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ORIGINAL CONTRIBUTION

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Small bowel absorption of magnesium and calcium sulphate from a natural mineral water in subjects with ileostomy

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Summary *Background* In many developed countries, magnesium and calcium intakes do not reach recommendations for a large part of the population. Mineral water may be a useful alternative source of dietary minerals, especially in groups of people at risk for developing deficiency due to low intakes. *Aim* To assess if the addition of a natural and mineral-rich water increased small bowel mineral absorption in people with ileostomy. *Methods* A controlled randomised crossover study with two periods of two days each and a minimum 5 days of washout was performed in six ileostomy subjects. Apparent mineral absorption from 0.5 L of natural mineral water with either a high or a low mineral content consumed in the fasting state was compared. The daily addition of minerals corresponded to 2.3 mmol magnesium, 6.9 mmol calcium and 7.7 mmol sulphate. Ileostomy effluents were sampled and analysed for magnesium, calcium and total sulphate. *Results* When compared with the control, the median absorbed amount of magnesium increased from 0.8 (0–1.34) mmol/d to 1.2 (0.8–1.9) mmol/d, which corresponded to a 30 % increase ($P = 0.028$). Median amount of calcium absorbed increased from 8.3 (6.7–13.6) mmol/d to 14.8 (8.3–20.4) mmol/d, i. e. a 45 % increase ($P = 0.027$). The sulphate absorption increased from 1.9 (1.3–2.2) mmol/d to 5.1 (4.2–6.8) mmol/d, which corresponded to 197 % increase ($P = 0.028$). *Conclusions* The mineral-rich water increased absorption of both magnesium and calcium and can therefore be used as an additional source of minerals. However, consumption with meals may be preferred.

Key words ileostomy – absorption – magnesium – calcium – sulphate

Introduction

Consumption of natural mineral water is an old tradition and remains a popular habit in many European countries. Mineral water is considered beneficial for the promotion of health due to their content of minerals and trace elements. They are not only used as a lactose-free alternative to milk and dairy products, but also as convenient magnesium and calcium sources for people who want to reduce and control their energy intake [1]. For people at risk of developing mineral deficiency due to low intakes and/or low absorption [2], mineral water could substantially improve the dietary intake of minerals.

Despite that both magnesium and calcium are essential minerals they are often consumed below the recommended daily intakes in Western countries [3, 4]. An increased magnesium intake has been shown to reduce blood pressure [5], insulin resistance [6] and increase

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bone density [7]. An increased calcium intake has been shown to be associated with increased bone mass or decreased fracture rates [8].

Absorption of magnesium and calcium sulphate from mineral-rich water have been assessed in both animal [9–11] and human studies [12–22]. Magnesium is absorbed throughout the intestine with a predominate site in the distal part of the small intestine with minor amounts absorbed actively in the descending colon [23]. As mineral absorption is a multi-site and complex process, which takes place in both the small and the large bowel, a study of ileostomy excretion might provide quantitative data on the fractional mineral absorption in the small bowel. Absorption of sulphate is also particularly interesting since so called Epsom salt ($MgSO_4$) is a well-known laxative when used in higher doses (> 2 g) and theoretically could limit mineral absorption [24]. A study of small bowel absorption of magnesium and calcium sulphate from a natural mineral water could therefore shed light on some different aspects of the tested water as a liquid dietary supplement.

The primary aim of the present study was to estimate the apparent small bowel absorption of minerals from a natural mineral water. Since mineral water frequently is consumed between meals, the water was ingested in a fasting state in the morning. A secondary aim was to compare the apparent magnesium absorption with predicted values from other human studies to assess whether mineral absorption from water and foods are different.

