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PRACA ORYGINALNA
ORIGINAL PAPER

The analysis of sodium butyrate content in various dietary supplements available on the Polish market

Analiza zawartości maślanu sodu w różnych suplementach diety dostępnych na polskim rynku

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ABSTRACT

INTRODUCTION: Sodium butyrate is a chemical compound which seems to have several beneficial properties that alleviate signs and symptoms of gastrointestinal diseases. As a result, it has become a frequent component of therapeutic regimens. However, all of the oral products containing sodium butyrate available on the Polish market are registered as dietary supplements and not approved for use as a medication, which might raise concerns about their quality.
MATERIAL AND METHODS: Sodium butyrate was extracted with diethyl ether from eight different dietary supplements available on the Polish market in April 2024. Subsequently, the diethyl ether was evaporated and the residue was dissolved in methanol in order to perform high-performance liquid chromatography. The resulting solutions were passed through a chromatography column to assess the exact content of sodium butyrate in each sample. Finally, the peaks were compared with standard samples and the content of sodium butyrate in each sample was calculated.
RESULTS: Sodium butyrate was detected in each of the analysed samples. However, the ratio of the amount detected to that declared by the producers varied from 45.51% to 106.15%.
CONCLUSIONS: In the majority of cases, the analysed dietary supplements contained sodium butyrate in quantities similar to those declared by the manufacturers; however, the variance between the manufacturer's declarations and the calculated amounts in certain products highlights the need for further research in this area.

KEYWORDS

HPLC, high-performance liquid chromatography, butyric acid, dietary supplement, sodium butyrate

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STRESZCZENIE

WSTĘP: Maślan sodu jest związkiem chemicznym, któremu przypisuje się liczne korzystne właściwości biologiczne, w tym zdolność łagodzenia objawów chorób przewodu pokarmowego. W konsekwencji stał się on częstym składnikiem terapii wspomagających. Wszystkie doustne preparaty zawierające maślan sodu dostępne na polskim rynku są jednak zarejestrowane jako suplementy diety, a żaden z nich nie posiada statusu produktu leczniczego, co może budzić wątpliwości co do ich jakości.
MATERIAŁ I METODY: Maślan sodu został wyekstrahowany z użyciem eteru dietylowego z ośmiu różnych suplementów diety dostępnych na polskim rynku w kwietniu 2024 r. Następnie eter odparowano, a pozostałość rozpuszczono w metanolu w celu przeprowadzenia wysokosprawnej chromatografii cieczowej. Otrzymane roztwory przepuszczono przez kolumnę chromatograficzną, aby oznaczyć dokładną zawartość maślanu sodu w każdej próbce. Uzyskane piki porównano ze wzorcami, a następnie obliczono zawartość maślanu sodu w poszczególnych próbkach.
WYNIKI: Zawartość maślanu sodu wykryto we wszystkich analizowanych próbkach. Jednak stosunek ilości wykrytej do ilości deklarowanej przez producenta wahał się od 45.51% do 106.15%.
WNIOSKI: W większości przypadków analizowane suplementy diety zawierały maślan sodu w ilościach zbliżonych do deklarowanych przez producentów; jednak rozbieżności między deklaracjami producentów a ilościami wyliczonymi dla niektórych produktów wskazują na potrzebę dalszych badań w tym zakresie.

SŁOWA KLUCZOWE

HPLC, wysokosprawna chromatografia cieczowa, kwas masłowy, suplement diety, maślan sodu

INTRODUCTION

Short chain fatty acids are produced during the fermentation of indigestible carbohydrates by commensal bacteria inhabiting the human digestive tract. One of these substances is butyric acid, which is known for its variety of functions in vivo, starting from being an energy source for colonocytes to activating G-protein coupled receptors, and thus exerting metabolic effects [1,2]. Its derivative, sodium butyrate, is a common component of dietary supplements, and these products are thought to be beneficial in patients with some gastrointestinal conditions such as irritable bowel syndrome [3,4] and inflammatory bowel disease [5,6]. Moreover, growing evidence confirms the correlation between gut dysbiosis and metabolic disorders such as obesity and type 2 diabetes mellitus, which may suggest the advantageous effects of sodium butyrate supplementation in the course of these illnesses [7,8].

