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RESEARCH ARTICLE

Bowel health, defecation patterns and nutrient intake following adoption of a vegan diet: a randomized-controlled trial

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ABSTRACT

Background: The beneficial effects of a plant-based diet on gut microbiota diversity are well documented, however, its impact on clinical bowel health and defecation patterns are less well understood. Vegetarian diets have been associated with a higher bowel movement (BM) frequency as well as softer stools in cross-sectional studies. The effects of the de-novo adoption of a vegan diet on bowel health, however, have never been investigated in a randomized-controlled trial.

Materials and Methods: The present study examined bowel health and defecation patterns in relation to diet and nutrient intake in a young and healthy sample of $n = 65$ physically-active German university students who were randomly assigned to either a vegan or a meat-rich diet for eight weeks. Bowel health assessment included the Bristol Stool Form Scale (BSFS), the Gastrointestinal Quality of Life Index (GIQLI) and the Cleveland Clinic Fecal Incontinence Score (CCFIS). Nutrient intake was assessed using weighed food diaries. The study was prospectively registered at the German Clinical Trial Register (DRKS00031541).

Results: Weekly BM frequency slightly increased in vegans, whereas it remained unaltered in participants assigned to a meat-rich diet. Fiber intake increased significantly in vegans (34.89 (18.46) g/d) whereas it decreased in those assigned to the meat-rich group (22.79 (12.5) g/d). No significant intergroup differences in BSFS and CCFIS patterns were observed. Adoption of a vegan diet neither resulted in a transient increase in abdominal discomfort nor in a decreased gastrointestinal quality of life, which was comparable across the diet groups.

Conclusions: The short-term de-novo adoption of a vegan diet did not negatively affect markers of bowel health in this study.

Introduction

Plant-based diets enjoy increasing popularity in many modern societies for their potential benefits to human and environmental health [1,2]. The adoption of vegetarian and vegan diets has been associated with a lower risk of incident type-2-diabetes [3], cardiovascular disease [4] and cancer [5]. Both diets also offer benefits for gastrointestinal health [6], and have been linked to gut microbiota diversity and improved Short Chain Fatty Acid (SCFA) levels as well as protective effects against cancers of the digestive system [7,8].

While the beneficial effects of plant-based nutrition on the gut microbiota composition are now well documented [9,10], its impact on clinical bowel health,

defecation patterns and stool morphology are less well understood [11]. Often attributed to a more optimal fiber intake [11,12], vegetarian diets have been associated with a higher Bowel Movement (BM) frequency as well as softer stools with fewer superficial cracks [11,13]. As such, vegetarians appear to be less frequently affected from chronic constipation [14] – A debilitating condition with a high economic and social burden in many Western societies [15]. Vegetarians also avoid meat, for which an inverse dose relationship with the frequency of defecation has been reported [16].

Bowel health and BM frequency in plant-based populations seem to be strongly associated with fiber intake [17], and our previous work suggested that the

overall quality of a plant-based diet plays an important role for both [11]. The number of studies in this field, however, is severely limited [11,13,16–18], and the majority of information was derived from cross-sectional studies.

Randomized-controlled trials in this scientific area are scarce but could greatly enhance our understanding of the role of plant-based nutrition for defecation and bowel health *beyond* the gut. This applies in particular to the '*de-novo*' adoption of plant-based diets, e.g. when individuals suddenly transition from a meat-based diet to a diet that restricts or completely avoids animal products and that is more abundant in high-fiber foods.

Here, we report the results of an eight-weeks dietary intervention in healthy German omnivores who were randomized to either a meat-rich diet or a plant-based vegan diet. Defecation patterns, stool morphology and self-perceived gastrointestinal quality of life were assessed with validated tools and compared between both groups.

We hypothesized that the vegan diet would increase BM frequency and soften stools in comparison to the meat-rich diet. In addition to that, we put forward the hypothesis that the expected increase in the intakes of fibers and certain phytochemicals (such as phytic acid) could transiently result in abdominal discomfort, bloating and cramping in vegans, and thus in a temporary reduction of gastrointestinal quality of life during the first weeks of the new diet. Finally, we used dietary intake data to examine potential associations between bowel movement frequency and selected nutrients and foods.

Materials and methods

Study design

The study was conducted between April and June 2023 and extends our series of studies on plant-based nutrition [19–21]. The methodology and design have been adopted from our previous trial [19], but were modified in some relevant aspects and supplemented with new features and study outcomes. The study was a monocentric, randomized-controlled trial performed at the Centre for Complementary Medicine at Freiburg University Medical Center, Germany. This multi-topic study was primarily designed to investigate the impact of a vegan dietary intervention on immune system-related outcomes. The primary study outcome focused on changes in measured neutrophilic granulocytes. Given the comprehensive scale and outlay of the study, however, multiple secondary endpoints were included.

Immune-system related outcomes will be discussed at a later stage in another publication specifically dedicated towards blood count alterations subsequent to the dietary modification. Here, we focused on a detailed study description and on multiple secondary endpoints including alterations in nutrient intake patterns, defecation patterns and overall bowel health.

Figure 1 schematically displays the course of the study. After a one-week run-in phase, participants were randomized to either a vegan or meat-rich diet for eight consecutive weeks. The meat-rich diet was defined by a consumption of at least 150 g of meat daily, whereas the vegan diet was defined by the exclusion of all animal-based products. Any kind of meat including poultry, pork or beef were permitted whereas fish and fish products were not considered to fulfill this criterion in the context of our study.

Recruitment

We recruited participants by placing advertisements on notice boards at local universities, in student residences and in public places in Freiburg, Germany. In addition, we posted advertisements on social media platforms. Potentially eligible individuals were screened by phone and subsequently invited for an in-person appointment. In- and exclusion criteria were checked on both occasions.

Eligibility criteria

Healthy individuals aged 18–60 years and with a BMI between 21 and 30 kg/m^2 were considered. Criteria of exclusion included: chronic health conditions, a regular intake of medication, pregnancy or lactation, clinically-relevant allergies, eating disorders as well as alcohol (>20 g/d) and daily nicotine intake. Individuals who reported any kind of infection two weeks prior to their screening appointment were not considered. Those who already indicated a plant-based diet prior to their study participation were excluded, as well. The same applied to individuals who considered themselves as 'flexitarians' and to individuals who consumed meat less than 3 times a week on average prior to the study. Eligible participants had to be able to speak and understand German as the study included multiple nutrition-related group seminars and dietary counseling sessions.

