d) for 5– 8 yshowed an increase in the numbers of deaths from haemorrhagic stroke among male smokers. Although the number of haemorrhagic stroke cases with 50 mg α-tocopherol was 66 compared to 44 in the control group (total n = 29,133) no statistical significance was observed. A more recent analysis of this study indicated that there was an increased risk of sub-arachnoidal hemorrhage in hypertensive men (RR 2.45; CI 1.08-5.55) and a significantly higher mortality.

HOPE Study: The Heart Outcomes Prevention Evaluation Study (Yusuf et al 2000; Johannes et al 2004;) was an evaluation of the effects of angiotensin-converting enzyme inhibitor ramipril, vitamin E given at 400 IU/d, or both in 9541 patients with multiple CVD risk factors.The HOPE study investigators concluded that vitamin E was "well tolerated" because the number of adverse events with the treatment was not significantly greater than that with the placebo over the mean follow-up of 4.5 years.

SPACE Study: The effect of high-dose vitamin E supplementation on cardiovascular disease outcomes in haemodialysis patients with pre-existing cardiovascular disease was investigated (Boaz et al 2000). Haemodialysis patients with pre-existing cardiovascular

disease (n=196) aged 40-75 years at baseline from six dialysis centres were enrolled and randomised to receive 800 IU/day vitamin E or matching placebo. Patients were followed for 519 days. The primary endpoint was a composite variable consisting of: myocardial infarction (fatal and non-fatal), ischaemic stroke, peripheral vascular disease (excluding the arteriovenous fistula), and unstable angina. Secondary outcomes included each of the component outcomes, total mortality, and cardiovascular-disease mortality. In haemodialysis patients with prevalent cardiovascular disease, supplementation with 800 IU/day vitamin E was shown to reduce composite cardiovascular disease endpoints and myocardial infarction.

CHAOS Study: In Cambridge Heart Antioxidant Study (Stephens *et al* 1996), 2002 patients with symptomatic and angiographic CVD were randomly assigned to receive placebo or vitamin E at 400 or 800 IU/d. Over a median follow-up of 510 d, no significant adverse effects of vitamin E supplementation were reported among these patients. The slight numerical excesses of fatal myocardial infarction and total deaths with vitamin E treatment were not statistically significant

ASAP study: Antioxidant Supplementation in Atherosclerosis Prevention (ASAP) study (Salonen *et al* 2000) showed that a combined supplementation with reasonable doses of both vitamin E and slow-release vitamin C can retard the progression of common carotid atherosclerosis in men. This may imply benefits with regard to other atherosclerosis-based events.

GISSI Study: Dietary supplementation with n-3 polyunsaturated fatty acids and vitamin E after myocardial infarction: results of the GISSI-Prevenzione trial (1999). Dietary supplementation with n-3 PUFA but not vitamin E led to a clinically important and statistically significant benefit in patients who had myocardial infarction.

DATATOP (Parkinson Study Group 1998). The DATATOP clinical trial, which followed 800 subjects for 8.2 years, found no adverse effects of 2000 IU of vitamin E per day. This study supports the safety of very high intakes of vitamin E over a long period.
Several frequently cited literature reviews meticulously document the very consistent absence of adverse effects of vitamin E at intakes well above the RDA. The cited research includes randomized, double-blind, placebo-controlled trials with large patient populations. Kappus and Diplock (1992) observed that many scientifically reliable studies showed no consistently significant adverse effects associated with vitamin E supplementation at intakes ranging up to 3200 IU/d. A recent meta-analysis of 19 clinical trials of vitamin E supplementation for various diseases, including heart disease,

end-stage renal failure, and Alzheimer disease has shown that adults who took supplements of 400 IU/d were 6% more likely to die of any cause than those who did not take vitamin E supplements. However, further breakdown of the risk by vitamin E dose and adjustment for other vitamin and mineral supplements found that the increased risk of death was significant only at a dose of 2000 IU/d, which is higher than the UL for adults. Furthermore, 3 other meta-analyses that combined the results of randomized controlled trials designed to evaluate the efficacy of vitamin E supplementation for the prevention or treatment of cardiovascular disease (CVD) found no evidence that vitamin E supplementation up to 800 IU/d significantly increased or decreased CVD mortality or all-cause mortality. At present, the evidence is not convincing that vitamin E supplementation up to the UL increases the risk of death due to CVD or other causes.

Double blind control studies with oral vitamin E in humans*:* Controlled double blind studies on vitamin E toxicity in humans reveal that vitamin E has low toxicity and no consistent adverse effects (**Appendix 8**).

European Commission, Scientific Committee on Food (EC SCF 2003). The EC SCF reviewed all available evidence and found no adverse effects of oral vitamin E in humans. Declaring the evidence at higher intakes to be insufficient, the EC SCF selected the clinical study by Meydani and colleagues (1998) to identify a NOAEL of 800 IU per day, or approximately 540 mg per day. Judging the database to be only moderately robust, the EC SCF applied a UF of 2, converting from IU to mg to derive a UL of 270 mg per day, rounded up to 300 mg per day.

Recommendation:
Quantitative Summary for Vitamin E TUL

CRN UL, supplemental intake	1,000 mg (1600 IU)/day
IOM UL, total intake	1,000 mg/day
EC SCF UL,	300 mg/day
EC supplement maximum	Not determined
EVM SUL, supplemental intake	540 mg (800 IU)/day

EXCERPTED FROM: Vitamin and Mineral Safety 3rd Edition (2013) Council for Responsible Nutrition (CRN) www.crnusa.org. CRN - Council for Responsible Nutrition: IOM; Institute of Medicine (IOM). 2000: EC SCF - European Commission, Scientific Committee on Food (EC SCF 2003).

CRN identified a vitamin E UL of 1,600 IU from clinical trial data that showed no adverse effects at that level of intake (Gillilan et al. 1977). Correspondingly, CRN considers 1,600 IU as the upper limit to have a very low level of uncertainty because of the absence of adverse effects at the higher intake of 3,200 IU (Anderson and Reid 1974). With the

conversion to mg alpha-TE as performed by the EVM, the CRN upper limit for supplements of 1,600 IU is equivalent to 1,073 mg, a value very similar to that identified by the IOM through extrapolation from animal data. The CRN upper limit for supplements applies to healthy adults who are not taking any anticoagulant drug. In line with international recommendations, the Vitamin E TUL for adults is 1000 mg/day.

Evidence and Bibliography provided in Appendix 8.

8j. B vitamins

The TUL considerations for some of the B vitamins are presented below. According to the report of the Institute of Medicine (IOM), TUL has not been determined for some B-vitamins (1). However, for the record, information on other B-vitamins as reported in some Indian and other studies are summarized below and hence, there is a need to develop improved methods for estimating intakes of micronutrients from fortified foods and food supplements in future dietary surveys.

Evidence(Tables) and bibliography for all B vitamins provided in Appendix 9.

i) Thiamine

In the case of vitamin B1 (thiamine), while a few studies in India used 1-3 mg/ day involving 5-15 year boys/ girls or women (refer to the Table, B1-Indian Studies), other studies have used 100-600 mg/day on adults under various conditions.

Recommendation: No TUL identified

ii) Riboflavin

While some studies in India used 1-3 mg/ day of vitamin B2 (riboflavin) involving 5-15 year boys/ girls or women (Table: B2-Indian Studies), other studies used 60-400 mg/day on adults under various conditions.

Recommendation: No TUL identified

iii) Pyridoxine

A few studies in India used 1-10 mg/ day of vitamin B6 (pyridoxine) involving 5-15 year boys/ girls or women (Table: B6-Indian Studies).

Recommendation: Adult TUL of 100 mg/day

iv) Biotin

A study in India used 10 µg of Biotin on healthy boys and girls between the ages of 7 and 11 yrs (Table: Biotin-Indian Studies) and studies elsewhere used up to 1 mg/day on adults (Table: Biotin-Other Studies).

Recommendation: No TUL identified

v) Niacin

While the studies from elsewhere reported using 0.5-6.0 g/day of Niacin (Table: Niacin-Other Studies), a few studies in India used 1-15 mg/day on 5-15 year boys/ girls or women (Table: Niacin-Indian Studies).

Recommendation:TUL for adults of 35 mg/day

vi) Folic Acid

The Folic Acid Sub-committee of the Food and Drug Administration (FDA) (2) first suggested in 1993 that the upper limit should be 1 mg per day in adults including the elderly, and this conclusion has been echoed by subsequent national or international advisory committees. The most thorough review of the literature was by the Institute of Medicine (IOM) in 1998, which conclude that excessive folate intake may precipitate or exacerbate neuropathy in vitamin B12-deficient individuals (1). In a recent review, Reynolds (3) reviewed the Reports of four Expert Advisory Committees in Europe and the USA, which also suggest that the safe upper tolerable limit (UL) for folic acid is 1 mg in adults. However, according to Reynolds these reports are unsound and there is already evidence of neurological harm from long-term exposure to doses of folic acid between 0.5 and 1 mg in the presence of vitamin B12 deficiency. Therefore, he suggests that there is an urgent need to review the safe TUL for folic acid and to consider the addition of vitamin B12 to folic acid fortification policies. Studies from India on adolescent boys/ girls and pregnant women used 0.5 mg folic acid per day (please refer to the Table, B9-Indian Studies).

