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Methodology for 4th Edition Nutrient Chapter Updates

Overview of Approach

This chapter provides an overview of the methodology used in this edition, the 4th, of Vitamin and Mineral Safety, originally authored by the late Dr. John Hathcock, developed by the Council for Responsible Nutrition (CRN), and last updated in 2014 (3rd edition). As described in previous editions, the premise of this book is that the safety evaluation for dietary supplements is best determined on a case-by-case basis through nutrient-appropriate risk assessment. As such, the current edition relied on the methodology and conclusions previously applied to derive upper levels for supplemental intake (ULs) by adults of the nutrients included. It should be noted that global regulatory and other authoritative bodies have derived varying types of upper intake limits for nutrients using differing methodologies – examples of these are also summarized within each nutrient chapter update for context and consideration by the user.

The rationale for and components of CRN's existing safety methodology are described in the section "The Risk Assessment Method" of the 3rd edition of the book, including the prioritization of direct safety evaluation of supplemental intakes, when data are available. The CRN approach to deriving ULs for supplemental intake is based on its principal points of departure for risk analysis, which include but are not limited to the following, as stated in the 3rd edition of the book:

- Preference to data on effects of supplemental intakes, rather than total intakes
- Stronger preference to use of human data over animal data
- Stronger preference to clinical trial data from human studies over other studies, if available, but also uses epidemiologic data
- Stronger preference to identifying no observed adverse effect level (NOAEL) values than to lowest observed adverse effect level (LOAEL) values
- Consideration only of effects that represent a true hazard (i.e., risk of impaired health), rather than nuisance effects
- Preferential use of direct evidence effects, rather than biochemical markers or other indirect indicators
- Use of history of use data, if necessary, to identify a highest observed intake (HOI) and UL when adverse effects in humans have not been identified
- Conservative selection of human NOAEL values that justify selection of an uncertainty factor (UF) of 1.0

The scope of the current update for each nutrient chapter was to determine whether more recent human clinical data are available that might change the conclusions and ULs published in the 3rd edition. Based on the data prioritization described above, the current chapter update also includes newly described approaches developed to employ a more systematic and transparent approach to identifying and evaluating available data based on CRN's methodology. As CRN's approach prioritizes clinical trial data from human studies, literature searching for and consideration of epidemiological and/or animal data were only performed if deemed necessary to fill gaps in available data or to ensure that critical endpoint(s) had been appropriately identified and assessed. As such, these nutrient chapters were not intended to be comprehensive literature reviews or systematic reviews. The methods used were intended to rapidly identify literature following the preferences outlined by CRN's methodology, which was specifically designed to assess supplemental intake of nutrients. As such, the methods described herein inherently have some uncertainties typical of any assessment in which not all available data for each nutrient were necessarily reviewed. Nevertheless, the approaches used for literature searching, study relevance screening and assessment, and study selection were developed to provide more structure and objectivity to the UL determination process and were integrated into a multi-step qualitative weighting system to assess study impact. Using this approach, key studies identified were then carried forward for consideration in the confirmation of the existing UL or derivation of a revised UL for supplemental intakes in adults for each nutrient.

Identification of Relevant Studies

Recent Human Clinical Trials

Literature searches were conducted in the PubMed and Embase databases using syntax specifically developed to capture human clinical trials and/or randomized controlled trials (RCTs) published on the nutrient of interest starting January 1, 2014. The results of these searches were combined, and duplicate studies removed. Title and abstract screening was then performed in which studies that met the minimum inclusion criteria were screened for relevance. To be included in the review, studies must have been a human clinical trial or RCT that evaluated oral supplementation with the nutrient of interest alone (i.e., at least one treatment group with no concomitant exposures) for greater than one consecutive week. Studies in all adult (sub)populations were considered to be relevant to the assessment, as the UL determination for supplemental intakes is intended for the general adult population.

Screening for relevance was conducted to determine to what extent the key components described above in CRN's principal points of departure were reported in each study. Three areas were assessed to determine strength of relevance of each study: 1) availability of data on supplemental intake independent of total nutrient intake; 2) availability of data to calculate a NOAEL, LOAEL, or basis of HOI; and 3) assessment of a true hazard (i.e., risk of impaired health), as described in the 3rd edition of the book. Studies that had information about associations of supplemental intake independent of total intake were assigned a value of "high", and studies where associations of supplemental intake cannot be separated from total intake were assigned a value of "low". Based on availability of data to calculate a UL (NOAEL or LOAEL), HOI, or neither, studies were assigned values of "high", "medium", "low", or "no", respectively. For true hazard assessment, studies were assigned values as follows: "high" for true hazard or clinical outcome, "medium" for strong biomarkers of hazard or clinical outcome, "low" for conclusion of no adverse effects, and "no" for no information. As described in the 3rd edition of the book, biochemical or other indirect indicators should be judged to represent a hazard only if they are surrogate markers for pathological conditions. To be considered relevant to a UL determination, studies needed to have a minimum score of "low" in each category, and higher scores were considered to indicate stronger relevance. In addition, key studies identified in the corresponding chapter from the most recent version of the Vitamin and Mineral Safety book (2014, 3rd edition) were considered for inclusion in the study assessment process and subsequently the development of the nutrient UL.

Studies from the 3rd Edition

Key studies considered in the development of the existing CRN UL for supplemental intakes by adults were identified and reviewed for continued inclusion in the study assessment process.

