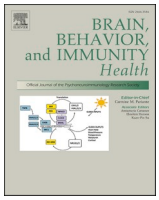
Brain, Behavior, & Immunity - Health 43 (2025) 100918

Contents lists available at ScienceDirect

Brain, Behavior, & Immunity - Health

journal homepage: www.editorialmanager.com/bbih/default.aspx



# A prebiotic intervention improves mood in everyday life in healthy women but not in men: Exploratory results from a larger double-blind placebo controlled cross-over study

Koen Hogenelst [a], Tanja Krone [b], Boukje Eveleens Maarse [c,d], Ines Warnke [e], Jessica Snabel [f], Tim J. van den Broek [g], Frank Schuren [g], Matthijs Moerland [c,d,*], Femke P.M. Hoevenaars [g]

[a] Department of Human Performance, The Netherlands Organisation for Applied Scientific Research (TNO), Soesterberg, the Netherlands
[b] Department of Risk Analysis for Products in Development, The Netherlands Organisation for Applied Scientific Research (TNO), Utrecht, the Netherlands
[c] Centre for Human Drug Research, Leiden, the Netherlands
[d] Leiden University Medical Center, Leiden, the Netherlands
[e] DSM -Firmenich, Kaiseraugst, Switzerland
[f] Department of Metabolic Health Research, The Netherlands Organisation for Applied Scientific Research (TNO), Leiden, the Netherlands
[g] Department of Microbiology and Systems Biology, The Netherlands Organisation for Applied Scientific Research (TNO), Leiden, the Netherlands

ARTICLE INFO

*Keywords:*
Prebiotics
Microbiome
Positive affect
Negative affect
Ecological momentary assessment
Cortisol
Cytokines
Everyday mood

ABSTRACT

Prebiotic dietary fiber (PDF) may reduce feelings of stress or improve mood in healthy individuals. Yet gut intervention studies that focus on mood in daily life are lacking and few studies include extensive biological sample analyses to gain mechanistic insights. As part of a larger randomized placebo-controlled crossover study including healthy individuals, we explored the effects of 12 weeks of PDF (acacia gum and carrot powder) on everyday mood, as measured with ecological momentary assessment (EMA). Microbiome composition and levels of microbial metabolites, endocrine, and inflammatory markers were determined prior to and after both intervention phases. Fifty-four participants completed the study. The intervention significantly increased daily positive affect (PA) and reduced daily negative affect (NA) in female but not male participants. The intervention-induced reduction in NA was associated with an increase in microbial diversity in female participants. The intervention did not significantly affect levels of fecal short chain fatty acids, cortisol, and inflammatory markers. This is one of the first studies to show that a dietary fiber intervention can positively alter mood as it is experienced in everyday life. Overall, our findings may stimulate more targeted gut-microbiome interventions and detection of its mental health effects in real life.

## 1. Introduction

The gut microbiome has an intricate relation with mental health and wellbeing (Desmedt et al., 2019; Liu et al., 2019; Berding and Cryan, 2022). Interventions with pre- or probiotics that modify the gut microbiome have shown symptom improvement in people with psychiatric conditions, including depression and anxiety (Noonan et al., 2020) as well as reductions in feelings of stress or improved mood in healthy individuals (Desmedt et al., 2019; Zhang et al., 2020). In their endeavor to elucidate involvement of the gut microbiota in gut-brain communication, researchers have explored possible underlying mechanisms including microbial-derived metabolites, and hormonal, immune, metabolic, and neuronal pathways (Berding et al., 2021a,b; Osadchiy et al., 2019; Spichak et al., 2021). Yet, how dietary interventions that modulate the gut microbiome exactly contribute to aforementioned mental health benefits in humans remains largely unclear. Filling some of the knowledge gaps requires well controlled intervention studies with extensive biological sample analyses (Chakrabarti et al., 2022) and reliable and sensitive prospective outcome assessments. We aimed to fill the gap by studying the effect of a prebiotic intervention on mood in everyday life, as well as on microbiome composition, and relevant microbial metabolites, endocrine, and immune markers.

Prebiotics are indigestible food ingredients (such as dietary fibres) that are selectively utilized by host microorganisms and stimulate growth or functioning of beneficial bacterial species (Gibson et al., 2017). Given the abundant and increasing levels of stress or mental

* Corresponding author. Centre for Human Drug Research, Leiden, the Netherlands
*E-mail address:* MMoerland@chdr.nl (M. Moerland).

https://doi.org/10.1016/j.bbih.2024.100918
Received 20 June 2024; Received in revised form 15 November 2024; Accepted 27 November 2024
Available online 28 November 2024

health problems among the general population (Twenge et al., 2019), it is relevant to highlight research on prebiotic interventions showing promising effects on mental health in healthy adults, including possible underlying mechanisms. A handful of studies show that prebiotics can have a positive impact on affect or other mental health parameters (Best et al., 2009; Talbott and Talbott, 2009; Lawton et al., 2013; Childs et al., 2014; Schmidt et al., 2015; Berding et al., 2021a,b; Berding et al., 2022), but caution is advised as there is little consistency among interventions and affected parameters and several studies show no clear effects on affect, mood, and anxiety (Desmedt et al., 2019; Johnstone et al., 2021). Furthermore, only one intervention study that used prebiotics included extensive biological sample analyses (Berding et al., 2022) underscoring the need for more comprehensive and rigorously designed studies.

One common denominator of performed studies is the use of retrospective questionnaires for mental health, usually administered once or few times, often only before and after a gut microbiome intervention. While these are often validated questionnaires and provide valuable insights, the obtained data may suffer from several biases, such as recall bias and mood congruent memory bias (Moskowitz and Young, 2006; Trull and Ebner-Priemer, 2009). To avoid this and to study mental health as it is experienced in daily life and across days, diary approaches including ecological momentary assessment (EMA) can be used as an alternative (Mehl and Conner, 2012). Using a correlational design, previous EMA studies have reported positive associations between eating fiber rich foods such as fruit and vegetables with feelings of well-being, curiosity and creativity (Conner et al., 2015) as well as positive affect (White et al., 2013). Furthermore, a 14-day intervention that stimulated fruit and vegetable intake resulted in improvements in daily well-being with increases in vitality, flourishing, and motivation as compared to a diet-as-usual group (Conner et al., 2017). In another EMA study, tryptophan supplementation in healthy individuals improved daily positive affect and reduced daily negative affect (Hogenelst et al., 2015). In addition, EMA may be used to capture momentary, yet relevant affect changes around daily positive and negative events. This affective reactivity is predictive of depressive symptoms in the general population (Booij et al., 2018). Given that attention to negative emotional information, which is often increased in people with depression (Beck, 2008), may be beneficially altered by prebiotics (Schmidt et al., 2015; Johnstone et al., 2021), prebiotics may also beneficially alter another depression feature, namely affective reactivity to daily life events.

The present study was part of a larger study primarily designed to investigate metabolic health outcomes of a dietary fibre mixture of acacia gum (AG) and carrot powder including pectin (KaroPRO) [ratio 10 + 3]. Microbiome composition, immune markers and fecal SCFAs analyses were all part of the initial protocol (see Eveleens Maarse et al., 2024). By adding mental health endpoints and saliva sampling we were able to exploratively assess the effects of the prebiotic mixture on mental health and potential underlying mechanisms. For the present study we explored the effects of prebiotic intake on daily mood (positive and negative affect) in healthy individuals. We hypothesized that prebiotic intake would increase positive affect and reduce negative affect. In addition, we explored microbial composition and function (SCFAs in faeces), endocrine (cortisol), and immune (e.g., cytokines) biomarkers to shed light on possible underlying gut-to-brain mechanisms. Lastly, we explored if intervention-induced daily affect responses could be linked to microbiome changes and what other factors characterize intervention response.