The terms “medication” and “dietary supplement” are defined by pharmaceutical law, in this particular case by the Polish one. Medications are substances used to treat or prevent specific diseases. Before they are launched on the market, they must be registered and approved by proper institutions; afterwards, they are subjected to constant monitoring. On the other hand, dietary supplements are designed to enrich and complete the diet, and institutional control over them is not as strict as in the case of medications [9,10]. Taking into account the increasing popularity of dietary supplements among patients, there are concerns about their quality and safety and the legitimacy of their usage [11,12]. Analyses of the composition of specific dietary supplements, including those containing sodium butyrate, conducted by independent scientific institutions is gaining increasing attention as evidence continues to emerge that many such products exhibit discrepancies between their declared and actual content. Regarding products containing sodium butyrate, to the authors’ knowledge, this is one of the first analyses of its kind worldwide.

In April 2024, all of the oral products containing sodium butyrate that were available on the Polish market were registered as dietary supplements. In order to compare the actual amounts of sodium butyrate they contain with the amounts declared by the manufacturers, the authors performed appropriate high-performance liquid chromatography (HPLC) analytical tests, utilizing modified methods for quantitative analysis of short chain acid in stool that had been previously described by De Baere et al. [13] and Eberhart et al. [14].

MATERIAL AND METHODS

Reagents and laboratory equipment

The following reagents were used during the experiment: methanol, diethyl ether, trifluoroacetic acid (TFA) [POCh, Gliwice, Poland], a standard solution of propionic acid and butyric acid [Sigma--Aldrich, Germany, Cat. No. 81910].

As far as equipment is concerned, the following devices were used: a Merck/Hitachi D-7000 HPLC system along with a Hitachi/Merck L-7350 column oven and a LaChrom L-7420 UV-Vis detector. The liquid chromatography analysis was performed on a C-18 chromatographic column [XBridge C18 5µm, Waters

Corporation, Milford, MA, USA]. A linear gradient separation was performed using a mobile phase consisting of 0.1% (v/v) TFA in water (A) and 0.1% (v/v) TFA in acetonitrile (B) running from 10% B to 20% B in 8 min (1.25% per min). The injection volume was 30 µL and the column oven temperature was set to 30 °C. The flow rate was 1.5 mL/min. All fatty acids were monitored at λ = 210 nm.

Analytical procedure

Eight different dietary supplements containing sodium butyrate were tested. The products were selected based on its availability in three different pharmacies in the Silesian Voivodeship. One of them was in the form of tablets and the others were capsules. Each supplement was randomly assigned a number from 1 to 8. Next, the following procedure was repeated for each of the tested products. The tablet or contents of the capsule was put in a mortar and ground into a fine powder. This was placed into tubes and then 100 mg of propionic acid and 2 mL of diethyl ether were added. The propionic acid was used as an internal standard. The mixture was then vortexed and centrifuged. After that the supernatant was decanted from the sediment. In order to evaporate the diethyl ether, a vaporizer was used. Afterwards, the residue was dissolved in 1.8 mL of methanol using an ultrasonic bath. Subsequently, solutions were passed through a syringe filter and put into vials appropriate for the autosampler. Finally, HPLC was conducted at 30 °C for 8 min at a flow rate of 1.5 mL per min. The UV-Vis detector was set at a wavelength of 210 nm. Chromatography was performed three times for each sample, and the results were averaged. Based on the chromatograms, the area under the curve of the peaks corresponding to propionic acid and butyric acid was read.

Calibration curves for butyric and propionic acids were prepared in parallel, using standard solutions at six different concentrations (Figure 1).

Using the calibration curves based on the area under the corresponding peaks, the contents of propionic acid and butyric acid in 30 µL of the analysed samples were determined. Then, using the proportions, the contents of these acids in the whole sample were calculated. Assuming that the extraction of both propionic and butyric acids was equally efficient and knowing the initial content of propionic acid, the initial content of butyric acid was quantified. Finally, because it was known that the butyrate residues came from the sodium salt of this acid, the masses of sodium butyrate corresponding to the previously calculated masses of butyric acid were measured using the stoichiometric formula.