Recommendation: TUL for adults of 1000 ug/day

vii) Vitamin B12

In the case of vitamin B12, studies used 500 µg on non-pregnant vegetarian women and 1-10 µg on children (Table: B12-Indian Studies).

Recommendation: No TUL identified

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2. Food and Drug Administration. Food labelling: health claims and label statements; folate and neural tube defects. Federal Register 1993; 58: 53254–53270.
3. EH Reynolds. What is the safe upper intake level of folic acid for the nervous system? Implications for folic acid fortification policies. Eur J Clin Nutr. (2016), 1–4. Advance online publication, 10 February 2016; doi:10.1038/ejcn.2015.231

8k. Vitamin C

Literature search on the safety concerns on high dose vitamin C intakes in humans including general population in India has not yielded any evidence. The literature presented in the table (Appendix 10) is global evidence and indicates that adverse effects of consumption of high dose vitamin C, including supplements providing 4-10 g/day is negligible.

The Food and Nutrition Board, Institute of Medicine USA (FNB, IOM, 2000) selected osmotic diarrhea and related GI disturbances as critical end points to formulate the UTL for vitamin C for apparently healthy individuals. A LOEAL of 3g/d was based on reported GI symptoms (transient colic, flatulent distension and diarrhoea) on ingestion of 3-4 g /d of vitamin C in normal healthy volunteers (Camron and Campbell, 1974) supported by evidence from graded dose study (8g/d in 4 divided doses for 3-7 days), with 1 in 3 persons reporting mild diarrhea (Stein et, al 1976) and the cross over study (1, 5, 10 g/d for 5 days), 2 in 15 developed diarrhea at 10g/d intake (Wandzilak et al 1994). Because of little uncertainty on the range of vitamin C intake to induce osmotic diarrhea, an uncertainty factor (UF) of 1.5 was selected to derive an NOEAL/UTL of 2g/d by extrapolating the LOEAL [3 (g/d)/1.5 = 2g/d] for apparently healthy adults (19 y and older).

Recommendation:
TUL for Vitamin C, (FNB, IOM, 2000)

Age/Physiological group	UTL (mg/d)	Age/Physiological group	UTL (mg/d)
0-12 mo infants	Not possible	19 y and older adults	2000
1-3 y children	400	14-18y adolescent pregnant women	1800
4-8 y children	650	19 y and older adult pregnant women	2000
9-13 y children	1200	14-18y adolescent lactating women	1800
14-18 y adolescents	1800	19 y and older adult lactating women	2000

The TUL values rounded off to the nearest 50mg for toddlers, children and adolescents is extrapolated based on body weight differences from that of adults using reference weights (Table). Due to lack of convincing evidence on adverse effects of excess vitamin C during pregnancy and lactation, their UTL values are not different from those of non-pregnantnon lactating adolescent and adult women.

Recommendation: TUL for adults of 2000 mg/day

Table of evidence and bibliography provided in Appendix 10.

	Nutrient	Children (6-9 y)	Adolescent (9-17 y)	Pregnant/ Lactating women	Adults	Remarks
1.	Iron (mg/d)	40	40	45	45	
2.	Zinc (mg/d)	12	30	40	40	
3.	Iodine (µg/d)	200	600	1100	900	
4.	Vitamin A (µg/d)	600 (1-3) 1700 (10-12y)	2800	3000	3000	
5.	Vitamin D (µg/d)	75	100	100	100	
6.	Vitamin E (mg/d)	200 (1-9y)	800	1000	1000	
7.	Calcium (mg/d)	1500 (1-3y) 2500 (4-9y)	3000	2500	2500	
8.	Thiamine (mg/d)	No Tolerable Upper Limits found on the data available				
9.	Riboflavin	No Tolerable Upper Limits found on the data available				
10.	Pyridoxine (mg/d)	40	80	100	100	
11.	Biotin (µg/d)	No Tolerable Upper Limits found on the data available				
12.	Niacin (mg/d)	15	30	35	35	Therapeutic dose – reported from Indonesia – 1000 µg/d
13.	Folic acid (µg/d as Folate equivalent)	300	600-800	1000	1000	Folate together with B12 and iron may have adverse effects in the long term. Hence the recommended TUL to be viewed with caution.
14.	Vitamin B12 (mg/d)	No Tolerable Upper Limits found on the data available				
15.	Vitamin C (mg/d)	400-650	1200-1800	2000	2000	TUL data to be viewed with greater caution.

9.Summary of TUL values

Conversion units
Vitamin A: 1 µg = 3.33 IU
Vitamin D: 1 µg = 40 IU
Vitamin E: 1 mg = 1.5 IU d-alpha-tocopherol, or 1.1 IU dl-alpha-tocopherol.
Folic Acid: 1 µg = 1.7 DFE (Dietary folate equivalent)

10. Suggestions for future perspectives

Exposure considerations: (On specific micronutrients: Vitamins and Minerals)

The several surveys conducted from NNMB and other agencies over the period 2008 – 2015 have clearly indicated that there is no clarity in the analysis and collection of data on diet and other sources, so that an unambiguous decision and recommendations can be arrived at, regarding the exposure considerations (for Indian Population at individual / household levels). For example, there are no data on Vitamin B12 or Vitamin D3 intakes, and in some cases, the distributions were very skewed, and the relative magnitudes of the standard deviation and the mean suggested that a transformation of the observed values to some other scale (e.g. logarithmic) might normalize the data and lead to better criteria. Hence the Committee strongly felt that keeping in view the constraints of the Report to address the three TORs for TUL, it is desirable to have a larger data set, and well spread out models; along with more meta-analyses from primary data from the NNMB, to arrive at any final conclusive values on *Exposure considerations*. It suggests that more work in this area be taken up.

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APPENDIXES

Appendix 1: Iron

Literature search on side-effects of excess iron supplementation - Indian Studies. The side effect of excess iron supplementation considered was GI related.

Study & Population (n)	Features	Dosage and Study Design	Duration (days)	Key Observations	NOAEL Considerations
Periara *et al* 1977 N=44	2-5 y	1st study: Expgp: 20mg Iron +5µg B12+200µg folic acid 2nd study: Exp gp:30mg iron +5µg B12+200µg folic acid. In both studies, control group received 5µg B12+200µg folic acid (Iron Source: ferrous sulphate)	1st study: 7 months 2nd study: 12 months	Fe supplemented children showed a significant increase in Hb, serum iron, % saturation of transferrin and decrease in iron binding capacity. Illness: Conjunctivitis was epidemic during study period, more affected children & recurrent attacks were observed in iron supplemented group. But authors were unsure if that could be linked to iron.	20 and 30 mg does not contradict the recommended UL of 40 mg of IOM
Vijayaraghavan *et al* 1990 n=5754 households (6 districts)	Pregnant women of 20 weeks of gestation (n=487, Hb estimation)	Program evaluation 60 mg iron +500 µg folic acid (adult) 20 mg ferrous +100 µg folic acid/ 2 ml Folifer liquid (child)	100 continuous days	Poor coverage: Only 19% pregnant women, 17% lactating women, >1% children received folifer tablets. 11% received > 60 tablets 64% received <30 tablets (this was due to irregular supply (79%) and side effects (10%)) Side effects: GI upset including constipation, nausea, vomiting Hb status % Anemic persons declined from 93% (<25 tablets) to 83% (>50 tablets)	60 mg in pregnant women does contradict the recommended UL of 45 mg
Shatrugna	Pregnant	Tolerance test:	Single dose	Liquid	Does

et al 1992 n=115	women (20th - 28th week of pregnancy)	Ferrous sulphate tablet Gp 1:60 mg Gp 2:120 mg, Gp 3: 180 mg Gp4:Pure ferrous sulpahte salt (60mg) Gp 5: Ferrous fumarate tablet(60mg) Gp6:syrup (60 mg) Gp7:Ferroussulpahte salt (60mg) with excipients Gp8:Ferroussuphate tablets powdered 60 mg Gp9:NNAPP tablet (60mg) Gp 10: Gelatin capsule (60 mg) All preparations had 500 µg folic acid.	study	formulations of iron had a better bioavailability, with ferrous fumarate syrup and gelatin capsules being the most superior. Increasing the dose 60 mg to 180 mg improved the bioavailability of iron, but was associated with unacceptable side-effects such as vomiting, nausea and Diarrhoea. 40% of the subjects given 180 mg of iron could not tolerate the tablet.	contradict the recommended UL of 45 mg
Shobha & Sharada 2003, n=244	13-15 y Adolescent girls [anemic (n=203) & non-anemic (n=41)]	Gp 1: 60mg iron daily + 0.5mg of folic acid Gp 2: 60 mg iron twice weekly + 0.5mg of folic acid. [3 hours after lunch and 3 hours before dinner]	12 weeks (84 days)	The Hb level increased in both groups. Twice weekly supplementation of iron is as efficacious as daily supplementation in adolescent girls. Side effects: Daily dose-abdominal pain (42%),nausea (10%), vomiting (5%) Weekly dose-abdominal pain (5%),nausea (1%)	Does contradict the recommended UL of 45 mg
Sharma *et al* 2004 N=254	Moderate anemic Pregnant women	Gp 1: oral dose of 100 mg elemental iron + 500µg folic acid	100 days	Hb& iron indicators improved sig. Side effects:	Does contradict the recommende

	(16-24 weeks of gestation)			Oral iron gp: GI (Dyspepsia (10%), constipation (5%), diarrhea (3%) and vomiting (2%) were seen .	d UL of 45 mg
Joshi *et al* 2013 N=120	Anemic Adolescent girls (10-19 y)	RCT: Gp 1: daily iron + folic acid Gp 2: weekly iron + folic acid [Capsule: hard gelatine capsule containing Ferrous Fumerate IP 300 mg; Vitamin B12, in gelatine, 15 mcg; Folic Acid IP 1.5 mg)]	3 months	Hb: Mean rise was 1 gm/dl in both groups Compliance: 1.3±3.15 (weekly) 6.1±10.98 (daily) Adverse effect: abdominal pain 8.3% (weekly) 13.35% (daily)	Does contradict the recommended UL of 45 mg

Legend: IOM -TUL for iron 0-13 y is 40 mg/d and for 14->70 y is 45 mg/d
http://www.nationalacademies.org/hmd/Activities/Nutrition/SummaryDRIs/~/media/Files/Activity%20Files/Nutrition/DRIs/ULs%20for%20Vitamins%20and%20Elements.pdf

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Appendix 2: Zinc

Side effects of excess zinc supplementation -Indian Studies. The side effect of excess zinc supplementation considered was lowering of Cu status.

