Sports Medicine and Health Science xxx (xxxx) xxx

Contents lists available at ScienceDirect

Sports Medicine and Health Science

journal homepage: www.keaipublishing.com/smhs

Regulation of the gut microbiota through meditation and exercise: potential for enhancing physical well-being across all ages in the twenty-first century

Veronica Volpini [a], Sofia Bricca [a], Francesca Ubaldi [a], Tehreema Ghaffar [a], Serena Platania [b], Federica Valeriani [a,*], Vincenzo Romano Spica [a]

[a] Department of Movement, Human, and Health Sciences, University of Rome Foro Italico, 00135, Rome, Italy
[b] Genes, Via Venti Settembre 118, 00187, Roma, Italy

ARTICLE INFO

Keywords:
Meditation
Wellness
Physical activity
Microbiota

ABSTRACT

Purpose: This study explores the gut microbiota of Aikido practitioners, a Meditation in Motion discipline, focusing on the integration of moderate-intensity physical activity with meditative practices. The innovative approach examines how these combined activities influence gut health and overall well-being.

Methods: Researchers collected data through anonymous questionnaires including personal information, dietary habits, physical activity levels, and meditation depth, assessed using the Meditation Depth Index. Fecal samples were analyzed to identify correlations between gut microbiota changes and participants biographical and behavioral factors.

Results: The analysis revealed significant alterations in gut microbiota among high-meditation practitioners, with increased levels of *Bacteroides* and *Faecalibacterium* and a slight rise in *Prevotella*. Moderate-intensity Aikido practice was linked to enrichment of Firmicutes. These findings suggest that Aikido promotes a balanced Bacteroidetes/Firmicutes ratio, supporting intestinal eubiosis. The dual focus of Aikido—movement and meditation contribute to improved gut health and enhanced well-being.

Conclusions: Aikido benefits not only physical fitness and muscular function but also cognitive and mental health, particularly stress management, a crucial factor in modern life. While the study has limitations, it provides preliminary insights into the relationship between gut microbiota and Meditation in Motion disciplines. Further research is needed for deep understanding of this topic.

1. Introduction

In the twenty-first century, there has been a profound and accelerated increase in the use of digital technologies across various aspects of society. This transformation has affected nearly every sector, from healthcare to education, business, and entertainment. The development of digital tools and platforms has brought about new ways of communicating, working, learning, and entertaining, creating both opportunities and challenges.[1] In addition, a wide population of both adults and adolescents spend a lot of time on smartphones or computers or watching television, even in their free time. So, considering the advancement of technology, screen time has become a daily activity for all of us.[2] Several studies have demonstrated that a sedentary lifestyle, prolonged screen time, is associated with increased risk for various diseases, such as cardiovascular disease,[3] obesity,[4] and diabetes.[5] Moreover, sedentary behavior also influences sleep patterns and other mental health aspects such as anxiety and depression.[6-8]

According to the World Health Organization (WHO), 28% of the world's adult population (older than 18 years) does not engage in sufficient physical activity (i.e., does not reach the levels of physical activity recommended for good health). Physical inactivity is mainly characterized by women (32% *vs*. 23% of men) and is more pronounced in high-income countries.[9,10]

The importance of physical activity comes from the evidence that it provides people with physical and mental health benefits like improved functional capacity, decreased risks of diseases, improved body composition, weight loss, improved mood, and reduced depression and anxiety.[11–13] In contemporary Western societies, characterized by their high prevalence of stress, cardiovascular and metabolic diseases have become significant public health concerns. This has led to a critical need for effective stress-reducing methods that can be easily utilized by large

* Corresponding author.
E-mail address: federica.valeriani@uniroma4.it (F. Valeriani).

Peer review under the responsibility of Editorial Board of Sports Medicine and Health Science

https://doi.org/10.1016/j.smhs.2025.07.007
Received 29 January 2025; Received in revised form 17 May 2025; Accepted 20 July 2025
Available online xxxx

Please cite this article as: Volpini V et al., Regulation of the gut microbiota through meditation and exercise: potential for enhancing physical well-being across all ages in the twenty-first century, Sports Medicine and Health Science, https://doi.org/10.1016/j.smhs.2025.07.007

Abbreviations

WHO	World Health Organisation
ISTAT	Istituto Nazionale di Statistica
BDNF	Brain-derived neurotrophic factor
CD	Crohn's disease
IL-10	Interleukin-10
IL-12	Interleukin-12
MEDI	MEditation Depth Index
IPAQ	International Physical Activity Questionnaire
MEDEQ	Meditation Depth Questionnaire
NGS	Next-generation sequencing
PCR	polymerase chain reaction
DNA	Deoxyribonucleic Acid
dsDNA	Double strand deoxyribonucleic Acid
OTUs	Operational taxonomic units
RDP	Ribosomal Database Project
BMI	Body Mass Index
SD	Standard deviation
PCoA	Principal coordinate analysis
ANOVA	Analysis of variance
PA	physical activity
MD	meditation depth
ANOSIM	analysis of similarity
F/B	Firmicutes/Bacteroidetes ratio
OR	odds ratio
CI	confidence interval
SCFA	producing short-chain fatty acids
ENS	Enteric Nervous System
GPCR	G-protein-coupled receptors
HDAC	Histone Deacetylase

numbers of people, readily available, inexpensive, and have minimal side effects. This need was articulated by Henriques et al. in their paper about the anxiety reduction in college students.[14] Physical activity meets all these requirements and additionally, practicing mindfulness meditation or, even better, a sport that involves a meditation part, are considered effective in addressing depression and in preventing diseases.[15]

Regardless of gender or sport type, mindfulness has been found to be positively associated with flow in athletes.[16] Mindfulness-based interventions have been demonstrated to enhance sport-associated physiological activations like salivary cortisol levels, immune responses, and psychological status indicators, as well as sport performances themselves (e.g., shooting and dart throwing performances).[17] Many studies have investigated the role of mindfulness in treating mainly psychological disorders (e.g., anxiety, depression, and stress) but also in relieving pain in chronic medical diseases.[18] From these data, we can conclude that Mindfulness meditation programs show small improvements in anxiety, depression, and pain with moderate evidence, and small improvements in stress and health-related quality of life with low evidence when compared to nonspecific active controls.[19] Recent studies have also correlated a long-term deep meditation practice to an enriched microbiota composition which is in turn related to a reduced risk of psychological issues like depression and anxiety.[20] The world of martial disciplines is rather heterogeneous from different points of view, among which there are the performance pattern, competition, geographical and historical descent, the involvement of the spiritual aspect, and the athleticism level of its practitioners. Studies in literature focus mainly on the best-known martial arts, namely those that involve the competitive aspect. This is the case of the study by Liang et al. (2019), in which the gut microbiota of professional martial arts athletes of different levels, but with similar diets, was analyzed in order to minimize potential confounding effects.[21] Data report that gut microbial diversity and structure of higher-level athletes is significantly higher than that of lower-level athletes. *Parabacteroides* and *Phascolarctobacterium* are the most abundant genera in higher-level athletes and are closely associated with cardiac, metabolic, and mood function.[22–24] In addition, higher level athletes exhibit overexpression of the microbial gene related to carbohydrate metabolism, and this implies greater metabolic capacity of the microbiota. Finally, higher level athletes had significantly higher microbiota capable of histidine metabolism, and this latter amino acid is a key component of muscle carnosine. A recent study by Kang et al. investigated the effects on gut microbiota of two groups of basketball players after 20 weeks of Tai Chi training.[25] Tai Chi can, like Aikido, in fact be counted among the Moving Meditation Disciplines as it is a type of physical and mental exercise that uses breath-controlled body movements. The health benefits of traditional Tai Chi are widely demonstrated in literature, although its role on gut flora appears to be an unexplored field. It is necessary to point out that gut microbiota is influenced by various factors, so data reported from Kang et al. may have been varied by the different exercise approaches. In any case, several data suggest that Tai Chi had a significant effect on the abundance and structure of the gut microbiota: in particular, the species richness of microorganisms was significantly higher in Tai Chi group than that of controls. In addition, the level of Proteobacteria is found to be decreased in the gut flora of the Tai Chi group. The fecal flora of the study group was enriched with butyrate-producing bacteria from the phylum Firmicutes (*Blautia* and *Roseburia*).

In recent years, research has increasingly focused on meditation as a possible influencing factor on gut microbial variability, with systemic effects on the overall health of practitioners. Kanchibhotla et al. investigated the health benefits of the meditation technique “Vaishvanara Agni Meditation”, focusing on the gastrointestinal tract.[26] The data suggested that gastrointestinal health improved significantly by practicing 50 days of meditation. In addition, further improvements in physical strength and psychological mastering were identified. Another study enrolled a cohort of adults who attended a 3-month meditation and yoga retreat.[27] This study assessed the various data like psychometric measures, brain-derived neurotrophic factor (BDNF), cortisol levels, and pro- and anti-inflammatory cytokines before and after the retreat. A decrease in self-reported anxiety and depression was recorded and also an increase in mindfulness. Test results showed an increase in both plasma BDNF levels and the magnitude of cortisol's response upon awakening. Also, levels of anti-inflammatory cytokine IL-10 were increased, while proinflammatory cytokine IL-12 was reduced. In contrast, levels of other proinflammatory cytokines were found to be increased: this could be due to additional influences such as exercise and diet, which varied during the retreat. A study conducted by Sun et al. evaluated the effects on gut microbiota regulation after long term deep meditation (monks).[20] The control group showed greater gut microbial richness, and this could be related to monk's sedentary lifestyle. In any case, several species differ significantly between the two groups: in general, in meditation group the most prominently expressed bacteria were associated with alleviation of mental illness; thus, meditation may play a role in mental health. In addition, the biosynthesis of glycans and lipopolysaccharides increases in the meditation group, which is related to decreased intestinal inflammation, improved barrier function, and reduced infection induced colitis. Several studies have examined the potential benefits of various types of meditations, including mindfulness, in relation to chronic inflammatory conditions such as inflammatory bowel diseases (IBD). The findings of these studies suggest that meditation may contribute to enhancing the quality of life for individuals affected by these conditions.[28] Particularly, several studies

reported a positive correlation between the mindfulness score and health-related quality of life in IBD patients.[28,29] It is important to note that these improvements are also documented in relation to other functional modifications, including levels of C-Reactive Protein and other modulators of inflammatory response.[29,30] Ilan et al.'s study examined the impact of a three-month intervention on Crohn's disease (CD) patients. The combination of cognitive behavioral therapy, mindfulness-based practices, and daily exercise regimens showed promising results. Before the intervention, CD patients showed a microbial profile mostly made up of Proteobacteria (17.71%), Firmicutes (65.56%), Actinobacteria (8.46%), and Bacteroidetes (6.24%). These levels were linked to markers of inflammation and distress. No major changes in diversity were detected after the intervention, but there were significant changes in bacterial abundance among the intervention group. The intervention led to a decrease in bacteria linked to inflammation, and a species of *Lachnospiraceae*, often associated with severe CD, was reduced.[31] Given all these statements and data results about meditation and physical activity on health benefits, the concept of "moving meditation" appears to be a powerful tool in psychophysical well-being. The mind-body approaches combine forms of body movements, typically described as slow and flowing, but may range from a high level of dynamic movement to quiescent static postures, with a focus on breathing and meditation that generate a state of mental and physical relaxation.[32]

The present cross-sectional study aims to evaluate the intestinal microbiota of Italian subjects who practice disciplines characterized by Meditation in Movement (Aikido). Indeed, Aikido may be considered a highly beneficial practice for promoting and maintaining health, due to its inherent sustainability. This discipline is distinguished by the integration of the motor components with the mental and spiritual ones, a process well delineated by meditation, which constitutes an indispensable element of practice. As an activity that can be practiced by anyone, it is also possible to maintain a low motor impact, which makes it suitable and adaptable even for the most vulnerable populations, who can benefit from the positive effects on the body and social relationships. Additionally, Aikido is accessible to all, as it can be practiced in any location, although a mat is the preferred setting. To investigate the impact of Aikido practice on the human intestinal microbiota, an anonymous questionnaire was administered to participants. This questionnaire enquired about personal data, eating habits, levels of physical activity and a literature-validated index of meditation effectiveness, the MEDI (Meditation Depth Index). Subsequently, faecal material was gathered for analysis of microbial composition. Each participant was responsible for collecting faecal samples using dedicated swabs with preservation liquid. The data were analyzed to identify any variations in the intestinal microbiota in relation to personal data, eating habits, levels of physical activity and the degree of depth of meditation. Moreover, the data were compared with data from the literature, which differs from the study population due to the implementation of a strict dietary regimen, participation in competitive sports, and the absence of meditation.