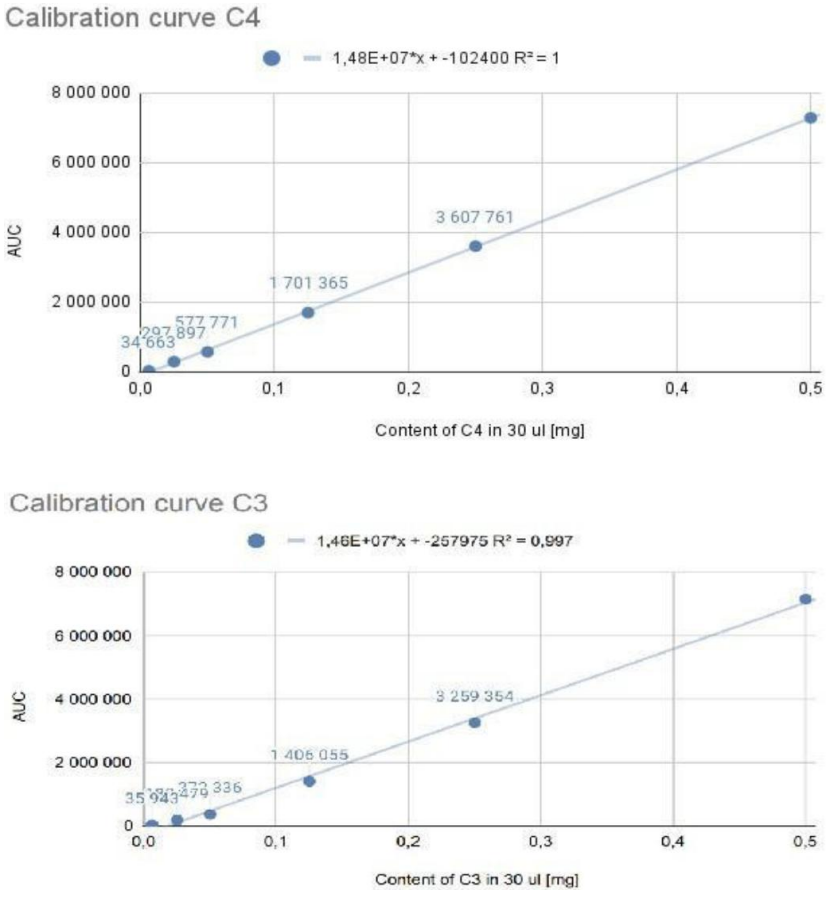

Fig. 1. Calibration curves for butyric acid (C4) and propionic acid (C3). AUC – area under the curve

RESULTS

Based on the analysis of the chromatograms (Figure 2) and the subsequent calculations, it was proven that all of the analysed products contained sodium butyrate, in the following amounts: 1) 235.11 mg, 2) 117.41 mg, 3) 159.22 mg, 4) 141.96 mg, 5) 136.53 mg, 6) 142.98 mg, 7) 186.52 mg and 8) 101.15 mg. The ratios of the determined content to that declared by the producers expressed as a percentage were 47.02%, 78.27%, 106.15%, 83.51%, 45.51%, 95.32%, 62.17% and 67.43%, respectively. These results are summarized in Table I and Figure 3.

