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# Quality assessment of selected vitamins and minerals in effervescent food supplements

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ABSTRACT

Effervescent tablets (ETs) are a popular form of food supplement due to their convenience and taste. However, their quality and regulatory compliance remain insufficiently investigated. This study assessed the composition and labelling quality of ETs, focusing on vitamins (D, B12, C) and the minerals zinc, calcium, magnesium, and iron. Altogether, 71 products were purchased in Slovenia, mostly international brands (87.3 %). The purchased samples were analysed, using standards applicable to food control authorities. The laboratory analyses revealed that overall 11 % of the products deviated from the declared nutrient values beyond permitted tolerance limits, with the highest non-compliance (24 %) observed in ETs containing vitamin D. Over half of the products (55 %) contained at least one micronutrient exceeding the reference daily intake values, and 18 % surpassed established Tolerable Upper Intake levels (ULs), raising safety concerns. All majority of these cases were linked to magnesium content. Although most products declared the chemical forms of nutrients, 14 % did not provide this information. Additionally, 13 % of the declared health claims failed to comply with EU regulatory standards. This study highlights major substantial shortcomings in the quality control, labelling transparency, and regulatory compliance of ET. The results call for stronger oversight, harmonised standards, and better consumer awareness.

## 1. Introduction

Food supplements (FS) have become heavily advertised (Sfodera et al., 2020; Temple, 2012) and increasingly popular, providing concentrated sources of essential nutrients to enhance dietary intake and address specific nutritional deficiencies. The global use of these supplements has surged, fuelled by heightened health awareness and the demand for convenient methods to improve the quality of diets (Djaoudene et al., 2023; Ipsos for Food Supplements Europe, 2022). Available in various forms, including tablets, capsules, drops, and effervescent tablets (ET), FS often resemble pharmaceutical products. However, it is crucial to distinguish FS from medication, which is intended to treat or cure diseases and is controlled by strict medical regulations (Tallon and Kalman, 2025); the purpose of FS, on the other hand, is to support health through the supplementation of the diet with concentrated sources of nutrients or other substances which have a physiological effect (European Parliament & European Council, 2002).

Of the available supplement formats, ETs have become a popular form of FS, due to their convenience, pleasant taste, and rapid dissolution, but they remain one of the least scientifically investigated types of FS. When dissolved in water, ETs produce a flavoured drink which eliminates swallowing difficulties, making them particularly suitable for older individuals and patients with dysphagia (İpci et al., 2016). Their technological characteristics, such as rapid disintegration and enhanced sensory acceptability, improve consumer compliance; however, these same characteristics introduce notable formulation and analytical challenges. The effervescent matrix – composed of organic acids, carbonates, and hygroscopic excipients – is highly sensitive to environmental factors such as humidity, temperature, and light, which can accelerate nutrient degradation. Water-soluble and labile vitamins (such as vitamins C, B12, D) are particularly prone to oxidation and loss, while matrix interference may affect extraction efficiency and instrumental detection (Chatzidopavlaki, Triantafyllopoulou, Pippa, Valsami, & Dallas, 2024; Lykkesfeldt & Tveden-Nyborg, 2019). These physicochemical constraints contribute to variability between the declared and measured nutrient content, underscoring the need for dedicated analytical

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approaches and systematic quality assessment of this supplement formulation.

Moreover, minerals such as magnesium and zinc, although chemically stable, exhibit variable bioavailability depending on their chemical form and interactions within the formulation matrix (EFSA Panel on Dietetic Products, Nutrition, & Allergies, 2015a, 2015b). Sources of calcium and iron can further complicate manufacturing due to precipitation or unfavourable reactions in effervescent solutions (EFSA Panel on Dietetic Products, Nutrition, & Allergies, 2015a, 2015b).

In the European Union, FS, including effervescent formulations, are regulated under Directive 2002/46/EC and Regulation (EU) 1169/2011, which define the requirements for composition, labelling, and consumer protection (European Parliament, & Council of the European Union, 2011, 2002). However, official controls typically prioritise acute safety risks – such as contaminants or unauthorised additives – over the verification of the declared nutrient content. Yet, substantial discrepancies between the declared and actual micronutrient levels may also pose important public health risks (Žmitek et al., 2021).

These regulatory and analytical gaps highlight the need for the systematic evaluation of ETs, which remain largely overlooked despite their increasing market share. This study, therefore, aimed to assess the composition and regulatory compliance of ETs available on the Slovenian market, the majority of which were international brands. By comparing the declared and analytically determined levels of selected vitamins (C, B12, D,) and minerals (zinc, calcium, magnesium, and iron), this work provides evidence on labelling accuracy and compliance, including the validity of nutrition and health claims, supporting improved analytical surveillance and consumer protection.

## 2. Materials and methods

### 2.1. Samples and declared (labelled) composition

The present study builds upon the comprehensive CLAS database (Pravst et al., 2022), which provides detailed information on the labelling and composition of FS available in the Slovenian food supply. The database was set up as part of the Composition and Labelling Information System monitoring in 2015, and is now updated every 3–4 years. While monitoring originally covered only traditional pre-packed foods, FS were added for the first time in 2023. The cross-sectional monitoring of FS was conducted in the two biggest general grocery stores; three pharmacies, one of which is also the biggest retailer of FS online; and two specialised retailers of FS. The monitoring was carried out by collecting photographs of the packaging using the CLAS mobile app, described elsewhere in detail (Pravst et al., 2022). The whole sample of FS included 2030 unique items.