## 2. Methods

The data reported here are part of a larger study that is described in greater detail by Eveleens Maarse et al. (2024). Here, we describe only the measurements and procedures used in the current sub-study. The study was approved by a local medical ethics board (Toetsingonline Registry NL71723.056.19), registered in the clinicaltrials.gov register (NCT04829396), and was executed according to Good Clinical Practice (GCP). Prior to any study-related activity, all subjects signed an informed consent form according to Declaration of Helsinki recommendations. The study was conducted at the Centre for Human Drug Research (CHDR) in Leiden, The Netherlands, between August 2020 and July 2021.

### 2.1. Study design and participants

This study was designed as a double-blind, randomized, placebo-controlled crossover study with two 12-week intervention periods separated by 8 weeks of wash-out. In each intervention period participants visited the study centre at baseline and again after 4, 8 and 12 weeks. Men and women could participate if they met the following inclusion criteria: age 45–70 years, BMI 25–30 kg/m2, no current major medical illness. Exclusion criteria were: use of antibiotics, antacids, laxatives, anti-diarrheal, or immunomodulatory medication within 3 months before the start of the study, and use of any concomitant medication, vitamins or dietary supplements within 7 days (or 5 half-lives) before start of the study or during the study, with exception of paracetamol and ibuprofen. A total of 65 participants were included in the study.

### 2.2. Intervention

The intervention consisted of a daily 13g fibre mixture or placebo powder, for 12 weeks each. Fibre and placebo intervention powders were dispensed in identical jars and were matching in appearance. The order was randomized across participants. The fibre mixture was prepared in a ratio ensuring that participants took 10g Acacia gum powder (Type 4880, A. Seyal, Willy Benecke, Germany) and 3g milled carrot powder (KaroPRO 1–26 SG, Food Solutions Team B.V., The Netherlands) with every dosing. See Eveleens Maarse et al. (2024) for further details. Placebo powder consisted of brown (Glucidex® 19) and white (Glucidex® 17) maltodextrin only (Roquette, France), to colour match the fibre powder.

### 2.3. Measurements

#### 2.3.1. Questionnaires for general health, gastro-intestinal symptoms and stool form

Health-related quality of life was assessed with the short form-12v2 (SF-12) questionnaire (Ware et al., 1996). The SF-12 uses 12 items tapping into eight health domains to assess physical and mental health. Scores range from 0 to 100. A higher score represents better health.

Gastro-intestinal symptoms were assessed with the gastro-intestinal symptoms rating scale (GSRS). The GSRS uses a 7-point Likert scale, based on the intensity and frequency of gastrointestinal symptoms experienced during the previous seven days (Svedlund et al., 1988). The 15 items combine into five symptom clusters: Reflux, Abdominal pain, Indigestion, Diarrhea and Constipation. A higher score represents more symptoms.

Stool form was assessed with the Bristol Stool Form Scale (BSFS). The BSFS is a 7-point scale used extensively in clinical practice and research for stool form measurement, with established validity and reliability, including in healthy participants (Blake et al., 2016).

#### 2.3.2. Ecological momentary assessment (EMA)

Participants used a smartphone-based EMA application (app) to record daily positive and negative affect, physical discomfort, and stool form. Because the EMA was an addition to a larger study and to avoid participant burden, we only measured for one period in each study phase. Starting in week 10 of each intervention phase and for a period of 10 d, the app prompted participants six times a day to answer a fixed set of EMA questions (explained in detail below). Prompts were randomly delivered between 09:30 and 21:30 h, with a minimum interval of 90

min. Following each prompt, the questionnaire was available for 30 min. A reminder prompt was sent 15 min after the initial prompt in case of non-response.

*Positive affect (PA), negative affect (NA), and other EMA items*: Momentary affect items were derived from the circumplex model of affect (Feldman Barrett and Russell, 1998). Each item started with "At this moment I feel …". PA items included relaxed, energetic, enthusiastic, content, calm, and cheerful. NA items included gloomy, anxious, nervous, irritable, dull, and tired. Each item was rated on a visual analogue scale (VAS) with 99 increments labeled from "Not at all" (0) to "Very much" (100). For each prompt, responses to PA and NA items were averaged into a PA and NA score (range 0–100), respectively. With each prompt, participants could also report if they experienced a *pleasant* and/or *unpleasant event* since the last prompt. If so, the event could be rated in terms of (un)pleasantness on a VAS labeled "not (un) pleasant" (0) to very (un)pleasant (100). To assess daily *Physical discomfort (PD)*, a single item was used: "To what extent to you currently experience physical complaints or physical discomfort?" rated on a VAS labeled "No discomfort at all" (0) to "A lot of discomfort" (100). For the last prompt of each day, the fixed set of EMA questions was expanded with questions for stool frequency and form. The first question inquired if people had bowel movements that day. If the answer was "yes", a next question inquired about frequency and for each bowel movement participants could indicate the stool form using the seven BSS visual scales that were incorporated in the app.

*Affective reactivity:* To investigate the extent to which the intervention also influenced affective responses to daily (un)pleasant events, prompt-to-prompt changes in PA and NA were computed. These changes were computed as PA(t0)-PA(t-1) for example. Ratings of (un)pleasant events of the first prompt of the day were excluded as the prompt prior to that was of the day before. This allowed us to determine PA increases to positive events, PA decreases to negative events, NA decreases to positive events, and NA increases to negative events.

#### 2.3.3. Microbiome profiling

Profiling of the microbiome composition was performed on fecal samples that were collected at baseline and weeks 4, 8 and 12 in each intervention phase. Microbial DNA was extracted from fecal samples and 16S rRNA gene sequencing was performed as described previously (Gart et al., 2018; see also supplementary material). Microbial diversity was either determined by inverse Simpson index or Shannon index. We used inverse Simpson instead of just Simpson index because it provides a more intuitive measure of the effective number of species making. The inverse form offers a linear response to changes in dominance so that the interpretation becomes more intuitive versus the Simpson index.

#### 2.3.4. SCFAs

Faecal SCFAs (iso-valeric acid, propionic acid, iso-butyric acid, butyric acid, valeric acid, acetic acid, 2-methylbutanoic acids) were measured by liquid chromatography-mass spectrometry (LC-MS) using a high-resolution mass spectrometer (Q-Exactive, Thermo, USA), as further described in the supplementary material (and by Eveleens Maarse et al. (2024).

#### 2.3.5. Cortisol awakening response

Participants could wake up according to their own schedule. Morning saliva samples on study visit days were collected for assessment of the cortisol awakening response (CAR) using the Salivette system (Sarstedt, Germany). Three samples were collected, the first upon wakening, the second and third 30 and 45 min later. Participants were instructed to not brush their teeth until after all saliva samples were collected, to not eat or drink anything prior to the first sample, and to avoid eating and drinking 15 min prior to the remaining samples. Participants stored collected saliva in their fridge prior to the study visit that same morning. Upon study site arrival, samples were centrifuged within 2 h, for 10 min at 2000 G and 4 °C, aliquoted, and stored at −80 °C for later analysis. Salivary cortisol was analyzed in duplicates using enzyme-linked immunosorbent assay (ELISA) kits (Cortisol parameter assay kit, R&D systems, Inc., Bio-Techne®, Abingdon, UK). The lower detection limit was 0.55 nmol/l. Inter- and intraassay variability coefficients were 13.6% and 7.0%. For subsequent analyses, the area under the curve of the cortisol assessments was computed with respect to ground (AUCg) and increase (AUCi) (Pruesner et al., 2003).