Consideration of Additional Study Types

As described in the 3rd edition of the book, the CRN methodology prioritizes human clinical trial data but may also use epidemiologic data, if needed. In addition, animal data are used only if appropriate human data are not available, as well as to guide the search for a hazard that might be identified in the human data. A targeted review of the most recent (post-2013) authoritative positions, or published secondary review articles if needed, was conducted to understand if any additional endpoint(s) of concern should be considered relevant to the UL assessment for the nutrient. If sufficient data from human clinical trials were determined not to be available, additional targeted searching in the primary literature for human epidemiological and/or animal studies was also conducted.

Key Study Classification

A tiered approach was used to rank or classify relevant studies based on CRN's principal points of departure for risk analysis. Only the most relevant studies based on these preferences were carried forward to data extraction and study selection below. For example, if human trials were available that identified a true hazard (as defined

by CRN), the most conservative of these studies were carried forward. In the absence of data identifying a true hazard, other studies, such as trials that incorporate a range of standard safety outcomes (e.g., complete blood count, liver enzymes, kidney function, adverse event monitoring) were carried forward.

Data Extraction
For key studies carried forward based on the classification approach described above, data necessary to sufficiently describe each study and potentially calculate a UL for the nutrient were extracted. Study descriptive information followed the Participant, Intervention, Comparator, and Outcome (PICO) framework, and included design, study population information (age, gender, geographic location, and health status), number of participants, supplement information (formulation, dose), intervention duration, comparator (placebo information), outcome assessment, and information regarding NOAEL and LOAEL values. This information was captured in tabular form and a subset included in the corresponding chapter update for each nutrient.

Overall Study Assessment
The final step in the qualitative weighting system and determination of studies most appropriate for development of the nutrient UL considered the relevance assessment, study size, study duration, and whether the study specified safety-related outcome measures *a priori* (as opposed to those that only monitored adverse event reports). Because appropriate study sizes and durations are dependent on the specific outcome or hazard being evaluated, expert judgement was used to rank these studies stratified by outcome or hazard being evaluated. When deemed necessary to adequately differentiate between studies, a quality assessment was also conducted, on a case-by-case basis.

Quality Assessment
Quality assessment was conducted on a case-by-case basis and only when additional criteria were needed to differentiate between studies. The Nutrition Quality Evaluation Strengthening Tools (NUQUEST) framework and tools were utilized to evaluate study quality based on risk of bias assessment (Kelly et al., 2022). NUQUEST tools are designed to evaluate study designs commonly used in human nutrition, while retaining assessment components from existing tools for study quality. The NUQUEST RCT tool was utilized to create a quality scoring system and includes quality assessment for four areas: selection of participants, comparability of study groups, assessment of outcomes, and nutrition-specific considerations. Within each of these four areas, there are questions to assess study quality that have a response of “yes”, “probably yes”, “probably no”, “no”, and “not applicable”. Based on the responses to each of these questions an overall of assessment of “good”, “neutral”, or “poor” was assigned, where good reflects that almost all criteria are met and little or no concern about that area, neutral reflects that most criteria are met and there is some concern about that area, and poor reflects that most or all criteria are met and significant flaws are noted about that area. An overall study rating of “good”, “neutral”, or “poor” was then generated based on the responses to the study quality assessment, using the approach and definitions described in Kelley et al. (2022). Studies were initially assessed by an epidemiologist and a second scientist conducted a quality control (QC) check of a subset of the assessments. When necessary, disagreements in an assessment were resolved by a third reviewer. As designed, this quality assessment system accounts for factors such as randomization that support preference for RCT data over open label or uncontrolled studies.

Similarly, if epidemiological and/or animal toxicological studies were considered for a nutrient, assessment of quality (e.g., based on NUQUEST Tools or Klimisch et al. [1997], as appropriate), was conducted on a case-by-case basis.

Safety Review and UL Determination
Based on a detailed review of the data identified, assessed, and extracted, expert judgement was used to identify the most appropriate study to carry forward to UL development. Standard toxicological principles were

followed, including but not limited to consideration of relevant routes of exposure and selection of the most conservative value for the point of departure, where relevant. CRN's methodology as previously described was implemented for confirming the existing UL or deriving a revised UL for supplemental intake by adults of the nutrient of interest. These methods are described in detail in the 3rd edition book, as well as in publications by Hathcock and Shao (2008) and Shao and Hathcock (2006). Briefly, this risk analysis method based on select point of departure (e.g., from most relevant human clinical trial) included identification of the critical effect, determination of the effect level (e.g., NOAEL, LOAEL), evaluation of uncertainty and selection of uncertainty factors for risk analysis, and calculation of the UL (or HOI, where relevant).

The UL of a vitamin or mineral may be calculated through risk assessment in the following way:

UL = NOAEL ÷ UF (or UL = LOAEL ÷ UF).

As previously described in the methodology chapter of the 3rd edition of the book, if the NOAEL or LOAEL value is identified from animal data, an appropriate UF is assigned to the extrapolation to UL values for humans. If the UL is derived from an HOI and the HOI is based on sparse data, a similar procedure may be used to adjust for uncertainty in that value; however, if the total dataset is extensive, the absence of any adverse effect at any intake supports the argument that no correction for uncertainty is needed (i.e., the UF should be 1.0). For all nutrients with large datasets that include multiple clinical trials involving administration of a range of doses, the uncertainties may be addressed by arranging the data in decreasing order of intake and then selecting downward until confidence in the data is sufficient to justify the selection of a NOAEL or HOI with a UF of 1.0.

References

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