Study & Population	Features	Dosage and Study Design	Duration (days)	Key Observations	NOAEL considerations
Sazawal *et.al*	Infant (5-36mo). N=115 Zinc Gr n=61 Control n=54	10 mg of elemental zinc	120 d	Post-supplementation Zn was significantly higher (93.0 ± 3.6 vs C 60.6 ± 8.0) in the Zn group. There was no significant difference in the mean/median copper levels between the zinc and control groups.	10 mg does not contradict the recommended UL of 5-7 mg
Bhandari *et .al*	RCT, Delhi 6-30 mo with severe recurrent diarrhoea Zn gr n=1093, placebo n=1133	zinc gluconate (elemental zinc 10 mg to infants and 20 mg to older children) 2XRDA (high phytate)	4 mo	Small but significant increase in the average number of days with vomiting in the zinc group (4.3 [SD 5.8] vs 2.6 [SD 3.9] days. 8 children (0.3%), all in the zinc group, reported vomiting immediately after the supplement on each day during the first 2 weeks of supplementation. Mean Cu levels were substantially lower in the zinc group (difference in means: -15.5 ug/dL; 95% CI: -19.9 to -11.1).The proportion of children at the end of the study who had plasma Cu <80 g/dL was 4.8% in the Zn compared with 0.6% in the placebo group (difference in proportions: 4.2%; 95% CI:2%–6.2%).	10/20 mg does contradict the recommended UL of 5-7mg Toxicity observed
Hemalatha *et.al*	Severely malnourished children(n=33)	40 mg of Zn		Plasma copper showed a significant improvement in both zinc supplemented ($P < 0.01$) and placebo ($P < 0.025$) groups.	Does not contradict the recommended UL of 5-7 mg
KV Radhakrishna *et.al*	infants 4-18 mo, Zn150 or placebo 149	5mg Zn +0.5 mg Riboflavin	190 days	Mean Hb, serum Zn, Cu and vitamin A levels were similar between groups; Zn deficiency (<,60 mg/dl) prevalent in lesser proportion (26.5%) of Zn group compared to	5 mg Zn does not contradict the recommended UL of 5-7mg

				placebo group (44.1%)	
Archana P*et.al*	Children 6-59 Mo Placebo Gr = 239 Zn Gr =224 Zn+Cu Gr = 253	ZnSO4 equivalent to 20 mg/5 ml of elemental zinc, For Zn+Cu, CuSO4 equivalent to 2 mg/5 ml elemental copper	2 weeks	Mean absolute difference from baseline in Cu : Placebo -79.4 ± 429.2, Zinc -41.2 ± 418.8, Zn+Cu 15.6 ± 439.8.	
Kumar A *et.al*	Neonates with idiopathic neonatal hyperbilirubinemia Placebo = 33 Zn Gr. = 40	zinc sulfate (10 mg/d) or placebo	7 days	At the end of intervention, serum zinc levels were significantly higher in the zinc supplemented group while serum copper levels were comparable between the two groups. The adverse events were comparable between the two groups (vomiting 3 vs. 2; skin rash 3 vs. 3; diarrhea 4 vs. 3; excessive cry 2 vs. 2 in placebo and zinc group, respectively). Post-intervention serum copper levels (µg/dL) : Placebo 48.6 ± 15.2 ; Zn Gr. 46.2 ± 16.0 ; Mean Difference -2.468 (-10.214, 5.279) P value 0.527	Neonates with idiopathic neonatal hyperbilirubinemia
Lodha R *et.al*	Children 6 mo to 15 y of age with newly diagnosed intrathoracic tuberculosis.	20 mg elemental zinc and placebo	6 months	Serum copper (mg/dL) Level : Month 0 -Zn Gr. 146.5 (141.9, 151.0) ; +Zn Gr. 147.0 (142.5, 151.5) Month 6 -Zn Gr. 112.3 (107.6, 116.9) ; +Zn Gr. 108.3 (103.6, 113.0) Change -Zn Gr. - 34.2 (-39.7, -28.8) ; +Zn Gr. -38.7 (-44.3, -33.2) Difference -4.5(-12.3, 3.3), *P* 0.26	Children with intrathoracic tuberculosis
Zaka-ur-	3-60 mo	RDA Zinc	2 weeks	Decline in Fe status	5 mg Zn showed

Rab Z *et.al*	single arm N=62	gluconate dose			reduction in iron status.

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Appendix 3: Calcium

Relevant Indian studies on calcium supplementation

Study & Population (n)	Features	Dosage and Study Design	Duration (days)	Key Observations	NOAEL considerations
(10) n= 173	Healthy young women	Calcium 1000 mg/day + Vitamin D 60,000 IU/week for 8 week followed by 60,000 IU/fortnight (dietary calcium intake was 578 ± 259 mg/day) Randomized Controlled Trial	6 months	Serum calcium and urinary calcium /creatinine ratio did not change 11% participants had hypercalcemia (serum total calcium adjusted for albumin>10.6 mg/dl), 2 participants had severe Hypercalcemia (serum calcium > 11 mg/dl) 9% participants had hypercalciuria (Urinary calcium/creatinine ratio higher than 0.2 mg/mg)	Hypercalciuria observed at total calcium intake 2100 mg/day contradicts the recommended UL of 2500 mg/day
(11) N =48	Non-diabetic adults with vitamin D deficiency	Supplementation with Calcium 1000 mg + Cholecalciferol 9,570 IU/day	5 months	Serum calcium concentration reduced from 9.11 ± 0.45 mg/dl to 8.73 ± 1.01 mg/dl ($p<0.05$). Urinary calcium excretion was not assessed.	Does not contradict the recommended UL of 2500 mg/day.
(12) N=40	Healthy volunteers 24M/16F	Supplementation with Calcium 1 g + Vitamin D (60 000 IU D3/week for 8 weeks followed by 60 000 IU/month for 4 months)	6 months	Serum calcium adjusted for albumin did not differ in relation to calcium supplementation.	Does not contradict the recommended UL of 2500 mg/day.
(13) N=29	Healthy volunteers	'Calcium load test' with 1 g of oral elemental calcium	One dose after 8 weeks of cholecalciferol supplementation	Mean urinary calcium /creatinine ratio of the study subjects increased from 0.030 +/- 0.024 mg /mg under fasting conditions to 0.059 +/- 0.045 mg /mg	Does not contradict the recommended UL of 2500 mg/day.

				after calcium loading (delta change = 96.6%, P = 0.008)	
(14) N=214	Pre-menarchal girls	Calcium 500 mg/day with multivitamins including zinc 15 mg/day	1 year	Serum ionized calcium increased from1.03 nmol/l at baseline to 1.16 nmol/l post-supplementation.	Does not contradict the recommended UL of 2500 mg/day.
(15)	Young children (age 2.7±0.52 years)	Calcium supplement 405 mg/day + vitamin D 30000 IU per month	1 year	Serum ionized calcium increased from 0.91 mmol/l to 1.08 mmol/l after supplementation.	Does not contradict the recommended UL.

Possible considerations in the Indian context:

1. A few studies on calcium supplementation have been reported with no studies on long term follow up after calcium supplementation.
2. High oxalate content of the plant based diets may increase risk of nephrolithiasis.
3. Calcium supplementation may interfere with absorption of zinc and iron and may exacerbate their deficiency.

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Appendix 4: Potassium and Sodium

EFSA, Opinion of the Scientific Panel on Dietetic Products, Nutrition and allergens and (TUL from Sodium) The EFSA Journal, *209,* 1 – 26, (2005)

EGVM (Expert Group on Vitamins and Minerals) Report on safe upper levels for vitamins and minerals. London. May 2003. Available on the Internet at : http://www.foodstandards.gov.uk/multimedia/pdfs/vitamin2003.pdf(2003).

National Academies of Science, Dietary Reference Intake from Water, Potassium, Sodium, Chloride and Sulphate published by Standing Committee on the Safety Evaluation of Dietary Reference Intakes, FNB, Institute of Medicine of the National Academies The National Academies, Press, Washington DC, USA (www.nap.edu), (P.1-639) (2005).

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WHO (World Health Organization) Food and Health in Europe: a new basis for action. WHO Regional Publications, European Series No.96. (2004).