Methods

Subjects

Six subjects, four men and two women, all proctocolectomized for ulcerative colitis, participated in the study (Table 1). The subjects had conventional ileostomies due to ulcerative colitis, with less than 10 cm of terminal ileum removed, and adequately ileostomy functions. Median time since operation was 14 years (5–27 years). Subjects 2 and 3 used medications for hypertension, subject 3 also for hypothyroidism, and subject 5 for sacroilitis. Median age was 64 years (38–74 years) and median BMI was 25.9 kg/m^2 (22.5–30.6 kg/m^2). The two women studied were postmenopausal. The subjects were otherwise healthy, showing no signs of anaemia, inflammation, hepatic or thyroid disease judged by history and standard laboratory tests. They were asked to temporarily stop their medications during the study. The local Ethical Committee at the Medical Faculty of Göteborg University approved the study. Informed consent was given by all participators.

Design

The study was a randomised controlled cross-over study with two periods of two days each with a minimum washout period of 5 days in between. The participants received the same controlled diet during both periods. The dietary intervention started each day at 7.30 with ingestion of 0.5 L of a mineral water. Patients were then fasting and were later allowed to eat their breakfast at 9h00. The intervention water was a mineral-rich natural water (Hépar®, Vittel, France) while the control water had a low mineral concentration (Valvert®, Etalle, Belgium). Half a litre of Hépar contained 2.3 mmol magnesium, 6.9 mmol calcium and 7.7 mmol sulphate. Half a litre of the control water contained 0.1 mmol magnesium, 0.8 mmol calcium and 0.1 mmol sulphate. The volume of 0.5 L was chosen in order to obtain a substantial mineral intake from the intervention water and to avoid any potential gastrointestinal symptoms. Subjects who are not used to drinking water with a high mineral content may observe certain side effects. Among them is an increased fluidity of the ileostomy excreta, which might cause discomfort when too short of an adaptation period is used prior to ingestion.

Table 1 Detailed information about the participators of the study

Subject	Age	Gender	BMI	Years since operation	Mineral balance during control period[1]		
					Mg (mmol/d)	Ca (mmol/d)	Sulphate (mmol/d)
1	38	Male	30.7	13	0.8	11.1	1.7
2	74	Male	26.3	5	0.0	8.7	2.2
3	55	Male	25.3	11	0.8	8.0	2.1
4	70	Female	28.3	27	0.5	6.7	1.8
5	70	Male	25.5	20	1.3	7.0	1.3
6	59	Female	22.5	15	1.0	13.6	2.0
Median (range)	64 (38–74)		25.9 (22.5–30.7)	14 (5–27)	0.8 (0–1.3)	8.3 (6.7–13.6)	1.9 (1.3–2.2)

Balance is here defined as the difference between the total excretion – total intake. A positive value indicates a positive balance

Ileal effluents were not collected during the first day, which was considered an adaptation period to the experimental diet. On the second day, ileostomy effluents were collected every two hours to minimise bacterial degradation of sulphate. The collected bags were immediately sealed and deep-frozen on dry ice. The ileostomy effluents were weighed, freeze-dried, weighed again and homogenised. Ileostomy bags were weighed and individually pooled for analysis of energy, nitrogen, starch, fibre, calcium, magnesium and sulphate.

Diet

The diet was designed to mimic a typical Swedish diet, which usually has a very high calcium content and relatively lower magnesium content. Trained staff members of the metabolic ward prepared the diet. During preparation of the basal diet, all food items were weighed on an electronic scale within the margin of ± 1 g. The subjects had breakfast at 9h00, lunch at 12h00, coffee break at 14h00, dinner at 17h00, and an evening snack at 20h00. For breakfast, the subjects consumed orange juice, coffee, sandwiches and sweetened yoghurt. Lunch consisted of a vegetarian lasagne and a salad. In the afternoon the subjects had coffee and a chocolate muffin. Dinner included a pie with minced meat, fresh peppers and a low alcohol beer. The evening snack was sandwiches and tea. No addition of sugar in coffee or tea was allowed. The nutrient content of the diet was calculated with software (Dietist, Näringsdata, Bromma, Sweden). Analysis of the dietary content of oxalic acid, phytic acid (inositole phosphates), calcium, magnesium, and sulphate were performed. The composition of the controlled diet is presented in Table 2.