#### 2.3.6. Inflammatory parameters

With each site visit, blood was sampled by venipuncture. To explore the role of inflammatory marker in the present study, we included data of cytokines (interleukin [IL]-1β, IL-2, IL-4, IL-6, IL-8, IL-10, IL-12p70, IL-13, interferon [IFN]-γ, tumor necrosis factor [TNF] and C-reactive protein (CRP). Details regarding quantification are provided in the supplementary material.

### 2.4. Study procedures

Participants were instructed to take in 13g of powder daily with their breakfast, stirred into 200–400 mL milk, yoghurt or smoothie. They received a measuring spoon for 13g and instructions to make sure intake was identical every day. On the first and last day of every intervention period of 12 weeks, participants visited the study site (4 times in total). During those visits the SF-12 and GSRS were completed and blood was sampled. Participants handed in fecal samples during the visits. With each fecal sample participants completed a BSFS rating. To determine intake compliance, participants were required to hand in the empty fibre/placebo jars at the end of each intervention period. By weighting these jars, total fibre or placebo intake could be calculated. Intake of minimal 80% of the total dosing was considered as compliant. In week 10 of each intervention period, the EMA app procedure started as described previously.

### 2.5. Statistical analyses

Data analysis was carried out using R statistics v4.3.1 (R Core Team, 2023). For the SF-12, the raw scores were submitted to the software program 'ProCore' (v1.5; ProCore, 2019) and the resulting mental and physical component scores were used. The GSRS gave five different outcomes (Diarrhea syndrome, indigestion syndrome, constipation syndrome, abdominal pain syndrome, reflux syndrome) after averaging the combined questions for each of these dimensions. Missing data was imputed as the average score per scale where it pertained 50% or less than the raw scores for that dimension. Where used, differences between post- and pre-intervention scores were calculated by subtracting the pre-intervention score from the post-intervention score. The BSS was calculated as the absolute deviation from the most optimal score of 4 for the first scored BSS per measurement day subtracted from 4. As such, a score of 6 would result in 2, but so would a score of 2, while a 4 would remain a 4. Analyses were done using a linear mixed effects model with the subject as random effect to account for individual difference that were persistent over the measurement period.

Outcome variables of EMA were PA, NA, and prompt-to-prompt affect changes and physical discomfort. For intervention effects on affect we considered the within-subjects factor intervention (prebiotic vs placebo), the between-subjects factor sex (male, female), and their interaction as possible predictors. For assessing affective reactivity, we considered event (pleasant, unpleasant, no event reported) as an additional within-subjects factor. To allow for unintended time effects, we checked the effect of treatment order (i.e., prebiotic first or placebo first) and the effect of time as overall variable. These effects were non-significant or negligible, and are as such not included in the final analyses (results available upon request). The effect of treatment and sex on PA, NA and discomfort were studied using linear mixed effect models with maximum likelihood estimation using the package LME4 (Bates et al., 2015). Subject was included as a random effect, to account for

individual differences in baseline assessment of PA, NA and discomfort. Sex and treatment were included as separated and as interaction effects for the combined model, the models stratified by sex only took intervention as independent effect. Due to the relatively large number of tests for the multilevel modelling, the level of statistical significance was set at $p = 0.01$. Potential changes in affect (delta between fiber and placebo) were associated with changes in microbial diversity (delta between fiber and placebo) using spearman correlation. Analyses regarding the microbiota were not corrected for multiple comparisons testing.

Figure composition and statistical analysis of the microbiome was performed using R statistics (v4.1) and is reported in supplementary material as well as in the primary publication of the larger study (Maarse et al., 2024). All SCFA data were adjusted for the amount of bacterial 16S rRNA gene per sample (see supplementary material).

## 3. Results

### 3.1. Baseline characteristics

In total, 65 individuals were included in the study and the 54 participants that completed the study were included in the present analyses. Table 1 shows characteristics of the 54 participants and of the 33 participants that were included in the EMA analyses (see below).

### 3.2. General health, gastro-intestinal symptoms and stool form

For both the physical and mental component of the SF-12 questionnaire, there were no significant effects of intervention, sex or intervention by sex interaction ($p$'s $> 0.35$). Similarly, for the five gastrointestinal symptom clusters as measured with the GSRS and stool form as measured with the BSFS there were no significant effects of intervention, sex or intervention by sex interaction ($p$'s $> 0.1$).

**Table 1**
Participant baseline characteristics.

| | Per protocol | EMA analyses | | |
|---|---|---|---|---|
| | All | All | Males | Females |
| N | 54; (46% female) | 33 | 17 | 16 |
| Age. years (mean ± SD) | 57.8 ± 5.8 | 56.5 ± 6.0 | 58.6 ± 6.8 | 54.3 ± 4.0 |
| BMI. kg/m$^2$ (mean ± SD) | 27.3 ± 1.4 | 27.2 ± 1.4 | 26.9 ± 1.1 | 27.5 ± 1.6 |
| Baseline daily fiber intake (g) | 18.7 ± 5.9 | 18.9 ± 5.9 | 19.5 ± 5.9 | 18.2 ± 6.1 |
| Physical health (SF-12) | 38.67 ± 2.57 | 38.84 ± 1.2 | 38.57 ± 1.22 | 39.13 ± 1.15 |
| Mental health (SF-12) | 51.49 ± 4.23 | 51.35 ± 3.35 | 51.9 ± 3.06 | 50.77 ± 3.65 |
| Diarrhea syndrome (GSRS) | 1.21 ± 0.44 | 1.32 ± 0.57 | 1.37 ± 0.62 | 1.27 ± 0.53 |
| Indigestion syndrome (GSRS) | 1.53 ± 0.72 | 1.66 ± 0.59 | 1.68 ± 0.62 | 1.64 ± 0.57 |
| Constipation syndrome (GSRS) | 1.32 ± 0.68 | 1.27 ± 0.55 | 1.17 ± 0.29 | 1.38 ± 0.73 |
| Abdominal pain syndrome (GSRS) | 1.2 ± 0.4 | 1.16 ± 0.22 | 1.19 ± 0.24 | 1.14 ± 0.2 |
| Reflux syndrome (GSRS) | 1.15 ± 0.42 | 1.22 ± 0.33 | 1.26 ± 0.36 | 1.18 ± 0.3 |
| Stool Form (BSFS) | 3.17 ± 0.84 | 2.66 ± 0.53 | 2.61 ± 0.46 | 2.7 ± 0.61 |
| Positive Affect (mean ± SD) | 71.80 ± 12.41 | 72.74 ± 13.48 | 76.51 ± 14.05 | 68.73 ± 12.0 |
| Negative Affect (mean ± SD) | 17.56 ± 10.54 | 15.46 ± 9.89 | 9.84 ± 7.16 | 21.43 ± 8.96 |
| Physical Discomfort (mean ± SD) | 13.81 ± 13.60 | 10.56 ± 10.69 | 8.62 ± 8.58 | 12.61 ± 12.52 |

EMA: Ecological Momentary Assessment; N: sample size; SD: standard deviation; BMI: Body Mass Index; SF-12: Short Form Health Survey – 12 items; GSRS: Gastro-intestinal Symptoms Rating Scale; BSFS: Bristol Stool Form Scale.