WHO Guideline. Sodium Intake from adults and children, Geneva, Switzerland (2012)

WHO/FAO (World Health Organization/Food and Agriculture Organization) Diet, nutrition and the prevention of chronic diseases. WHO Technical Report Series 916. World Health Organization, Geneva, Switzerland (2003).

Appendix 5: Iodine

Study & population (n)	Features and study population	ICMR, RDA µg/kg/d	Dosage & study	Duration (days)	Key observations
Martinez TA, Bozkaya D, and Yurdakök M. Accidental oral administration of povidone iodine in a newborn: case report. Arch Argent Pediatr 2016;114. n=1	8 day old female (newborn)	6-30	Two teaspoons of povidone iodine accidentally given	2000 mg of elemental iodine (Single dose)	Showing no signs or symptoms of toxicity after ingestion On the 15th day of life, the baby was healthy and the thyroid function tests were within normal range.
Chow CC, Phllllps DIW, Lazarus JH and Parker AB. Effect of low dose Iodide supplementation on thyroid function in potentially susceptible subjects: are dietary iodide levels in Britain acceptable? Clinical Endocrinology 1991;34:413-416. n=225 women	Healthy women and women with underlying thyroid abnormalities	2	A placebo controlled trial- daily approx 750 µg/d for 28 days	28 DAYS	Serum concentrations of FT4 were significantly decreased and serum TSH concentrations were significantly elevated in the women who received the iodide supplements, relative to a placebo control group. Dietary Iodide Intakes of 750 pg/day or more may adversely affect thyroid function, especially In individuals wlth borderline hypothyroidism.

Paul T, Meyers B, Witorsch RJ, Pino S, Chipkin S, Ingbar SH, Braverman LE. The effect of small increases in dietary iodine on thyroid function in euthyroid subjects. *Metabolism* 1988;37:121–124. n=18	Healthy euthyroid adults Males =9 Females =9	2	Received daily oral doses of 250, 500, or 1,500 µg I/day as sodium iodide for 14 days	14 DAYS	Those receiving 1,500 µg/day of iodide showed a significant increase in baseline and TRH-stimulated serum TSH Effects not seen in the two lower doses. The conclusion would be that an iodine intake of about 1,700 µg/ day increased TSH secretion.
Pearce EN, Gerber AR ,Gootnick DB , Khan LK ,Ruowei LI, Pino S, Braverman LE. Effects of Chronic Iodine Excess in a Cohort of Long- Term American Workers in West Africa. The Journal of Clinical Endocrinology & Metabolism 2002; 87(12):5499–5502 n=93	Adult (predominantly were female 75%) Mean age (25.5± 3.0 year)	2	Mean concentration of 10mg iodine/liter to the drinking water No evidence of a high concentration of iodine in the diet, salt, medications, or nutritional supplements. Subjects consumed at least 50mg Iodine daily	30 weeks	During prolonged excess iodine exposure there was marked increases in serum total iodine concentration and the prevalence of goiter, elevated serum TSH values, and elevated serum Thyroid peroxidase antibody values increased. Prevalence of abnormalities decreased after removal of excess iodine.

Rebagliato M, Murcia M, Espada M, Alvarez-Pedrerol M, Bolumar F, Vioque J, Basterrechea M, Blarduni E, Ramon R, Guxens M, Foradada CM, Ballester F, Ibarluzea J, Sunyer J. Iodine intake and maternal thyroid function during pregnancy. Epidemiology 2010;21:62-9. n=1844	Pregnant women (gestational age range 8–23 weeks) was carried out in 3 areas in Spain	4.5	Cross sectional study Women received 200µg/d and 100µg/day iodine supplements	DURING PREGNANCY	Iodine supplement intake in the first half of pregnancy may lead to maternal thyroid dysfunction in iodine-sufficient or mildly iodine-deficient populations
Connelly KJ,Boston BA, Pearce EN, Sesser D, Snyder D,Braverman LE,Pino S and LaFranchi SH. Congenital Hypothyroidism Caused by Excess Prenatal Maternal Iodine Ingestion. J Pediatr. 2012 October ; 161(4): 760–762. n=3	Infants	6-30	Excess maternal iodine ingestion-12.5mg/day during pregnancy	DURING PREGNANCY	Congenital Hypothyroidism detected among infants. Elevated iodine level in urine and breast milk sample.
Laurberg P, Pedersen KM, Hreidarsson A, Sigfusson N, Iversen E, Knudsen PR. 1998. Iodine intake and the pattern of thyroid disorders: A comparative epidemiological study of thyroid abnormalities in the elderly in Iceland and in Jutland, Denmark. *J ClinEndocrinolMetab 83*:765–769 n=423 (Jutland and Denmark) n=100 (Ice land)	Elderly adults	2	Iodine intake of residents of Jutland and Denmark = 40-60µg/day Iodine intake of residents of Iceland= 300-350µg/d		Thyroid abnormalities in population with low iodine intake and those with high iodine intake develop in opposite directions. Goiter and thyroid hyperfunction when iodine intake is relatively low. Impaired thyroid function when iodine intake is high.

http://www.ign.org/cm_data/2004_WHO_Iodine_in_Vitamin_and_mineral_requirements_chap16.pdf ACCESSED ON 29TH JULY 2016

ed dietary intakes of iodine and upper limits, by group

	Recommended intake (µg/kg/day)	Upper limit[a] (µg/kg/day)
ldren		
	30	100
	15	150
s	15	140
	6	50
	4	50
nd adults (13+ years)	2	30
en	3.5	40
en	3.5	40

from reference (18).

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Appendix 6: Vitamin A

Study & population (n)	Features and study population	ICMR , RDA Vitamin A(µg/d) Retinol	Dosage & study	Duration (days)	Key observations
Baqui AH, Francisco AD, Arifeen SE, Siddique AK, Sack RB. Bulging fontanelle after supplementation with of 25000 IU of Vitamin A in infancy using immunization contacts. ActaPaediatr 1995;84: 863-6. n=167	Infant (6.5, 11.8 and 17.0 weeks of age) 86 infants received Vitamin A and the remaining 81 received placebo.	350	Double-blind, randomized, placebo-controlled trial was conducted in Bangladesh. 167 infants received three doses of either 25 000 IU of vitamin A or a placebo at about 6.5, 11.8 and 17.0 weeks of age	3 dose of Vitamin A 25000 IU about 6.5,11.8 and 17.0 weeks of age	In total, there were 14 episodes of bulging of the fontanelle, 12 of which occurred in infants supplemented with vitamin A together with vaccine. Thus, the higher incidence of bulging of the fontanelle in the vitamin A group relative to the placebo group. Vitamin A dose: 25000 IU palmitate in peanut oil Placebo: soybean oil
West KP , Sommer A. Vitamin A programme in Assam probably caused hysteria. BMJ 2002;324:791	6 months-59 months	0-12 months -350 1-6 years-400	Received mega doses (500 000 IU) of Vitamin A	Single dose	A large number of children reported symptoms of side effects within a few hours after receiving the dose of vitamin A solution. About 953 children showed adverse effects like vomiting after administration of Vitamin A. >30 children died

Shah D. Does Vitamin A supplementation help in preventing pneumonia? Indian Pediatrics 2009; 46:403-404. n- 33,179	Children		Randomized controlled trials Mega dose trial- 100,000 to 200,000 IU vitamin A Low dose trial- 5,000 to 45,000 IU Vitamin A	Daily to every two months	Increased lower respiratory tract infection (LRTI) in normal weight children. Megadose of vitamin A failed to lower the incidence of acute LRTI.
ADULTS (MALE/FEMALE)					
Geubel AP, Galocsy CD, Alves N, Rahier J and Dive C. Liver damage caused by therapeutic Vitamin A administration : estimate of dose-related toxicity in 41 cases . Gastroenterology1991;100:1701-1709. n=41	Patient with Vitamin A hepatotoxicity Male =21 Female =20 (age not been mentioned)	600	Develop cirrhosis - 25,000 IU/day over 6 years and 100000 IU/day over 2 years and 6 months resulted similar histological lesions.	25,000IU- 6 years 100,000 IU-2 years and 6 months	Chronic Vitamin A consumption might cause of chronic liver disease. Prolonged and continuous consumption of doses in the low therapeutic range can result in life-threatening liver damage.
PREGNANT WOMEN					
Rothman KJ, Moore LL, Singer MR, Nguyen US, Mannino S, Milunsky A. Teratogenicity of high vitamin A intake. N Engl J Med. 1995 Nov 23;333(21):1369-73. n=22,748	Pregnant women	800	15,000 IU of preformed Vitamin A from food and supplements and 10,000 IU from supplements compared with baby's mothers consuming 5,000 IU	During pregnancy	Ratio of prevalence of defects associated with cranial-neural-crest tissue to be 4.8 for mothers who had consumed ≥3030 RE Vitamin A/d (10000 IU/d) compared with those who had consumed ≤ 1515 RE Vitamin A/d (5000 IU/d). They further suggested a threshold risk near an intake of 3030 RE Vitamin A/ d.
ELDERLY (AGE 60 YEARS AND ABOVE)					

Cartmel B, Moon TE, Levine N. Effect of long term intake of retinol on selected clinical and laboratory indexes. The American Journal of Clinical nutrition 1999 ;69 :937-43 n= 2297	Median age 63 year old men and women residing in Arizona	600	1157(50.4%) were randomly assigned to receive retinol 7576 RE/d and 1140(49.6%) to receive a placebo that contained vegetable oil.	49 months (4 years)	Significant differences in alkaline phosphatase (P< 0.0001), triacylglycerol (P<0.0001), cholesterol (P=0.04),and HDL(P=0.01) were observed over time between the 2 groups. After 49 mo of follow-up, alkaline phosphatase was 7% higher, tri-acylglycerol was 11% higher, cholesterol was 3% higher, and HDL was 1% lower in the retinol group than in the placebo group. Study reported 1% increase in cholesterol concentrations has been associated with a 2% increase in coronary artery disease risk, long-term ingestion of 7576 RE vitamin A/d should be considered with caution.
Minuk GY, Kelly JK, Hwang WS. Vitamin A hepatotoxicity in multiple family members. Hepatology 1988; 8: 272-5. n=3	Father – 62 year old Mother - 63 year old Son- 33 year old	600	vitamin A (20,000 to 45,000 IU per day)	Daily for 7 to 10 years	vitamin A hepatotoxicity occurring in multiple family members. Finding emphasizes that even moderate levels of vitamin A ingestion, when taken over prolonged periods of time, are capable of causing significant liver disease.