To avoid a negative energy balance, subjects were offered extra bread and butter during the study. Tap water was allowed during the experimental days. The subjects kept a special water diary, where they recorded the number of glasses they consumed per day. The subjects were initially asked to drink the same amount during both periods and to collect samples of tap water for analysis of calcium, magnesium and sulphate concentrations.

Analysis

The diet was collected with the double-portion technique. Foods eaten during one day were freeze-dried, weighed, homogenised and analysed. Ileostomy effluents were analysed with the same methods as the diet. Energy was analysed using a bomb calorimeter (Gallencamp Automatic Adiabatic Bomb Calorimetry, Loughborough, Leicestershire, UK). Inositol phosphates IP3-IP6 were separated with ion-exchange chromatography, and thereafter quantified with ion-pair high pressure liquid chromatography [25, 26]. Samples of diet and ileostomy excreta were wet-ashed at 300 °C in a Tecator system, dissolved in de-ionised water and calcium and magnesium were analysed by atomic absorption spectrophotometry (Perkin-Elmer, Model 360).

Sulphate and oxalate were analysed according to a modified anion exchange chromatography method by Florin et al. [27]. For the analysis of free sulphate, samples of 100 mg of freeze-dried diet or ileostomy excreta were mixed with 10 ml of de-ionised water. The samples were centrifuged at 2000 *g* for 10 min. For analysis of the total sulphate, an acid hydrolysis was added to the procedure, and samples of either diet or ileostomy excreta were prepared by mixing 100 mg freeze-dried matter with 10 ml of 0.8 M HCl and incubated at 95 °C for 4 hours and thereafter centrifuged at 2000 *g* for 10 min. Then, 0.5 ml of the supernatant was mixed with 0.5 ml propanol, and centrifuged at 2000 *g*. A dilution of both sets of prepared aliquots (free and total sulphate) was made at the end of the preparation with de-ionised wa-

Table 2 Content of macronutrients, micronutrients and bioactive substances

Basal diet	Intake/day			Energy %
Energy[a] (kJ)	9400			–
Fat[a] (g)	96			38
Protein[a] (g)	88			16
Carbohydrate[a] (g)	244			44
Dietary fibre[a] (g)	17			–
Oxalic acid[a] (mg)	129 (120–135)			–
Phytic acid[a, b] (mg)	387 (320–412)			–
Mineral intake	**Basal diet**	**Basal diet + control water**	**Basal diet + mineral rich water**	
Magnesium[a] (mmmol/d)	7.5 (7.1–7.7)	7.7 (7.3–8.4)	9.9 (9.5–10.7)	–
Calcium[a] (mmol/d)	34.3 (31.2–37.8)	35.7 (32.4–39.9)	41.7 (38.4–46.0)	–
Total sulphate[a] (mmol/d)	2.9 (2.6–2.9)	3.4 (2.9–3.7)	11.0 (10.5–11.3)	–