### 3.3. Daily PA, NA, and physical discomfort

Of the 54 participants, 33 completed at least 50% of the questionnaires (≥30 affect records in 10 days) in each intervention phase. A exploratory analyses did not find significant differences in the mean score per person for the EMA variables between the included and excluded group. The EMA results reported in this manuscript refer to 33 included participants.

For PA, the effect of treatment was significant (2.36, 95% CI: 1.41–3.32, $p < 0.001$) for the full model, taking into account the effect of sex (Male: 9.54, 95% CI: 0.57–18.52, $p < 0.04$) and their interaction effect (Male * Treatment: 3.49, 95% CI: 4.82 to −2.15, $p < 0.001$). Separate analyses within sex groups showed that prebiotic treatment significantly improved PA in females (2.36, 95% CI: 1.39–3.34, $p < 0.001$, $d = 0.12$) but not in males (−1.12, −2.04 to −0.2, $p < 0.02$) (Fig. 1). For NA, there was a mean effect of treatment (−3.99, 95% CI: 5.01 to −2.97, $p < 0.001$), sex (−13.93, 95% CI: 19.50 to −8.36, $p < 0.001$) and their interaction effect (Male * Treatment: 4.63, 95% CI: 3.20–6.07, $p < 0.001$). Stratification showed significantly lower NA levels in the prebiotic treatment condition compared to placebo for females (−3.99, 95% CI: 5.11 to −2.87, $p < 0.001$, $d = 0.28$), but not for males (0.64, 95% CI: 0.26 – 1.54, $p > 0.15$) (Fig. 1). For physical discomfort, there was a mean effect of treatment (−5.01, 95% CI: 6.12 to −3.89, $p < 0.001$) and for treatment with sex (Male * Treatment: 3.61, 95% CI: 2.04–5.17, $p < 0.001$) but not for sex alone (Male: 5.87, 95% CI: 13.21 – 1.47, $p > 0.11$). Stratification showed significantly lower discomfort levels in the prebiotic treatment condition compared to placebo for females (−5.01, 95% CI: 6.29 to −3.73, $p < 0.001$, $d = 0.26$) and for males (−1.40, 95% CI: 2.32 to −0.48, $p = 0.003$, $d = 0.09$).

### 3.4. Affective reactivity

Changes in PA and NA affect in response to pleasant or unpleasant events (compared to affect changes when no event was reported) were explored for a possible effect of prebiotic intervention. In response to pleasant events, the increase in PA did not reach significance (1.38, 95% CI: 0.16–2.59, $p = 0.03$), whereas for NA an interaction effect with sex was observed, such that NA decreased for females (−2.76, 95% CI: 4.09 to −1.43, $p < 0.001$) but not for males (−0.71, 95% CI: 2.03 to −0.61, $p > 0.25$). In response to unpleasant events, PA decreased (−9,44, 95% CI: 11.25 to −7.63, $p < 0.001$), and NA increased significantly (9.21, 95% CI: 7.31–11.11, $p < 0.001$). However, for both PA and NA there was no significant interaction with intervention for these analyses.

### 3.5. Changes in NA scores and microbial diversity

In female participants, the reduction in NA was negatively related to an increase in microbial diversity only at 12 weeks (Fig. 2), either determined by inverse Simpson index ($r = -0.60$; $p = 0.003$) or Shannon index ($r = -0.51$; $p = 0.013$). The association between the change in PA in women and change in microbial diversity was not significant, nor were the associations in men (data not shown).

### 3.6. SCFAs, cortisol, and inflammatory markers

The prebiotic intervention did not significantly affect levels of fecal SCFAs, CAR, and inflammatory markers (data not shown).

## 4. Discussion

This study explored effects of a prebiotic intervention on mood in everyday life in healthy individuals, on microbial composition, and on a range of biomarkers implicated in gut to brain signalling. Prebiotics improved PA and decreased NA in daily life, but only in female participants. These changes could not be attributed to changes in general health, gastro-intestinal symptoms, or changes in stool form.

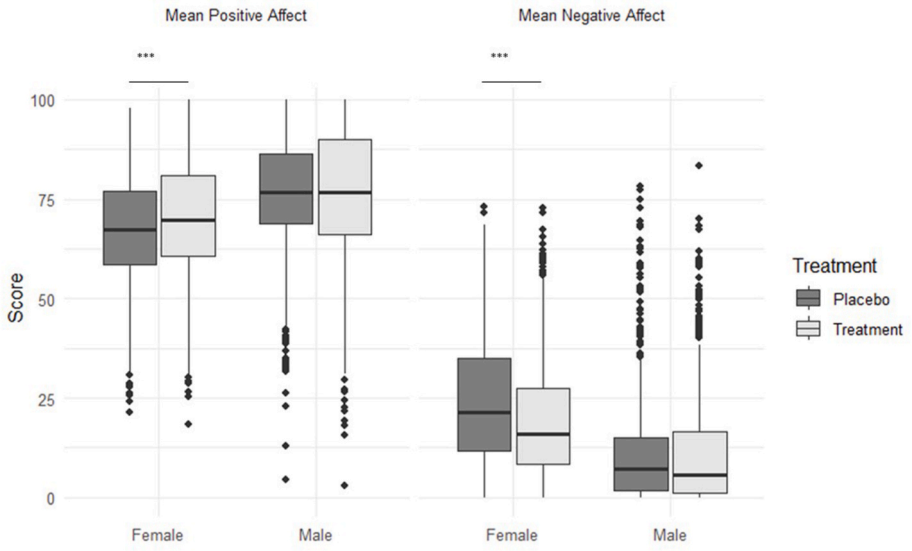



**Fig. 1.** Changes in positive and negative affect during placebo and during intervention, separated for female ($n = 16$) and male (n = 17) participants, analyzed using linear mixed effect models with maximum likelihood estimation; ***p < 0.001.