					A daily for 7-10 years developed nausea and weakness and were found to have liver test abnormalities [bilirubin 0.3-1.0 mg/dL, ALT 71-265 U/L, Alk P 76-258 U/L], resolving in 6-12 months on normal diet).

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Appendix 7: Vitamin D

Literature search on side effects of excess Vitamin D intake.

Study & population	Features	Dosage & study Design	Duration	Key observations	NOAEL consideration
Byrne et al., Calcif Tissue Int. 1995 Jun;56(6):518-20. n = 449	Elderly people	low-dose daily oral administration and intermittent high-dose administration	4-6 months	Mean levels of 25-OHD was 57 to 105 nmol/L in low dose daily vs 55 to 87 nmol/Lin high dose supplementation Hypercalcemia occurred in only 3 subjects and was associated with a predisposing cause in 2 of 3 subjects.	low dose continuous supplementation (10 to 20 micrograms daily OR high-dose intermittent supplementation (2.5 mg six monthly
Honkanen et al., J Am Geriatr Soc. 1990 Aug;38(8):862-6. 66 independently living and 73 institutionalized elderly women	Elderly women	45 micrograms (equal to 1,800 IU) of vitamin D administered daily	11 months (winter)	No group or individual side effect was observed	
Johnson et al., Age Ageing. 1980 May;9(2):121-7. n = 63	elderly	2000 IU	6 months	Two out of 63 individuals on vitamin D developed hypercalcaemia	Concluded that, although there appears to be improvement in the phosphate status of treated patients over the short term of this trial, hypercalcaemia after vitamin D administration precludes the continuous prophylactic use of vitamin D at the levels employed in this trial
Stamp et al., Lancet. 1977 Jun 25;1(8026):1341-3.		short-term and long-term oral treatment with 25-	About 5 years	Ten times more vitamin D than 25-OHD3 was required to produce equivalent plasma-25-OHD	

n = over 200		hydroxycholecalciferol (25-OHD3), or with vitamin D		concentrations	
Davie et al., ClinSci (Lond). 1982 Nov;63(5):461-72.	Normal and anti-convulsant treated subjects from England	measured u.v. irradiation applied thrice weekly	10 weeks	Plasma 25-(OH)D3 concentrations reached a steady state after 5-6 weeks of u.v irradiation or of oral intake within the usual intake range	
Schwartzman & Frank, Am J Med. 1987 Feb;82(2):224-30.	Patients with osteoporosis or osteomalacia	pharmacologic doses of Vit D		Data on the use of 25-OH D show no greater benefit than for vitamin D Hypervitaminosis D developed in four patients with osteoporosis or osteomalacia	Pharmacologic doses of vitamin D cannot be recommended for any form of osteoporosis
Davies & Adams Lancet. 1978 Sep 16;2(8090):621-3. n = 8					Large doses of vitamin D should only be used when strictly indicated and on the understanding that close biochemical and clinical supervision is necessary
Selby et al., ClinEndocrinol (Oxf). 1995 Nov;43(5):531-6.		Vitamin D metabolites were measured in six patients with vitamin D intoxication		In each case the serum 25-hydroxyvitamin D was grossly elevated and there was a more modest elevation in serum 1,25-dihydroxyvitamin D.	Hypercalcaemia of vitamin D intoxication is mediated by increased bone resorption and bisphosphonates have a role in its management.
Rizzoli et al., Bone. 1994 Mar-Apr;15(2):193-8. n = 7		7 cases of vitamin D overdose (25-hydroxyvitamin D: 710 +/- 179 nmol/l; normal range: 20-90).		An intravenous administration of a single infusion of the bisphosphonate clodronate to 3 patients led to a correction of hypercalcemia/hypercalciuria	Enhanced bone resorption encountered in vitamin D intoxication could be favorably influenced by bisphosphonate treatment.
Klontz KC N Engl J Med	58 year old	186,906 IU of vitamin D_3	2 months	25-hydroxyvitamin D, 1171 nmol per	recommended safe upper limit

2007; 357:308-309July 19, 2007DOI: 10.1056/NEJMc063341	woman with diabetes and arthritis	per capsule (per day)		liter (normal range, 22 to 135); 1,25-dihydroxyvitamin D, 305 pmol per liter (normal range, 36 to 144);	2000 IU per day

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Appendix 8: Vitamin E

Study & Population (n)	Features	Dosage and Study Design	Duration (days)	Key Observations	NOAEL Considerations
Farrell PM & Bieri JG 1975 N =28 adults	Possible toxic and/or beneficial effects of vitamin E supplementation	67-537 mg TE*/d 100 to 800 IU/day of tocopherol	4months to 3years	Megavitamin E supplements produced no apparent toxic side effects and that subjective claims for beneficial effects were highly variable	
Ernst E and Matrai A 1985 N = 16 adults		536 mg TE/d (800 mg/d all rac -α-tocopheryl-acetate)	4 wk	No adverse effects by clinical chemical blood analysis	
Corrigan 1982 N=12	Warfarin treated cardiology patients	67-269 mg TE/d (100-400 IU all rac α-tocopherol)	4 wk	Warfarin effect was intensified	
Anderson *et al*, 1974 N = 38	Angina pectoris patients	2362 mg TE*/d (3200 mg RRR-α-tocopheryl succinate)	9 wk	No adverse effects except some gastrointestinal disturbance (diarrhoea: 3 subj; intestinal spasm)	
Bierenbaum *et al*,1985 N = 25	Diabetic subjects	1820 mg TE/d (2000 mg all rac-α-tocopheryl acetate)	2-6wks	No adverse effects by clinical chemical blood analysis (cholesterol, T3, T4, blood coagulation	
Gillilian *et al*, 1997 N = 52 6 mo (	Angina pectoris patients	1322 mg TE/d 1600 IU RRR-α-tocopheryl succinate)	6 mo	No adverse effects in cardiac function parameters, urinalysis, blood count, blood chemistry,	

				prothrombin time	
Kitagawa and Mino, 1989 N = 19 adults	Megadosage of free RRR-alpha-tocopherol in healthy college student volunteers.	600mg TE/d (600mg α-tocopherol)	12 wk	No objective or subjective adverse effects	
Stampfer *et al*, 1983 N = 30 volunteers	randomized, double-blind, placebo-controlled clinical trial of vitamin E administration	550mg TE/d (800 IU α-tocopherol)	16 w	No group differences	
Tsai *et al*, 1978 N = 202 volunteers	healthy college student volunteers	441 mg/d TE (600 IU α-tocopheryl acetate)	4 w	Serum T3 and T4 lower; no adverse effect	
Meydani *et al*, 1998 N = 88	healthy volunteers aged >65 years divided between control and three dose groups (17-19 per group)	60, 200 or 800 IU/d	4 months	No subjective side effects No effect on GSH PX, SOD, immunoglobulin, anti-DNA or thyroglobulin antibodies, body weight, total plasma proteins, albumin, glucose, lipids or lipoprotein profile, total bilirubin, serum liver enzymes, blood count, platelet number, bleeding time, Hb, haematocrit, urinary or serum creatinine	NOAEL established in this study was 540 mg/day.