To identify the most relevant micronutrients, we examined the frequency of those declared on effervescent tablet labels and found that, among vitamins, the most commonly declared were C, B1, B6, B12, and D; while among minerals, the most frequently declared were magnesium, calcium, zinc, sodium, and selenium. For this study, we aimed to include the three vitamins and three minerals most commonly found in ETs. Based on this, we selected vitamins C, D, and B12, and magnesium, calcium, and zinc. Vitamin B12 was chosen as the most relevant of the B-group vitamins, given the notable prevalence of the deficiency in Slovenia (Lavriša et al., 2022a). In addition to these six micronutrients in ETs, we additionally included iron, because of its public health relevance and previous national findings indicating suboptimal iron intake in specific population groups (Lavriša et al., 2022b), despite its lower frequency in ETs. For samples where different flavours and/or packaging sizes of the same food supplement brand were available, we only selected one flavour in the smallest packaging size. Altogether, 71 samples were selected for further analysis. One product (one series/LOT) of each selected ET was purchased from the stores. International brands accounted for the vast majority of these samples (N = 62; 87 %), while only nine samples (13 %) were national brands. The declared content of the selected vitamins and minerals per single tablet, the ingredient list, any nutritional and health claims, and the expiry date were extracted from the packaging.

### 2.2. Laboratory analysis

Laboratory analysis encompassed the quantification of the selected micronutrients. All the purchased samples were weighed, and the average number of single ETs in the packaging was calculated to enable comparison with the labelled composition. The analyses were conducted by an independent laboratory (Mérieux NutriSciences, Via Fratta 25, Resana, Italy), using standards applicable for food control authorities. For all the investigated samples, we used laboratory methods accredited for use with the appropriate matrix (food supplements). Vitamin D was quantified using liquid chromatography coupled with tandem mass spectrometry (AOAC International, 2012) [accreditation reference MP 1570 revision 3/2021], which can distinguish between cholecalciferol (vitamin D3) and ergocalciferol (vitamin D2). Vitamin B12 was quantified using liquid chromatography and UV detection (AOAC International, 2018, 2019b) [accreditation reference MP 2347 revision 3/2022], which enables the detection of cyanocobalamin. Ascorbic acid was determined using an adapted method (Fontannaz, Kilinç, & Heudi, 2006), with liquid chromatography and UV detection [accreditation reference MP 2174 revision 3/2019]. Calcium and magnesium were quantified using inductively coupled plasma atomic emission spectroscopy (ICP-AES) (AOAC International, 2019a) [accreditation reference MP 1289 revision 18/2023], while iron and zinc were determined using inductively coupled plasma mass spectrometry (ICP-MS) (European Committee for Standardisation, 2014) [accreditation reference MP 1288 revision 23/2024]. The following data for the coefficient of variation (CV) as a precision measure of the laboratory methods are provided in the accreditation documentation for the methods used: vitamin D (CV 7.2 %); vitamin B12 (CV 3.9 %); vitamin C (CV 7.7 %); calcium and magnesium (CV 3.8 %); iron and zinc (CV 3.7–7.4 %).

### 2.3. Data analysis

The results of the laboratory quantification of the micronutrients were compared to the values declared on the packaging. Differences between the declared and analysed content were assessed using the Wilcoxon signed-rank test. Prior to testing, data distribution was assessed using the Shapiro-Wilk test, confirming non-normal distribution, which justified the use of non-parametric methods. Statistical significance was set at $p < 0.05$. Afterwards, we established the number and proportion of the tested micronutrients which were outside the tolerance limits. We considered two tolerance limits for the allowable deviations in the composition of FS, as defined by the European Commission (2012b): firstly, we used a tolerance limit for permitted deviations in FS,- 20 % to + 50 % for vitamins, and −20 % to + 45 % for minerals (including measurement uncertainty); and secondly, we used stricter regulatory tolerance limits for permitted deviations in FS with labelled nutrition and health claims, where the (analysed) content of micronutrients in FS should be at least the same as the declared content (within the measurement uncertainty) and not higher than + 50 % for vitamins, and + 45 % for minerals. To assess whether the quantity of micronutrients was appropriate for the FS product type, we compared the declared and analysed amounts of the nutrients in the recommended dosage of FS to the Recommended Daily Intake (RDI) of the nutrient, as defined by Regulation (EU) No 1169/2011 (European Parliament, & Council of the European Union, 2011), to Tolerable Upper Intake Levels (ULs), as defined by EFSA (2024), and to Reference Values (RV) for recommended daily intake, as defined by the German Nutrition Society (German Nutrition Society (DGE) & Austrian Nutrition Society (ÖGE)), which are also used in Slovenia as an intake reference (NIJZ, 2020) listed in Supplemental Table 1.

Each nutrient was evaluated both individually and by nutrient group (vitamins and minerals). The results were summarised using descriptive statistics (median, mean, range), and visualised as box plots and categorical distributions to illustrate compliance with tolerance limits and reference values. The use of nutrition and health claims was evaluated in line with EU regulations (European Parliament & Council of the European Union, 2006, 2011). This assessment accounted for all the labelled health claims and all the labelled nutrition claims, with the exception of statements referring to “source of vitamin/mineral” which we suspected were present on most products. The chemical forms of added vitamins/minerals and sweeteners were evaluated from ingredient lists, according to European regulations (Comission Regulation, 2012a; European Parliament & European Council, 2002).

For data processing and analysis, we used Microsoft Excel 2019 (Redmond, Washington, USA) and Python (Version 3.12.4), along with the libraries Pandas, NumPy, Matplotlib, and Statistical Functions.

Data organisation was carried out in Excel, and Python was used for data analysis, statistics, and visualisation creation.

## 3. Results

All the ETs available on the Slovenian market containing at least one of the target micronutrients (vitamin B12, vitamin C, vitamin D, calcium, iron, magnesium, or zinc) were systematically collected and analysed for their declared and actual nutrient content, as well as labelling accuracy and the use of nutrition and health claims.