2. Material and methods

2.1. Ethical approval

The study protocol was approved by the Institutional Review Board of the University of Rome "Foro Italico" (approval number 204/2024). Aikido practitioners were invited to participate after their scheduled training sessions. During the recruitment phase, the researchers clearly explained the study's objectives, procedures, and ethical safeguards, including data anonymity and confidentiality. All participants were required to provide written informed consent prior to inclusion in the study, in accordance with the Declaration of Helsinki.

2.2. Study design

Each participant was provided with a faecal swab (Copan Italia S.P.A., Brescia, Italy), accompanied by instructions for the collection of a faecal sample. Participants were requested to deliver the swab on a scheduled day within 2 h of collection. Subsequently, the samples were stored at 4–8 °C in a refrigerated container and transported within 24 h to the laboratory of the University of Rome 'Foro Italico', where they were processed using a previously validated protocol for DNA extraction from faecal traces. Additionally, a salivary swab was delivered and stored by the procedure above. The study targeted adult Aikido practitioners attending regular classes. A total of 28 individuals were approached and provided with standardized sampling kits and detailed instructions. Of these, 25 consented to participate and completed all the required procedures, including informed consent, questionnaire submission, and biological sample collection. Two samples were excluded from the final analysis due to protocol non-compliance (e.g., improper labeling or delayed sample delivery). Therefore, the final dataset consisted of 23 fully eligible and analyzable participants. Inclusion criteria were: regular Aikido practice, consent to provide samples and complete the survey, and absence of antibiotic/probiotic use or gastrointestinal procedures in the previous three months. All data were anonymized using a code derived from non-identifiable personal information and matched across sample and questionnaire entries (Fig. 1).

2.3. Questionnaire

The participants were requested to complete a questionnaire, administered via the EUsurvey platform, which requested information regarding their age, sex, weight, height and any specific dietary regimes they may adhere to (e.g. vegetarian or vegan). It was made clear that providing this information would not lead to the exclusion of any participants. Additionally, intercurrent chronic diseases, pregnancy, food intolerances, concomitant infections (and the associated use of antibiotics and probiotics) or gastrointestinal surgical procedures in the previous three months were investigated and represented exclusion criteria. The 9-item questionnaire developed by Martínez-González et al. and the short version of the International Physical Activity Questionnaire (IPAQ) were employed to assess participants' dietary habits and usual physical activity levels.[33,34] Furthermore, a 35-item questionnaire, designated the Meditation Depth Questionnaire (MEDEQ), was administered for the purpose of evaluating the depth of meditation practiced by each participant.[35] The initial questions of the questionnaire pertain to the methodology and duration of meditation, encompassing both the number of years spent practising and the minutes dedicated to each session. The second part of the questionnaire comprises 30 questions designed to elicit information about the physical and mental sensations experienced by the subject during the meditation session. This data is then used to calculate the MEDI, a quantitative measure of the depth of meditation.[35] The MEDI and MEDEQ are two instruments developed by Piron (2008).[36] For each question, respondents were required to indicate their level of agreement using a scale ranging from 0 (indicating a lack of agreement) to 4 (indicating a high level of agreement). The total score was obtained by summing the factors that were perceived to be conducive to meditation.[3,4,6–9,11,12,15,16] The total score is obtained by summing the scores for factors 16, 17, 19–30, and items 1, 4, 9, 12, 13, and 18, which are inversely related to meditation. It should be noted that the total score is not related to the time factor, but rather to the physical and mental sensations perceived during meditation.[35]

2.4. Collection data

The sample size ($n = 25$) provided 84% power to detect a difference with an effect size of 0.7 at alpha 0.05 using paired two-tailed *t*-tests.[37]

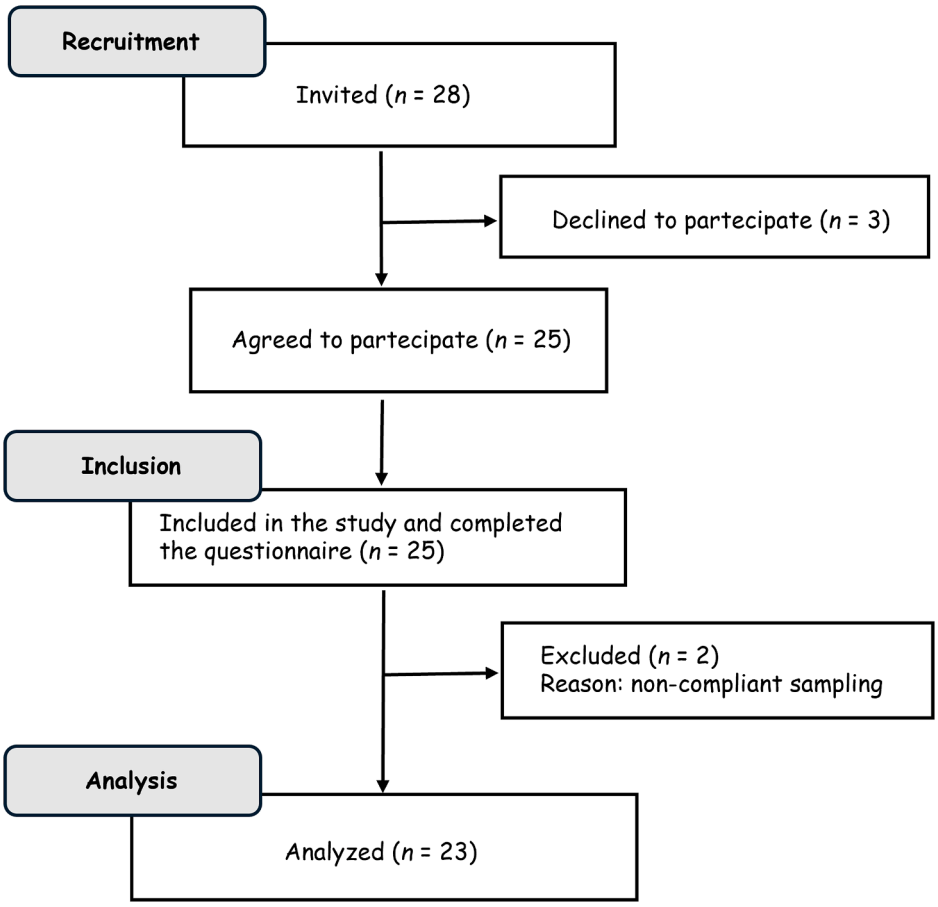

Fig. 1. Flowchart of the study. Recruitment, inclusion, and analysis phases of participants enrolled in the Aikido and meditation-based microbiota study. Of 28 individuals invited, 25 agreed to participate. Two were excluded due to non-compliant sampling, resulting in 23 participants in the final analysis.

Data was collected following an invitation proposed at the end of class. Those who accepted the invitation were provided with the material on pre-determined days. The material consisted of: information letter, informed consent form, instructions, test tube for faecal sampling, test tube for saliva sampling. Labels were placed on the test tubes and linked to complete the questionnaire. The faecal and salivary material for analysis was collected independently, following detailed instructions on the collection method, and kept refrigerated until delivery on pre-established days and times. To ensure anonymity, each participant was identified by a code consisting of the first three letters of the mother's name and the father's birth year (e.g. FRA1967), which was unknown to researchers. The identification code was placed on each tube using the provided labels and inserted into the questionnaire, which allowed the cross-analysis of data. The questionnaire, which included both open and closed questions, was administered via a link on the EUSurvey platform, the official European Commission tool for managing online surveys.

2.5. Data analysis

A faecal swab (Copan Italia S.P.A., Brescia, Italy) was provided to each participant, along with written instructions for stool sample collection. Participants were instructed to deliver the sample on a pre-determined date, within 2 h from collection. The samples were temporarily stored at 4–8 °C in a refrigerated container and transported within 24 h to the laboratory at the University of Rome "Foro Italico". Upon arrival, faecal samples were weighed, and DNA was extracted using a previously validated protocol for microbial DNA isolation from faecal traces. DNA was then purified and normalized prior to sequencing. Amplicon libraries targeting the V1–V3 region of the 16S rRNA gene were prepared following the *16S Metagenomic Sequencing Library Preparation Guide* (part# 15044223 Rev. A; Illumina, San Diego, CA, USA).[38,39] Tagged polymerase chain reaction (PCR) products were generated by two-step PCR using unique barcoded primer pairs. In this strategy, the adapter-containing target primers were used in the first PCR reaction to amplify the target gene (16S rDNA), and this product was then used in the second PCR using the barcode-containing primers to identify the individual sample. Each amplification reaction had a total volume of 25 μL, containing 12.5 μL of KAPA HiFi Hot Start Ready Mix (Roche, Pleasanton, CA, USA), 5 μL of each primer (1 μM) and 2 μL of template deoxyribonucleic Acid (DNA). Reactions were performed on a Techne® TC-PLUS thermal cycler (VWR International, LLC, Radnor, PA, USA). After amplification, 5 μL of PCR product from each reaction was used for agarose gel electrophoresis (1%) to confirm amplification. The final concentration of clean DNA amplicon was determined using the Qubit PicoGreen dsDNA BR Assay Kit (Invitrogen, Grand Island, NY, USA). Libraries were prepared using the MiSeq Reagent Kit Preparation Guide (Illumina, San Diego, CA, USA) and the iSeq100 sequencing platform (Illumina, San Diego, CA, USA). Raw sequencing data were processed using a customized bioinformatic pipeline based on the Galaxy platform, integrating quality control and taxonomic assignment tools. Initial quality assessment of FASTQ files was performed using FASTA/Q Information Tools and Mothur. Reads were filtered to exclude low-quality sequences (length $<$ 200 nt), ambiguous base calls, and chimeric sequences. High-quality sequences were clustered into operational taxonomic units (OTUs) at 97% sequence identity, using a de novo clustering approach. Representative sequences from each OTU were taxonomically classified using the Ribosomal Database Project (RDP)

Classifier (version 2.5), which assigns taxonomic ranks based on a naïve Bayesian algorithm. OTUs defined at 97% similarity were considered species-level units, while 95% similarity was used for genus-level classifications.