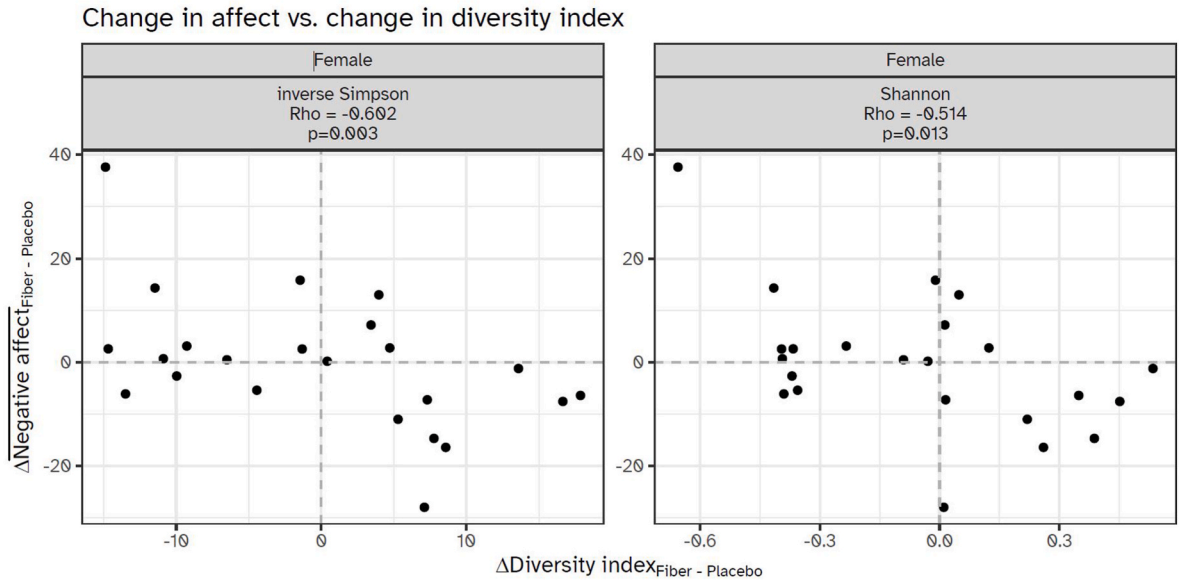



**Fig. 2.** Spearman correlations between the change in negative affect (delta fiber compared to placebo) versus the change in microbial diversity index (delta fiber compared to placebo) in females ($n = 16$), expressed by inverse Simpson (left) and Shannon index (right).

Furthermore, prebiotics reduced feelings of physical discomfort in both men and women. The decrease in NA was associated with an increase in microbial diversity. Improvement of daily mood occurred in the absence of significant main changes in SCFAs in faeces, CAR, and immune markers.

### *4.1. Discussion of the main findings*

The changes in daily affect in women, albeit in a relatively small group, confirmed our hypothesis and are in line with previous research in which a prebiotic intervention improved aspects of mental health in healthy individuals (Schmidt et al., 2015; Berding et al., 2021a,b). Desmedt et al. (2019) reviewed prebiotic intervention studies that investigated affective variables (experienced emotions, mood, well-being, anxiety, and depression). Among the six studies with prebiotic interventions in healthy participants, four studies found a beneficial effect of prebiotics on affective variables (Best et al., 2009; Talbott et al., 2009; Lawton et al., 2013; Childs et al., 2014) with effect sizes (Cohen's *d*) ranging from 0.11 to 0.5 (compared to respectively 0.12 and 0.28 for the effect of treatment in women on increased PA and reduced NA in the present study). These previous four studies included interventions of four weeks of beta-glucan (Talbott et al., 2009), 12 weeks of a combination of polysaccharides (Best et al., 2009), two weeks of breakfast high in wheat bran fiber (Lawton et al., 2013), and three weeks of xylooligosaccharides (XOS) (Childs et al., 2014). In the latter study, XOS intake significantly increased bifidobacteria and had immunostimulatory effects, but the functional consequences of this remained unclear. On the contrary, one study with three weeks of fructooligosaccharides (FOSs) or galactooligosaccharides (GOSs) as an intervention found no effects on self-rated mood, anxiety, and stress (Schmidt et al., 2015). In a computer task that assesses affective information processing, participants in the GOS group showed a positive

attentional bias effect. This result must nonetheless be interpreted with caution, as the other objective measures of emotional processing were not affected by GOS, no correction for multiple testing was performed for the emotion processing outcomes and groups sample sizes were small. The authors also reported a decreased CAR, but this was not associated with task performance, suggesting that the hypothalamic–pituitary–adrenal (HPA) axis was not involved in the emotional processing effect seen (Schmidt et al., 2015). A recent intervention study with GOS (4 weeks) showed no significant effects on self-reported anxiety, mood, depression, emotion regulation or attention to emotional stimuli in young healthy females. Yet, in further post-hoc analyses, in females classified as high anxious there were trend effects of GOS for reduced anxiety, positive attentional bias, and microbial composition difference at follow-up (Johnstone et al., 2021). Also recently, Berding et al. (2021) reported on a four week dietary fibre (polydextrose) supplementation in healthy females that did not affect levels of anxiety, depression, perceived stress, or psychopathological symptoms. Neither CAR nor stress responses were affected by polydextrose supplementation. In 2022, another study from this group was published that employed a psychobiotic diet (high in prebiotic and fermented foods) versus a control diet and included healthy adults, though with overweight (Berding et al., 2022). Perceived stress scores reduced in the psychobiotic diet group, though not between groups. Whereas the psychobiotic diet dit not change anxiety scores, nor the CAR, immune markers or short chain fatty acids (SCFAs) in faeces, significant changes in 40 specific fecal lipids and urinary tryptophan metabolites were observed. Among participants in the psychobiotic diet group, those with less volatility (more stable microbiome) had greater decrease in perceived stress. Although the above mentioned mental health outcomes differ compared to the present study, experienced NA affect is strongly related to perceived stress (Montpetit et al., 2010). The finding that intervention effects were observed only in women is in line with prior research (Patterson et al., 2020). No clear sex differences were observed in age, or baseline BMI, fibre intake, general health, gastro-intestinal symptoms, stool form, fecal SCFAs, CAR, and inflammatory markers. However, among the male participants in the present study PA ratings were higher and NA ratings were lower compared to female participants. They had thus less room for improvement, which may have dampened potential intervention effects in males.

The decrease in NA was associated with an increase in microbial diversity at week 12. This is interesting as other scholars have suggested that a reduction in microbial diversity may affect gut-brain communication leading to psychological abnormalities that underlie mental illness (Foster and Neufeld, 2013; Wu et al., 2020). Furthermore, improvements in daily affect have been shown to be predictive in remission from depression (Geschwind et al., 2011). The association was only present for microbial diversity at week 12, not weeks 4 and 8 suggesting that a change in microbial diversity did not necessarily precede changes in affect. Nevertheless, the observed association raises the question whether prebiotics may increase microbial diversity and lower NA in everyday life in populations known to exhibit both gut dysbiosis and higher levels of NA, such as individuals with major depression (Dinan and Cryan, 2019), hence representing a promising research avenue in gut-brain research.

Though speculative, it is further worth noting that the magnitude and direction of affect changes in the present study are similar to previous research investigating the effect of tryptophan suppletion on daily PA and NA in individuals with a familial risk of depression (Hogenelst et al., 2015) as well as individuals with high trait irritability and hostility (aan het Rot et al., 2006). Tryptophan metabolism has been suggested to be one of the candidate mechanisms in gut to brain communication, with implications for mood regulation (Kennedy et al., 2017; Berding et al., 2022) and may have played a role in the mood effects observed in the present study. Unfortunately, this candidate mechanism could not be tested as tryptophan metabolites were not analyzed in the present study. We did however analyse levels of fecal SCFAs, CAR, and a range of inflammatory markers to gain insight into possible gut to brain communication. Yet, none of the included analytes was significantly affected by the prebiotic intervention. The absence of an intervention effect on fecal SCFAs does not rule out a possible intervention effect on SCFA production by the microbiota, as other researchers have estimated that due to absorption, use by other microorganisms, and depending on transit time, fecal SCFAs represent only about 5% of SCFAs produced in the intestine (den Besten et al., 2013). Thus far, the limited amount of human research on SCFAs as a candidate mechanism in gut-brain research relies heavily on fecal SCFA analyses (e.g., Childs et al., 2014; Berding et al., 2021a,b). Currently, only rodent studies provide direct evidence that prebiotics can increase colonic SCFA production as well as plasma SCFA levels and affect behaviours thought to reflect aspects of human psychological functioning (Dalile et al., 2019; Desmedt et al., 2019). Two previous prebiotic or fibre intervention studies that reported mental health effects also included cortisol analyses, with one study reporting a GOS-intervention induced reduction in the CAR using five saliva samples and up to 1 h of awakening (Schmidt et al., 2015) and the other study reporting no effect of a psychobiotic diet on saliva cortisol with four saliva samples and up to 45 min after awakening (Berding et al., 2022). It is unclear if participants in these studies woke up at a specified time or not. But in the present study participants could wake up according to their own schedule. It is possible that with the three saliva samples and sampling up to 45 min after awakening, we were unable to detect a CAR in some instances. It is possible that the difference between studies can be at least partly attributed to intervention differences. Like the present study, the study by Berding et al. (2022) found no effects of increased fibre intake on the inflammatory markers CRP, IL-6, 8, 10, 12 p70, INF-a, and TNF-a. A reason might be that in a healthy population a gut intervention has little effects on inflammation. Perhaps individuals with a priori higher levels of inflammation may benefit more from higher fibre intake.