In total, 71 ETs were analysed for the selected micronutrients. Overall, we conducted 173 quantifications of micronutrients in the analysed products; in all cases, the measured concentrations were above the laboratory method detection and quantification limits. The number of declared active ingredients per product ranged from 1 to 22, indicating a wide diversity in formulation complexity. Ten (14 %) ETs contained only a single vitamin, and another ten (14 %) contained only a single mineral. Two products (3 %) included exclusively vitamins, and two (3 %) contained only minerals. The remaining 47 products (66 %) were multi-ingredient formulations combining both vitamins and minerals.

Of the 71 included products, 87.3 % originated from international brands. The most frequently added micronutrient was vitamin C (64 %), followed by magnesium (47 %) and vitamin B12 (32 %) (Table 1). The labelled recommended daily doses varied considerably, with the ratio between the lowest and highest values ranging from 1:5 for iron to as much as 1:300 for vitamin B12 (Table 1). In 89 % of the samples (N = 63), the recommended daily dose (RDD), as indicated on the product packaging, was one tablet. Two tablets were recommended in 10 % of the samples (N = 7), while one sample (1 %) recommended three tablets per day. The declared vitamin/mineral content matched the European daily reference intake (DRI) in 9 % of the samples containing vitamin B12; 17 % for vitamin C; 5 % for vitamin D; 44 % for zinc; 90 % for calcium; 59 % for magnesium; and 50 % for iron.

**Table 1**
Number and percentage of selected vitamins/minerals in effervescent tablets with declared recommended daily doses.

| Nutrient | Number (%) of samples | Median labelled RDD | Minimal labelled RDD | Maximal labelled RDD | Ratio labelled minimal/maximal RDD |
|---|---|---|---|---|---|
| **Vitamin B12** | 23 (32 %) | 2.5 μg | 1 μg | 300 μg | 1:300 |
| **Vitamin C** | 46 (64 %) | 120 mg | 19 mg | 1000 mg | 1:53 |
| **Vitamin D** | 21 (29 %) | 5 μg | 3.75 μg | 50 μg | 1:13 |
| **Calcium** | 21 (29 %) | 200 mg | 82 mg | 1000 mg | 1:12 |
| **Iron** | 10 (14 %) | 10.5 mg | 4 mg | 18 mg | 1:5 |
| **Magnesium** | 34 (47 %) | 193.8 mg | 37 mg | 500 mg | 1:24 |
| **Zinc** | 18 (25 %) | 10 mg | 2.7 mg | 15 mg | 1:6 |

Notes: RDD: Recommended Daily Dose, as declared by the manufacturer on the label

### 3.1. *Compliance of the analytically determined content with the declared content*

The contents of the selected micronutrients from the packaging of the FS were compared with the results of the laboratory quantification. The mean declared and analysed RDDs are presented in Table 2. Statistically significant differences between the declared and analytically determined mean contents were observed for vitamin B12, iron, magnesium, and zinc.

Overall, 11 % (N = 8) of the tested ET samples were non-compliant with the regulatory tolerance limits applicable to effervescent tablets without nutrition or health claims, for at least one micronutrient; 10 % (N = 7) were below, and one (1 %) was above the tolerance limits. However, when applying the stricter tolerance limits defined for FS bearing nutrition or health claims—a category that includes the majority of products in our sample (as detailed in the Results section)—the non-compliance rate increased to about one-fifth (N = 14; 20 %), with 18 % (N = 13) of the samples below and 1 % (N = 1) above the allowed limits.

To better understand which micronutrients contributed most to the observed non-compliance, we compared the ETs based on their micronutrient content. The highest ratio of non-compliance (22.4 %; N = 5) was found in products containing vitamin D, with 19.0 % (N = 4) of samples falling below and 3.4 % (N = 1) exceeding the tolerance limits (Table 2). When applying the stricter limits for products with claims, the highest prevalence of samples outside the tolerance limits was observed for vitamin B12 (26.1 %; N = 6). For all the mineral-containing samples, the proportion outside the tolerance limits remained below 10 %, and no such cases were observed for vitamin C. Except for one vitamin D sample which exceeded the upper tolerance limit, all the deviations were due to lower-than-declared contents. Overall, 62 % of products showed deviations from the declared values, most frequently in vitamin C and B12.

**Table 2**
Mean declared and analysed recommended daily dose and number of food supplements outside the regulatory tolerance limits.

| Nutrient | Mean declared RDD (SD) | Mean analytically determined RDD (SD) | Outside the tolerance limits using minimal standards [N] [a] | Outside the tolerance limits using stricter standards [N] [b] |
|---|---|---|---|---|
| **Vitamin B12** | 18.6 (61.8) μg* | 17.1 (57.3) μg* | 1 (4.3 %) | 6 (26.1 %) |
| **Vitamin C** | 259.1 (307.1) mg | 257.7 (300.0) mg | 0 (0 %) | 0 (0 %) |
| **Vitamin D** | 10.5 (11.2) μg | 10.7 (12.0) μg | 5 (22.4 %) | 5 (22.4 %) |
| **Calcium** | 326.5 (280.4) mg | 316.2 (266.4) mg | 0 (0 %) | 2 (9.5 %) |
| **Iron** | 10.4 (4.9) mg* | 9.4 (4.6) mg* | 1 (10.0 %) | 1 (10.0 %) |
| **Magnesium** | 211.3 (140.9) mg* | 193.0 (123.6) mg* | 1 (2.9 %) | 3 (8.8 %) |
| **Zinc** | 8.1 (3.2) mg* | 7.7 (3.5) mg* | 0 (0 %) | 1 (5.6 %) |

Notes: RDD: Recommended Daily Dose; * $p < 0.05$ difference between declared and analysed recommended dose; SD: standard deviation; N: number; [a]EU regulatory (2012b) tolerance limit for permitted deviations in FS −20 % to + 50 % for vitamins, and −20 % to + 45 % for minerals (including measurement uncertainty); [b]EU regulatory (2012b) tolerance limit for permitted deviations in FS with labelled nutrition and health claims: analysed content at least as declared content (within the measurement uncertainty) and not higher than + 50 % for vitamins, and + 45 % for minerals.