Course of the study

Individuals who met the inclusion criteria were assigned to a balanced mixed diet for one week
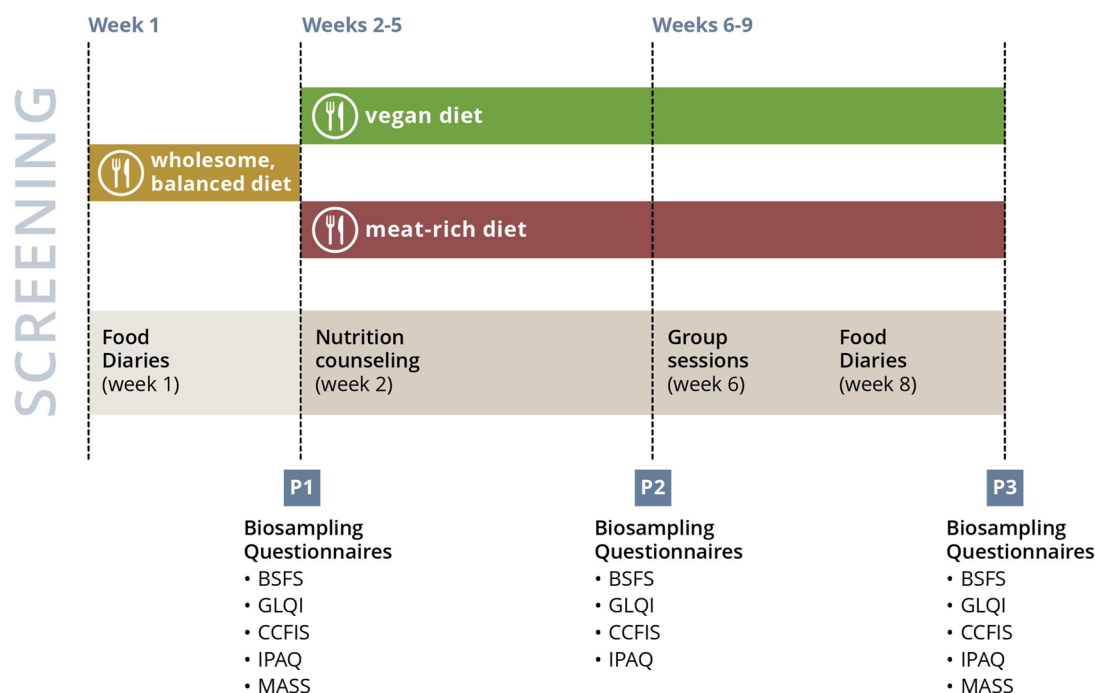
Figure 1. Course of the study: a schematic overview. BSFS: Bristol Stool Form Scale; GLQI: Gastrointestinal Life Quality Index; CCFIS: Cleveland Clinic Fecal Incontinence Score; IPAQ: International Physical Activity Questionnaire; MASS: MacArthur Scale of Subjective Social Status.

(hereafter referred to as 'run-in phase'). Participants were asked to prepare their own meals in accordance with the recent recommendations of the German Nutrition Association (DGE) for the German general population [22]. For this, participants were instructed in individual coaching sessions and received a hand-out summarizing the key points. Although these recommendations were considered obvious by many participants, the coaching session was mandatory to partake in the study. The key aspects of these recommendations are summarized in Supplementary Table 1. Moreover, participants were required to complete a weighted food diary at three consecutive days during the run-in phase.

After the first week, participants were invited for a second appointment where randomization and biosampling took place. Prior to that, we captured sociodemographic data and participants were asked to complete several questionnaires as discussed below. An independent person who was unfamiliar with the study aims created a block-wise randomization list (block size: 6) using Stata software. Sealed envelopes were used for implementation.

Blood samples were taken early in the morning between 7 and 9:30 am after a fasting period of at least 10 h. We subsequently instructed all participants on how to implement their assigned diet in individual coaching sessions of at least 30 min. The same group-specific scheme was used for all participants

who also received a study- and diet-specific booklet for their assigned diet. The coaching ended with a brief capstone session and participants were invited to a mandatory group seminar for their diet at the end of the first study week (see below). The information in the booklet for the vegan group was mainly compiled of specifically tailored advice for individuals who just recently adopted a plant-based diet. It was mainly based on two sources: (I) the Physician Committee for Responsible Medicine's literature resource hub and (II) on information from the German website of the Physicians Association of Nutrition [23,24]. The diet thus focused on whole plant-based foods and aimed to minimize processed food items. The information in the booklet for the meat-rich group was based on a brochure published and developed by the German QS GmbH [25].

Blood results were checked on the same day and participants with abnormal values (e.g. due to iron-deficiency anemia or elevated inflammation markers suggesting a recent infection) were excluded despite having indicated good health in the screening examination.

Biosampling was repeated after 4 weeks and 8 weeks, respectively. Weighed food diaries were again collected during the last study week. We asked participants to complete various questionnaires at both stages. Supplementary Table 2 summarizes the collected data for each time point. Participants received

a financial remuneration of 400€ upon successful completion of the study (as defined by adhering to the assigned diet for eight weeks, by providing complete nutritional protocols and by partaking in all bio-sampling sessions and at least one nutrition-related group seminar).

Group seminars, coaching sessions and newsletters

A hallmark of this study was our regular and close contact with the participants to increase motivation and to ensure high dietary adherence in both groups. A study-specific hotline and email-address were installed to allow for a close exchange between the study personnel and the participants throughout the study and with regard to all study aspects, e.g. for diet-related questions or administrative issues.

Two diet-specific group seminars (a mandatory one in the first week after randomization and an optional one in week 6) of 45 min each were offered. These seminars included important contextual information on the assigned diets, as well as cooking and shopping hints. The small seminar group sizes (6–10 persons) allowed for an interpersonal exchange between participants, who were also encouraged to share both their positive and negative experiences with other members. Potential solutions were discussed in a constructive and open-end style. Each seminar was led by at least two persons from the study personnel and the head of the study was always available for individual queries.

In addition to that, we offered weekly newsletters that covered study-related information in an FAQ-style. A specific micronutrient (e.g. iron, zinc or calcium) was discussed in every newsletter, detailing its relevance for human health and listing potential foods to ensure an adequate intake in both groups (Supplementary Table 3). The newsletter was created by an experienced physician supported by a registered dietician. It also included discounts for selected restaurants in Freiburg, where study participants of both groups received benefits upon showing their individualized certificate of participation (Supplementary Figure 1). Supplementary Table 4 summarizes these discounts and offers.

Bowel health and defecation pattern assessment

Bowel health and defecation pattern assessment have been described previously [11,26], and included the Bristol Stool Form Scale (BSFS) [27], the Gastrointestinal Quality of Life Index (GIQLI) [28], and the Cleveland Clinic Fecal Incontinence Score/Jorge-Wexner Score (CCFIS/JW Score) [29]. The BSFS is a reliable and validated tool to evaluate the stool form of healthy adults [30], and a surrogate marker for its consistency [31]. The BSFS ranges from 1 (separate hard lumps) to 7 (water, no solid pieces) and previous studies suggested that values of 1 or 2 could be used as surrogate for a delayed Colonic Transit Time (CTT) in Western populations [32]. Study participants were asked to define their stool by recording the number type that corresponded to their usual/most common stool type within the last week. Following Wilson's method, BSFS stool types 1 and 2 were defined as constipation, whereas stool types 6 (fluffy pieces with ragged edges) and stool type 7 (described above) were defined as diarrhea [33].

We assessed stool frequency with the question: 'How many times do you have a BM per week?' [11,26]. To screen for potential anal incontinence leakage, we used the CCFIS (Cleveland Clinic Fecal Incontinence Score)/Jorge-Wexner (JW) score [29], which is a widely used and accepted instrument that applies a type x frequency matrix to obtain a participant's perception of symptom. The overall scores ranges from 0 (indicating perfect continence) to 20 (suggesting complete incontinence) and was mainly used for the assessment of gas leakage in our study [34].

Finally, we used the GIQLI developed and validated by Eypasch et al. in Germany [28] to assess alterations in gastrointestinal life quality subsequent to dietary modifications. This reliable and validated questionnaire encompasses 36 items covering 5 sub-domains, such as abdominal symptoms (e.g. bloating and fullness) as well as physical and mental functioning associated with bowel health. There are 5 answer options for each item and scoring ranges from 0 points (the least desirable answer option) to 4 points (the most desirable answer option). Following the instructions of Eypasch et al. [28], the modified (and validated) version for healthy individuals was used [28].

Nutrient intake assessment and weighed food diaries

Dietary intake was estimated based on a 3-day weighed food diary. We instructed participants to quantify all consumed foods and beverages (including tap water) to the nearest 0.1 g or ml, respectively. The participants received detailed instructions in filling in the food diaries by qualified medical personnel and completed protocols were provided as templates. We provided participants with a calibrated precision scale

(Wedo Elektronische Universalwaage Optimo5000, Dieburg, Germany or a comparable model) for a security deposit of 20 Euros.