### 4.2. *Strengths and weaknesses of the present study*

The present cross-over study employed a strong design with well-controlled study conditions, high level of rigorous control and a long intervention of 12 weeks. A strength of the design is the within subject comparison of effects. Moreover, the study had a relatively long washout period of eight weeks to avoid any carry-over effects of prebiotics-induced microbial changes and function, including systemic effects. To our knowledge, the present gut microbiome intervention study is the first to include a measure of affect or mood in daily life with repeated and frequent assessments such as EMA. As affect was assessed as it occurs in everyday life the study had high ecological validity. Furthermore, in contrast to using surveys that ask participants to rate their mood or stress 'in general' or over a specific retrospective period, which is prone to retrospective bias, with EMA the data were collected close in time to experience, limiting such bias. For example, the study by Berding et al., (2022) measured perceived stress only at the start and end of the intervention period. While they found a significant decrease of perceived stress within the dietary intervention group, there was no significant difference between the groups post-intervention. Using EMA, we were able to detect an effect both within and between groups of intervention on PA and NA. Few gut microbiome intervention studies that focus on mental health include a comprehensive set of biochemical assessments as the present study. Assessments of microbiome composition and function and biomarkers including plasma cytokines and saliva cortisol allowed us to investigate some of the proposed gut microbiome-to-brain mechanisms underlying the prebiotics-induced mood improvements (Dalile et al., 2019). We hope these insights will stimulate more targeted gut-microbiome interventions in the future.

At the same time, the study also has several limitations. Whereas we consider the use of EMA a strength, no traditional mood or well-being questionnaires were included in order to participant burden. It therefore remains unclear if intervention effects would have been identified

by questionnaires such as the positive and negative affect schedule (Watson et al., 1988) or the perceived stress scale (Cohen et al., 1994) that are frequently used in gut-brain interventions studies. Yet, we observed no changes in experienced (mental) health or (mental) health related quality of life, as measured with the SF-12. Furthermore, previous research in individuals without mental health complaints (Hogenelst et al., 2015) has shown everyday mood improvements following tryptophan supplementation in the absence of changes in depressive symptoms (QIDS -SR, Rush et al., 2003) or rumination (LEIDS-R, Van der Does, 2002). This suggests that significant changes in daily affect as measured with EMA, may occur in the absence of significant changes in mental health questionnaire outcomes administered only prior and after the intervention. Moreover, in addition to self-report measures a more objective measure such as computer tasks measuring emotional information processing may be used (see for example Schmidt et al., 2015). A limitation that is inherent to naturalistic investigations such as EMA, is missing data. The daily affect analyses were restricted to participants with at least 50% of questionnaire completions (≥30 affect records in 10 days) in each intervention phase, resulting in a data sub-set with 33 participants. However, our compliance and retention rates are within common ranges for EMA studies (Vachon et al., 2019). Moreover, intervention effects regarding daily affect were comparable when a compliance threshold of 25% (lower compliance but higher participant retention) or 75% (higher compliance but lower participant retention) was used (data not shown). Further analyses showed no differences in baseline mood in EMA responders (included in our analyses) and EMA non-responders (not included in our analyses). It is therefore unlikely that our results are affected by poor compliance or retention. Nevertheless, it should be noted that the reported mood effect of prebiotics was observed in a sample of 16 female participants. This warrants replication with larger sample sizes. Another limitation is the assessment of fecal SCFAs. Due to differences in stool sample quantities and composition, the dilution across stool samples differed markedly and therefore the fecal SCFA concentrations. Although we tried to correct for this using the concentration of 16s rRNA in the samples, the results should be interpreted with caution. Together with the notion that faecal SCFA is an indirect measure of SCFAs produced in the intestine as previously mentioned, it is hard to determine differences in response to a prebiotic intervention. Alternatively, SCFAs may be analyzed in plasma. Despite known challenges of such quantitative analysis including low concentrations of circulating SCFAs and their volatile nature, changes in plasma SCFAs in response to high fiber rye have recently been reported (Iversen et al., 2022) and technological advances to aid plasma SCFA quantification continue to be made (e.g., Yao et al., 2022). Future gut-brain studies may therefore additionally analyse SCFAs in plasma.

### 4.3. Implications/future perspectives

Our results suggest that prebiotics (acacia gum and carrot powder) can improve mood in everyday life in overweight but otherwise healthy middle-aged to older women (45–70 years old). The prevalence of low mood is known to be higher among women than men in general, and in women the prevalence of low mood correlates with hormonal changes (Albert, 2015). Understanding why effects were only observed in women and understanding the mechanisms which led to mood improvements in women should be explored in future studies. As the present study was part of a larger study with a primary focus on metabolic health, the sample was not specifically selected to exhibit room for improvement with regard to mental health, which may have yielded even larger intervention effects. Future studies may therefore include individuals with pre-existing mental health complaints and/or higher inflammation levels and investigate the extent to which a gut intervention can improve mood and other mental health aspects in everyday life.

## 5. Conclusion

The present study is among the first to report an effect of a gut microbiome intervention on mental health outcomes in everyday life, be it in relatively small group of female participants. Negative affect changes were associated with an increased microbial beta-diversity, but the intervention did not affect faecal SCFAs, salivary cortisol, nor a range of inflammatory parameters. Future studies may include larger samples and further elucidate the gut intervention-induced changes in daily mood in women and/or focus on (sub) clinical population samples and diary methods should be used more often in gut-brain research.

## Disclosure statement and acknowledgments

All authors declare that they have no relevant financial interests or disclosures to report. This study was part of the collaboration project No Guts No Glory. The project is co-funded by the PPP Allowance made available by Top Sector Agri & Food to stimulate public-private partnerships, and Top Sector Life Sciences & Health. Funding for the study was received from the Centre for Human Drug Research, Leiden, The Netherlands, dsm-firmenich, Kaiseraugst, Switzerland, and TNO Netherlands Organisation for Applied Scientific Research, Leiden, The Netherlands. We are very grateful to the participants of our study. We thank Ferry Jagers, Martien Caspers, and Bas van Kraaij for their research assistance.