[a] Analysed value; [b] ∑IP3-IP6, *IP* inositol phosphates

ter in a 1:25 ratio. For the oxalic acid analysis, samples were prepared by mixing 500 mg of freeze-dried matter from the diets with 10 ml de-ionised water, stirred on a magnetic stirrer for 15 min and thereafter incubated in an ultrasonic bath for 30 min. This procedure was repeated 3 times. One millilitre of the sample was transferred to an Eppendorf tube and was centrifuged for 5 min at 10,000 g. A portion of 4.5 ml of de-ionised water was then added to 0.5 ml of the supernatant. The anion-exchange chromatographic analyses were performed by a Dionex, model 4500i (Sunnyvale, CA, USA) equipped with a 50 µl loop injector, PAX-100 guard and analytical column, Anion Micro Membrane Suppressor and a conductivity detector. For the sulphate analysis, the samples were eluted using a three-phase gradient, 200 mM NaOH, de-ionised water and 50 % (by weight) isopropanol, respectively. The straight gradient of a 200 mM NaOH was infused with de-ionised water, and 50 % (by weight) isopropanol in the following percentage ratios at 0 min: 6:92:2, 15 min: 20:78:2, 30 min: 20:78:2, 35 min: 48:50:2, and 36 min: 6:92:2. For the oxalate analysis, the percentage ratios for NaOH, H_2O, and isopropanol were at 0 min: 6:92:2, 15 min: 30:68:2, 30 min: 50:48:2, 25 min: 48:50:2, and 36 min: 6:92:2. Flow rate was 0.8 ml/min. Standards were prepared from sodium sulphate and oxalate acid per analysis grade (Merck, Darmstadt, Germany). Concentrations of 2.5, 10, 15, 20, and 50 nmol/ml were used for sulphate analysis, and 5, 10, and 20 nmol/ml for oxalate analysis.

Calculations and statistics

Esterified sulphate excretion from the ileum was calculated by subtracting the amounts of excreted free sulphate from the excreted total sulphate. Net intestinal absorption of magnesium and calcium was calculated as the difference between total intake and ileal excretion. All descriptive data are presented as median, minimum and maximum values for the studied 24 h period. Statistical differences between periods were evaluated by the non-parametric Wilcoxon's signed ranks test for pair wise comparisons. Potential carryover effects on mineral absorption were estimated according to Kuehl [28]. A level of $P < 0.05$ was chosen as a limit for statistical significance. The statistics were performed with SYSTAT for Windows, version 7.0 (Evanston, IL: Systat Inc. 1998).

Results

There were no carry-over effects based on the order of treatment, which indicated that the length of the washout period was sufficient. The intervention with the mineral-rich water increased calcium intake by 17 %, magnesium intake by 29 % and total sulphate intake by 226 % (Table 2). The water intake recorded from the water diaries was 1.1 L/day (0.7–2.1 L/day). Tap water did not contribute to any substantial amounts of minerals, as the median concentrations of minerals were 0.2 (0.1–0.3) mmol magnesium/L and 0.5 (0.4–0.7) mmol calcium/L. The total calcium intake from tap water contributed with 0.5 (0.3–1.3) mmol calcium/d, which corresponded to 1.5 % of the total intake in the basal diet period, and 1.2 % in the intervention period. The magnesium intake from tap water was 0.2 (0.1–0.5) mmol magnesium/d, which corresponded to levels of 2.2 % of the total magnesium intake in the control period, and 1.7 % in the intervention period.

The amount of magnesium absorbed increased from 0.8 (0–1.34) mmol/d to 1.2 (0.8–1.9) mmol/d with the mineral-rich water, which corresponded to a median increase of 30 % (Fig. 1, $P = 0.028$). The fractional magnesium absorption was 9.9 % (–0.2–16.7 %) during the control period and increased to 11.8 % (7.9–18.1 %) during the intervention period with the addition of the mineral-rich water ($P = 0.20$).

Median apparent calcium absorption increased from 8.3 (6.7–13.6) mmol/d to 14.8 (8.3–20.4) mmol/d with the addition of the mineral-rich water, which corresponded to a median increase of 45 % (Fig. 2, $P = 0.027$). The fractional calcium absorption corresponded to 23.7 % (19.3–39.1 %) during the control period and increased to 35.2 % (19.8–48.9 %) during the intervention period ($P = 0.13$).