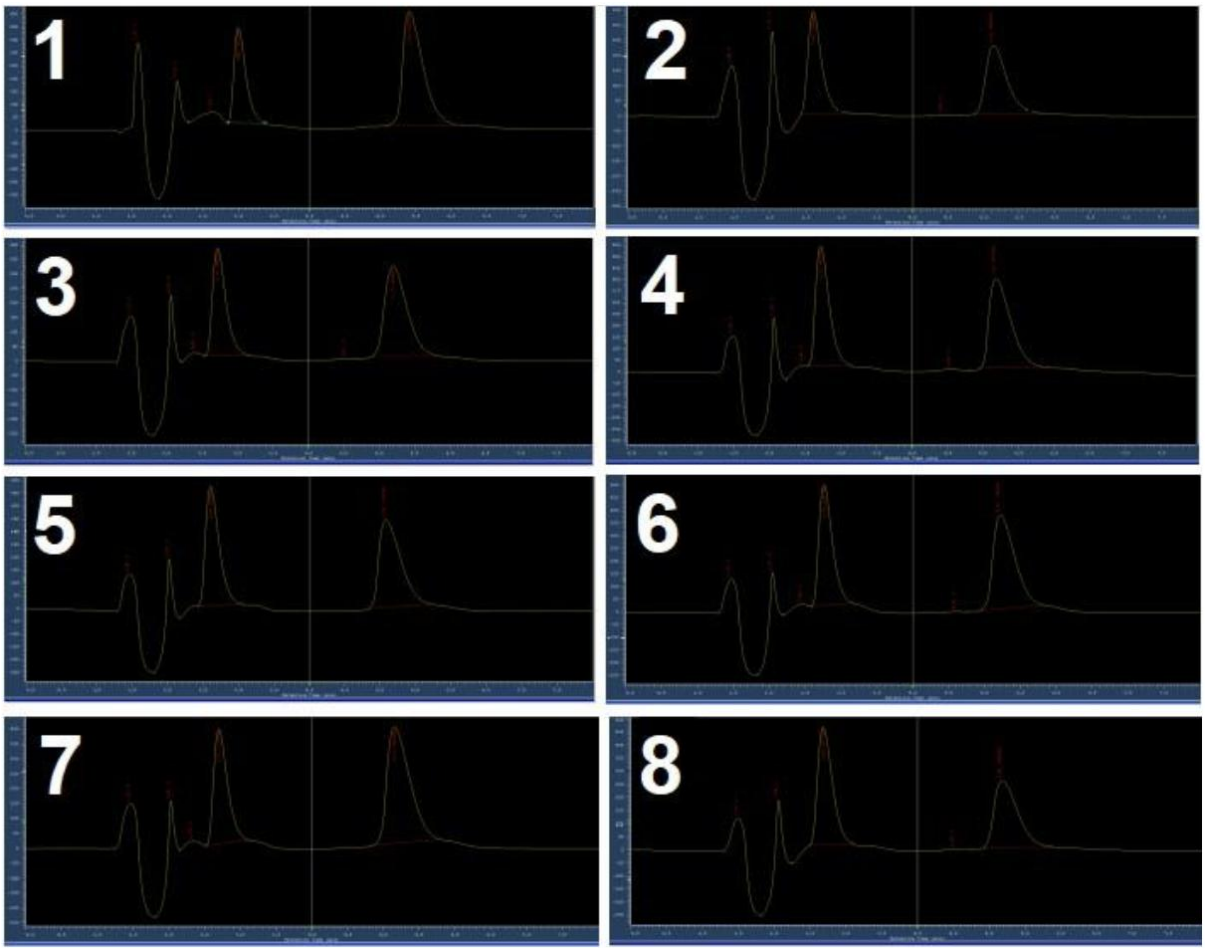

Fig. 2. Chromatograms obtained from sample numbers 1–8

Table I. Declared and empirically determined content of sodium butyrate in the analysed samples

Sample number	Declared content of sodium butyrate [mg]	Actual content of sodium butyrate [mg]	Percentage of declared content of sodium butyrate [%]
1	500	235.11 ± 9.75	47.02
2	150	117.41 ± 3.34	78.27
3	150	159.22 ± 9.79	106.15
4	170	141.96 ± 9.79	83.51
5	300	136.53 ± 5.06	45.51
6	150	142.98 ± 6.11	95.32
7	300	186.52 ± 8.95	62.17
8	150	101.15 ± 5.28	67.43