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Appendix 9: B Vitamins

Vitamin B1 (Thiamine) - Indian Studies

Study & Population (n)	Features	Dosage and Study Design	Duration (days)	Key Observations	NOAEL considerations
Kumar M.V.2007, (n= 129)	7-11 yr old school residential school children were the experimental group whereas day scholars were the control group	A salt fortified with multiple micronutrients was developed containing chelated ferrous sulphate, microencapsulated vitamin A, **B1 (2 mg/10g)**, B2, B6, B12, folic acid, niacin, calcium pantothenate and iodine. The average salt consumption was about 10 g per child per day. Pre-post test design	1 year (365 d)	Multiple micronutrient fortified salt was effective in improving multiple micronutrient status and cognition in children	No adverse affects
Kumar M.V.2009, (n= 245)	5-15 yr old schoolchildren	Salt fortified with multiple micronutrients was developed containing chelated ferrous sulfate and microencapsulated vitamins A, **B1 (2 mg/10g),** B2, B6, B12, folic acid, niacin, calcium pantothenate and iodine. The average salt consumption was about 10 g per child per day. Pre-post test design	1 year (365 d)	Salt fortified with multiple micronutrients was stable during cooking and storage and effective in combating multiple micronutrient deficiencies.	No adverse affects
	Indian women of	Vitamin A 5000 I.U, **thiamin 3**	3-6	The delivery system for oral contraceptive	No adverse

Mahtab S. Bamji et al., 1985, (n=300)	low-income group, receiving a low-dose oral contraceptive	**mg**, riboflavin 3 mg, folic acid 300 ug, vitamin B_{12} 2 ug, pyridoxine 10 mg and vitamin C 50 mg. Longitudinal study	months. (90-180 d)	can be effectively used for giving vitamin supplements	affects
Mario Vaz. 2011, (n=300)	Healthy boys and girls between the ages of 7 and 10.5 yrs	40 g of the nutritional beverage micronutrient fortified powder containing **thiamin (1.1 mg/40g).** Double-blind, placebo-controlled, randomization design	120 d	Multiple micronutrient supplementations may be beneficial in improving micronutrient status and enhancing aerobic capacity and endurance in children.	No adverse affects
Sivakumar B et al. 2006, (n= 869)	Apparently normal healthy children,6 to 16 yrs of age	Micronutrient-enriched beverage served containing **vitamin B1-0.7mg.** Double-blind, placebo-controlled, matched-pair, cluster, randomization design	14 months (420 d)	Daily intake of a micronutrient-enriched beverage showed a significant positive shift in the status of nutrients such as vitamins A, C, B2, and B12, folic acid, vitamin D, and calcium across the age groups of 6 to 16 y	No adverse affects
Bamji MS. 1979, (n=407)	Rural school boys, 5-13 yrs of age	2 tablets of B-complex vitamins **thiamin-2 mg/tablet,** riboflavin, pyridoxine, calcium pantothenate, niacin.	1 month (30 d)	Treatment with B-complex vitamins showed significant reduction in the prevalence of glossitis but had no effect on angular stomatitis.	No adverse affects

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Vitamin B2 (Riboflavin) - Indian Studies

Study & Population (n)	**Features**	**Dosage and Study Design**	**Duration (days)**	**Key Observations**	**NOAEL considerations**
Mahtab S. Bamji et al. 1985, (n=300)	Indian women of low-income group, receiving a low-dose oral contraceptive	Vitamin A 5000 I.U., thiamin 3 mg, **riboflavin 3 mg**, folic acid 300 ug, vitamin B_{12} 2 ug, pyridoxine 10 mg and vitamin C 50 mg. Longitudinal study	3-6 months. (90-180 d)	The delivery system for oral contraceptive can be effectively used for giving vitamin supplements	No adverse affects
Kumar M.V.2006, (n= 413)	Residential school children, 5 to 15 yrs of age	The experimental group received a 1 gm micronutrient supplement containing vitamin A, **B2**	9 months	Vitamin A, vitamin E, vitamin B12 , folic acid, and iron are bioavailable from the multiple micronutrient food supplement used in this study	No adverse affects

		(1 mg), B6 , B12(1µg), folic acid(100 µg), niacin, calcium pantothenate, vitamin C, E, iron, lysine, and calcium daily for 9 months. There was no nutritional intervention in the control group. Randomized, controlled trial with pre-post test design			
Kumar M.V.2007, (n= 129)	7-11 yr old residential school children were the experimental group whereas day scholars were the control group	A salt fortified with multiple micronutrients was developed containing chelated ferrous sulphate, microencapsulated vitamin A, B1, **B2 (2 mg/10g)**, B6, B12, folic acid, niacin, calcium pantothenate and iodine. The average salt consumption is about 10 g per child per day. Pre-post test design	1 yr (365 d)	Multiple micronutrient fortified salt was effective in improving multiple micronutrient status and cognition in children	No adverse affects
Kumar M.V.2008, (n= 123)	7-11 yr old school children	Multiple micro nutrient food supplement (MMFS) containing chelated ferrous sulphate and microencapsulated vitamin	1 yr (365 d)	MMFS was effective in improving the nutrition status and cognition in children.	No adverse affects

		A, **B2 (1 mg/g),** B6, B12, folic acid, niacin, calcium pantothenate, vitamin C, vitamin E, lysine and calcium. Pre-post test design			
Kumar M.V.2009, (n= 245)	5- 15 yr old school children	Salt fortified with multiple micronutrients was developed containing chelated ferrous sulfate and microencapsulated vitamins A, B1, **B2 (2 mg/10g),** B6, B12, folic acid, niacin, calcium pantothenate and iodine. The average salt consumption is about 10 g per child per day. Pre-post test design	1 yr (365 d)	Salt fortified with multiple micronutrients was stable during cooking and storage and effective in combating multiple micronutrient deficiencies.	No adverse affects
Sivakumar B et al 2006, (n= 869)	Apparently normal healthy children, 6- 16 yrs of age	Micronutrient-enriched beverage served containing **vitamin B2- 1.6mg.** Double-blind, placebo-controlled, matched-pair, cluster, randomization design	14 months (420 d)	Daily intake of a micronutrient-enriched beverage showed a significant positive shift in the status of nutrients such as vitamins A, C, B2, and B12, folic acid, vitamin D, and calcium across the age groups of 6- 16 yrs	No adverse affects

Mario Vaz. 2011, (n=300)	Healthy boys and girls between the ages of 7 and 10.5 yrs	40 g of the nutritional beverage micronutrient fortified powder containing **riboflavin (1.1 mg/40g).** Double-blind, placebo-controlled, randomization design	120 d	Multiple micronutrient supplementation may be beneficial in improving micronutrient status and enhancing aerobic capacity and endurance in children.	No adverse affects
Bamji MS. 1979, (n=407)	5-13 yrs rural school boys	2 tablets of B-complex vitamins thiamin, **riboflavin-2 mg/tablet**, pyridoxine, calcium pantothenate, niacin	1 month (30 d)	Treatment with B-complex vitamins showed significant reduction in the prevalence of glossitis but had no effect on angular stomatitis.	No adverse affects

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Table- Pyridoxine – Indian Studies

Study & Population (n)	Features	Dosage and Study Design	Duration	Key Observations	NOAEL considerations
Sivakumar B et al 2006; (n= 869)	Apparently normal healthy children,6 to 16 yrs of age	**Vitamin B6 2 mg** Vitamin B12 1 µg Folic acid 200 µg double blinded study	14 months	Daily intake of a micronutrient-enriched beverage showed a significant positive shift in the status of nutrients such as vitamins A, C, B2, and B12, folic acid, vitamin D, and calcium across the age groups of 6 to 16 y	No adverse affects
Kumar M.V.2006; (n= 413)	Residential school children 5 to 15 yrs of age	The experimental group received a 1 gm micronutrient supplement containing vitamin A, B2, **B6 (1mg)** , B12(1µg), folic acid(100 µg), niacin, calcium pantothenate, vitamin C, E, iron, lysine, and calcium daily for 9 months. There was no nutritional intervention in the control group. Randomized controlled trial with pre-post test design	9 months	Vitamin A, vitamin E, vitamin B12 , folic acid, and iron are found to be bioavailable from the multiple micronutrient food supplement used in this study .	No adverse affects
Kumar M.V.2007; (n= 129)	7-11 yr old residential school children were the experimental group whereas day scholars	A salt fortified with multiple micronutrients was developed containing chelated ferrous sulphate, microencapsulated vitamin A, B1, B2, **B6 (2 mg/day)**, B12(400 µg),	1 year	Multiple micronutrient fortified salt is effective in improving multiple micronutrient status and cognition in children .	No adverse affects

	were the control group	folic acid(5 mg), niacin, calcium pantothenate and iodine.pre-post test design			
Kumar M.V.2008; (n= 123)	7-11 yr old school children	Multiple micronu trient food supplement (MMFS) containing chelated ferrous sulphate and microencapsulat ed vitamin A, B2, **B6 (1 mg/g)**, B12(1 µg /gm), folic acid(100 µg /gm), niacin, calcium pantothenate, vitamin C, vitamin E, lysine and calcium.pre-post test design	1 yr	MMFS was effective in improving the nutrition status and cognition in children.	No adverse affects
Kumar M.V.2009; (n= 245)	5 -15 yr old schoolchil dren	Salt was fortified with multiple mi cronutrients and 10 gms of salt contained chelated ferrous sulfate and microencapsulat ed vitamins A, B1, B2, **B6 (2mg)**, B12(4 µg), folic acid(50 µg), niacin, calcium pantothenate and iodine	1 yr	Salt fortified with multiple micro nutrients was stable during cooking and storage and effective in combating multiple micronutrie nt deficiencies.	No adverse affects
Mahtab S. Bamji et al., 1985; (n=300)	Indian women of low-income group, receiving a low-dose oral contracep tive	vitamin A 5000 I.U., thiamin 3 mg, riboflavin 3 mg, folic acid 300 µg, vitamin B 2µg, **pyridoxine 10 mg** and vitamin C 50 mg	3-6 months.	The delivery system for oral contraceptive can be effectively used for giving vitamin suppl ements	No adverse affects

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Table- Biotin - Indian Studies