Participants were asked to document the following information for all foodstuffs and beverages: time and place of consumption, brand name, exact product name, packaging details, condition at the time of purchase, weighed quantity and remaining quantity. In case an exact weighing was impossible, semi-quantitative household recording with measures (e.g. spoon, cups, etc.) was allowed following an established approach by Alexy et al. [35]. NutriGuide® plus software (Version 4.9, Nutri-Science GmbH, Hausach, Germany) was used to analyze the protocols.

Biosampling details

We obtained 130 mL of blood from each participant by venipuncture in the early morning hours. Blood samples were used to measure various laboratory parameters, including a differential blood count, a standard biochemistry profile, a lipid panel as well as for peripheral blood mononuclear cell isolation. For this sub-analysis, we restricted ourselves to reporting high-sensitive C-reactive protein (hs-CRP) and albumin levels, which were deemed to be potentially relevant when discussing nutrient and bowel health related outcomes, and which were measured by the accredited Central Laboratory of the University Medical Center of Freiburg.

Covariates

We measured social status using the German version of the MacArthur Scale of Subjective Social Status (MASSS) [36], which is a pictorial representation that employs a symbolic ladder developed to capture the common sense of social status based on usual socioeconomic status indicators [37]. Physical activity was assessed with the International Physical Activity Questionnaire - Short Form (IPAQ-SF), a reliable method that showed associations with an individual's cardiorespiratory fitness but that also has been reported to overestimate physical activity [38,39]. Supplementation behavior was meticulously captured and participants were asked to bring supplements taken within the last 12 months prior to the baseline examination as described by Kristensen et al. [40]. Supplement names and content as well as intake frequency and quantity were noted. Participants were requested not to take any supplements during the study. Nevertheless, we inquired about supplement/ medication usage during the study period at week 8 to assess for all potentially relevant confounders.

Statistical analysis

We used Stata 14 software (StataCorp. 2015. Stata Statistical Software: Release 14. College Station, TX: StataCorp LP) for the statistical analysis. Box plots and histograms were used in conjunction with Stata's Shapiro-Wilk Test to test for normality. Normally distributed variables were described with their mean and standard deviation whereas categorical variables were described as follows: number of observations (percentage).

Depending on the data distribution, parametric (paired and unpaired two-tailed t-tests) and non-parametric tests (Wilcoxon rank-sum test, Mann–Whitney-U-Test) were used to test for statistically significant intergroup differences in continuous variables. For the intergroup comparison of the proportions of categorical variables, the chi-squared test, Fisher's exact test, McNemar-test, and McNemar-Bowker test were used where applicable. Statistical significance was determined at $\alpha = 0.05$. We used scatterplots, Pearson's correlation coefficients, and Spearman's correlations coefficients to examine potential associations between the estimated fiber intakes and BM frequency as well as the GIQLI. Stripplots were used to visualize the relationship between macronutrient distributions and diet groups.

To control the family-wise error rate when performing multiple hypothesis tests, we used Stata's command 'wyoung'. The command enables the user to calculate adjusted p-values using the free step-down resampling methodology of Westfall and Young [41]. A precursor to the Romano-Wolf procedure, this command allows for dependence amongst p-values. The Westfall-Young correction was performed using 1000 and 10,000 bootstraps where applicable. Familywise error rate-adjusted Westfall-Young p-values were reported for all significant crude outcomes and may be obtained from the legend of the respective tables.

Finally, a repeated measures analysis of variance (ANOVA) was performed for selected outcomes of interest to test for the between-subject factor of diet (two levels: meat-rich diet vs vegan diet), and the within-subject factor of time (three levels: week 0, week 4 and week 8).

Results

The sample encompassed $n = 65$ individuals who completed the study. Figure 2 displays a participant inclusion flowchart, providing an explanation for inclusion and exclusion of participants in our study.

Table 1 shows baseline sample characteristics. The sample included 60 % males and 40% females. Participants had a median age of 23 years and a median BMI of 23.45 kg/m². Approximately 2/3 of the sample were highly active in terms of physical activity (health enhancing physical activity, HEPA). The sample was mainly composed of students, and all participants had finished secondary education with the German Abitur (highest national high-school diploma). No significant differences in baseline sample characteristics were observed between those participants randomized to the vegan diet and those randomized to the meat-rich diet.

Weight slightly decreased in both groups over the course of the study. We observed no significant inter-group differences in the median/mean weight change scores when comparing baseline to week 8 (−1.25 (1.7) kg vs. −1.08 ± 1 kg; p=.295). Likewise, participants did not significantly alter their physical activity levels over the course of the study.
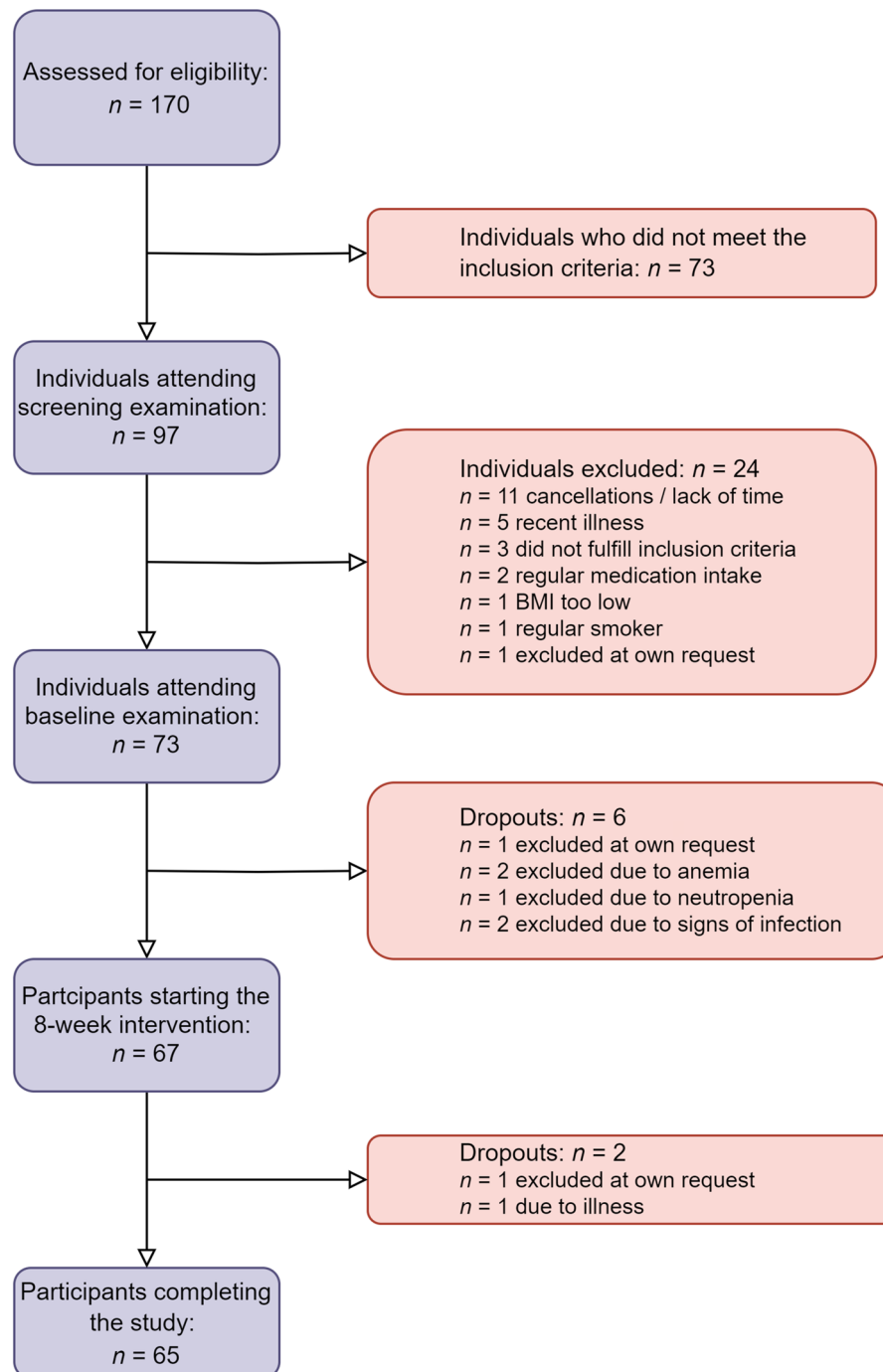

Figure 2. Participant inclusion flowchart.