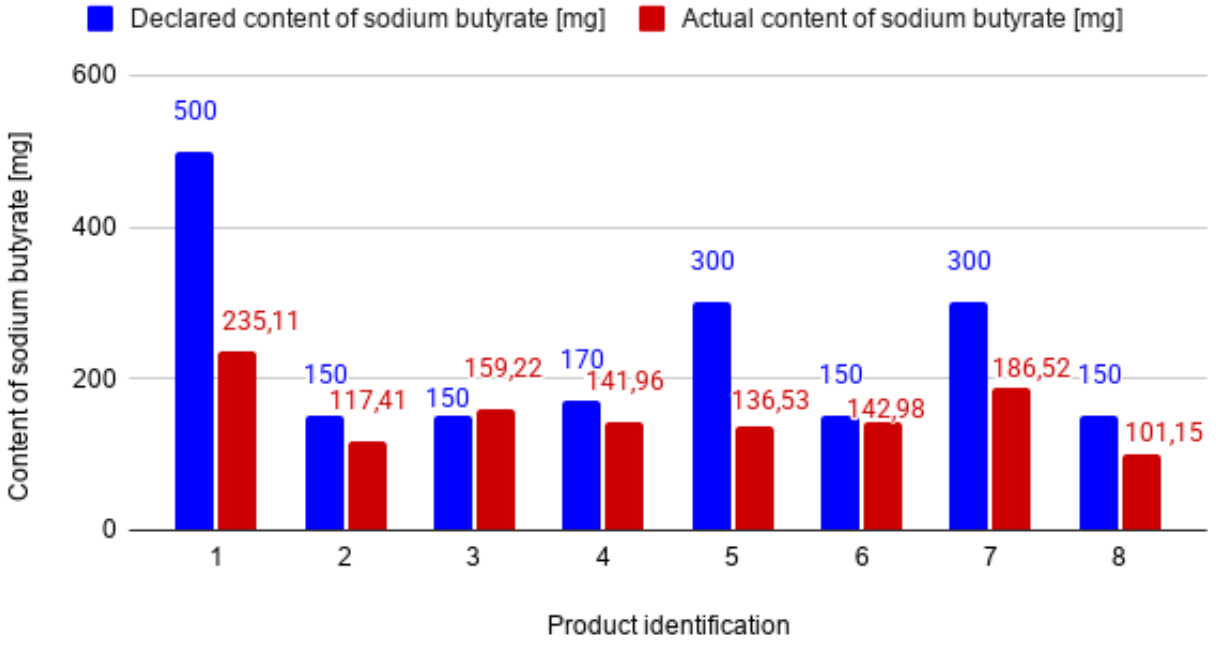

Fig. 3. Comparison of the actual and declared content of sodium butyrate in the analysed products. The whisker plot represents the standard deviation calculated based on three measurements.

DISCUSSION

In order to compare results with any similar studies, the PubMed database was searched with the following terms: "sodium butyrate quality", "sodium butyrate dietary supplements" and "sodium butyrate products". No matching results were found.

This study confirmed that the products in question did contain sodium butyrate, although the percentage of the declared content varied. Formulations with a higher declared content in a single dose (300–500 mg) in fact had a relatively lower content of sodium butyrate (45%–62%) than those with lower declared amounts (150–170 mg) of the active ingredient. It is worth noting that at the time of the experiments (April 2024), many more supplements containing butyric acid derivatives were available on the Polish market than the eight analysed herein. Moreover, the same products might potentially vary between different batches or even between tablets or capsules in the same batch. Given these considerations, further studies are needed to draw general conclusions about the quality of these types of dietary supplements.

Another factor that might have influenced the final results was the fact that the analysed products varied in terms of the concipients used to prepare them. The exact list of excipients declared by the manufacturers in each of the tested products is presented in Table II. Due to the unique nature of the method used, data on the potential impact of these substances on the extraction efficiency and chromatography results are limited. As a limitation of the study, it should be stressed that only one batch of each dietary supplement was evaluated. Furthermore, the method used to detect butyric acid was primarily designed for stool testing, and designing new and more selective methods could control the influence of concipients on the results.