## CRediT authorship contribution statement

**Koen Hogenelst:** Writing – original draft, Methodology, Conceptualization. **Tanja Krone:** Writing – review & editing, Methodology, Formal analysis. **Boukje Eveleens Maarse:** Writing – review & editing, Methodology, Investigation, Data curation, Conceptualization. **Ines Warnke:** Writing – review & editing, Methodology, Funding acquisition, Conceptualization. **Jessica Snabel:** Writing – review & editing, Methodology, Formal analysis. **Tim J. van den Broek:** Writing – review & editing, Visualization, Formal analysis. **Frank Schuren:** Writing – review & editing, Supervision, Methodology, Funding acquisition, Conceptualization. **Matthijs Moerland:** Writing – review & editing, Supervision, Project administration, Methodology, Investigation, Funding acquisition, Conceptualization. **Femke P.M. Hoevenaars:** Writing – review & editing, Supervision, Methodology, Investigation, Funding acquisition, Conceptualization.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bbih.2024.100918.

## Data availability

The authors do not have permission to share data.

## References

aan het Rot, M., Moskowitz, D.S., Pinard, G., Young, S.N., 2006. Social behaviour and mood in everyday life: the effects of tryptophan in quarrelsome individuals. Journal of Psychiatry and Neuroscience 31 (4), 253–262.

Albert, P.R., 2015. Why is depression more prevalent in women? J. Psychiatr. Neurosci. 40 (4), 219–221.

Bates, D., Mächler, M., Bolker, B., Walker, S., 2015. Fitting linear mixed-effects models using lme4. J. Stat. Software 67 (1), 1–48. https://doi.org/10.18637/jss.v067.i01.