### *3.2. Comparison of the analytically determined contents with reference values*

Further, we investigated the potential overuse of micronutrients in FS by comparing the analytically determined contents of each selected vitamin/mineral with (i) the Daily Reference Intake (DRI) used for food labelling in the European Union (European Parliament, & Council of the European Union, 2011); (ii) the reference values commonly used in

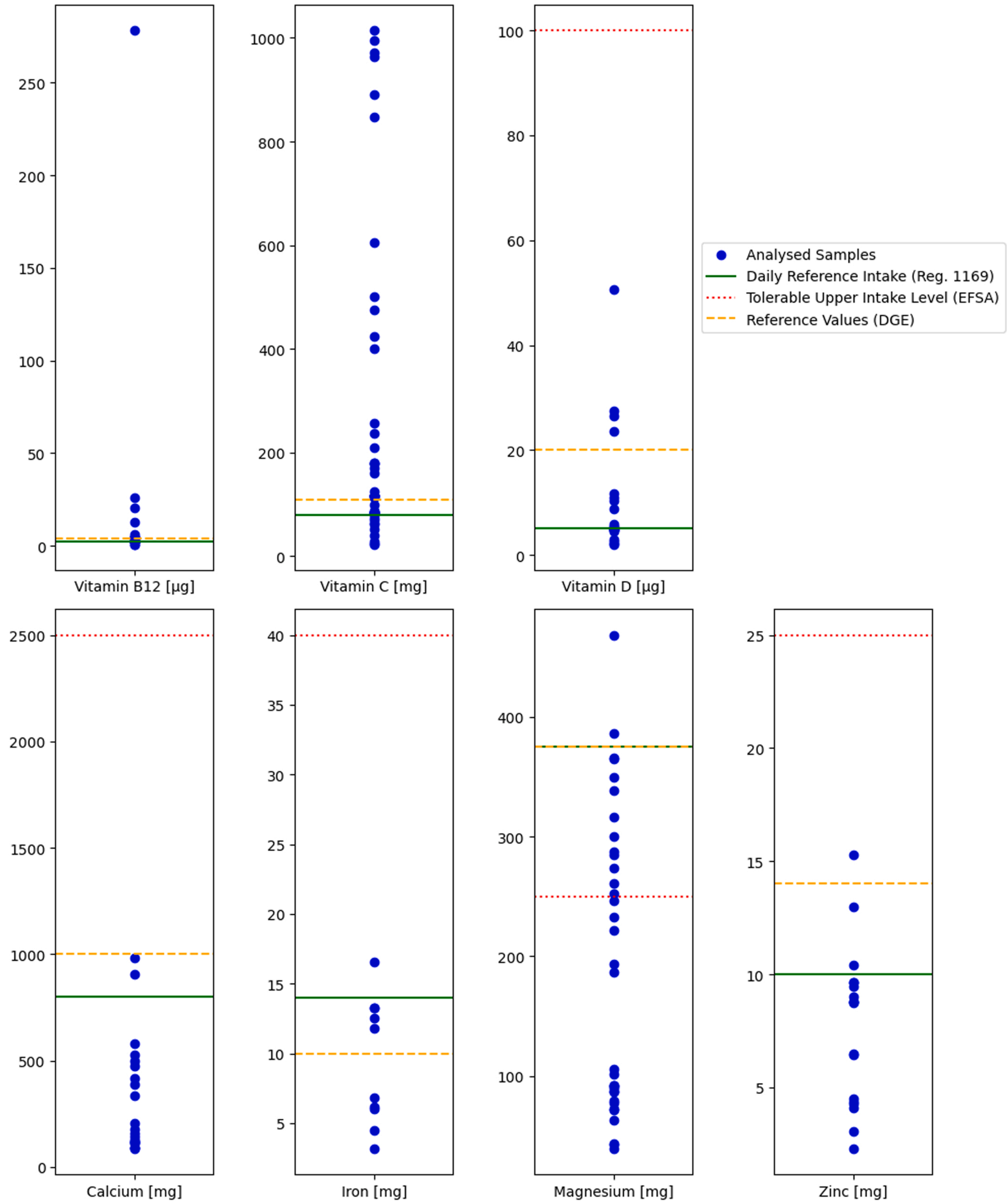

**Fig. 1.** Distribution of the analytically determined contents of the selected micronutrients in the investigated food supplements in comparison with the thresholds for Daily Reference Intake (EU reg. 1169), Tolerable Upper Intake Level (EFSA), and Reference Values (D-A-CH).

clinical practice in Europe (D-A-CH) (German Nutrition Society (DGE) & Austrian Nutrition Society (ÖGE)); and (iii) the ULs defined by the European Food Safety Authority EFSA (EFSA, 2024), which represent the upper safety limit for the use of micronutrients in FS in the EU (Fig. 1 and Fig. 2).

The distribution of the results for the analysed samples is presented in Fig. 1. For vitamin B12 and vitamin C, the ULs have not been established; this means that the range of their concentrations in the analysed samples is wider than the other investigated micronutrients. Except for calcium, iron, and magnesium, most samples clustered around the EU-defined DRI.

As shown in Fig. 2, 66 % of the products exceeded the DRI for at least one nutrient. Furthermore, 52 % of the products exceeded the D-A-CH reference values, and 18 % exceeded the EFSA's UL. Of the vitamins, the highest proportion of samples exceeding the DRI was observed for vitamin C, followed by vitamin B12 and vitamin D. Of the minerals, magnesium was particularly problematic, with nearly 40 % of samples exceeding the UL. This is noteworthy, as the UL for magnesium is set below both the DRI and D-A-CH values, given that the total intake should primarily come from the diet rather than FS. Iron also stood out, with 50 % of the samples exceeding the D-A-CH reference values.