The daily apparent absorption of total sulphate in-

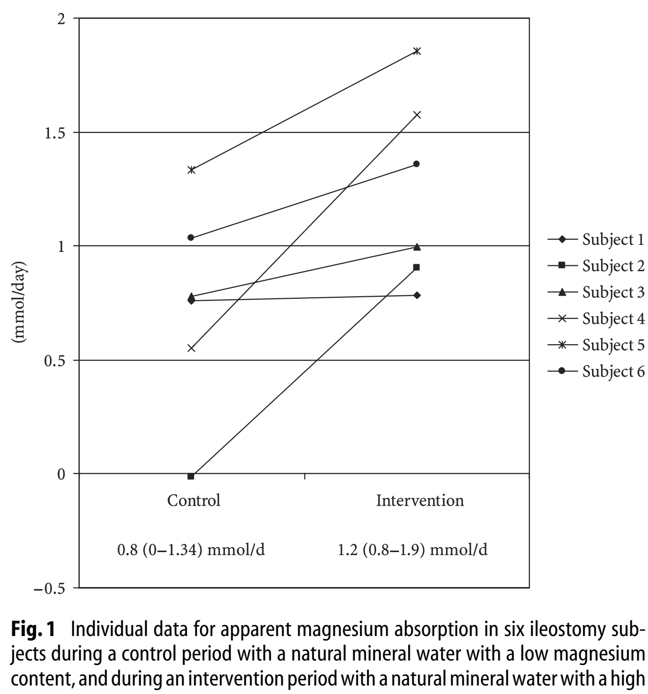

Fig. 1 Individual data for apparent magnesium absorption in six ileostomy subjects during a control period with a natural mineral water with a low magnesium content, and during an intervention period with a natural mineral water with a high magnesium content. Median and range of the apparent absorption is presented under each period

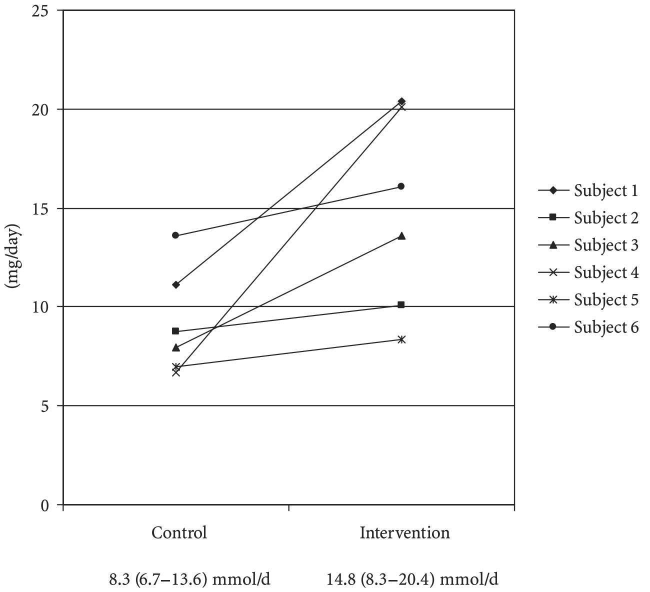

Fig. 2 Individual data for apparent calcium absorption (mg/day) in six ileostomy subjects during a control period with a natural mineral water with a low calcium content, and during an intervention period with a natural mineral water with a high magnesium content. Median and range of the apparent absorption is presented under each period

creased from 1.9 (1.3–2.2) mmol/d during the control period to 5.1 (4.2–6.8) mmol/d during the intervention, which corresponded to a 197 % increase (P = 0.028). The fractional absorption was 56.4 % (39.3–67.6 %) with the control water and 58 % (50.9–68.8 %) with the mineral rich water (P = 0.48). The tested water gave a significantly increased apparent daily absorption of free sulphate; during the control period the absorption was 2.5 (2.4–2.8) mmol/d free sulphate compared to 8.9 (7.3–10.8) mmol/d during the intervention period (P = 0.028). For esterified sulphate, the mean amount absorbed during the control period was 0.6 (0.4–1.1) mmol/d, which was not significantly different from the amount of 0.8 (0.2–1.1) mmol/d during the intervention period.