Beck, A.T., 2008. The evolution of the cognitive model of depression and its neurobiological correlates. Am. J. Psychiatr. 165 (8), 969–977.
Berding, K., Vlckova, K., Marx, W., Schellekens, H., Stanton, C., Clarke, G., et al., 2021a. Diet and the microbiota–gut–brain axis: sowing the seeds of good mental health. Adv. Nutr. 12 (4), 1239–1285.
Berding, K., Cryan, J.F., 2022. Microbiota-targeted interventions for mental health. Current opinion in psychiatry 35 (1), 3–9.
Berding, K., Long-Smith, C.M., Carbia, C., Bastiaanssen, T.F., van de Wouw, M., Wiley, N., et al., 2021b. A specific dietary fibre supplementation improves cognitive performance—an exploratory randomised, placebo-controlled, crossover study. Psychopharmacology 238, 149–163.
Berding, K., Bastiaanssen, T.F., Moloney, G.M., Boscaini, S., Strain, C.R., Anesi, A., et al., 2022. Feed your microbes to deal with stress: a psychobiotic diet impacts microbial stability and perceived stress in a healthy adult population. Mol. Psychiatr. 1–10.
Best, T., Kemps, E., Bryan, J., 2009. Saccharide effects on cognition and well-being in middle-aged adults: a randomized controlled trial. Dev. Neuropsychol. 35 (1), 66–80.
Blake, M.R., Raker, J.M., Whelan, K., 2016. Validity and reliability of the Bristol Stool Form Scale in healthy adults and patients with diarrhoea-predominant irritable bowel syndrome. Aliment Pharmacol. Therapeut. 44 (7), 693–703.
Booij, S.H., Snippe, E., Jeronimus, B.F., Wichers, M., Wigman, J.T., 2018. Affective reactivity to daily life stress: relationship to positive psychotic and depressive symptoms in a general population sample. J. Affect. Disord. 225, 474–481.
Chakrabarti, A., Geurts, L., Hoyles, L., Iozzo, P., Kraneveld, A.D., La Fata, G., et al., 2022. The microbiota–gut–brain axis: pathways to better brain health. Perspectives on what we know, what we need to investigate and how to put knowledge into practice. Cell. Mol. Life Sci. 79 (2), 80.
Childs, C.E., Röytiö, H., Alhoniemi, E., Fekete, A.A., Forssten, S.D., Hudjec, N., et al., 2014. Xylo-oligosaccharides alone or in synbiotic combination with Bifidobacterium animalis subsp. lactis induce bifidogenesis and modulate markers of immune function in healthy adults: a double-blind, placebo-controlled, randomised, factorial cross-over study. Br. J. Nutr. 111 (11), 1945–1956.
Cohen, S., Kamarck, T., Mermelstein, R., 1994. Perceived stress scale. Measuring stress: A guide for health and social scientists 10 (2), 1–2.
Conner, T.S., Brookie, K.L., Richardson, A.C., Polak, M.A., 2015. On carrots and curiosity: eating fruit and vegetables is associated with greater flourishing in daily life. Br. J. Health Psychol. 20 (2), 413–427.
Conner, T.S., Brookie, K.L., Carr, A.C., Mainvil, L.A., Vissers, M.C., 2017. Let them eat fruit! The effect of fruit and vegetable consumption on psychological well-being in young adults: a randomized controlled trial. PLoS One 12 (2), e0171206.
Dalile, B., Van Oudenhove, L., Vervliet, B., Verbeke, K., 2019. The role of short-chain fatty acids in microbiota–gut–brain communication. Nat. Rev. Gastroenterol. Hepatol. 16 (8), 461–478.
Den Besten, G., Van Eunen, K., Groen, A.K., Venema, K., Reijngoud, D.J., Bakker, B.M., 2013. The role of short-chain fatty acids in the interplay between diet, gut microbiota, and host energy metabolism. J. Lipid Res. 54 (9), 2325–2340.
Desmedt, O., Broers, V.J., Zamariola, G., Pachikian, B., Delzenne, N., Luminet, O., 2019. Effects of prebiotics on affect and cognition in human intervention studies. Nutr. Rev. 77 (2), 81–95.
Dinan, T.G., Cryan, J.F., 2019. Gut microbes and depression: still waiting for godot. Brain Behav. Immun. 79, 1–2.
Eveleens Maarse, Eggink, H.M., Warnke, I., Bijlsma, S., van den Broek, Oosterman, J.E., Hoevenaars, F.P., 2024. Impact of fibre supplementation on microbiome and resilience in healthy participants: A randomized, placebo-controlled clinical trial. Nutrition, Metabolism and Cardiovascular Diseases 34 (6), 1416–1426.
Feldman Barrett, L., Russell, J.A., 1998. Independence and bipolarity in the structure of current affect. J. Pers. Soc. Psychol. 74 (4), 967.
Foster, J.A., Neufeld, K.A.M., 2013. Gut–brain axis: how the microbiome influences anxiety and depression. Trends Neurosci. 36 (5), 305–312.
Gart, E., Souto Lima, E., Schuren, F., De Ruiter, C.G., Attema, J., Verschuren, L., et al., 2018. Diet-independent correlations between bacteria and dysfunction of gut, adipose tissue, and liver: a comprehensive microbiota analysis in feces and mucosa of the ileum and colon in obese mice with NAFLD. Int. J. Mol. Sci. 20 (1), 1.
Geschwind, N., Nicolson, N.A., Peeters, F., van Os, J., Barge-Schaapveld, D., Wichers, M., 2011. Early improvement in positive rather than negative emotion predicts remission from depression after pharmacotherapy. Eur. Neuropsychopharmacol 21 (3), 241–247.
Gibson, G.R., Hutkins, R., Sanders, M.E., et al., 2017. Expert consensus document: the International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics. Nat. Rev. Gastroenterol. Hepatol. 14 (8), 491–502.
Hogenelst, K., Schoevers, R.A., aan het Rot, M., 2015. The effects of tryptophan on everyday interpersonal encounters and social cognitions in individuals with a family history of depression. Int. J. Neuropsychopharmacol. 18 (8), pyv012.
Iversen, K.N., Dicksved, J., Zoki, C., Fristedt, R., Pelve, E.A., Langton, M., Landberg, R., 2022. The effects of high fiber rye, compared to refined wheat, on gut microbiota composition, plasma short chain fatty acids, and implications for weight loss and metabolic risk factors (the RyeWeight study). Nutrients 14 (8), 1669.
Johnstone, N., Milesi, C., Burn, O., van den Bogert, B., Nauta, A., Hart, K., et al., 2021. Anxiolytic effects of a galacto-oligosaccharides prebiotic in healthy females (18–25 years) with corresponding changes in gut bacterial composition. Sci. Rep. 11 (1), 8302.
Kennedy, P.J., Cryan, J.F., Dinan, T.G., Clarke, G., 2017. Kynurenine pathway metabolism and the microbiota-gut-brain axis. Neuropharmacology 112, 399–412.
Lawton, C.L., Walton, J., Hoyland, A., Howarth, E., Allan, P., Chesters, D., Dye, L., 2013. Short term (14 days) consumption of insoluble wheat bran fibre-containing breakfast cereals improves subjective digestive feelings, general wellbeing and bowel function in a dose dependent manner. Nutrients 5 (4), 1436–1455.
Liu, Y., Ajami, N.J., El-Serag, H.B., Hair, C., Graham, D.Y., White, D.L., Jiao, L., 2019. Dietary quality and the colonic mucosa–associated gut microbiome in humans. The American journal of clinical nutrition 110 (3), 701–712.
Mehl, M.R., Conner, T.S., 2012. Handbook of Research Methods for Studying Daily Life. Guilford Press, New York, NY.
Montpetit, M.A., Bergeman, C.S., Deboeck, P.R., Tiberio, S.S., Boker, S.M., 2010. Resilience-as-process: negative affect, stress, and coupled dynamical systems. Psychol. Aging 25 (3), 631.
Moskowitz, D.S., Young, S.N., 2006. Ecological momentary assessment: what it is and why it is a method of the future in clinical psychopharmacology. J. Psychiatr. Neurosci. 31 (1), 13–20.
Noonan, S., Zaveri, M., Macaninch, E., Martyn, K., 2020. Food & mood: a review of supplementary prebiotic and probiotic interventions in the treatment of anxiety and depression in adults. BMJ nutrition, prevention & health 3 (2), 351.
Osadchiy, V., Martin, C.R., Mayer, E.A., 2019. The gut–brain axis and the microbiome: mechanisms and clinical implications. Clin. Gastroenterol. Hepatol. 17 (2), 322–332.
Patterson, E., Griffin, S.M., Ibarra, A., Ellsiepen, E., Hellhammer, J., 2020. Lacticaseibacillus paracasei Lpc-37® improves psychological and physiological markers of stress and anxiety in healthy adults: a randomized, double-blind, placebo-controlled and parallel clinical trial (the Sisu study). Neurobiology of Stress 13, 100277.
ProCore [Computer software], 2019. Retrieved from. https://www.qualitymetric.com/.
Pruessner, M., Hellhammer, D.H., Pruessner, J.C., Lupien, S.J., 2003. Self-reported depressive symptoms and stress levels in healthy young men: associations with the cortisol response to awakening. Psychosom. Med. 65 (1), 92–99.
R Core Team, 2023. _R: A Language and Environment for Statistical Computing_. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org/.
Rush, A.J., Trivedi, M.H., Ibrahim, H.M., Carmody, T.J., Arnow, B., Klein, D.N., et al., 2003. The 16-Item Quick Inventory of Depressive Symptomatology (QIDS), clinician rating (QIDS-C), and self-report (QIDS-SR): a psychometric evaluation in patients with chronic major depression. Biol. Psychiatr. 54 (5), 573–583.
Schmidt, K., Cowen, P.J., Harmer, C.J., Tzortzis, G., Errington, S., Burnet, P.W., 2015. Prebiotic intake reduces the waking cortisol response and alters emotional bias in healthy volunteers. Psychopharmacology 232, 1793–1801.
Spichak, S., Bastiaanssen, T.F., Berding, K., Vlckova, K., Clarke, G., Dinan, T.G., Cryan, J. F., 2021. Mining microbes for mental health: determining the role of microbial metabolic pathways in human brain health and disease. Neurosci. Biobehav. Rev. 125, 698–761.
Svedlund, J., Sjödin, I., Dotevall, G., 1988. GSRS—a clinical rating scale for gastrointestinal symptoms in patients with irritable bowel syndrome and peptic ulcer disease. Dig. Dis. Sci. 33, 129–134.
Talbott, S., Talbott, J., 2009. Effect of BETA 1, 3/1, 6 GLUCAN on upper respiratory tract infection symptoms and mood state in marathon athletes. J. Sports Sci. Med. 8 (4), 509.
Trull, T.J., Ebner-Priemer, U.W., 2009. Using experience sampling methods/ecological momentary assessment (ESM/EMA) in clinical assessment and clinical research: introduction to the special section [Editorial]. Psychol. Assess. 21 (4), 457–462.
Twenge, J.M., Cooper, A.B., Joiner, T.E., Duffy, M.E., Binau, S.G., 2019. Age, period, and cohort trends in mood disorder indicators and suicide-related outcomes in a nationally representative dataset, 2005–2017. J. Abnorm. Psychol. 128 (3), 185.
Vachon, H., Viechtbauer, W., Rintala, A., Myin-Germeys, I., 2019. Compliance and retention with the experience sampling method over the continuum of severe mental disorders: meta-analysis and recommendations. J. Med. Internet Res. 21 (12), e14475.
Van der Does, W., 2002. Cognitive reactivity to sad mood: structure and validity of a new measure. Behav. Res. Ther. 40 (1), 105–119.
Ware Jr, J.E., Kosinski, M., Keller, S.D., 1996. A 12-Item Short-Form Health Survey: construction of scales and preliminary tests of reliability and validity. Med. Care 220–233.
Watson, D., Clark, L.A., Tellegen, A., 1988. Development and validation of brief measures of positive and negative affect: the PANAS scales. J. Pers. Soc. Psychol. 54 (6), 1063.
White, B.A., Horwath, C.C., Conner, T.S., 2013. Many apples a day keep the blues away–Daily experiences of negative and positive affect and food consumption in young adults. Br. J. Health Psychol. 18 (4), 782–798.
Wu, M., Tian, T., Mao, Q., Zou, T., Zhou, C.J., Xie, J., Chen, J.J., 2020. Associations between disordered gut microbiota and changes of neurotransmitters and short-chain fatty acids in depressed mice. Transl. Psychiatry 10 (1), 350.
Yao, L., Davidson, E.A., Shaikh, M.W., Forsyth, C.B., Prenni, J.E., Broeckling, C.D., 2022. Quantitative analysis of short-chain fatty acids in human plasma and serum by GC–MS. Anal. Bioanal. Chem. 414 (15), 4391–4399.
Zhang, N., Zhang, Y., Li, M., Wang, W., Liu, Z., Xi, C., et al., 2020. Efficacy of probiotics on stress in healthy volunteers: a systematic review and meta-analysis based on randomized controlled trials. Brain and Behavior 10 (9), e01699.