Table 1. Sample characteristics including sociodemographic, anthropometric and physical activity data at baseline, after 4 weeks and after 8 weeks of the dietary intervention.

Variable	Complete sample (n=65)	Vegan group (n=33)	Meat-rich group (n=32)	p-value
		Baseline		
Sex				.919
Male	n=39 (60%)	n=20 (60.61%)	n=19 (59.38%)	
Female	n=26 (40%)	n=13 (39.39%)	n=13 (40.62%)	
Age (years)	23 (3)	22 (2)	24 (3.5)	.152
Weight (kg)	74.03±9.88	74.15±9.64a	73.91±10.26	.924
Height (cm)	176.44±8.94	177.2±9.03[a]	175.67±8.93	.498
BMI (kg/m^2)	23.45 (3.03)	23.58 (2.77)[a]	23.87±2.1	.582
MASSS	6.86±1.2	6.85±1.3	6.88±1.1	.930
Physical activity level				
Inactive	n=2 (3.08%)	n=2 (6.06%)	n=0 (0%)	.592
Minimally active	n=24 (36.92%)	n=12 (36.36%)	n=12 (37.5%)	
HEPA active	n=39 (60%)	n=19 (57.58%)	n=20 (62.5%)	
Highest education level				
German 'Abitur'	n=50 (76.92%)	n=28 (84.85%)	n=22 (68.75%)	.124
University degree	n=15 (23.08%)	n=5 (15.15%)	n=10 (31.25%)	
		Week 4		
Weight (kg)	73.67±9.72	73.85±9.3	73.48±10.27	.879
Height (cm)	176.75±8.72	177.62±8.66	175.86±8.82	
BMI (kg/m^2)	23.31 (2.65)	23.35±1.77	23.67±2.06	.502
Physical activity level				
Inactive	n=1 (1.54%)	n=0 (0%)	n=1 (3.125%)	.897
Minimally active	n=24 (36.92%)	n=12 (36.36%)	n=12 (37.5%)	
HEPA active	n=40 (61.54%)	n=21 (63.64%)	n=19 (59.375%)	
		Week 8		
Weight (kg)	72.99±9.88	73.14±9.73	72.83±10.19	.879
Height (cm)	177±8.74	177.92±8.75	176.04±8.77	
BMI (kg/m^2)	22.96 (2.87)	23.03±1.84	23.13 (3.24)	.495
MASSS	7.01±1.34	7±1.39	7.03±1.31	.926
Physical activity level				
Inactive	n=1 (1.54%)	n=1 (3.03%)	n=0 (0%)	.352
Minimally active	n=20 (30.77%)	n=8 (24.24%)	n=12 (37.5%)	
HEPA active	n=44 (67.69%)	n=24 (72.73%)	n=20 (62.5%)	

Continuous data displayed as mean±SD if normally distributed or as median (IQR) if not normally distributed. Categorical data displayed as n=x (%).
HEPA: Health enhancing physical activity.
[a]Based on n=32 observations (no anthropometric data available for one person at baseline).

Table 2 shows bowel health outcomes at baseline and over the course of the study. Median weekly BM frequency was 9 for the entire sample and no significant baseline differences between the two groups were detected. The BSFS type 4 was the most prevalent type in both groups at baseline. The mean GIQLI score was slightly higher in those randomized to the vegan group, yet again no significant intergroup differences were found at baseline. Over the course of the study, median BM frequency slightly increased in the vegan group whereas it remained almost unchanged in the meat-rich group (Electronic Supplementary Figure 2). The differences, however, were not statistically significant. No significant intergroup differences were observed in the overall BSFS patterns at week 4 and 8, respectively. Four weeks into the study, vegan participants scored significantly better on the GIQLI emotional subscale. Although the intergroup difference remained significant after adjustments, it appeared marginal from a clinical viewpoint. Gas leakage did not appear to be an issue in any of the groups.

Nutrient intakes at baseline and at week 8 are displayed in Table 3. We observed no significant intake differences at baseline, except for total moisture intake which was significantly higher in those assigned to the vegan group (p=.017). Following the eight-week dietary intervention, significant intergroup intake differences were observed for the following nutrients: protein, fiber, calcium, sodium, phosphorus and zinc. Significant changes in comparison to the baseline-values are marked with an asterisks in Table 3. This table also lists the local dietary recommendations published by the Societies for Nutrition in Germany (DGE), Austria (Österreichische Gesellschaft für Ernährung, ÖGE) and Switzerland (Schweizerische Gesellschaft für Ernährung, SGE), the so-called D-A-CH (D—Deutschland, Germany, A—Austria, CH—Confederation Helvetica, Switzerland) [42].

At baselines no significant differences in mean energy-adjusted fiber intakes were found between the meat-rich and the vegan group (12.75±4.59 g/1000 kcal vs 11.12±4.25 g/1000 kcal). At week 8, significant differences were found (10.88±3.46 g/1000 kcal vs 15.96±4.40 g/1000 kcal) at a p-value <.001.

A pre-post comparison of macronutrient-intake profiles may be obtained from Figure 3.

Table 2. Bowel movement frequency, defecation patterns and gastrointestinal life quality at baseline, after 4 weeks and after 8 weeks of the dietary intervention.