Additionally, it is worth considering that because the presented amounts of sodium butyrate were detected in vitro, there is no information concerning both bioavailability of these products or the exact amounts that reach the colon as the site of action.

Finally, regarding the popularity of dietary supplements and their potential unfavourable impact on regular treatment administered by physicians, more studies on the quality of dietary supplements may contribute to their safety. To provide context on supplement quality, illustrative evidence can be drawn from studies examining other dietary supplements available on the Polish market. For instance, Maćkiewicz et al. [15], in a study published in 2025, analysed the composition of 24 dietary supplements containing iron. Among the products they assessed, only two contained iron in amounts that were consistent with the manufacturers' declarations; two contained lower amounts and as many as 20 contained higher amounts than declared. Notably, one product contained only 0.062 mg of iron instead of the declared 27 mg, whereas another contained 214% of the declared iron content. Another Polish study conducted in 2019 examined the caffeine content in dietary supplements intended to support weight loss. Five dietary supplements and one medicinal product were assessed.

The labelled caffeine content in the medicinal product and one of the dietary supplements matched the information on the packaging. Three products contained less caffeine than expected, and one contained more. The ratio of labelled caffeine to declared caffeine in the supplements tested ranged from 58.16% to 103.27% [16]. In addition, a Polish study assessing the magnesium content of 116 dietary supplements demonstrated that only 41.3% contained the declared dose or an acceptably similar amount (defined as up to 20% less or up to 45% more than declared). By contrast, 39.7% of the products contained less magnesium than declared, and 19.0% contained higher amounts [17]. Data from scientific studies showing the scale of discrepancies between the declared composition of dietary supplements and the actual content emphasize the importance of conducting further extensive research in this area as well as the need for vigilance on the part of consumers, doctors and pharmacists.

Table II. Excipients declared by the producers of the tested products

Sample number	Excipients used
1	Hydroxypropyl methylcellulose, dicalcium phosphate, anionic methacrylate copolymer, polysorbate 80, mono- and diglycerides of fatty acids, triethyl citrate, magnesium salts of fatty acids, colloidal silica
2	Fully hydrogenated triglycerides of fatty acids from palm oil, hydroxypropyl methylcellulose
3	Microcrystalline cellulose, maltodextrin, magnesium salts of fatty acids, hydroxypropyl methylcellulose, titanium dioxide
4	Hydroxypropyl methylcellulose, cellulose, maltodextrin, corn starch, ethyl cellulose
5	Palm oil, hydroxypropyl methylcellulose, magnesium salts of fatty acids
6	Microcrystalline cellulose, hydroxypropyl methylcellulose, palm oil, magnesium salts of fatty acids
7	Maltodextrin, hydroxypropyl methylcellulose, gellan gum, magnesium salts of fatty acids, silicon dioxide
8	Fully hydrogenated palm oil, hydroxypropyl methylcellulose, sodium alginate, calcium carbonate, zinc oxide, starch, maltodextrin, gum arabic, mono- and diglycerides of fatty acids, carrageenan, potassium chloride

CONCLUSIONS

Our results show that each of the analysed dietary supplements contained sodium butyrate. The measured content, as a percentage of the amount declared by manufacturers, ranged from 45% to 106% in the tested samples. This range indicates that, for some products, the discrepancies between the declared and actual sodium butyrate content in dietary supplements were significant, while other supplements contained an amount of sodium butyrate that was very close to the declared amount. Further research in this area is warranted to enhance the quality control and safety of dietary supplements.

Conflict of interest

The authors declare that they have no conflict of interest.

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Authors' contribution

Study design – M. Borówka, M. Nicze, G. Machnik, A. Bołdys, Ł. Bułdak
Data collection – M. Borówka, M. Nicze, G. Machnik
Data interpretation – M. Borówka, G. Machnik, A. Bołdys, Ł. Bułdak
Statistical analysis – M. Borówka, M. Nicze, G. Machnik, Ł. Bułdak
Manuscript preparation – M. Borówka, M. Nicze, A. Bołdys, Ł. Bułdak
Literature research – M. Borówka, A. Bołdys

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