Additionally, sequencing reads were analyzed in parallel using the Illumina BaseSpace 16S Metagenomics App (version 1.0.1), which applies an alternative taxonomic classification pipeline based on the curated May 2013 release of the Greengenes reference database. Only taxa with a relative abundance above 0.1% were included in downstream statistical analyses.

2.6. Statistical analysis

A descriptive analysis was conducted on the information collected. Weight and height values were used to calculate the BMI (Body Mass Index). BMI, an anthropometric indicator, is calculated as the ratio between body weight (expressed in kilograms) and the square of height (expressed in meters). Other parameters that define the cohort are age, type of diet and levels of habitual physical activity and expressed as mean ± standard deviation (*SD*) and ranges. Participants were also grouped according to their meditation score with 3 score levels: 11 with a low score (0–60 – MED 0) (44%), 7 with a medium score (60–80 – MED 1) (28%), 7 with a high score (80–120 – MED 2) (28%) and level of Aikido practice. The meditation score is related to the physical and mental sensations perceived during practice, rather than the time factor. Relative abundances of community members were determined with sparse data and summarized at each taxonomic level. The proportion of the gut microbiome at each taxonomic rank, such as phylum, order, class, family, and genus, was determined using the RDP classifier and the Greengenes database. Briefly, the relative abundance of the bacterial community was determined across 29 phylotypes (%abundance = [number of sequences per phylum or genus/total sequences per sample] × 100). Subsequent mathematical analyses excluded the 24 rarest phyla (i.e. those with a relative abundance lower than 0.1% across all samples). Alpha and beta diversity were 56 calculated using Primer7 software at a sequence similarity level of 97%. For alpha diversity, the Shannon index and species-level evenness index were calculated. Principal coordinate analysis (PCoA) was performed using the Primer7 software platform to investigate dissimilarity between the groups.[40–42] Student's *t*-test was used to compare bacterial diversity and relative abundance of major bacterial phyla and genus (representing 90% of the fecal bacterial component) of samples from participants with different Aikido levels and meditation levels. Analysis of variance (ANOVA) with a Bonferroni post hoc test was used to assess differences between participants grouped according to habitual physical activity (PA) levels and to compare lifestyle subgroups obtained by combining meditation depth (MD) adherence and PA. The statistical significance of factors potentially contributing to compositional differences between microbiota samples were also tested with PRIMER software (version 7) using the nonparametric permutation analysis of similarity (ANOSIM) function and the default setting. To identify possible determinants of microbiome composition, two linear regression analyses were performed with age, gender, BMI, diet, and Aikido level as independent variables and Shannon index or Firmicutes/Bacteroidetes ratio (F/B) as outcomes. A stepwise procedure was used to adjust for possible confounders. A p-value < 0.05 was considered statistically significant. Data were analyzed with IBM SPSS version 25 for Windows (SPSS, Chicago, IL, USA).

3. Results

Cohorts are particularly homogeneous, and consistent with these results, also *t*-test produced no significant dissimilarity for BMI, sex, age, level of education, smoking, alcohol, diet, and Aikido level ($R = -0.011$, $p = 0.5$; Table 1).

A total of 1 688 571 sequences were produced by sequencing 23 samples (one sample was not in line with prerequisites required by the analysis). The number of sequences for each sample ranged from 20 000 to 92 000, leading to the identification of 471 OTUs defined at 99% identity. Regression analyses showed that only meditation score was inversely proportional to the Shannon index ($p = 0.02$, odds ratio (*OR*) $= -0.08$, 95% confidence interval (*CI*) $= [-0.16$ to $-0.01]$), as shown in Fig. 2 a where on the x-axis 0 = low levels of meditation, 1 = medium levels of meditation, and 2 = high levels of meditation. Data analysis showed that alpha diversity of practitioner's gut microbiota was similar according to the Aikido level (Fig. 2b).

Alpha diversity indices Chao1 ($p = 0.796$) and ACE ($p = 0.886$) were not significantly different when compared between group 1 (high level of Aikido) and group 2 (insufficient level of Aikido). However, the alpha diversity of gut microbiota was lower in higher levels of meditation (Fig. 2). From data analysis, training intensity and significant differences in biodiversity richness based on Chao1 ($p = 0.006$) and ACE ($p = 0.004$)

Table 1
Socio-demographic characteristics of the sample ($n = 24$).

Variable	Value	Percentage
Gender		
female	9	37.5
male	15	62.5
Age		
18–30	1	4.2
31–65	20	83.3
> 65	3	12.5
Educational level		
Middle school graduation	1	4.2
High school degree	9	37.5
Bachelor's degree	1	4.2
Master's degree	9	37.5
PhD	4	16.7
Smoking		
No	16	66.7
Yes	8	33.3
Smoking frequency		
At least once/day	6	75
4-6 times/week	1	12.5
Maximum 3 times/week	1	12.5
BMI		
Normal weight	17	70.8
Overweight	6	25
Obese	1	4.2
Diet		
No diet	10	41.7
Mediterranean	8	33.3
Vegetarian	3	12.5
Requested by physician	1	4.2
Caloric restriction	1	4.2
Other	1	4.2
Alcohol consumption		
Often (> 5 times/month)	11	45.8
Sometimes (3–4 times/month)	10	41.7
Occasionally (1–2 times/month)	3	12.5
Aikido practice		
< 300 min/week	14	58.3
> 300 min/week	10	41.7
General physical activity		
Partially active	4	16.7
Physically active	20	83.3
MEDI score		
Low (0–60)	10	41.7
Medium (60–80)	7	29.2
High (80–120)	7	29.2
Meditation time		
< 60 min/week	13	54.2
> 60 min/week	11	45.8

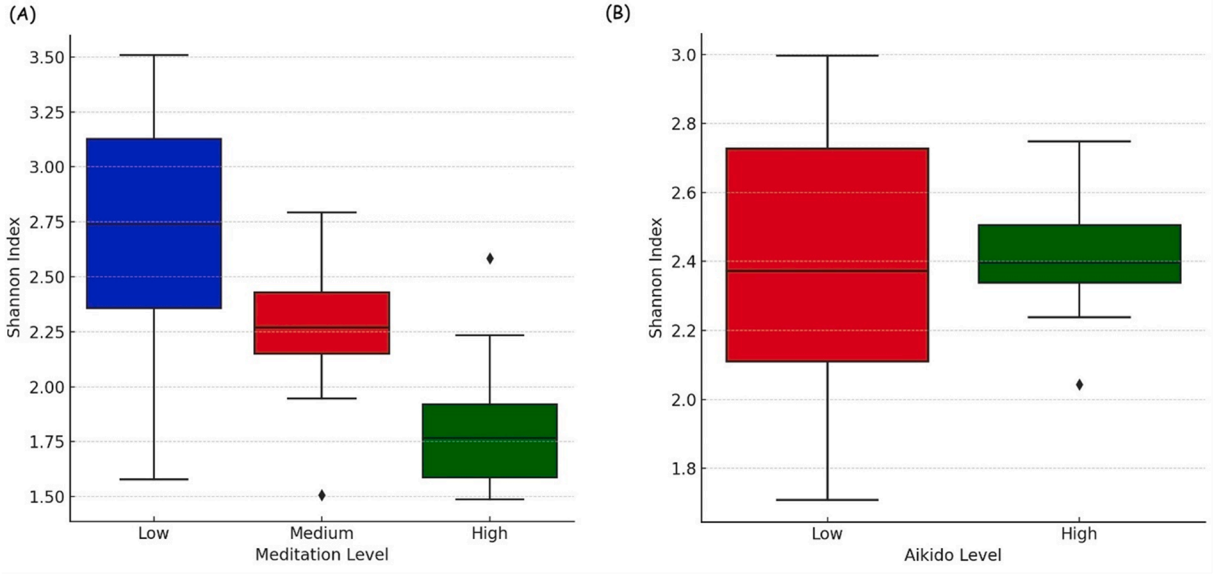

Fig. 2. Shannon Index Distribution by Meditation and Aikido Practice Levels. Box plot showing the distribution of the Shannon index in relationship between Aikido levels and Meditation depth. The box border closest to zero indicates the 25th percentile, the line inside the box represents the median, and the box border farthest from zero indicates the 75th percentile. The whiskers above and below the box indicate the 10th and 90th percentiles. A) Shannon value trend in three levels of Meditation. A significant reduction in alpha diversity is observed in the high meditation group, suggesting a potential compositional shift in gut microbiota associated with deeper meditative practices. B) Shannon value trend in two levels of Aikido. The Shannon index appears slightly higher in the low Aikido practice group compared to the high practice group, although the medians are similar (~2.4). The "Low" group shows greater variability (wider range and presence of outliers), while the "High" group appears more homogeneous. However, the difference is not statistically significant, suggesting that the level of Aikido practice may not substantially influence gut microbiota alpha diversity.

indices. Beta diversity was demonstrated as PCoA based on Bray Curtis algorithm by clustering for different levels of meditation (Fig. 3). Statistically significant differences were found in gut microbiota composition based on Aikido and Meditation levels ($p = 0.001$). Differences were recorded for meditation level ($p < 0.1$). Principal Coordinates Analysis (PCoA) based on Bray–Curtis dissimilarity revealed distinct clustering patterns among the three meditation groups, with partial overlap between groups 0 and 1, while group 2 appeared more dispersed along the first principal coordinate (Fig. 3). The variable representing Aikido practice (AIKIDO) did not lead to a complete separation of the samples, although some directional shifts were noticeable, particularly within meditation group 1. This pattern suggests a potential interaction between physical-meditative activity and gut microbiota structure, although not strong enough to drive a dominant separation. These observations are further supported by ANOSIM analysis for the Meditation score, which showed a moderate but significant effect ($R = 0.350$, $p = 0.001$).

The first two PCoA axes explained approximately 33.62% and 19.4% of the total variance in microbial composition.

The biplot includes vectors representing the top 10 most variable genera, indicating their correlations with the PCoA axes. For example, *Roseburia* and *Blautia* vectors point along the PCoA1 axis, suggesting a higher abundance of these genera in samples with positive PCoA1 scores—possibly associated with MED groups 0 or 1. In contrast, *Anaerostipes* and *Lachnospira* align more strongly with PCoA2, contributing to variation particularly within group 2. These directional trends highlight that specific genera may act as key microbial drivers of group-level differences, and potentially serve as biomarkers in future interventional studies.