Variable	Range	Complete sample (n=65)	Vegan group (n=33)	Meat-rich group (n=32)	p-values
			Baseline		
BM frequency (n/week)	3–20	9 (5)	8 (5)	9.44±3.32	.695
BSFS					
Type 1		n=1 (1.54%)	n=1 (3.03%)	n=0 (0%)	.703
Type 2		n=8 (12.31%)	n=5 (15.15%)	n=3 (9.38%)	
Type 3		n=21 (32.31%)	n=10 (30.3%)	n=11 (34.38%)	
Type 4		n=28 (43.08%)	n=15 (45.45%)	n=13 (40.63%)	
Type 5		n=5 (7.69%)	n=2 (6.06%)	n=3 (9.38%)	
Type 6		n=2 (3.08%)	n=0 (0%)	n=2 (6.25%)	
Type 7		n=0 (0%)	n=0 (0%)	n=0 (0%)	
JW Score					
Total score	0–20	0 (1)	0 (0)	0 (1)	.498
Solid leakage	0–4	0 (0)	0 (0)	0 (0)	.696
Liquid leakage	0–4	0 (0)	0 (0)	0 (0)	.461
Gas leakage	0–4	0 (1)	0 (0)	0 (1)	.507
GIQLI					
Total score	0–144	132 (8)	132.18±5.93	131.5 (8.5)	.273
Symptoms	0–76	69 (5)	68.81±3.92	68.25±3.8	.555
Emotions	0–20	18 (2)	18 (2)	17.5 (3.5)	**.038**
PF	0–28	26 (2)	25.7±1.57	26 (3.5)	.501
SF	0–16	16 (1)	16 (0)	16 (1.5)	.174
MT	0–4	4 (0)	4 (0)	4 (0)	.564
			Week 4		
BM frequency (n/week)	1–23	9 (6)	9 (7)	9.5±3.84	.755
BSFS					
Type 1		n=1 (1.54%)	n=1 (3.03%)	n=0 (0%)	.748
Type 2		n=1 (1.54%)	n=0 (0%)	n=1 (3.13%)	
Type 3		n=20 (30.77%)	n=10 (30.3%)	n=10 (31.25%)	
Type 4		n=28 (43.08%)	n=15 (45.45%)	n=13 (40.63%)	
Type 5		n=10 (15.38%)	n=4 (12.12%)	n=6 (18.75%)	
Type 6		n=4 (6.15%)	n=3 (9.09%)	n=1 (3.13%)	
Type 7		n=1 (1.54%)	n=0 (0%)	n=1 (3.13%)	
JW Score					
Total score	0–20	0 (0)	0 (0)	0 (1)	.140
Solid leakage	0–4	0 (0)	0 (0)	0 (0)	.639
Liquid leakage	0–4	0 (0)	0 (0)	0 (0)	.983
Gas leakage	0–4	0 (0)	0 (0)	0.31±0.69	.319
GIQLI					
Total score	0–144	130 (10)	130.39±6.21	130.5 (15)	.264
Symptoms	0–76	67 (7)	66.97±4.92	67.5 (9.5)	.703
Emotions	0–20	18 (2)	18 (2)	17±1.78	**.001**
PF	0–28	26 (3)	25.52±1.64	25.5 (4)	.371
SF	0–16	16 (1)	16 (0)	16 (1)	.214
MT	0–4	4 (0)	4 (0)	4 (0)	.237
			Week 8		
BM frequency (n/week)	3–21	9 (7)	9 (7)	9.53±3.48	.701
BSFS					
Type 1		n=1 (1.54%)	n=1 (3.03%)	n=0 (0%)	.490
Type 2		n=8 (12.31%)	n=3 (9.09%)	n=5 (15.63%)	
Type 3		n=21 (32.31%)	n=9 (27.27%)	n=12 (37.5%)	
Type 4		n=22 (33.85%)	n=13 (39.39%)	n=9 (28.13%)	
Type 5		n=9 (13.85%)	n=6 (18.18%)	n=3 (9.38%)	
Type 6		n=4 (6.15%)	n=1 (3.03%)	n=3 (9.38%)	
Type 7		n=0 (0%)	n=0 (0%)	n=0 (0%)	
JW Score					
Total score	0–20	0 (0)	0 (0)	0 (0)	.094
Solid leakage	0–4	0 (0)	0 (0)	0 (0)	NaN
Liquid leakage	0–4	0 (0)	0 (0)	0 (0)	NaN
Gas leakage	0–4	0 (0)	0 (0)	0 (0)	.094
GIQLI					
Total score	0–144	134 (9)	134 (7)	133 (14.5)	.172
Symptoms	0–76	69 (5)	69.76±3.35	69 (9.5)	.218
Emotions	0–20	19 (2)	19 (2)	18 (3)	.101
PF	0–28	26 (2)	26 (2)	26 (3)	.323
SF	0–16	16 (1)	16 (0)	16 (1)	.279
MT	0–4	4 (0)	4 (0)	4 (1)	.340

Continuous data displayed as mean±SD if normally distributed or as median (IQR) if not normally distributed. Categorical data displayed as n=x (%). BM: Bowel Movements; BSFS: Bristol Stool Form Scale; JW: Jorge-Wexner; GIQLI: Gastrointestinal Quality of Life Index; PF: Physical Functioning; SF: Social Function; MT: Medical Treatment; NaN: Not a number. The familywise error rate-adjusted Westfall-Young p-values for the GIQLI emotional subscales at baseline and week 4 were .219 and .017, respectively. Bold indicates significant p-values.

Table 3. Nutrient intakes at baseline and after 8 weeks of the dietary intervention.

Nutrient	Complete sample (n = 65)	Vegan group (n = 33)	Meat-rich group (n = 32)	p-values	D-A-CH recommendations
Baseline					
Energy (kcal/d)	2331.67 (929)	2391.68 ± 719.36	2402.73 ± 752.85	.952	
Fat (g/d)	82.23 (40.4)	78.87 (42.38)	87.83 ± 31.63	.844	30% of tE intake
Carbohydrate (g/d)	267.11 (108.38)	268.14 ± 94.27	268.29 ± 101.26)	.582	>50% of tE intake
Protein (g/d)	87.8 (47.88)	95.54 ± 30.78	88.83 ± 26.8	.353	0.8 g/kg bodyweight/d
Fiber (g/d)	28.20 ± 12.1	26.52 ± 12.02	29.94 ± 12.12	.257	≥30 g/d
Moisture (ml/d)	2685.12 ± 886.03	2939.99 ± 865.27	2422.28 ± 841.09	**.017**	2650 ml/d
Calcium (mg/d)	693.87 (338)	723.35 (359.9)	678.95 ± 229.99	.463	1000 mg/d
Potassium (mg/d)	2167.3 (1244.43)	2105.03 (919.71)	2208.88 (1377.96)	.803	4000 mg/d
Sodium (mg/d)	2351.57 (1160.22)	2448.5 (1462.3)	2333.15 ± 870.32	.358	1500 mg/d
Iron (mg/d)	9.16 (6.23)	8.67 (4.48)	10.17 ± 4.5	.694	M: 10 mg/d; F: 15 mg/d
Magnesium (mg/d)	277.27 (158.88)	235.41 (142.31)	302.63 (202.17)	.600	M: 350 mg/d; F: 300 mg/d
Phosphorus (mg/d)	944.62 (622.95)	894.67 (718.34)	1016.4 ± 355.35	.885	700 mg/d
Zinc (mg/d)	7.94 (5.66)	8.7 ± 4.39	7.81 (5.39)	.990	M: 14 mg/d; F: 8 mg/d
Week 8					
Energy (kcal/d)	2367 (1120)	2328.33 (1033.67)	2405.67 (1336)	.564	
Fat (g/d)	80.68 (50.63)	76.18 (54.05)	82.96 (46.69)	.618	30% of tE intake
Carbohydrate (g/d)	267.08 (172.31)	275.64 (133.64)*	242.62 (169.33)	.096	>50% of tE intake
Protein (g/d)	86.16 (44.6)	75.79 (34.53)*	101.61 (45.35)*	**.003**	0.8 g/kg bodyweight/d
Fiber (g/d)	29.15 (15.69)	34.89 (18.46)**	22.79 (12.5)*	**<.001**	≥30 g/d
Moisture (ml/d)	3168.68 ± 1098.61*	3252.82 ± 1007.96	2882.32 (1032.03)*	.203	2650 ml/d
Calcium (mg/d)	565.07 (319.62)	525.32 (159.88)*	653.36 (547.71)	**.004**	1000 mg/d
Potassium (mg/d)	2169.48 (959.74)	2337.08 (977.88)	2030.52 (907.93)	.582	4000 mg/d
Sodium (mg/d)	2267.48 (1914.65)	2040.33 (1003.01)	2850.55 (2344.52)*	**.014**	1500 mg/d
Iron (mg/d)	9.06 (3.81)	8.95 (4.09)	9.15 (4.03)	.927	M: 10 mg/d; F: 15 mg/d
Magnesium (mg/d)	277.29 (145.9)	306.15 (147.99)*	247.7 (142.59)	.068	M: 350 mg/d; F: 300 mg/d
Phosphorus (mg/d)	858.89 (586.37)	720.84 (408.40)*	1081.39 (644.58)	**.002**	700 mg/d
Zinc (mg/d)	6.31 (5.27)	4.91 (1.96)**	9.38 (6.37)*	**<.001**	M: 14 mg/d; F: 8 mg/d

Continuous data displayed as mean ± SD if normally distributed or as median (IQR) if not normally distributed. The familywise error rate-adjusted Westfall-Young p-values for protein, fiber, calcium, sodium, phosphorus and zinc intake were 0.016, 0.005, 0.007, 0.006, 0.005, and 0.001 respectively.
tE: total energy; d: day; M: male; F: female. D-A-CH (D—Deutschland, Germany, A—Austria, CH—Confederation Helvetica, Switzerland) recommendations modified from [42].
*indicates significant differences in comparison to baseline at a p-value < .05; ** indicates significant differences in comparison to baseline at a p-value < .001.