Study & Population (n)	Features	Dosage and Study Design	Duration (days)	Key Observations	NOAEL considerations
Mario Vaz, 2011. (n=300)	Healthy boys and girls between the ages of 7 and 10.5 yrs	40 g of the nutritional beverage micronutrient fortified powder containing **biotin (10 µg/40g).** Double-blind, placebo-controlled, randomization design	120 d	Multiple micronutrient supplementations may be beneficial in improving micronutrient status and enhancing aerobic capacity and endurance in children.	No adverse affects

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Niacin & Pantothenic acid- Indian Studies

Study & Population (n)	Features	Dosage and Study Design	Duration (days)	Key Observations	NOAEL considerations
Kumar M.V.2007, (n= 129)	7-11 yr old school children. Residential school children were the experimental group whereas day scholars were the control group	A salt fortified with multiple micronutrients was developed containing chelated ferrous sulphate; microencapsulated vitamin A, B1, B2, B6, B12, folic acid, **niacin (30 mg/10g), calcium pantothenate (2 mg/10g)** and iodine. The average salt consumption is about 10 g per child per day. Pre-post test design	1 yr (365 d)	Multiple micronutrient fortified salt is effective in improving multiple micronutrient status and cognition in children	No adverse affects
Mario Vaz, 2011. (n=300)	Healthy boys and girls between the ages of 7 and 10.5 yrs	40 g of the nutritional beverage micronutrient fortified powder containing **niacin (12 mg/40g) and pantothenic acid (2 mg/40g).** Double-blind, placebo-	120 d	Multiple micronutrient supplementation may be beneficial in improving micronutrient status and enhancing aerobic capacity and endurance in children.	No adverse affects

		controlled, randomization design			
Kumar M.V.2009, (n= 245)	5-15 yr old schoolchildren	Salt fortified with multiple micronutrients was developed containing chelated ferrous sulfate and microencapsulated vitamins A, B1, B2, B6, B12, folic acid, **niacin (30 mg/10g), calcium pantothenate (2 mg/10g)** and iodine. The average salt consumption is about 10 g per child per day. Pre-post test design	1 yr (365 d)	Salt fortified with multiple micronutrients was stable during cooking and storage and effective in combating multiple micro nutrient deficiencies.	No adverse affects
Kumar M.V.2006, (n= 413)	Residential schoolchildren, 5 to 15 yrs of age	The experimental group received a micronutrient supplement containing vitamin A, B2, B6, B12, folic acid, **niacin (15 mg/g), calcium pantothenate (1 mg/g)**, vitamin C, E, iron,	9 months (270 d)	Vitamin A, vitamin E, vitamin B12 , folic acid, and iron are found to be bioavailable from the multiple micronutrient food supplement used in this study	No adverse affects

		lysine, and calcium daily for 9 months. Randomized, controlled trial with pre-post test design			
Kumar M.V.2008, (n= 123)	7-11 yr old school children	Multiple micronutrient food supplement (MMFS) containing chelated ferrous sulphate and microencapsulated vitamin A, B2, B6, B12, folic acid, **niacin (15 mg/g), calcium pantothenate (1 mg/g)**, vitamin C, vitamin E, lysine and calcium. Pre-post test design	1 yr (365 d)	MMFS is effective in improving the nutrition status and cognition in children.	No adverse affects
Bamji MS, 1979 (n=407)	5-13 yrs rural school boys	2 tablets of B-complex vitamins thiamin, riboflavin, pyridoxine, **calcium pantothenate-2 mg/tablet, niacin-20 mg/tablet**	1 month (30 d)	Treatment with B-complex vitamins showed significant reduction in the prevalence of glossitis but had no effect on angular stomatitis.	No adverse affects
Sivakumar B et al 2006,	Apparently normal	Micronutrient-enriched beverage	14 months	Daily intake of a micronutrient-	No adverse affects

n= (869)	healthy children,6 to 16 yrs of age	served containing **niacin-0.9 mg.** Double-blind, placebo-controlled, matched-pair, cluster, randomization design	(420 d)	enriched beverage showed a significant positive shift in the status of nutrients such as vitamins A, C, B2, and B12, folic acid, vitamin D, and calcium across the age groups of 6 to 16 yrs	

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Folic Acid

Indian Studies

Study & Population (n)	Features	Dosage and Study Design	Duration	Key Observations	NOAEL considerations
Shah SP et al, 2016 , (n=244)	Adolescent boys and girls(10-19 yrs)	100 mg elemental iron and **500 µg folic acid**. Community based intervention study	52 weeks	A significant association was found in change in hemoglobin before and after intervention	No adverse effects
Bansal PG et al, 2016, (n=373)	Mild and moderate anaemic adolescent girls	Group A : iron (100 mg), **folic acid (500 µg)** and placebo; Group B (n=224): iron (100 mg), **folic acid (500 µg)** and cyanocobalamin (500 µg for 6 weeks and 15 mcg for 20 weeks).Community-based randomized controlled trial	26 weeks	IFA supplementation with or without vitamin B12 is an effective measure to cure anaemia	No adverse effects
Dhir V et al., 2015; (n=100)	18-75 yrs rheumatoid arthritis patients	Methotrexate started at 10 mg/week and escalated to 25 mg/week by 12 weeks, **Folic acid 10 mg or 30 mg** per week. Double-blind randomized controlled	24 weeks	Even with the high doses of Methotrexate used in current practice, there was no additional benefit (or harm) of a higher dose of folic acid (30 mg/week) over a usual dose (10 mg/week).	No adverse effects

		trial			
Swaminathan S et al, 2015, This Review covered the following studies	Women and children in low-income populations	B-vitamin interventions		Interventions with B vitamins were found to be efficacious in improving the status in women and children. The optimum composition of the supplement needs to be determined.	The deleterious effect of high folate intakes with low B12 intakes needs to be explored further
Krishnaveni et al. 2014 (n=654) mothers and children (n= 578 at 5 yrs, 533 at 9.5 yrs and 539 at 13.5 yrs)	Follow-up of children in a birth cohort at 5.0, 9.5 and 13.5 yrs of age	Routine iron and folic acid supplements given to the mother during pregnancy. Prospective observational birth cohort study.		1.Higher maternal homocysteine was associated with smaller birth size, and higher post-load glucose concentrations at 5 and 9.5 yrs 2. Higher maternal serum folate was associated with higher IR at 9.5 and 13.5 yrs 3. Maternal serum B12 was not associated with birth size or IR 4. The prevalence of low birth weight and preterm births was higher among those born to mothers with hyper-homocysteinaemia compared with those born to mothers with normal homocysteine level	
Sucharita et al. (n=79)	children born to urban mothers in a pregnancy cohort Follow-up of children aged 3–8 yrs	Supplementation of mothers first trimester: **5 mg folate**; second and third trimester: **0.5 mg folate**, 150mg FeSO4, 1000 mg Ca, 250 IU vitamin D		1. Low-frequency power spectra, which is an indicator of cardiac sympathetic activity, was significantly reduced in children born to mothers with low B12 status	

Dwarakanath et al. , (n=1838)	Follow-up of urban healthy pregnant women aged 17–40 yrs from <13 weeks of gestation until delivery Prospective observational cohort study	Dietary folate along with first trimester: **5 mg folate**; second and third trimester: **0.5 mg folate**, 150mg FeSO4 (_45mg elemental Fe), 1000mg Ca, 250 IU vitamin D		1. Inadequacy of B12 intake in the first, second and third trimester of pregnancy was 25, 11 and 10%, respectively 2. Low B12 and folate intakes in the first	
Duggan et al. (n=366)	Urban healthy pregnant women 18 yrs of age	Oral supplementation of 50 mg vitamin B12 from <14 weeks of gestation till 6 weeks postpartum along with routine daily iron (60 mg) and **folic acid (500µg)** during pregnancy Randomized placebo controlled clinical trial		1. B12-supplemented women had significantly higher plasma B12 concentration at second trimester and third trimester compared with the placebo group 2. Incidence of delivering an IUGR infant was 25% in the supplemented group whereas it was 34% in the placebo group 3. At 6 weeks ostpartum, median breast milk B12 concentration was significantly higher in the supplemented group 4. In a subset of infants, those whose mothers were supplemented (N=43) had significantly higher plasma B12 concentrations, lower MMA and plasma homocysteine than infants of un-supplemented mothers	