With regard to Aikido practice, subtle compositional differences were also observed and supported by ANOSIM analysis ($R = 0.250$, $p = 0.001$). At the phylum level, the most abundant taxa across all groups were Firmicutes (53%) and Bacteroidetes (34%), followed by subdominant phyla including Proteobacteria (6.7%), Actinobacteria (3.2%), and Verrucomicrobia (2.0%), together accounting for over 98% of the gut microbiome (Fig. 5).

When microbial composition was stratified by meditation level, phylum-level distributions showed notable trends (Fig. 4). In group 0, the predominant phylum was Firmicutes (63%)—now classified under the revised nomenclature as *Bacillota*—followed by Bacteroidetes (25%) (*Bacteroidota*), Actinobacteria (5%), and Proteobacteria (6%). In group 1, the abundance of Firmicutes decreased to 50%, with Bacteroidetes increasing to 42%. This trend continued in group 2, with Firmicutes at 48% and Bacteroidetes rising to 45%. Actinobacteria and Proteobacteria showed a corresponding decrease across groups, further confirming a shift in microbial dominance profiles with increasing meditation engagement. At the taxonomic genus level, 103 taxa were present with a relative abundance greater than 0.1%, where *Bacteroides* (18.4%), *Alistipes* (7.0%), *Faecalibacterium* (4.5%), *Blautia* (2.4%), *Ruminococcaceae* UCG-002 (2.4%) and *Roseburia* (2.2%) were the most abundant bacteria in the intestine. *Bacteroides*, *Faecalibacterium*, *Prevotella*, *Ruminococcus* and *Collinsella* represented the top five genera (whose proportions were greater than 5%). Heatmap, in Fig. 5, shows the relative abundance (%) of the top 20 most represented bacterial genera across three meditation levels (Low, Medium, and High). The most dominant genus overall was *Bacteroides*, with relative abundances ranging from 26.98% (Medium) to 34.39% (High), followed by *Faecalibacterium*, *Prevotella*, *Ruminococcus*, and *Collinsella*. Notable shifts in microbial composition were observed across meditation levels: for example, *Prevotella* peaked at Medium meditation (17.01%), while *Faecalibacterium* and *Bacteroides* showed a progressive increase from Low to High meditation. Other genera such as *Sutterella*, *Collinsella*, and *Ruminococcus* showed higher relative abundance in the Medium group, whereas genera like *Streptococcus*, *Lachnospira*, and *Alistipes* remained relatively stable across all levels. Fig. 5 illustrates how specific microbial genera may fluctuate in relation to the depth of meditative engagement, suggesting a potential link between meditation practice and gut microbiota composition.

A comparison of the most abundant bacterial genera (relative abundance > 1%) between the *Aikido* and *Meditation* groups revealed both overlapping and distinct microbial patterns (Fig. 6). In total, 10

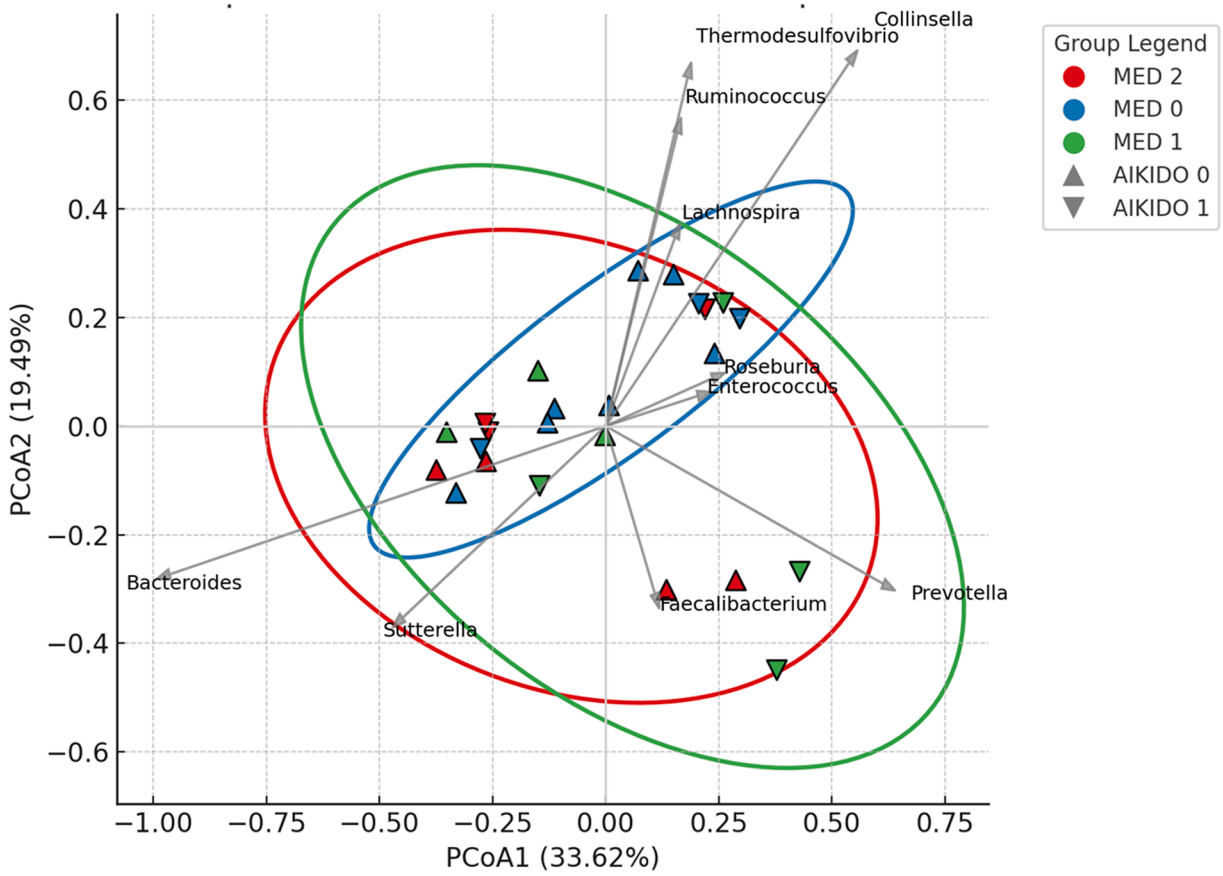

Fig. 3. Principal Coordinates Analysis of Gut Microbiota by Meditation and Aikido Practice. Principal Coordinates Analysis (PCoA) biplot based on Bray–Curtis dissimilarity of genus-level microbial profiles. Samples are color-coded by MED groups—red for group 0, green for group 1, and blue for group 2—and shaped by AIKIDO practice status (▲ for non-practitioners, ▼ for practitioners). Ellipses represent 95% confidence intervals for each MED group, delineating the dispersion of samples within each group. Arrows indicate the Spearman correlation vectors of the 10 most variable genera, illustrating their directional associations with the first two principal coordinates. PCoA1 and PCoA2 explain XX% and YY% of the total variance, respectively.

genera exceeded the 1% threshold in the Aikido group, while 13 genera did so in the Meditation group. Notably, seven genera were found in both groups: *Prevotella*, *Bacteroides*, *Roseburia*, *Enterococcus*, *Ruminococcus*, *Faecalibacterium*, and *Alistipes*, all of which are common and relevant members of the human gut microbiota. By analyzing the various distributions and fluctuations of the percentages of these genera in various levels of meditation depth and Aikido frequency, we observed that some of these genera tend to change within Meditation group. Also, these changes are statistically significant ($p = 0.000\ 1$), while lower fluctuations are recorded for Aikido practice levels. In contrast, three genera were exclusive to the Aikido group (*Phascolarctobacterium*, *Oscillibacter*, *Dialister*), and six were unique to the Meditation group (*Collinsella*, *Sutterella*, *Lachnospira*, *Parabacteroides*, *Butyrivibrio*, *Lachnoclostridium*). These differences suggest that distinct microbial profiles may be associated with specific lifestyle or practice-related factors, such as physical movement, postural stability, and meditative engagement. *Prevotella* was found slightly increased compared to the group with the lowest meditation score, and these results are also shown in Fig. 6.

4. Discussion

Human gut microbiota comprehends a large community of commensal, symbiotic and pathogenic microorganisms like bacteria, fungi, virus and protozoa. This serves a lot of important functions like physiologic ones, immune function, nutrition and metabolism of the host and thus. Its composition is crucial in keeping a state of health. It is therefore considered an influential “hidden organ”. Indeed, numerous studies have shown the correlation between a microbiota alteration and a range of disorders such as obesity, inflammatory bowel diseases, autism, cardiovascular disease, osteoporosis and cancer.[43–47] The human gut microbiota's composition is influenced by numerous factors such as environment, diet, lifestyle, health and disease status, and drugs.[48] Recent studies have shown that exercise is also an important modulating factor.[49,50] Studies concerning the effects of physical activity on gut microbiota are an entirely new field, and while mechanisms of influence are not yet entirely clear. Preliminary studies have revealed a bidirectional relationship that has to be further explored. These first studies were conducted on animal models and reported beneficial changes in microbiota composition of trained rats compared with controls.[51] In addition, a protective function of exercise against negative consequences of high-fat diets has been identified.[52] In addition to animal experiments, studies on professional athletes are also increasing to investigate the unique characteristics, benefits and diversity of their gut microbiota.[53] Indeed, regular physical activity should be considered as a valid method to maintain microbiota eubiosis (or rebalance any dysbiosis), thus improving health status. Further and more in-depth studies on the specific changes produced by physical activity on the composition of gut microbiota could be useful to explore new approaches for the treatment of metabolic and inflammatory diseases, in which the microbiota plays a key role. In this regard, data reported in literature show that moderate-intensity physical activity can provide greater benefits to gut microbiota than that given by high intensity or not at all exercise. In fact, physical activity leads to increased abundance of genera producing short-chain fatty acids (SCFA) and lactic acid-producing bacteria and, concomitantly, improves gut microbial diversity resulting in increased *Akkermansia muciniphila* and *Oscillospira*.