Excretions of total, free, and esterified sulphate are presented in Table 3. The results were in agreement with the calculated absorption of total, free and esterified sulphate. There was a significantly increased excretion of total and free sulphate, while there is no change in esterified sulphate excretion. There was a significant increase in wet weight of ileal effluents (P = 0.028) with mineral-rich water but no change in dry weight (P = 0.075). Although wet weight increased, no adverse effects or discomfort were reported by any of the participators. There was no personal preference between the waters, which suggests that blinding was successful.

Table 3 Daily ileostomy excretion

	Control	Intervention
Wet weight (g)	742 (449–830)	857 (540–1330)[a]
Dry weight (g)	59.7 (48.4–64.5)	62.3 (53.8–68.9)
Total sulphate (mmol)	1.5 (1.1–2.1)	5.8 (4.4–6.9)[a]
Free sulphate (mmol)	0.5 (0.3–0.6)	4.5 (3.1–6.3)[a]
Esterified sulphate (mmol)	1.0 (0.7–1.5)	1.2 (0.5–1.5)

[a] Results are significantly different from the control period at a P-value < 0.05. During the intervention period, the subjects received 0.5 L of Hépar® (Vittel, France), a natural mineral water with high calcium and magnesium sulphate concentrations. Valvert® (Etalle, Belgium) is characterised by a low mineral content, and was used as a control water

Discussion

To our knowledge, this is the first time that the apparent absorption of magnesium and calcium sulphate from a natural mineral water has been studied in ileostomy subjects. The study shows that minerals were well absorbed as a sulphate salt in the fasting state.

Mineral balance in normal people varies considerably, with reliable cumulative balances often not reached for many weeks. In contrast, patients with ileal resection exhibit a low day-to-day variation in calcium and magnesium balances, irrespective of formula diet composition [29]. Short-term cumulative balances can therefore be performed with good precision in patients with ileostomy. As previously described, balance studies in ileostomy subjects show that a new steady state is possible to induce after only 24 hours [29], which is crucial for the validity of the results in the present study, as the design included a single day of adaptation to reach this new steady state. Overall, the ileostomy model seems reliable for estimates of apparent mineral absorption, though some minor overestimation of absorption cannot be ruled out. Reasons may include that ileostomy subjects might have a somewhat increased small bowel absorption as an adaptive response to a lack of a colon.

With intakes of 0.3–47 mmol/L, fractional magnesium absorption has ranged between 11–76 % with decreasing fractional absorption with increased intakes [19]. Using a predictive formula based on the active and passive dose-dependent absorption mechanisms described for magnesium [30], total magnesium absorption calculated from the intakes should be 0.8 mmol during the control period and around 2.0 mmol during the intervention period. The measured magnesium absorbed during the control period corresponded thereby to what was expected. Addition of the mineral-rich water, however, did not increase the absorbed amount of magnesium to as much as expected.

There are a few factors, which have been suggested to limit magnesium absorption to less than what would be expected. The first concern is that magnesium sulphate in high oral doses (> 2 g/d, popularly called Epsom Salt, $MgSO_4 \cdot 7H_2O$) have well known laxative effects [24]. Magnesium sulphate given orally at a dose of 125 mmol accelerates intestinal transit both in the fasting and fed state [31]; however, this dose is about 12 times higher than what subjects consumed in this study. Studies on lower doses (1.7–10.0 mmol) in people with an intact colon also support increased transit time but no signs of diarrhoea [32, 33]. In our study, transit time was increased but despite this, mineral absorption increased substantially with the addition of magnesium and calcium sulphate.

Another factor, which theoretically may limit the mineral absorption is the salt anion that the minerals are associated with, since sulphate has been suggested to limit the solubility of magnesium in the intestine [34]. Although associated with calcium rather than magnesium, Martin et al. tested the bioavailability of fortified calcium sulphate in white bread in healthy women [35], and showed that fractional absorption of calcium absorption was similar to that of calcium carbonate and lactate. When added to bread, calcium sulphate had a similar fractional absorption to whole milk [35].