### 3.3. *Chemical forms of added micronutrients*

European regulations only allow the addition of vitamins and minerals to FS in specific chemical forms (European Parliament & European Council, 2002). Compliance with this rule was examined using ingredient lists data extracted from the labels of the investigated FS. In all cases where the chemical form of the added micronutrient was declared, it complied with the permitted forms listed in the Directive. Of the 71 ET samples, we found that 14 % (N = 10) did not specify the chemical form of the included micronutrient. Notably, 40 % of the products lacking information on chemical form originated from the same brand. For minerals, the laboratory method used did not allow the evaluation of the compliance of the used chemical form. Vitamin B12 was quantified as cyanocobalamin in all the analysed samples, meaning that this allowed form was also used in the five samples in which the chemical form of this vitamin was not declared. Ascorbic acid, the permitted form of vitamin C, was quantified in all the samples which declared this vitamin, including in nine samples where the chemical form was not explicitly provided. Similarly, cholecalciferol was quantified in all the samples which declared vitamin D, including one sample where the chemical form was not provided on the product label.

### 3.4. *Use of sweeteners*

Effervescent tablets are typically used for the preparation of a drink, and therefore contain various food constituents and additives for improving its organoleptic properties; a particularly important product property is taste, which is commonly achieved by adding sweeteners. Of the analysed ET samples, the most commonly added sweeteners were saccharin (N = 51; 72 %), followed by cyclamates (N = 41; 58 %) and aspartame (N = 11; 15 %). Acesulfame K and sucralose were each present in 13 % of samples (N = 9), while the least frequently added sweeteners were polyols (N = 5; 7 %) and steviol glycosides (N = 3; 4 %). Only one of the 71 samples did not contain a sweetener, while 76 % of the products contained two or more sweeteners. It should be mentioned that while polyols were present in 48 % of the samples (N = 34), in only five samples (7 %) were these declared with the additive functional class of sweeteners.

### 3.5. *Use of nutrition and health claims on labels*

To provide a comprehensive overview of product presentation, the use of nutrition and health claims on ET labels was evaluated. At least one nutrition or health claim was present on 85 % (N = 60) of the 71 FS samples; 79 % (N = 56) were labelled with health claims, and 21 % (N = 15) with nutrition claims. Nutrition claims referring to a "source of vitamin/mineral" were not considered, as they are ubiquitous for this product category. Of the nutrition claims, those related to sugar were predominant: "no added sugars" (N = 8), "no sugars" (N = 5), and "reduced energy value" (N = 2), collectively accounting for 87 % (N = 13) of all nutrition claims.

A total of 337 individual health claims were identified, the most common being those related to the immune system (N = 59), general health (N = 50), and metabolism or the endocrine system (N = 49). Vitamin C was the most frequently mentioned nutrient (N = 69), with the authorised claim "Vitamin C contributes to the normal function of the immune system" appearing 18 times, followed by magnesium (N = 64), vitamin D (N = 22), and vitamin B12 (N = 17). Fewer claims

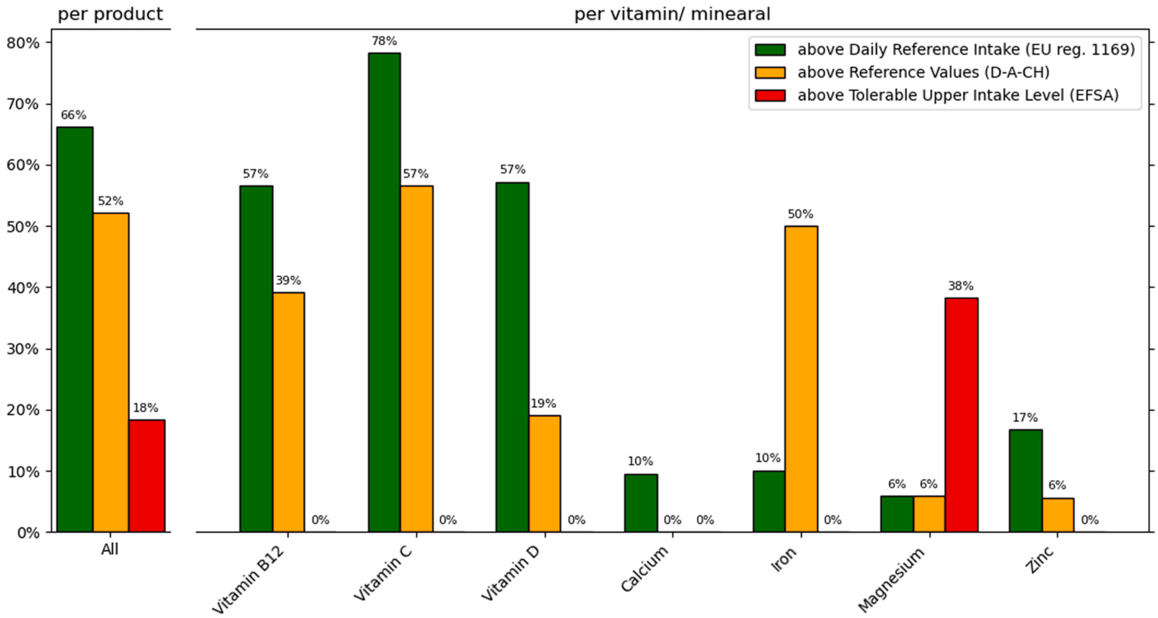

**Fig. 2.** Proportion of samples of food supplements (FS) above the thresholds for Daily Reference Intake (EU reg. 1169), Tolerable Upper Intake Level (EFSA), and Reference Values (D-A-CH) – for all selected micronutrients (per product), and for sub-groups of FS containing the specific vitamin/mineral.

were associated with zinc (N = 10) and iron (N = 9). Multiple claims were often present on a single product – the most extreme example contained 22 distinct health claims, covering all authorised statements for its active ingredients in addition to suggestive imagery and a brand name implying health benefits.