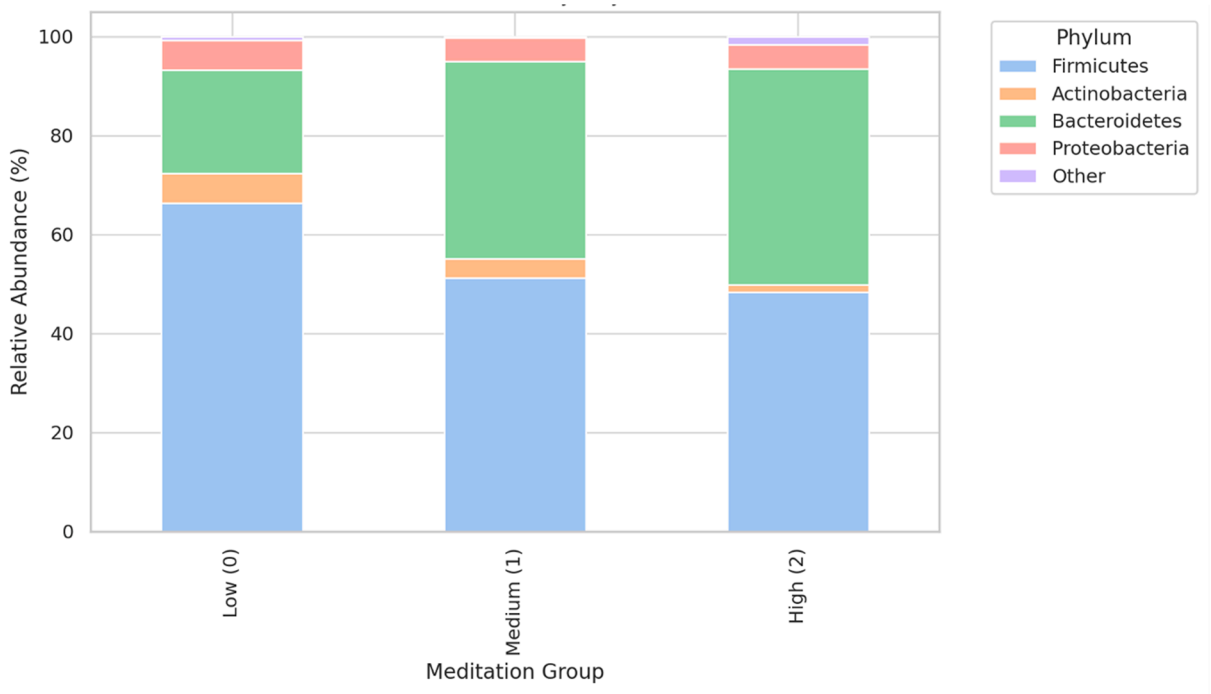

Fig. 4. Normalized relative abundance of bacterial phyla across meditation levels. Stacked bar plot showing the average composition of bacterial phyla in participants grouped by meditation depth: low (0), medium (1), and high (2). Only phyla with a mean relative abundance $\geq$ 1% across all samples are displayed individually; remaining taxa are grouped under "Other".

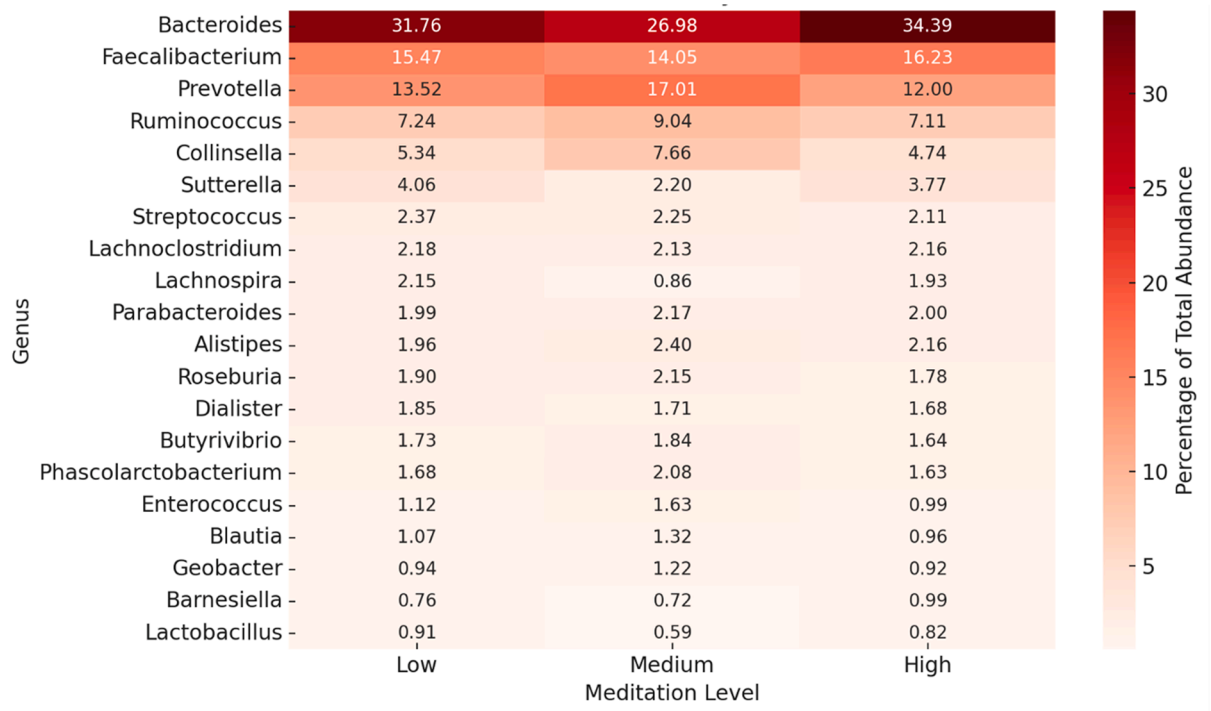

Fig. 5. Relative contribution of bacterial genera to total community abundance across meditation levels. This heatmap shows the percentage contribution of selected bacterial genera to the total abundance of gut microbiota in participants stratified by meditation level (Low, Medium, High). Values represent the proportion of each genus within the total bacterial load of the corresponding group, not normalized by row. Notably, *Bacteroides* consistently dominates the microbial composition, while genera such as *Faecalibacterium*, *Prevotella*, and *Collinsella* show variations across meditation levels, suggesting potential microbiota shifts linked to meditative practice intensity.

In contrast, intense exercise appears to have a negative effect on the gut microbiota.[54] This is an important finding for the present work, as it lends even more relevance to the use of Aikido as a possible tool for a positive influence on gut microbiota, with systemic effects on overall health. Indeed, Aikido is a discipline suitable for everyone, offering an extremely flexible performance that can be adapted to moderate

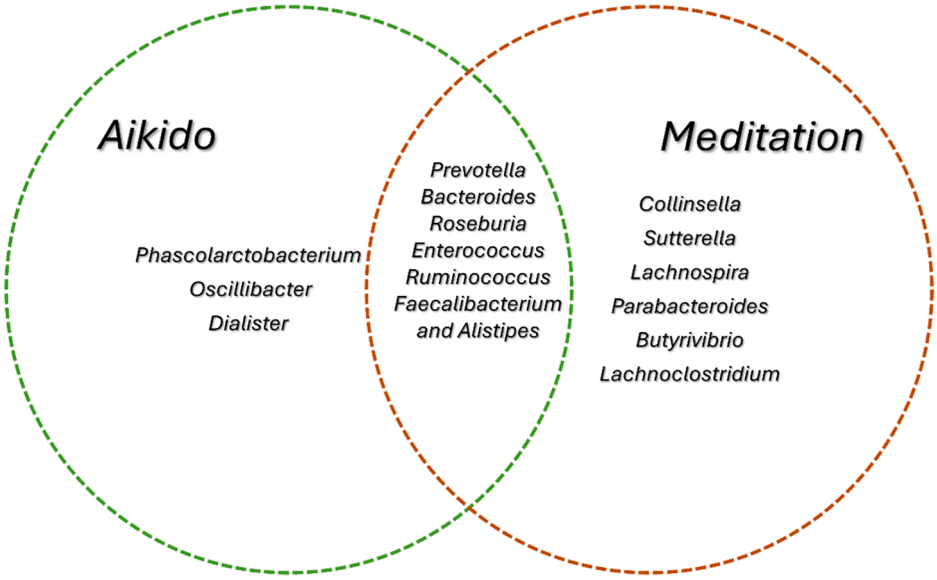

Fig. 6. Shared and Unique Bacterial Genera in Aikido and Meditation Groups. Venn diagram showing the distribution of bacterial genera (cutoff > 1% relative abundance) between the Aikido and Meditation groups. Seven genera—*Prevotella*, *Bacteroides*, *Roseburia*, *Enterococcus*, *Ruminococcus*, *Faecalibacterium*, and *Alistipes*—were shared between the two groups. The Aikido group uniquely exhibited *Phascolarctobacterium*, *Oscillibacter*, and *Dialister*, while the Meditation group was characterized by *Collinsella*, *Sutterella*, *Lachnospira*, *Parabacteroides*, *Butyrivibrio*, and *Lachnoclostridium*. These patterns may reflect the influence of physical and/or meditative practices on the gut microbiota composition.

intensity regimen. Furthermore, meditation practices, ranging from mindfulness to deep breathing exercises, induce a state of relaxation and focused attention. While forms of meditation have been present in many Eastern cultures for millennia, these practices are recently gaining increasing interest also in Western cultures. In addition to the spiritual benefits, meditation practice leads to physiological changes such as improved brain function, immune response and inflammatory processes. Contextually we may appreciate a positive effect on the control of diabetes, fibromyalgia, hypercholesterolemia, psychological disorders and a reduction in heart rate and blood pressure which promotes stress reduction and general well-being.[55] Stress is a well-known aggravator of gut dysfunction, affecting gut motility, barrier function, and microbiota. Meditation, through its stress-reducing effects, can positively influence gut health; in particular, it helps restore the balance of gut bacteria, improves gut barrier function, and reduces inflammation.[56] Thus, the combination of mindfulness with daily exercise can significantly impact the psychological symptoms and inflammation through microbial changes in chronic pathologies like inflammatory bowel diseases, diabetes, cancer, psoriasis, etc.[31,57,58] New research underscores the positive impact of regular meditation on gut health. Specifically, one study evaluated effects of years of deep meditation practiced by Tibetan monks on gut microbial regulation. In this study, the phyla Bacteroidetes and Firmicutes (currently referred to as Bacteroidota and Bacillota) were dominant in both groups, as would be expected. However, Bacteroidetes were significantly enriched in high level of depth meditation (29% *vs.* 4%), which also contained abundant *Prevotella* (42% *vs.* 6%) and a high number of *Megamonas* and *Faecalibacterium*. It seemed that the bacteria present in subjects performing meditative exercises would be associated with genera implicated in recovery from psychological or neurological disorders.[59] Based on this scientific evidence, in this work we focused on the assessment of gut microbiota in a sample of subjects practicing Meditation in Motion (Aikido) disciplines. The interest in this discipline is related to its conciliation of both motor and spiritual components, which make it a widely used practice in population of all ages. Being an activity "for everyone" means the possibility of practicing it in a low impact mode, making it suitable even for the most fragile populations, who can find benefits in wellness and social relationships. Aikido is also versatile in places of practice because it can be carried out anywhere, although the mat appears to be the best location. In this cross-sectional study, in addition to microbiota analysis, have been collected anthropometric data, biographical data, eating habits, physical activity levels and meditation practice have been evaluated through MEDI, (MEditation Depth Index) and MEDEQ questionnaire (MEditation Depth Questionnaire; Piron, 2008).[36] Data of microbial composition from faecal and salivary material was analyzed in relation to biographical aspects, dietary habits, physical activity and level of meditation depth. Subsequently, the data obtained were compared with data from literature and other disciplines, but only in relation to physical activity, as data in literature on diet and meditation was poor. More specifically, the distributions and fluctuations of microbial composition according to depth levels of Meditation and frequency of Aikido were analyzed. These new data show that some genera tend to change in a statistically significant way in Meditation groups ($p = 0.000\ 1$), while less fluctuations are recorded for Aikido practice levels. Overall, the bacteria enriched in those who practice meditation have previously been associated with improved mental health, behavior and mood. This, in addition to physiological effects, suggests the potentially central role of meditation in mental and physical well-being.[60] Moreover, different bacterial genera differ significantly in various depth levels of Meditation. At the genus level, *Bacteroides*, *Faecalibacterium*, and *Prevotella* are found to be predominant in all groups, but the two first were found to be significantly enriched in Meditation group. *Prevotella* was found to be slightly increased compared to the group with lowest Meditation score (Fig. 6). *Prevotella* is a marker bacterium of intestinal good health, but it is believed that, if not accompanied by good *Bacteroides* and *Akkermansia muciniphila*, it may promote pro-inflammatory processes, infections and dysbiosis.[61] Subjects practicing deep meditation and aikido show that both *Prevotella* and *Bacteroides* are enriched. Scientific evidence supporting this shows that *Prevotella* is deficient in subjects with depressive disorders,[62,63] indicating its connection to the gut-brain axis.[60,64] Also consistent with present data, *Feacalibacterium prausnitzii* and *Roseburia* are increased in moderately active individuals,[64] while vigorous exertion appears to decrease their abundance.[65,66] *Ruminococcus*, a marker of gut dysbiosis, is decreased in exercise intervention studies.[67,68] *Alistipes*, one of the newest genera in *Bacteroidetes* phylum, was found to be decreased in some exercise intervention studies,[69,70] although its role deserves further investigations. Relative abundance of *Enterococcus* spp. is higher after moderate intensity and long duration exercise, when compared to higher intensities and shorter durations.[71] *Megamonas* and *Faecalibacterium* are abundant in meditation practice. *Faecalibacterium* seems to be associated with better quality of life, in fact it decreases under stressful conditions.[72,73] These results may support this