Among the most likely causes to a lower than expected magnesium absorption in this particular study is that magnesium absorption is lowered in the fasting state. Sabatier et al. have shown that magnesium bioavailability from mineral water is enhanced when the water is consumed with carbohydrate-rich meal [17]. The composition of the meal is indeed important since certain meal constituents inhibit mineral absorption directly. Fractional magnesium absorption is decreased at amounts of phytic acid similar to those naturally present in whole-meal and brown bread [36] as well as for oxalate in spinach [37]. Mineral water may therefore especially be an excellent carrier for magnesium in contrast to fiber or oxalate rich diets, which inhibit magnesium absorption. It could be recommended to drink with carbohydrate rich foods or drink in between meals if fiber-rich foods are to be consumed.

Calcium absorption in our ileostomy participants was within the range of what has been found in healthy individuals [12]. Fractional absorption of calcium increased from 23 % to 35 % with the 17 % increased calcium intake. Bone density is slightly lower in patients with resected colon compared to people with an intact colon, but they have no significant differences in plasma levels of calcium, phosphate, magnesium, parathyroid hormone, calcitonin and 25-hydroxy-vitamin D nor in the urinary excretion of calcium and phosphate [38]. Ileostomists retain significantly more calcium than expected, which may be in line with an adaptive response.

Sulphate excretion from mixed diets has previously been evaluated in six healthy ileostomy subjects and three healthy normal subjects [27]. Diets containing sulphate in the range of 1.6–16.6 mmol/day gave a net absorption with the capacity reaching a plateau at 5 mmol/day in the ileostomy subjects and exceeding 16.6 mmol/day in the normal subjects. In food, sulphate exists mainly in the free anionic form, while sulphate from intestinal secretions is esterified with glycoproteins (mainly mucin) but also with steroids and glycolipids. As there is a low sulphatase activity in the mucosa of the gastrointestinal tract [39], the free sulphate in the ileostomy excreta comes from diet, while the esterified sulphate has an endogenous origin. Normal dietary intakes of sulphate might vary between 140 and 1500 mg/day, depending on the intake of food items with a high sulphate content such as commercial bread, dried fruits, nuts, tomato juice, fermented beverages and vegetables, particularly brassica vegetables [40]. The basal diet in the present study was designed to have a low content of sulphate. The addition of the mineral-rich water increased the intake considerably. Using the dose-response curve of sulphate absorption from diet made by Florin et al. [27], the apparent sulphate absorption observed in the present study after a single water dose of 0.5 L was similar to that obtained when the sulphate was consumed from food spread out over the whole day [27]. However, in contrast to the finding of Florin et al. the endogenous excretion of sulphate was not altered. According to the data of Florin et al. one might expect an increase in endogenous sulphate excretion by 0.06 mmol/mmol of increased sulphate ingestion, at high sulphate intakes. This corresponds to an amount of 0.46 mmol, or 44 mg. To our knowledge, no study has confirmed the finding by Florin et al. This difference could be related to an absence of increase in endogenous secretion of sulphate, when a single bolus dose is ingested, in contrast to an increased excretion when sulphate is consumed over the whole day.

In summary, this study showed that small bowel absorption of magnesium in ileostomy subjects is similar to that of people with an intact colon, as indicated by applying our absorption data during the control period to a formula based on other human studies [30]. However, by adding water with high levels of magnesium and calcium associated with sulphate, absorption increased considerably but not as much as what would be expected extrapolating from other human data. Mineral-rich water may be useful as a mineral supplement but should preferably be consumed with foods as shown by others. The absorption of sulphate, where a major part came from the mineral-rich water, corresponds with earlier estimates of the absorption of dietary sulphate. It was concluded that a mineral-rich water increased absorption of both magnesium and calcium. However, consumption with meals may be preferred.

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