Of the 56 products carrying health claims, 12.5 % (N = 7) did not meet the conditions for authorised use, accounting for a total of 10 non-compliant claims. Non-compliance involved either general, non-specific claims not accompanied by the required authorised statements, or brand-related elements conveying health connotations. Specifically, among the 10 non-compliant cases, one product featured an illustrative graphic of a human head with reference to brain performance, two products included health-related elements in their logos, three products contained health-related wording in the brand name, and four products displayed textual health claims which did not fulfil the conditions for authorised use. The health claims were most frequently presented on the front of the packaging.

## 4. Discussion

The present study provides the first systematic evaluation of the ETs available on the Slovenian market. A total of 71 products were analysed, covering formulations containing vitamins D, B12, and C, and the minerals zinc, calcium, magnesium, and iron.

Most of the analysed products (66 %) were multi-ingredient formulations combining both vitamins and minerals. The bioavailability of such complex formulations may be affected by competitive interactions between trace elements, which can decrease solubility and prolong dissolution time (Yetley, 2007). This distribution reflects a broader market trend towards complex, multi-nutrient FS, which may further increase the risk of compositional variability, labelling inconsistencies, and challenges in maintaining product stability. These observations underscore the need for strengthened quality control measures and harmonised analytical verification of ET.

Firstly, we investigated the differences between the analysed and declared content for the selected micronutrients, using the regulatory tolerance limits defined by the European Commission (2012b).

### 4.1. Compliance of the analytically determined content with the declared content

The present findings indicate notable discrepancies between the declared and the analytically determined contents of micronutrients in ETs available on the Slovenian market. Although most products complied with the EU tolerance limits for FS, 11 % of the samples deviated beyond the permitted range. This increased to 20 % when the stricter criteria for products bearing nutrition or health claims were applied. The vast majority of these discrepancies involved lower-than-declared values. Similar findings have been reported in other studies, conducted within the framework of harmonised European Union legislation, which assessed the quality and regulatory compliance of various supplement types (Niedzielski et al., 2016; Poniedziałek et al., 2018; Puścion-Jakubik et al., 2021). In studies on the Slovenian market, particularly high variability was reported for resveratrol supplements, with 95 % of samples deviating from the declared values (Bensa et al., 2023). Similarly, substantial discrepancies were identified in the analysis of mineral supplements (iron, manganese, calcium, zinc, and phosphorus), where 87 % of products were found to be non-compliant (Masten Rutar et al., 2022). Other studies reported moderate levels of deviation, such as 67 % for folate-containing supplements (Žmitek et al., 2021) and 55 % for various vitamin and coenzyme Q10 products (Temova Rakuša, 2021). Earlier work by Pravst and Žmitek (2011) similarly found that 54 % of coenzyme Q10 supplements deviated from the labelled values. Lower non-compliance rates were observed in iodine supplements (43 %)(Osterc and Stibilj, 2006) and probiotic products (12 %)(Mohar Lorbeg et al., 2021), while high compliance rates were very recently reported for collagen-based supplements (Kržišnik et al., 2024).

Previous studies addressing effervescent formulations have mainly focused on pharmaceutical or technological aspects, such as moisture sensitivity, degradation of active ingredients, and formulation challenges (Chatzidopavlaki et al., 2024). However, systematic evaluations of nutrient content accuracy and regulatory compliance in ETs are still lacking. By addressing these aspects, the present study fills an important research gap and contributes new evidence to the scientific understanding of quality assurance in food supplements, with specific emphasis on this underexplored product category.

Deviations were most pronounced in products containing vitamin D, where nearly one quarter (22 %) of the samples failed to meet tolerance requirements. This may reflect the inherent instability of vitamin D in effervescent matrices, which are sensitive to humidity and oxidative degradation during storage or handling. Previous work has shown that vitamin D degradation is accelerated by moisture and light exposure (Temova and Roškar, 2017), and such losses are particularly relevant in hygroscopic formulations such as ETs. The observed incompliance rate was notably higher than the 8 % reported in a previous study (Žmitek et al., 2021), which was, however, focused specifically on market-leading vitamin D-containing FS and did not include ET. Supplementation with vitamin D is particularly relevant in populations at risk of deficiency, an issue well-documented in Slovenia (Hribar et al., 2020). The higher variability observed for vitamin B12 may similarly be attributed to its chemical instability in multivitamin blends and its sensitivity to low pH, both of which are typical of effervescent systems (Temova Rakuša et al., 2022). Given the high prevalence of discrepancies observed in our study, there is a risk that consumers may mistakenly believe they are adequately supplementing their vitamin D intake, as some FS contain considerably less vitamin D than the declared amounts. In contrast, mineral-containing formulations demonstrated relatively stable performance, with non-compliance rates below 10 %. This aligns with the generally higher chemical stability of inorganic mineral salts compared to organic vitamin compounds.

Taken together, these results emphasise the fact that effervescent formulations present unique technological and stability-related challenges which can affect nutrient accuracy. Their moisture-sensitive matrices and frequent combination of multiple active ingredients complicate formulation control, and may account for the higher rates of non-compliance for certain vitamins. These aspects highlight the need for more targeted manufacturing quality standards and improved post-market analytical verification to ensure accurate labelling and consistent nutrient delivery to consumers.

Good manufacturing practices and regular independent monitoring are essential to safeguard consumers and ensure the accuracy of supplement labelling. In the light of the observed discrepancies, the actual content of micronutrients in food supplements in the form of ETs is particularly problematic for vitamins (especially for vitamin D), but not for minerals. Stricter manufacturing controls and enhanced regulatory oversight are crucial to maintain product quality and protect public health. These measures will support informed policy decisions and foster greater consumer trust in food supplements.