cross-sectional study data showing that this genus increases as meditation practice increases. Furthermore, in subjects who practice aikido and reach high levels of depth meditation, *Roseburia* is enriched as shown in other work on the topic.[59] So, scientific evidence seems to support this cross-sectional study data. In fact, there are works in literature that demonstrate specific changes in gut microbiota of subjects practicing physical activity, martial arts, or meditation and seem in line with these data.[73,74] Specifically, regarding physical activity, studies show that exercise intensity has an influence on *Fimicutes* abundance. Regarding meditation, *Bacteroidetes*, particularly *Prevotella* and *Faecalibacterium*, appear to be enriched in subjects who practice a lot of it. This opens the idea that practicing a Moving Meditation activity, such as Aikido, may result in an eubiotic balance in practitioners. Specifically, *Firmicutes* phylum includes genera producing SCFAs, which are widely recognized for their involvement in crucial processes such as maintaining colonocyte integrity, improving barrier function and increasing mucin expression regulating specific gene expression.[75–79] Particularly, SCFAs (e.g. butyrate, acetate and propionate) exert their physiological activities by binding the G-protein-coupled receptors (GPCRs) and modulating gene expression by inhibiting histone deacetylases (HDACs).[80] GPCRs are cell surface receptor proteins that regulate various physiological and pathological processes, while HDACs are part of the epigenetic regulatory mechanisms that control the expression of genes.[81,82] The defensive cells of microglia in the enteric nervous system (ENS), stimulated by bacterial LPS, trigger an inflammatory response, but this has been shown to be inhibited by acetate and butyrate through the inhibition of HDAC activity and NF-kB activation.[83] Furthermore, scientific evidence supports the notion of an inverse relationship between type and intensity of physical activity and the amount of fecal bile acids influencing *Firmicutes* growth (mainly Clostridia spp.). Besides short-chain fatty acids and bile acids, some recent studies show that gut microbiota also have a key role in neurotransmitter's production such as glutamate, GABA, serotonin, and dopamine.[84,85] These bacteria derived neurotransmitters precursors can arrive to the brain passing the blood-brain barrier and finalize the neurotransmitter synthesis.[86] Moreover, gut bacteria metabolites can interact with enteroendocrine cells and thus acting locally, or transmit to brain via the fast vagus nerve way, to regulate the synthesis and release of neurotransmitters, the so called gut-brain axes.[87,88] On the other hand, it seems that *Bacteroides* play a crucial role in neurotransmitters modulation along the gut-brain axis. It appears to be involved in tryptophan metabolism, a precursor of brain serotonin.[70] So, in general, these observations reinforce the notion that gut bacteria play an active role in metabolic homeostasis and are influenced by practices involving both physical and mental aspects. This study is intended to be a contribution to the characterization of gut microbiota of adults practicing Aikido, a Moving Meditation Discipline. It provides insight into the relative abundances of major bacterial phyla in gut in a sample of 23 participants, suggesting the influence of different Meditation levels and Aikido on the composition of gut microbiota. A results summary of the present work is schematized in Fig. 7 where we may see two different, but complementary situations. On one hand (A), the intensity of exercise seems to be positively correlated with growth in relative abundance of Firmicutes phylum, on the other hand (B) the level of Meditation depth seems to be positively correlated with the increase in relative abundance of Bacteroidetes phylum. Consequently, it follows that Aikido (C), that organically involves a performance in both aspects just described, can balance the Bacteroidetes/Firmicutes ratio, with satisfactory effects on the health and well-being of practitioners.

4.1. Limitations

This cross-sectional study has limitations. First of all, data collected are meager to paint a general picture of gut microbial variation in a population such heterogeneous as that of Aikido practitioners. As the latter is a highly individualized discipline, it can be practiced at different intensities. Nevertheless, in this study, only time of practice was assessed (minutes per week), but it was absent an instrumental assessment of training intensity. Regarding Meditation, the MEDI score concerns only the physical and mental sensations perceived during practice and does not consider the time factor. In addition, although the sample appears to be quite homogeneous, diet and smoking were not considered as possible confounding factors. Despite these limitations, this study represents the first attempt to characterize the effects of Moving Meditation Disciplines, particularly Aikido, on the composition of the gut microbiota and opens new perspectives for future research on exercise strategies involving the mind-body unicum.

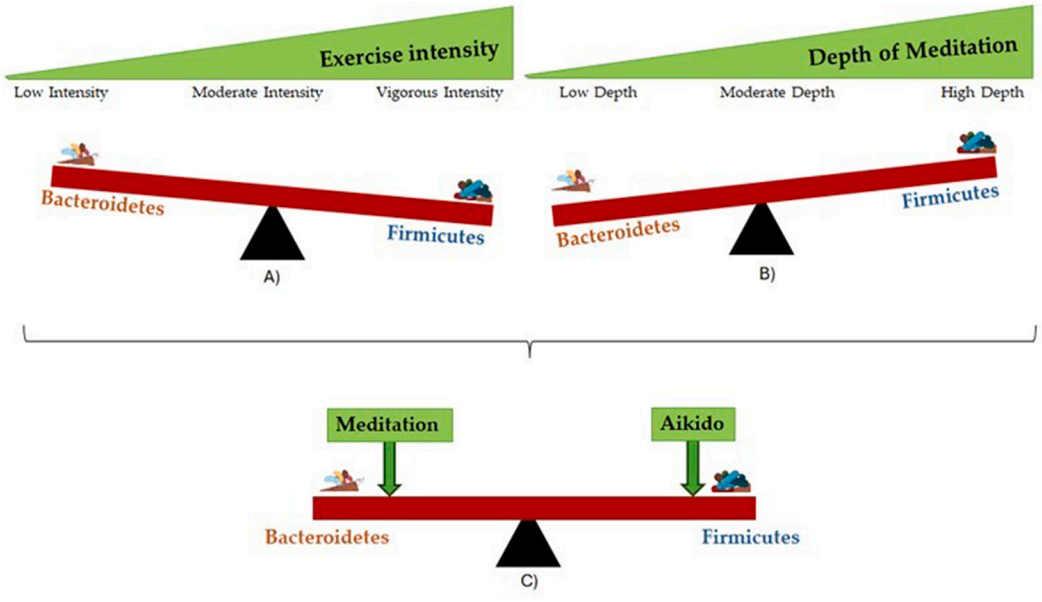

Fig. 7. Firmicutes/Bacteroidetes Ratio in Relation to Moving Meditation Disciplines. Representation of the balance Firmicutes/Bacteroidetes ratio through the practice of Moving Meditation Disciplines.

5. Conclusions

This work represents the first attempt to characterize the gut microbiota of a cohort of adult Aikido practitioners, a Moving Meditation Discipline derived from the integration of ancient Samurai arts with mental and spiritual aspects typical of Japanese world. So, Aikido proposes the systematic union of spiritual and physical components, which can also be proposed at low impact making it suitable or adaptable activity for everyone, even the most fragile populations. Moreover, it can be practiced anywhere and proposed to populations of all ages and conditions, with application to contexts of strong social value such as schools and workplaces. For these reasons, Aikido could represent a discipline of the highest value in health prevention and promotion because of its high sustainability. The results that we obtained showed significant alterations in gut microbiota composition among high-meditation practitioners. In this group, we noticed increased levels of *Bacteroides* and *Faecalibacterium* and a slight rise in *Prevotella*. For what concerns the Aikido level, the moderate-intensity group was linked to an enrichment in Firmicutes. These findings suggest that Aikido, a meditation in movement practice, promotes a balanced Bacteroidetes/Firmicutes ratio, supporting intestinal eubiosis and thus contributing to gut health and well-being.

Considering the current epidemiological scenario of western society, there is a clear and urgent need to spread physical activity as a tool for health promotion and prevention of chronic degenerative diseases. In addition to this, it is increasingly relevant to use simple, sustainable and well-tolerated methods to ensure benefits in a new perspective that considers the body-mind as a unicum. In this sense, Aikido can be effectively employed and considered as a well-being supportive tool in preventing chronic and degenerative diseases. In fact, this present work has shown how this discipline can be helpful and supportive for gut homeostasis, especially when coupled with adequate depth of meditation. The health benefits are massive, and they are not only about cardio-respiratory fitness and muscle function. Instead, they also involve cognitive and mental aspects, with positive consequences on stress control, now a daily component of contemporary society. In fact, meditation has proven to be an excellent and effective tool for improving stress management and individual responses to stressful situations. The present study, with all its critical issues, is meant to be a preliminary work on the interaction between gut microbiota, wellness and Moving Meditation Disciplines. It is desirable that this study would be used as basis for future intervention studies of Moving Meditation in order to verify a role in stress management and chronic disease prevention.

CRediT authorship contribution statement

Veronica Volpini: Writing – review & editing, Writing – original draft, Methodology, Investigation. **Sofia Bricca:** Writing – original draft, Investigation, Conceptualization. **Francesca Ubaldi:** Writing – review & editing, Writing – original draft, Methodology, Investigation. **Tehreema Ghaffar:** Writing – review & editing, Visualization, Investigation. **Serena Platania:** Visualization, Investigation. **Federica Valeriani:** Writing – review & editing, Writing – original draft, Validation, Software, Methodology, Investigation, Conceptualization. **Vincenzo Romano Spica:** Writing – review & editing, Supervision, Conceptualization.