Table 4 shows the intake of selected food groups that have known associations with BM frequency (e.g. coffee, legumes) over the course of the study. Consumption of legumes as well as tofu and other soy products was significantly more prevalent in the vegan group as compared to the meat-rich diet group after 8 weights.

Figure 4 displays scatterplots visualizing the relationship between BM frequency and fiber intake (top) and GIQLI and fiber intake (bottom). While no significant association between fiber intake and BM frequency was detected in this particular sample, the Spearman's correlation coefficient for fiber intake and GIQLI was significant (p = .03), revealing a weak correlation (Spearman's rho = 0.27) between both variables.

Table 5 shows the repeated measures ANOVA results. No significant diet × time interactions were found for the three investigated endpoints. A significant effect of 'diet' on GIQLI was found (p = .039), yet a closer inspection of the pooled within-subject covariance matrix casted doubt on the validity of the required compound symmetry assumption. Thus, the results must be discussed with great caution. Moreover, the overall effect appeared clinically irrelevant and the p-value was no longer significant after adjustment for multiple testing.

The electronic Supplementary Table 5 shows hs-CRP levels and albumin levels over the course of the study. Although hs-CRP levels increased in the meat-rich group, no significant differences were detected between both groups. Finally, electronic Supplementary Table 6 summarizes bowel health-related adverse events which participants reported over the course of the study. Events occurred more frequently in the meat-rich diet group and largely included postprandial abdominal pain, constipation and two gastrointestinal infections with self-limiting diarrhea and vomiting.

Discussion

The present study investigated bowel health and defecation patterns related to diet and nutrient intake in a young and healthy sample of German university students who were randomly assigned to either a vegan or a meat-rich diet for eight weeks. Our results did not support the hypothesis of a significant increase in BM frequency in vegans. The de-novo adoption of a vegan diet neither resulted in a transient increase in abdominal discomfort nor in a decreased gastrointestinal quality of life, which could have occurred subsequent to the sudden increase in dietary fibers and selected phytochemicals (mainly phytic acid). These results require a thorough
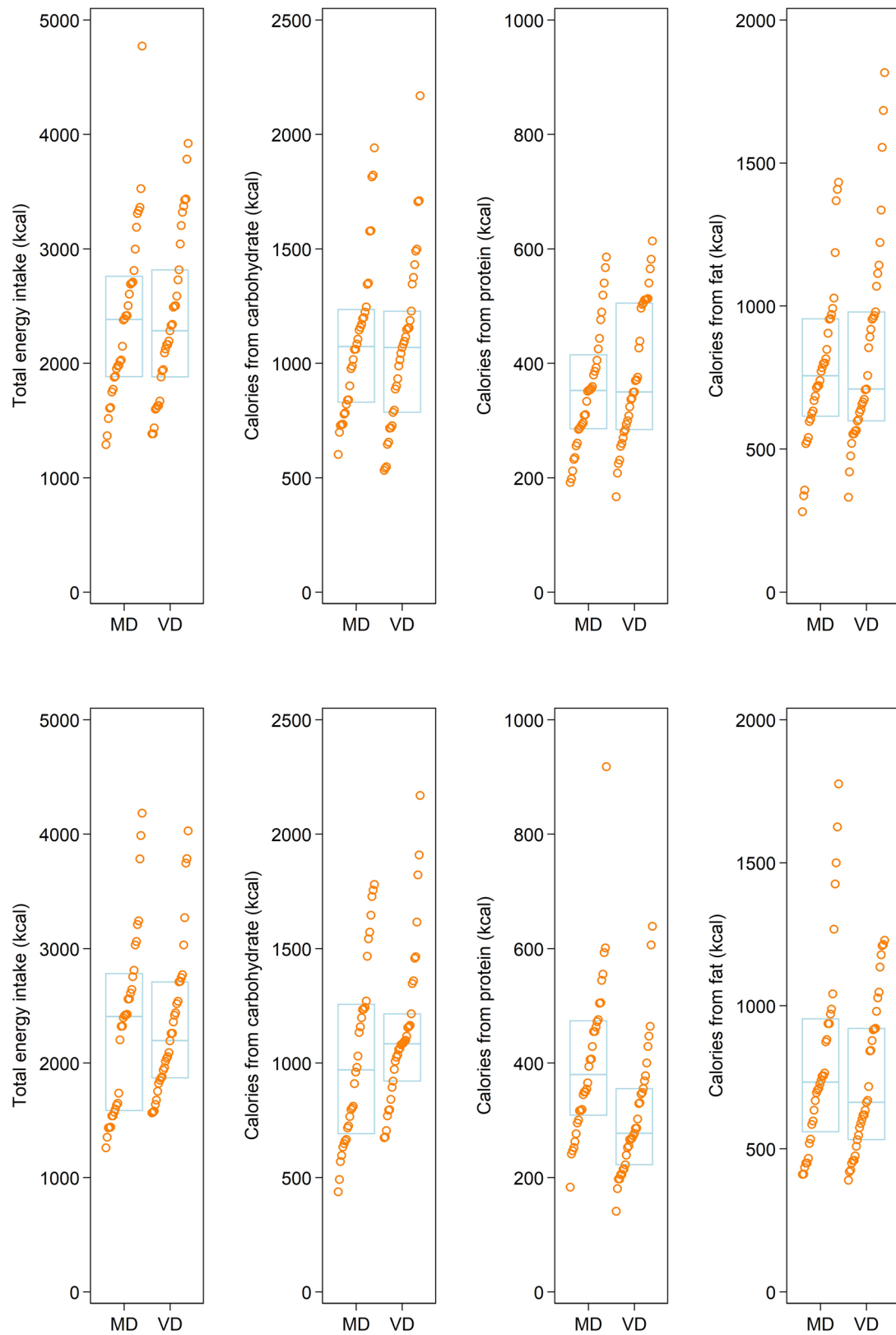
Figure 3. Macronutrient profile in both groups before and after the intervention. Stripplot showing macronutrient profiles for both groups at baseline (top) and after eight weeks (bottom). MD: Meat-rich diet; VD: Vegan diet.

Table 4. Intakes of selected food groups related to bowel health and dietary group at baseline and after 8 weeks of the dietary intervention.

Food group	Complete sample (n=65)	Vegan group (n=33)	Meat-rich group (n=32)	p-values
		Week 0		
Coffee intake				
Yes	n=33 (50.77%)	n=17 (51.52%)	n=16 (50%)	.903
No	n=32 (49.23%)	n=16 (48.48%)	n=16 (50%)	
Espresso intake				
Yes	n=7 (10.77%)	n=3 (9.09%)	n=4 (12.5%)	.708
No	n=58 (89.23%)	n=30 (90.91%)	n=28 (87.5%)	
NDMA intake				
Yes	n=33 (50.77%)	n=14 (42.42%)	n=19 (59.38%)	.172
No	n=32 (49.23%)	n=19 (57.58%)	n=13 (40.62%)	
Legumes intake				
Yes	n=29 (44.62%)	n=17 (51.52%)	n=12 (37.5%)	.256
No	n=36 (55.38%)	n=16 (48.48%)	n=20 (62.5%)	
Tofu intake				
Yes	n=21 (32.31%)	n=10 (30.3%)	n=11 (34.38%)	.726
No	n=44 (67.69%)	n=23 (69.7%)	n=21 (65.63%)	
Soy yoghurt intake				
Yes	n=4 (6.15%)	n=2 (6.06%)	n=2 (6.25%)	
No	n=61 (93.85%)	n=31 (93.94%)	n=30 (93.75%)	
		Week 8		
Coffee intake				
Yes	n=30 (46.15%)	n=16 (48.48%)	n=14 (43.75%)	.702
No	n=35 (53.85%)	n=17 (51.52%)	n=18 (56.25%)	
Espresso intake				
Yes	n=12 (18.46%)	n=3 (9.09%)	n=9 (28.13%)	.061
No	n=53 (81.54%)	n=30 (90.91%)	n=23 (71.88%)	
NDMA intake				
Yes	n=40 (61.54%)	n=24 (72.73%)	n=16 (50%)	.06
No	n=25 (38.46%)	n=9 (27.27%)	n=16 (50%)	
Legumes intake				
Yes	n=39 (0.6%)	n=27 (81.82%)	n=12 (37.5%)	**<.001**
No	n=26 (0.4%)	n=6 (18.18%)	n=20 (62.5%)	
Tofu intake				
Yes	n=24 (36.92%)	n=18 (54.55%)	n=6 (18.75%)	**.003**
No	n=41 (63.08%)	n=15 (45.45%)	n=26 (81.25%)	
Soy yoghurt intake				
Yes	n=17 (26.15%)	n=17 (51.52%)	n=0 (0%)	**<.001**
No	n=48 (73.85%)	n=16 (48.48%)	n=32 (100%)	