### 4.2. Comparison of the analytically determined contents with reference values

Our comparative analysis of the manufacturer's recommended daily intakes of micronutrients with different reference values revealed that a substantial proportion of the investigated food supplements exceeded recommended intakes, especially for certain vitamins and minerals. Two-thirds (66 %) of the products surpassed the Daily Reference Intake (DRI) defined for labelling purposes, while over half (52 %) exceeded the D-A-CH reference values used in clinical and public health practice. We should highlight the most critical observation: 18 % of the products exceeded the ULs defined by EFSA, raising potential safety concerns.

Particularly alarming are results for magnesium, as nearly 40 % of the magnesium-containing ETs exceeded the UL, which is notably set lower than the DRI or D-A-CH recommendations, as magnesium from supplements can cause gastrointestinal side effects, including diarrhoea, when taken in excessive doses (EFSA, 2024). These findings are consistent with concerns raised by other researchers with regard to excessive doses in commercially available food supplements (Andrews et al., 2017; Christensen et al., 2023; Flynn et al., 2009; Gernand, 2019; Sichert-Hellert et al., 2006).

Although most analysed products exceeded UL values only when considering the full recommended daily dose, such overages may still become critical when combined with dietary intake or concurrent supplement use. Evidence from European modelling studies indicates that frequent users of multiple supplements can easily surpass ULs for minerals such as magnesium and vitamin D (Flynn et al., 2009; Gernand, 2019; Sichert-Hellert et al., 2006). Therefore, the observed deviations, while moderate on a per-product basis, could contribute to excessive total intake in real-world-use scenarios, especially among consumers who use multiple fortified products or high-dose supplements daily.

This concern is further supported by data on consumer behaviour. A recent European survey reported that 68 % of supplement users take more than two products simultaneously (Ipsos for Food Supplements Europe, 2022), increasing the risk of nutrient overexposure. Similarly, our previous research among military personnel showed widespread multiple-product use (Pravst et al., 2023), compounding the likelihood of excessive intakes. ETs may present an additional risk, as their beverage-like format and pleasant taste can encourage casual use and underestimation of their potency. Unlike conventional tablets or capsules, ETs are often consumed for refreshment rather than targeted supplementation, which may inadvertently lead to regular intake of excessive micronutrient doses and associated adverse health effects.

### 4.3. Chemical forms of added micronutrients

In addition to quantitative deviations, the omission of the chemical forms of added micronutrients in 14 % of the samples could raise further concerns. According to Directive 2002/46/EC, only the permitted chemical forms listed in the Annex may be used in the production of food supplements (European Commission, 2002). The use of non-listed forms is not authorised, and the absence of this information on the label makes it challenging to evaluate regulatory compliance. Moreover, different chemical forms of micronutrients can differ substantially in bioavailability and safety. For instance, magnesium citrate and magnesium oxide differ significantly in absorption rates (Pardo et al., 2021; Werner et al., 2019), and similar variability applies to forms of vitamin A (retinyl acetate vs. beta-carotene) (Reboul, 2013), folate (folic acid vs. 5-MTHF) (Pietrzik et al., 2010), and iron (ferrous sulfate vs. bisglycinate) (Duque et al., 2014). ULs are sometimes form-specific, as in the case of niacin, where different ULs apply for nicotinic acid and nicotinamide – further reinforcing the need for precise labelling (EFSA, 2024). Such omissions pose challenges for healthcare professionals in accurately assessing intake and potential interactions, while consumers, especially those using multiple supplements or with health conditions, face safety risks and reduced efficacy.

In our study, we were only able to analytically verify the use of the chemical forms of the analysed vitamins, not the minerals, which were quantified using ICP-AES/ICP-MS methods. While the chemical form of vitamin B12 was not provided for 5 samples, permitted cyanocobalamin was found in all samples. Nine samples did not label the chemical form of vitamin C, but contained permitted ascorbic acid. This was also the case for one sample with an unspecified form of vitamin D, where permitted cholecalciferol was quantified. While for the analysed vitamins we can say that the used chemical forms were compliant with the regulation in all cases, we could not assess this for the minerals. To address this challenge, regulatory bodies should strengthen requirements for clear declaration of added vitamins and minerals in the labelled ingredient lists, ensuring transparency, safety, and informed consumer choices.

### 4.4. Use of sweeteners

Sweeteners are commonly added to ETs to enhance palatability and mask the often-unpleasant taste of certain vitamins and minerals. In the EU, sweeteners are considered food additives, and are regulated by EU Regulation 1333/2008 (European Parliament & Council of the European Union, 2008). Almost all the investigated supplements contained at least one added sweetener, most often in combination.

Saccharin emerged as the most frequently used sweetener in our sample, likely due to its high stability under acidic conditions and at elevated temperatures, which makes it particularly suitable for long-shelf-life products such as ETs (Shankar et al., 2013). Cyclamates, found in 58 % of samples, are also highly stable sweeteners, but due to their relatively low sweetness intensity compared to other sweeteners, larger quantities are required to achieve the desired taste (Kroger et al., 2006). Their flavour profile is broadly similar to sucrose, with a sweetening power approximately 35–50 times greater. However, they can leave a characteristic bitter and salty aftertaste, particularly sodium cyclamate, the most common form identified in our sample of ETs. Polyols were present in nearly half (48 %) of the products in our sample, a finding consistent with their known functional roles. Owing to their lower sweetening intensity compared to high-intensity sweeteners, polyols are frequently used not only for sweetness, but also as bulking agents – to increase product volume with minimal contribution to the total energy content (Piekara et al., 2020).