Ethical approval statement

The study was carried out respecting the principles of the Declaration of Helsinki. The protocol of the study was approved by the Ethical Board of the University of Rome "Foro Italico" (CAR approval number 204/2024).

Funding

This study was funded by Project grant of University of Rome "Foro Italico" - PNRR Grant DM 737 and PNRR Grant DM-351.

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.

Acknowledgements

The authors would like to thank the participants of this study.

References

1. Yang Y, An R, Zhu W. Physical activity and prolonged sedentary behavior in US working adults. *Arch Environ Occup Health*. 2016;71(6):362–365. https://doi.org/10.1080/19338244.2016.1151853.
2. Liu M, Wu L, Yao S. Dose-response association of screen time-based sedentary behaviour in children and adolescents and depression: a meta-analysis of observational studies. *Br J Sports Med*. 2016;50(20):1252–1258. https://doi.org/10.1136/bjsports-2015-095084.
3. Ford ES, Caspersen CJ. Sedentary behaviour and cardiovascular disease: a review of prospective studies. *Int J Epidemiol*. 2012;41(5):1338–1353. https://doi.org/10.1093/ije/dys078.
4. Mitchell JA, Rodriguez D, Schmitz KH, Audrain-McGovern J. Greater screen time is associated with adolescent obesity: a longitudinal study of the BMI distribution from ages 14 to 18. *Obesity (Silver Spring)*. 2013;21(3):572–575. https://doi.org/10.1002/oby.20157.
5. An R, Yang Y. Diabetes diagnosis and screen-based sedentary behavior among US adults. *Am J Lifestyle Med*. 2016;12(3):252–262. https://doi.org/10.1177/1559827616650416.
6. Aadahl M, Andreasen AH, Hammer-Helmich L, Buhelt L, Jørgensen T, Glümer C. Recent temporal trends in sleep duration, domain-specific sedentary behaviour and physical activity. A survey among 25–79-year-old Danish adults. *Scand J Publ Health*. 2013;41(7):706–711. https://doi.org/10.1177/1403494813493151.
7. Teychenne M, Costigan SA, Parker K. The association between sedentary behaviour and risk of anxiety: a systematic review. *BMC Public Health*. 2015;15:513. https://doi.org/10.1186/s12889-015-1843-x.
8. Hamer M, Stamatakis E. Prospective study of sedentary behavior, risk of depression, and cognitive impairment. *Med Sci Sports Exerc*. 2014;46(4):718–723. https://doi.org/10.1249/MSS.0000000000000156.
9. World Health Organization. Physical activity. https://www.who.int/en/news-room/fact-sheets/detail/physical-activity. Accessed November 22, 2024.
10. Gallè F, Sabella EA, Ferracuti S, et al. Sedentary behaviors and physical activity of Italian undergraduate students during lockdown at the time of COVID-19 pandemic. *Int J Environ Res Public Health*. 2020;17(17):6171. https://doi.org/10.3390/ijerph17176171.
11. Penedo FJ, Dahn JR. Exercise and well-being: a review of mental and physical health benefits associated with physical activity. *Curr Opin Psychiatr*. 2005;18(2):189–193. https://doi.org/10.1097/00001504-200503000-00013.
12. Vogel T, Brechat PH, Leprêtre PM, Kaltenbach G, Berthel M, Lonsdorfer J. Health benefits of physical activity in older patients: a review. *Int J Clin Pract*. 2009;63(2):303–320. https://doi.org/10.1249/MSS.0000000000000156.
13. Fox KR. The influence of physical activity on mental well-being. *Public Health Nutr*. 1999;2(3A):411–418. https://doi.org/10.1017/s1368980099000567.
14. Henriques G, Keffer S, Abrahamson C, Horst SJ. Exploring the effectiveness of a computer-based heart rate variability biofeedback program in reducing anxiety in college students. *Appl Psychophysiol Biofeedback*. 2011;36(2):101–112. https://doi.org/10.1007/s10484-011-9151-4.
15. Anderson SA, Haraldsdottir K, Watson D. Mindfulness in athletes. *Curr Sports Med Rep*. 2021;20(12):655–660. https://doi.org/10.1249/JSR.0000000000000919.
16. Cathcart S, Galatis N, Immink M, Proeve M, Petkov J. Brief mindfulness-based therapy for chronic tension-type headache: a randomized controlled pilot study. *Behav Cogn Psychother*. 2014;42(1):1–15. https://doi.org/10.1017/S1352465813000234.
17. Bühlmayer L, Birrer D, Röthlin P, Faude O, Donath L. Effects of mindfulness practice on performance-relevant parameters and performance outcomes in sports: a meta-analytical review. *Sports Med*. 2017;47(11):2309–2321. https://doi.org/10.1007/s40279-017-0752-9.
18. Bohlmeijer E, Prenger R, Taal E, Cuijpers P. The effects of mindfulness-based stress reduction therapy on mental health of adults with a chronic medical disease: a meta-analysis. *J Psychosom Res*. 2010;68(6):539–544. https://doi.org/10.1016/j.jpsychores.2009.10.005.
19. Goyal M, Singh S, Sibinga EM, et al. Meditation programs for psychological stress and well-being: a systematic review and meta-analysis. *JAMA Intern Med*. 2014;174(3):357–368. https://doi.org/10.1001/jamainternmed.2013.13018.
20. Sun Y, Ju P, Xue T, et al. Alteration of faecal microbiota balance related to long-term deep meditation. *Gen Psychiatr*. 2023;36(1):e100893. https://doi.org/10.1136/gpsych-2022-100893.
21. Liang R, Zhang S, Peng X, et al. Characteristics of the gut microbiota in professional martial arts athletes: a comparison between different competition levels. *PLoS One*. 2019;14(12):e0226240. https://doi.org/10.1371/journal.pone.0226240.