Toteja GS, (n=446)	Adolescent girls 11–18 yrs, mildly and moderately anaemic	Group A=iron 100 mg and **folic acid 500 µg** and placebo for 26 weeks Group B=iron 100 mg and **folic acid 500µg** vitamin B12 500mg for 6 weeks and 15 mg Community based randomized control trial	20 weeks	1. Anaemia decreased by 35.9% in Group A and by 39.7% in Group B 2. A significant reduction in prevalence of ferritin deficiency from 36.5 to 6.4% in Group B compared with a reduction from 39.1 to 15.2% in Group A was observed	
Taneja et al. 2013 (n= 1000)	6–30 month old children	5g of supplement provided if between 6 and11 months of age and 10 g if >12 months. Each 10 g contained energy 54.1 kcal, protein 0.7 g, fat 3.3 g along with folic acid group =**150 µg folic acid**, B12 group=1.8mg vitamin B12 Double-blind placebo controlled trial	6 months	1. Children in the folic acid group had more episodes of diarrhea and persistence of diarrhea 2. No significant difference between the groups in incidence of acute lower respiratory tract infections 3. In both folic acid and B12 groups, there was was a significant decrease in plasma homocysteine concentration compared with the placebo group, the median decrease being 5.3	
Thankachan et al. 2013 (n= 246)	school-aged children, 6–12 yrs of age	Children randomized to receive unfortified or fortified MMN	8 weeks	1. The prevalence of ID, IDA, vitamin C and vitamin B12 deficiencies significantly reduced by 42, 18, 21 and 5%, respectively, in the	

		containing per 18 g/180 ml serving of 243 mg of vitamin A, 27 mg vitamin C, 0.63 mg riboflavin, 1.27 mg of B12, **35 µg folic acid,** 5.9 mg iron and 1.2 mg zinc. Double-blind placebo controlled trial Intervention group,		intervention arm (P<0.01) as compared with the control arm at the end of the study 2. The concentration of hemoglobin, serum ferritin, vitamin A, vitamin B12, vitamin C and body iron stores were significantly higher in the intervention arm in comparison with the control arm. Red cell folate was also significantly higher (P¼0.04) in the intervention arm 3. The prevalence of ID, IDA, vitamin C and vitamin B12 deficiencies reduced by 42, 18, 21 and 5%, respectively, in the intervention arm (P<0.01) as compared with the control arm at the end of the study 4. The concentration of haemoglobin, SF, vitamin A, vitamin B12, vitamin C and body iron stores were significantly higher in the intervention arm in comparison with the control arm	
Central Technical Co-ordinating Unit, 2000, (n=466)	Pregnant women	**4 mg of folic acid** for 4 months (one month before conception and 3 months after conception) Blind, placebo-controlled randomized trial	4 months.	The study seems to support the role of periconceptional folic acid supplementation in prevention of recurrence of NTDs in the Indian population.	No adverse affects

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Vitamin B12 (Tolerable Upper Limit) Indian Studies

Study & Population (n)	Features	Dosage and Study Design	Duration	Key Observations	NOAEL considerations
Singh C et al 2016,(n=40)	Tinnitus patients	Intramuscular therapy of 1 ml **B12 (2500 µg)** weekly randomized, double-blind pilot study	6 weeks	Therapeutic role of B12 in B12-deficient patients of tinnitus.	no adverse effects
Strand T.A et al 2015; (n=1000)	Children, 6 to 35 months of age	All children across study groups were supplemented with (FA/ **B12) 5 g i**f they were 6 to 11 months, and 10 g if they were 12 months and above. For the intervention groups folic acid only, B12 only or B12/folic acid, the 10 g supplement also contained 150 µg folic acid or **1.8 µg B12** (as cyanocobalamin), or the combination of both. For the younger children, the 5 g supplement contained half of the vitamin doses of the older children. Randomized; placebo controlled double-blind trial	6 months	This study provide evidence that poor B12 status contributes to poor growth	no adverse effects

Sarkate P et al,2007; (n=48)	Pregnant women <14 wk of gestation	In group A combination of sodium feredetate (33 mg of elemental iron) along with **B12 (15 µg**) and folic acid (1.5 mg) was administered twice a day. In group B combination of sodium feredetate (66 mg of elemental iron) along with **B12 (15 µg)** and folic acid (1.5 mg) was administered twice a day. In group C combination of ferrous fumarate (100 mg of elemental iron) along with **B12 (15 µg)** and folic acid (1.5 mg) was administered twice a day. Randomized double-blind study.	75 days	effective in improving haemoglobin profile in pregnant anaemic women and is tolerated well	no adverse effects
Yajnik CS et al,2007, (n=42)	Non-pregnant vegetarian women (age 20-50 years)	Oral **B12 (500 µg)** and/or 100 gm cooked green leafy vegetables (GLV).2x2 factorial design	6 weeks	GLV supplementation did not alter plasma folate or tHcy. B12 supplementation increased plasma B12 concentration and reduced tHcy concentration	no adverse effects
DeshmukhUS et al,2010, (n=349)	Children and patients.	**2 or 10 µg B12** capsules, with or without 200 µg folic acid. Cluster randomized, placebo-controlled, double-blind, 2	12 months	Daily oral supplementation with physiological doses of B12 is an effective community intervention to reduce tHcy.	no adverse effects

		x 3 factorial trial			

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Appendix 10: Vitamin C

International Studies

Study & Population (n)	**Features**	**Dosage and Study Design**	**Duration (days)**	**Key Observations**	**NOAEL considerations**
Stein et al n = 3	Healthy Volunteers	8 g/d, over 4 divided doses; observational	3–9 d	Urinary uric acid increased 41–51% 1 subject developed mild diarrhoea	Does contradict the recommended UL of 2000 mg/day.
Johnston et al *n= 9*	Healthy Volunteers	500–2000 mg/d	28 d	Diarrhea 3 subjects reported in and nosebleeds in 2 subjects	Does not contradict the recommended UL of 2000 mg/day.
Johnston &	Healthy	Graded	70 d	Plasma	Diarrhea reported in 1

Cox *n* =10	Men & Women (26.1 ±2.1 y) Abstract	doses from 75–2000 mg/d; placebo controlled RCT		vitamin C increased by 55% in vitamin C-supplemented subjects by the end of the ten-week treatment ($p < 0.05$), Measures of oxidative stress decreased 60% to 90% for total lipid hydroperoxides and Heinz bodies, respectively. Significant decreases in markers of oxidative stress were noted at the 500 mg, 1,000 mg and 2,000 mg dosages versus placebo. Antioxidant protection was similar at the 1,000 mg and 2,000 mg dosage.	subject Does not contradict the recommended UL of 2000 mg/day.
Ludvigsson et al *n* = 24	Healthy women	Placebo or 1000 or 4000 mg/d; double blind RCT	28 d	Serum vitamin C increased	No clinical or biochemical adverse effects noted Does contradict the recommended UL of 2000 mg/day.
Tsao and Salimi *n* = 6	Healthy Volunteers	10 g/d; observational	12–20 d	Urinary oxalate increased 16% but remained in reference range	Does contradict the recommended UL of 2000 mg/day.

Omaye et al *n* =11	Healthy Men	600 mg/d; metabolic trial(depletion-supplementation - depletion design)	92 d	Accelerated plasma losses of vitamin C or conditioned deficiency noted after second depletion period Leukocyte ascorbic acid decreased 45% in 1st depletion and 77% in the 2nd depletion period	Does not contradict the recommended UL of 2000 mg/day.
Hunt et al *n* _=25	Healthy Women (25-45 y) Abstract	1500 mg/d, divided dose; Double blind cross-over	35 d	No effect on biochemical indexes of iron status	Does not contradict the recommended UL of 2000 mg/day.
Wandzilak et al *n* =15 reference range	Healthy Volunteers Abstract	1, 5, 10 g/d, divided dose; observational	5 d	Urinary oxalate increased 18–40% but remained in reference range 2 subjects had diarrhea at 10g/d	Does contradict the recommended UL of 2000 mg/day.
Levine et al *n* = 7	Healthy Volunteers	30–2500 mg/d, divided dose; controlled metabolic study	~40 d	Urinary oxalate increased 0–40% but remained in reference range; urinary uric acid increased 0–20% (above reference range);	No clinical adverse effects noted Does contradict the recommended UL of 2000 mg/day.
Vojdani et al *n* = 20	Healthy Volunteers	Placebo, 500, 1000,	14 d	A 0 - 40% increase in cellular	Does contradict the recommended UL of 2000 mg/day.

		5000 mg/d; RCT		absorption of ascorbic acid was observed at daily doses of 500 mg beyond which there was no further increase Doses up to 5,000 mg neither induced mutagenic lesions nor negative effects on NK cell activity, apoptosis, or cell cycle.	
Levine et al *n* = 15	Healthy young women	30–2500 mg/d, divided dose; depletion/repletion study	45 d	Biomarkers of endogenous oxidative stress were unchanged by vitamin C at all doses	Does contradict the recommended UL of 2000 mg/day.
Hajjar et al *n* = 54	Volunteers	500, 1000, 2000 mg/d, divided dose double blind RCT	8 mo	Both SBP and DBP improved No changes in lipids after 6 months of treatment No additional benefit beyond 500mg	No clinical intolerance or adverse effects noted Does contradict the recommended UL of 2000 mg/day.
Lenton et al *n* = 48	Healthy men and women; smokers &Non smokers (25 - 64y)	Placebo, 500, 1000 mg/d ; controlled trial (plasma vitamin C< 33 mol/L)	91 d	The changes in lymphocyte ascorbate were strongly associated with changes in lymphocyte glutathione after supplementation. For every 1-mol change in ascorbate a change of 0.5	No adverse effects reported Does not contradict the recommended UL of 2000 mg/day.

				molapprox in glutathione.	
Aghdassi et al *n* = 57	Crohn disease patients	1000 mg/d (combined with vitamin E); RCT	28 d	Plasma vitamin C and alpha tocopherol increased and oxidative stress indices decreased after supplementation Disease activity remained stable	No significant difference in clinical adverse effects noted Does not contradict the recommended UL of 2000 mg/day.
Aghdassi et al *n* = 57	Crohn disease patients	1000 mg/d (combined with vitamin E); RCT	28 d	Plasma vitamin C and alpha tocopherol increased and oxidative stress indices decreased after supplementation Disease activity remained stable	No significant difference in clinical adverse effects noted Does not contradict the recommended UL of 2000 mg/day.

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