Growing scientific debate surrounds the long-term health effects of intense sweeteners. Recent WHO guidelines advise against their prolonged use (excluding polyols) for weight control, due to limited benefits and potential health risks (World Health Organization et al., 2022). Our findings underscore the need for clearer labelling and stronger regulatory oversight to ensure transparency and to protect consumers, particularly those with health conditions or additive sensitivities.

### 4.5. Use of nutrition and health claims

Nutrition and health claims were present on 85 % of the investigated products, with health claims being far more common than nutrition claims (79 % vs. 21 %). Among the nutrition claims not referring to added vitamins and minerals, sugar-related statements were predominant, particularly “no added sugars” and “no sugars”. Such claims may help to guide consumers towards products that align with their dietary goals. The most frequently used health claim referred to vitamin C and its role in immune function (Hieke et al., 2015), followed by claims referring to magnesium, vitamin D, and vitamin B12.

The widespread use of health claims is consistent with earlier findings from the CLYMBOL project and other European studies, which highlighted a high prevalence of such claims in foods with a health-related positioning (Hieke et al., 2015; Klopčič eta l., 2020; Rodríguez-Hernández et al., 2025). However, 12.5 % of the investigated products with labelled health claims did not fully comply with the regulatory requirements, often due to vague or generalised messages unsupported by authorised specific health claims. Such inconsistencies can mislead consumers, particularly when presented ambiguously or in a foreign language.

Research shows that simple, familiar, and visually supported claims – especially those referring to well-known nutrients – are more likely to be noticed, trusted, and acted upon (Hieke et al., 2015; Steinhauser et al., 2019).

### 4.6. Study strengths and limitations

To our knowledge, this is one of the most comprehensive studies addressing the quality of FS in the form of ETs. While some previous studies addressed very broad aspects of the quality of food supplements, studies are typically not focused on ETs or even lack any such products, even though ETs are widely distributed and commonly sold in pharmacies, online shops, and even ordinary food stores. The strength of our study is not only that we were focused on this specific sub-category of ETs, but also in our sampling approach. While quality monitoring studies typically establish a sampling approach which limits the number of analysed samples, we examined all the ETs that were found in the national food supply.

Some limitations should also be considered. In our study, we focused on selected micronutrients, namely vitamins C, D, and B12, and the minerals calcium, magnesium, iron, and zinc. This means that the study does not provide information about ETs not containing any of these nutrients, and that the study results cannot be generalised to other nutrients. It should be noted that the selected micronutrients were very prevalent in the CLAS database, and that only 5 % of the unique ETs in the CLAS database were not selected for the analyses. We did not sample ETs which did not contain any of the selected micronutrients, were duplicates, or were similar products with different flavours. It should also be noted that the study did not evaluate the stability, bioavailability, or interactions of vitamins and minerals in the effervescent format, nor did it assess the potential health implications of regular consumption of such products, particularly concerning cumulative nutrient intake or additive exposure. The laboratory analyses were conducted on powdered ETs, and not on drinks prepared using the tablet, where additional nutrient-to-nutrient and nutrient-to-matrix interactions could occur. The content of the selected micronutrients was analytically quantified only when the content of such nutrients was declared on the label. While each analysis was repeated, we only analysed one LOT/series of the product. The samples were not analysed for potentially harmful ingredients (Costa et al., 2019; Rocha et al., 2016), which may pose an important risk for consumer health. The classification of labelled claims followed EU regulations, with a focus on authorised health and nutrition claims. Considering that all the investigated samples were sources of vitamins/minerals, statements referring to the content of such micronutrients (e.g. "source of vitamin C") were not included in the analysis of nutrition claims. This may have led to an underestimation of the use of nutrition claims.

## 5. Conclusions

This study provides comprehensive assessments of the quality and labelling accuracy of ETs available not only in Slovenia, but on the wider European market. Despite most products formally meeting basic regulatory requirements, notable inconsistencies were observed. Overall, 11 % of the analysed samples deviated from the permitted tolerance limits for nutrient content, while 18 % exceeded the ULs for at least one micronutrient – most frequently for magnesium. Such deviations raise concerns with regard to product standardisation, potential health risks, and the adequacy of current market surveillance.

Furthermore, 14 % of the examined products failed to declare the chemical forms of the added micronutrients, and 13 % of the displayed health claims were not compliant with EU legislation. These findings highlight gaps in labelling transparency and regulatory enforcement, particularly given the widespread use of health-related messages which may influence consumer perception.

Taken together, the results reveal substantial variability in the composition and labelling accuracy of ETs, underscoring the need for harmonised analytical verification, stricter quality control, and improved regulatory oversight across European markets. Beyond the regulatory dimension, the findings also emphasise the analytical complexity of effervescent matrices, where hygroscopic excipients, acid–base components, and reactive ingredients can compromise the stability and accurate quantification of labile micronutrients. Strengthening analytical methodologies and establishing matrix-specific validation protocols are therefore essential to ensure reliable nutrient determination, better comparability of results between laboratories, and, ultimately, improved consumer safety and product transparency.

## CRediT authorship contribution statement

**Igor Pravst:** Writing – review & editing, Supervision, Resources, Project administration, Methodology, Funding acquisition. **Špela Goljuf:** Writing – original draft, Investigation, Data curation. **Maša Hribar:** Writing – review & editing, Writing – original draft, Visualization, Methodology, Formal analysis, Data curation.

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## Declaration of Competing Interest

The authors declare no conflict of interest. The funder had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results. Igor Pravst has led and participated in various other research projects in the area of nutrition, public health and food technology, which were (co)funded by the Slovenian Research and Innovation Agency, Ministry of Health of the Republic of Slovenia, the Ministry of Agriculture, Forestry and Food of the Republic of Slovenia, and in case of specific applied research projects, also by food businesses.

## Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.jfca.2025.108803.

## Data availability

The data that support the findings of this study are available in the DiRROS repository at URL: https://dirros.openscience.si/IzpisGradiva.php?id=24809&lang=eng

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