Significant intake differences were found for legumes, tofu and soy yoghurt after the dietary intervention (week 8). The familywise error rate-adjusted Westfall-Young p-values for legumes and tofu intake were 0.001 and 0.019, respectively. Due to the unique data distribution of soy yoghurt intake in the meat-rich diet group (n=0 vs n=32 observations), a reliable familywise error rate-adjusted Westfall-Young p-value could not be properly computed for technical limitations. In light of the data, a significant p-value after adjustment is to be expected. NDMA: Non-Dairy Milk Alternative.

contextual discussion which considers previous publications on bowel health in plant-based diets.

Fiber-rich diets influence gut motility and defecation patterns [43]. Davies et al. reported more frequent bowel movements and softer stools in vegans when compared to vegetarians and omnivores [13]. In their cross-sectional study, vegans had, on average, 1.7 BM per day, whereas vegetarians and omnivores had 1.2 and 1.0, respectively. The authors reported that increasing fiber was associated with a shorter transit time, and – unsurprisingly – vegans had the highest intake (47 g), followed by vegetarians (37 g) and omnivores (23 g). Vegans in the Davies study consumed substantially more fiber than vegans in our study (47 g vs. 34.89 g/d), who barely met the minimum fiber intake recommendations of 14 g/1000 kcal [44]. This could be one of many possible factors explaining the lack of an increase in BM frequency in vegan participants in our study.

Another study that examined BM frequency in vegans used data from the European Prospective Investigation into Cancer and Nutrition Oxford cohort [17]. Sanjoaquin et al. reported a higher BM frequency in vegans, with sex-specific differences. Vegan men had on average 11.6 BM per week, as compared to 9.5 in male meat eaters. Vegan women on the other hand reported 10.5 BM per week, as compared to 8.2 in female meat eaters. The authors reported significant associations with fiber and moisture intake (restricted to non-alcoholic beverages). Vigorous exercise was also positively associated with BM frequency in women in this study, while results for men were less clear.

All of these factors play a pivotal role for bowel health and must be considered simultaneously, particularly because an adequate intake of fluid is necessary to enhance the positive effects of dietary fibers in stool frequency regulation [45]. Studies demonstrated
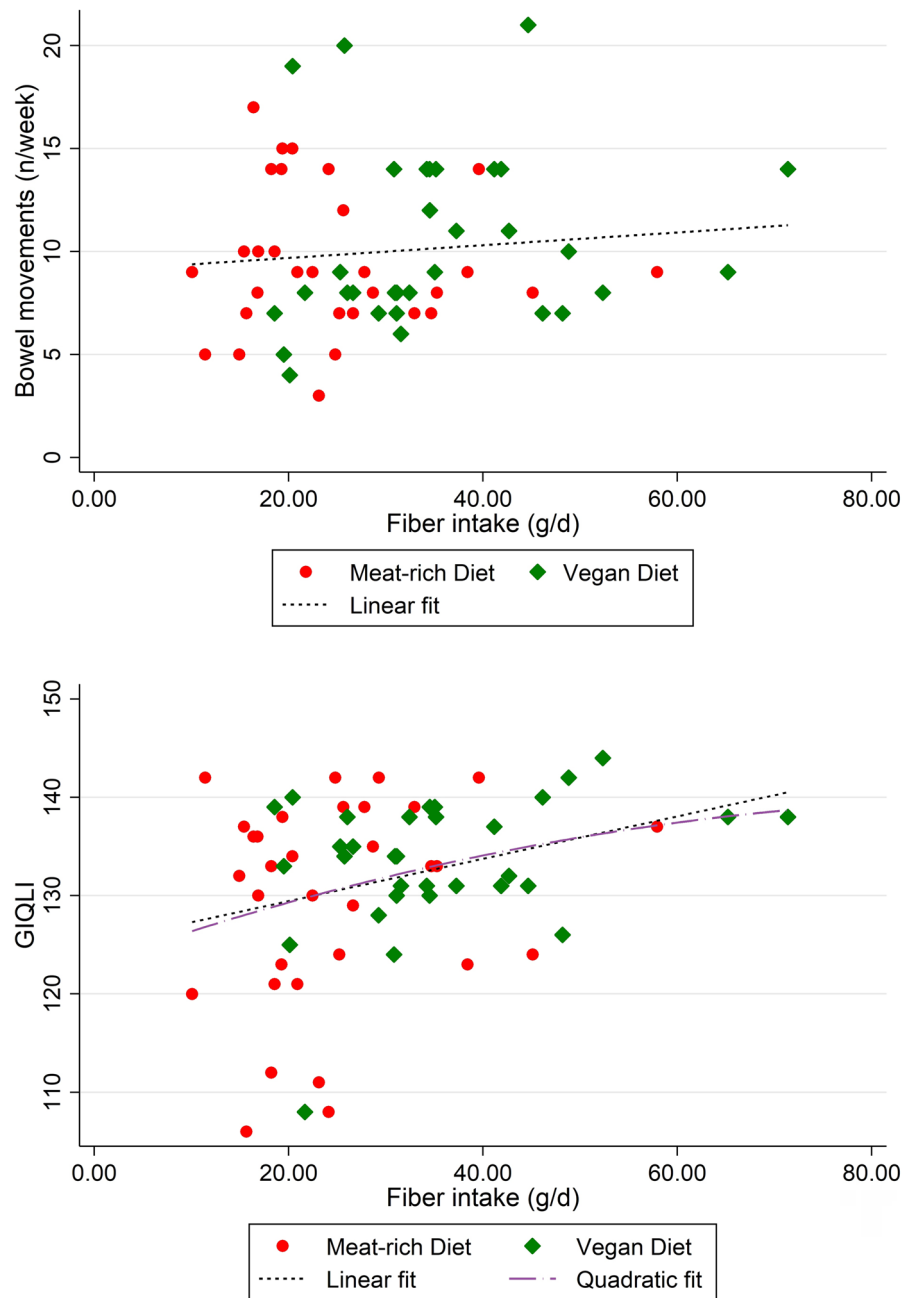
Figure 4. Scatterplots visualizing the relationship between BM frequency and fiber intake (top) and GIQLI and fiber intake (bottom). BM: Bowel Movement; GIQLI: Gastrointestinal Quality of Life Index. One outlier (fiber intake per day: 120 g) in the vegan group was removed to highlight full cohort behavior over the casuality of one outlier. Spearman's rho for fiber intake and GIQLI was 0.27 (p = .03), whereas it was not significant for fiber and BM frequency.

Table 5. Comparison of BM frequency, BSFS, and GIQLI across dietary groups.

| | Baseline | | Week 4 | | Week 8 | | p-value (Baseline – End) | | |
	Vegan Diet	MR Diet	Vegan Diet	MR Diet	Vegan Diet	MR Diet	Time	Diet	Time X Diet
BM frequency	8 (5)	9.44 ± 3.32	9 (7)	9.5 ± 3.84	9 (7)	9.53 ± 3.48	.078	.475	.142
BSFS (mean)	3.39 ± 0.9	3.67 ± 0.96	3.91 ± 1.04	3.97 ± 1.03	3.70 ± 1.07	3.59 ± 1.16	**.007**	.338	.378
GIQLI	132.18 ± 5.93	131.5 (8.5)	130.39 ± 6.21	130.5 (15)	134 (7)	133 (14.5)	**.001**	**.039**	.566

BM: Bowel movement; BSFS: Bristol Stool Form Scale; GIQLI: Gastrointestinal Quality of Life Index. BM frequency in number of BM/week. Table 5 shows unadjusted p-values. The p-value for the effect of 'Diet' on GIQLI was no longer significant after adjustment for multiple hypothesis testing.
Results of repeated measures ANOVA results are given for baseline vs 8 weeks, wherein the effects of vegan and meat-based diets were expectedly maximal.

that even a relatively short period of fluid deprivation may decrease BM frequency and could be an etiological factor for constipation [46,47]. While liquid deprivation was not an issue in our study cohort, it must be emphasized that the proportion of physically active participants was high. This could have partially offset the effects of a mild fiber reduction in the meat-rich diet group, as physical activity has been repetitively associated with BM frequency [48].

The unaltered BM frequency in vegans in our study still remains surprising, and, taking into account all potential etiological factors, might be the result of the only moderate increase in dietary fiber intake over the course of the study. While fiber intake increased in the vegan group compared to baseline, it still remained somewhat pale in comparison to other vegan cohorts across Europe. Clarys et al. reported a mean fiber intake of 41 g in Belgian vegans [49], while Kristensen et al. reported an intake of 56 g/d in vegan Danish men [40]. This again demonstrates that not all plant-based diets are created equal [1]. While vegans in our study consumed substantially less fiber, a reservation must be made that they were all naïve to the vegan diet, which requires considerable time and knowledge to adopt and master [50]. Another important difference with previously published studies is that we purposely selected participants who had been on a healthy omnivore diet at baseline, with an overall nutrient intake and level of physical activity that were health-promoting, prior to the dietary intervention. This may explain the relatively small effect size recorded for measures of bowel health, defecation patterns and gastrointestinal life-quality investigated in our study.

Particularly the regular intake and consumption of fiber-rich plant-foods, such as legumes and whole grains, requires habituation and practice [51]. While legume consumption increased in our study, vegans also often consumed fiber-depleted meat analogues and wheat gluten-based replacement products, which did not necessarily contribute to their overall fiber intake. Optimizing diet quality on a vegan diet for beginners is challenging and requires substantial nutritional knowledge and cooking skills. Acquiring these in a mere eight weeks seems unfeasible and could have theoretically contributed to the non-significant changes in BM frequency. While vegans optimized their intake of magnesium and fiber, their potassium intake remained worthy of improvement. The latter is an important indicator of fruit and vegetable intake [52], and reiterates that diet quality in vegans improved as part of the study but could have overall been much better.

It must also be emphasized that gastrointestinal quality of life did not decrease in the vegan group.

This is of paramount importance, since abdominal discomfort may be expected upon a sudden fiber intake increase when transitioning from an omnivorous to a vegan plant-based diet. Often used as an argument against plant-based dietary patterns, this phenomenon was not observed in our study. Significant improvements in the GIQLI though were not observed in either group, potentially because the baseline scores were already very high and because we exclusively recruited healthy young individuals.

Our study has strengths and limitations that warrant further discussion. As for the strengths, we compiled a homogenous study sample of young and healthy individuals. Rigorous exclusion criteria and a thorough consideration of relevant covariates and confounders (including physical activity, subjective social status and educational level) are an additional asset. Weighed food diaries are considered the 'gold standard' and the most precise method to quantify food intake, particularly in vegans who frequently consume foods that may not be found in commonly used food frequency questionnaires [53]. Participants of both groups received the same amount of attention and support in terms of group seminars, newsletters and individual interaction to reduce bias. To the best of our knowledge, we report the first randomized-controlled trial investigating the effects of a *de-novo* adoption of a vegan diet on selected parameters of bowel health.

Weaknesses include the modest sample size and potential reporting bias. Participants of both groups were meticulously instructed to maintain their weight as this parameter is known to influence other primary outcomes of the study (e.g. serum cholesterol levels, gut microbiota). While a slight weight loss occurred in both groups, the mean weight loss difference was not significant. It must be noted though that vegan diets are usually characterized by a higher nutrient density and reduced caloric density which both promote earlier satiety and could lead to subsequent weight loss [20]. To maintain their weight, some participants in the vegan group reported the consumption of granola bars, nuts and sweets to comply with their daily energy intake target of approximately 2000 kcal/d. Minimizing the odds for weight reduction was important for the study, as weight loss per se might have changed metabolomics parameters or well-being. Then again, energy intake in vegans in our study was slightly higher as expected compared to free-living vegans. The lack of gut microbiota richness analyses, which are associated with stool consistency, is another weakness that will be subject to future analyses [54].

Ultimately, we acknowledge that this randomized-controlled study was performed in free-living individuals

who were allowed to prepare their foods for themselves. No meals were provided and participants were free to select the foods they liked as long as it was in accordance with the study protocol and the requirements of the assigned dietary group. This lack of control may have introduced bias and confounders, which we intended to minimize with regular and close participant contact to ensure a high dietary adherence. Nevertheless, we cannot guarantee 100% dietary adherence in both groups. Then again, the sample was almost exclusively composed of students who preponderantly disclosed to have eaten at the local university cafeteria that provided both a hot meat-based and a hot vegan lunch. After all, controlling all meals is generally impossible in such a study design. While limiting the generalizability of our study, an inpatient setting was also deemed unfeasible in light of the particular study characteristics (study duration, extent, etc.) and the large amount of resources required for infrastructure, personnel and expense allowances in such cases.

To facilitate the recruiting process for a study with a potentially challenging dietary regimen, participants with a BMI between 21 and 30 kg/m^2 were considered. While a BMI > 25 kg/m^2 may no longer be considered normal or healthy as per definition [55], we decided to include such individuals to ensure continuous recruiting. We acknowledge that this could have introduced some important bias and limitations; then again the share of participants with a BMI ≥ 26 kg/m^2 was 'only' 15% in our study.

Conclusion

The short-term *de-novo* adoption of a vegan diet did not negatively affect bowel health. No clinically relevant differences in BM frequency and BSFS patterns were observed in vegan participants with a moderate increase in fiber intake.

Acknowledgements

Consent form

Written consent was obtained from all participants to publish the results of this study.

Ethics approval

The ethical committee of the University Medical Center of Freiburg approved this trial in January 2023 (approval number: 22-1474-S1). The study was prospectively registered at the German Clinical Trial Register (DRKS00031541). We obtained written and oral informed consent from all study participants.

Authors contributions

J. H. planned and conducted the study, supported the data analysis, performed PBMC isolation, and served as the main contact person for study participants.

F. S. conducted the study, analyzed the weighed food diaries, performed PBMC isolation, and was involved in the in-field organization.

V. L. was responsible for all laboratory-related aspects, performed and supervised the PBMC isolation and managed biosampling.

J.Z. supported the study related to organizational aspects and helped analyzing the weighed food diaries.

A.K.L. supported the study planning and the statistical analysis.

L.H. participated in experimental planning and data interpretation.

R. H. conceived, planned and supervised the clinical trial. R. H. was involved in funding acquisition and helped in the preparation of the manuscript.

M.A.S. served as the principal investigator, conceived, planned, conducted and supervised the clinical trial, analyzed data, analyzed the weighed food diaries, acquired funding and wrote the manuscript.

Disclosure statement

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Data availability statement

The datasets used and analyzed for the current study are available from the corresponding author on reasonable request.

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