22. Lin P, Ding B, Feng C, et al. Prevotella and klebsiella proportions in fecal microbial communities are potential characteristic parameters for patients with major depressive disorder. *J Affect Disord*. 2017;207:300–304. https://doi.org/10.1016/j.jad.2016.09.051.
23. Matziouridou C, Marungruang N, Nguyen TD, Nyman M, Fåk F. Lingonberries reduce atherosclerosis in Apoe(-/-) mice in association with altered gut microbiota composition and improved lipid profile. *Mol Nutr Food Res*. 2016;60(5):1150–1160. https://doi.org/10.1002/mnfr.201500738.
24. Li J, McArdle S, Gholami A, et al. CCR5+T-bet+FoxP3+ effector CD4 T cells drive atherosclerosis. *Circ Res*. 2016;118(10):1540–1552. https://doi.org/10.1161/CIRCRESAHA.116.308648.
25. Kang D, Wang X, Wang J. Intervention study of Tai chi training on the intestinal flora of college student basketball players. *Medicine (Baltim)*. 2023;102(36):e35044. https://doi.org/10.1097/MD.0000000000035044.
26. Kanchibhotla D, Sharma P, Subramanian S. Improvement in gastrointestinal quality of life index (GIQLI) following meditation: an open-trial pilot study in India. *J Ayurveda Integr Med*. 2021;12(1):107–111. https://doi.org/10.1016/j.jaim.2021.01.006.
27. Cahn BR, Goodman MS, Peterson CT, Maturi R, Mills PJ. Yoga, meditation and mind-body health: increased BDNF, cortisol awakening response, and altered inflammatory marker expression after a 3-month yoga and meditation retreat. *Front Hum Neurosci*. 2017;11:315. https://doi.org/10.3389/fnhum.2017.00315.
28. Christodoulou E, Mpali T, Dimitriadou ME, Koutelidakis AE. Mindfulness, gut-brain axis, and health-related quality of life: the paradigm of IBD patients. *Healthcare (Basel)*. 2024;12(12):1209. https://doi.org/10.3390/healthcare12121209.
29. Naude C, Skvarc D, Knowles S, et al. The effectiveness of mindfulness-based interventions in inflammatory bowel disease: a systematic review & meta-analysis. *J Psychosom Res*. 2023;169:111232. https://doi.org/10.1016/j.jpsychores.2023.111232.
30. Moreno JJ. Modulation of inflammatory response and pain by mind-body therapies as meditation. *Brain Behav Immun Health*. 2024;5:100036. https://doi.org/10.1016/j.bbii.2023.100036.
31. Ilan K, Motro Y, Nemirovsky A, et al. Cognitive behavioral and mindfulness with daily exercise intervention is associated with changes in intestinal microbial taxa and systemic inflammation in patients with crohn's disease. *Gut Microbes*. 2024;16(1):2337269. https://doi.org/10.1080/19490976.2024.2337269.
32. Larkey L, Jahnke R, Etnier J, Gonzalez J. Meditative movement as a category of exercise: implications for research. *J Phys Act Health*. 2009;6(2):230–238. https://doi.org/10.1123/jpah.6.2.230.
33. Meh K, Jurak G, Sorić M, Rocha P, Sember V. Validity and reliability of IPAQ-SF and GPAQ for assessing sedentary behaviour in adults in the european union: a systematic review and meta-analysis. *Int J Environ Res Public Health*. 2021;18(9):4602. https://doi.org/10.3390/ijerph18094602.
34. Martínez-González MA, García-Arellano A, Toledo E, et al. A 14-item mediterranean diet assessment tool and obesity indexes among high-risk subjects: the PREDIMED trial. *PLoS One*. 2012;7(8):e43134. https://doi.org/10.1371/journal.pone.0043134.
35. Piron H. Meditation depth questionnaire (MEDEQ) and meditation depth index (MEDI). In: Medvedev ON, Krägeloh CU, Siegert RJ, Singh NN, eds. *Handbook of Assessment in Mindfulness Research*. Springer; 2022. https://doi.org/10.1007/978-3-030-77644-2_41-1.
36. Piron H. The meditation depth index (MEDI) and the meditation depth questionnaire (MEDEQ) by. 2008. https://www.semanticscholar.org/paper/The-Meditation-Depth-Index-(-MEDI-)-and-the-Depth-(-Piron/4fb2f6294c0c3db35411fc70abfbd793863183ca [Last access 16.07.25].
37. Nieman DC, Wentz LM. The compelling link between physical activity and the body's defense system. *J Sport Health Sci*. 2019;8(3):201–217. https://doi.org/10.1016/j.jshs.2018.09.009.
38. Gallè F, Valeriani F, Cattaruzza MS, et al. Mediterranean diet, physical activity and gut microbiome composition: a cross-sectional study among healthy young Italian adults. *Nutrients*. 2020;12(7):2164. https://doi.org/10.3390/nu12072164.
39. Valeriani F, Agodi A, Casini B, et al. Potential testing of reprocessing procedures by real-time polymerase chain reaction: a multicenter study of colonoscopy devices. *Am J Infect Control*. 2018;46:159–164. https://doi.org/10.1016/j.ajic.2017.08.008.
40. Colwell RK, Chao A, Gotelli NJ, et al. Models and estimators linking individual-based and sample-based rarefaction, extrapolation and comparison of assemblages. *J Plant Ecol*. 2012;5:3–21. https://doi.org/10.1093/jpe/rtr044.
41. Magurran AE. Measuring biological diversity. *Curr Biol*. 2021;31(19):R1174–R1177. https://doi.org/10.1016/j.cub.2021.07.049.
42. Arndt D, Xia J, Liu Y, et al. METAGENassist: a comprehensive web server for comparative metagenomics. *Nucleic Acids Res*. 2012;40(Web Server issue):W88–W95. https://doi.org/10.1093/nar/gks497.
43. Gomaa EZ. Human gut microbiota/microbiome in health and diseases: a review. *Antonie Leeuwenhoek*. 2020;113(12):2019–2040. https://doi.org/10.1007/s10482-020-01474-7.
44. Chen Y, Zhou J, Wang L. Role and mechanism of gut microbiota in human disease. *Front Cell Infect Microbiol*. 2021;11:625913. https://doi.org/10.3389/fcimb.2021.625913.
45. Zhang YW, Wu Y, Liu XF, Chen X, Su JC. Targeting the gut microbiota-related metabolites for osteoporosis: the inextricable connection of gut-bone axis. *Ageing Res Rev*. 2024;94:102196. https://doi.org/10.1016/j.arr.2024.102196.
46. Zhang YW, Cao MM, Li YJ, et al. A narrative review of the moderating effects and repercussion of exercise intervention on osteoporosis: ingenious involvement of gut microbiota and its metabolites. *J Transl Med*. 2022;20:490. https://doi.org/10.1186/s12967-022-03700-4.
47. Zhang YW, Cao MM, Li YJ, et al. Fecal microbiota transplantation ameliorates bone loss in mice with ovariectomy-induced osteoporosis via modulating gut microbiota and metabolic function. *J Orthop Translat*. 2022;37:46–60. https://doi.org/10.1016/j.jot.2022.08.003.
48. El-Sayed A, Aleya L, Kamel M. Microbiota's role in health and diseases. *Environ Sci Pollut Res Int*. 2021;28(28):36967–36983. https://doi.org/10.1007/s11356-021-14593-z.
49. Campaniello D, Corbo MR, Sinigaglia M, et al. How diet and physical activity modulate gut microbiota: evidence, and perspectives. *Nutrients*. 2022;14(12):2456. https://doi.org/10.3390/nu14122456.
50. Pérez-Prieto I, Plaza-Florido A, Ubago-Guisado E, Ortega FB, Altmäe S. Physical activity, sedentary behavior and microbiome: a systematic review and meta-analysis. *J Sci Med Sport*. 2024;27(11):793–804. https://doi.org/10.1016/j.jsams.2024.07.003.
51. Matsumoto M, Inoue R, Tsukahara T, et al. Voluntary running exercise alters microbiota composition and increases n-butyrate concentration in the rat cecum. *Biosci Biotechnol Biochem*. 2008;72(2):572–576. https://doi.org/10.1271/bbb.70474.
52. Campbell SC, Wisniewski PJ, Noji M, et al. The effect of diet and exercise on intestinal integrity and microbial diversity in mice. *PLoS One*. 2016;11(3):e0150502. https://doi.org/10.1371/journal.pone.0150502.
53. Marttinen M, Ala-Jaakkola R, Laitila A, Lehtinen MJ. Gut microbiota, probiotics and physical performance in athletes and physically active individuals. *Nutrients*. 2020;12(10):2936. https://doi.org/10.3390/nu12102936.
54. Dziewiecka H, Buttar HS, Kasperska A, et al. Physical activity induced alterations of gut microbiota in humans: a systematic review. *BMC Sports Sci Med Rehabil*. 2022;14(1):122. https://doi.org/10.1186/s13102-022-00513-2.
55. Jamil A, Gutlapalli SD, Ali M, et al. Meditation and its mental and physical health benefits in 2023. *Cureus*. 2023;15(6):e40650. https://doi.org/10.7759/cureus.40650.
56. Househam AM, Peterson CT, Mills PJ, Chopra D. The effects of stress and meditation on the immune system, human microbiota, and epigenetics. *Adv Mind Body Med*. 2017;31(4):10–25.
57. Bartholomew E, Chung M, Yeroushalmi S, Hakimi M, Bhutani T, Liao W. Mindfulness and meditation for psoriasis: a systematic review. *Dermatol Ther*. 2022;12(10):2273–2283. https://doi.org/10.1007/s13555-022-00802-1.
58. Galante J, Friedrich C, Dalgleish T, Jones PB, White IR. Collaboration of mindfulness trials (CoMinT). Systematic review and individual participant data meta-analysis of randomized controlled trials assessing mindfulness-based programs for mental health promotion. *Nat Ment Health*. 2023;1:462–476. https://doi.org/10.1038/s44220-023-00081-5.
59. Jia W, Zhen J, Liu A, et al. Long-term vegan meditation improved human gut microbiota. *Evid Based Complement Alternat Med*. 2020;2020:9517897. https://doi.org/10.1155/2020/9517897.
60. Lin P, Ding B, Feng C, et al. Prevotella and klebsiella proportions in fecal microbial communities are potential characteristic parameters for patients with major depressive disorder. *J Affect Disord*. 2017;207:300–304. https://doi.org/10.1016/j.jad.2016.09.051.
61. Duan R, Liu Y, Zhang Y, et al. The impact of exercise on the gut microbiota in middle-aged amateur serious runners: a comparative study. *Front Physiol*. 2024;15:1343219. https://doi.org/10.3389/fphys.2024.1343219.
62. Chung YE, Chen HC, Chou HL, et al. Exploration of microbiota targets for major depressive disorder and mood-related traits. *J Psychiatr Res*. 2019;111:74–82. https://doi.org/10.1016/j.jpsychires.2019.01.016.
63. Kang DW, Park JG, Ilhan ZE, et al. Reduced incidence of prevotella and other fermenters in intestinal microflora of autistic children. *PLoS One*. 2013;8(7):e68322. https://doi.org/10.1371/journal.pone.0068322.
64. Bressa C, Bailén-Andrino M, Pérez-Santiago J, et al. Differences in gut microbiota profile between women with active lifestyle and sedentary women. *PLoS One*. 2017;12(2):e0171352. https://doi.org/10.1371/journal.pone.0171352.
65. Hampton HG, Watson BNJ, Fineran PC. The arms race between bacteria and their phage foes. *Nature*. 2020;577(7790):327–336. https://doi.org/10.1038/s41586-019-1894-8.
66. Karl JP, Margolis LM, Madslien EH, et al. Changes in intestinal microbiota composition and metabolism coincide with increased intestinal permeability in young adults under prolonged physiological stress. *Am J Physiol Gastrointest Liver Physiol*. 2017;312(6):G559–G571. https://doi.org/10.1152/ajpgi.00066.2017.
67. Jung Y, Tagele SB, Son H, et al. Modulation of gut microbiota in Korean navy trainees following a healthy lifestyle change. *Microorganisms*. 2020;8(9):1265. https://doi.org/10.3390/microorganisms8091265.
68. Craven J, Cox AJ, Bellinger P, et al. The influence of exercise training volume alterations on the gut microbiome in highly-trained middle-distance runners. *Eur J Sport Sci*. 2022;22(8):1222–1230. https://doi.org/10.1080/17461391.2021.1933199.
69. Sun K, Gao Y, Wu H, Huang X. The causal relationship between gut microbiota and type 2 diabetes: a two-sample Mendelian randomized study. *Front Public Health*. 2023;11:1255059. https://doi.org/10.3389/fpubh.2023.1255059.
70. Zhang Y, Fan Q, Hou Y, et al. Bacteroides species differentially modulate depression-like behavior via gut-brain metabolic signaling. *Brain Behav Immun*. 2022;102:11–22. https://doi.org/10.1016/j.bbi.2022.02.007.
71. Torquati L, Gajanand T, Cox ER, et al. Effects of exercise intensity on gut microbiome composition and function in people with type 2 diabetes. *Eur J Sport Sci*. 2023;23(4):530–541. https://doi.org/10.1080/17461391.2022.2035436.
72. Wang Z, Liu S, Xu X, et al. Gut microbiota associated with effectiveness and responsiveness to mindfulness-based cognitive therapy in improving trait anxiety.

Front Cell Infect Microbiol. 2022;12:719829. https://doi.org/10.3389/fcimb.2022.719829.

73. Ghaffar T, Ubaldi F, Volpini V, Valeriani F, Romano Spica V. The role of gut microbiota in different types of physical activity and their intensity: systematic review and meta-analysis. *Sports*. 2024;12(8):221. https://doi.org/10.3390/sports12080221.
74. Chao WC, Huang JC, Young SL, et al. Interplay of yoga, physical activity, and probiotics in irritable bowel syndrome management: a double-blind randomized study. *Compl Ther Clin Pract*. 2024;57:101892. https://doi.org/10.1016/j.ctcp.2024.101892.
75. Portincasa P, Bonfrate L, Vacca M, et al. Gut microbiota and short chain fatty acids: implications in glucose homeostasis. *Int J Mol Sci*. 2022;23(3):1105. https://doi.org/10.3390/ijms23031105.
76. Palmnäs-Bédard MSA, Costabile G, Vetrani C, et al. The human gut microbiota and glucose metabolism: a scoping review of key bacteria and the potential role of SCFAs. *Am J Clin Nutr*. 2022;116(4):862–874. https://doi.org/10.1093/ajcn/nqac217.
77. Verheggen RJHM, Konstanti P, Smidt H, et al. Eight-week exercise training in humans with obesity: marked improvements in insulin sensitivity and modest changes in gut microbiome. *Obesity (Silver Spring)*. 2021;29(10):1615–1624. https://doi.org/10.1002/oby.23252.
78. Rinninella E, Raoul P, Cintoni M, et al. What is the healthy gut microbiota composition? A changing ecosystem across age, environment, diet, and diseases. *Microorganisms*. 2019;7(1):14. https://doi.org/10.3390/microorganisms7010014.
79. Clarke SF, Murphy EF, Nilaweera K, et al. The gut microbiota and its relationship to diet and obesity: new insights. *Gut Microbes*. 2012;3(3):186–202. https://doi.org/10.4161/gmic.20168.
80. Dalile B, Van Oudenhove L, Vervliet B, Verbeke K. The role of short-chain fatty acids in microbiota-gut-brain communication. *Nat Rev Gastroenterol Hepatol*. 2019;16(8):461–478. https://doi.org/10.1038/s41575-019-0157-3.
81. Hauser AS, Attwood MM, Rask-Andersen M, Schiöth HB, Gloriam DE. Trends in GPCR drug discovery: new agents, targets and indications. *Nat Rev Drug Discov*. 2017;16(12):829–842. https://doi.org/10.1038/nrd.2017.178.
82. Wang K, Liu H, Hu Q, et al. Epigenetic regulation of aging: implications for interventions of aging and diseases. *Signal Transduct Targeted Ther*. 2022;7(1):374. https://doi.org/10.1038/s41392-022-01211-8.
83. Caetano-Silva ME, Rund L, Hutchinson NT, et al. Inhibition of inflammatory microglia by dietary fiber and short-chain fatty acids. *Sci Rep*. 2023;13(1):2819. https://doi.org/10.1038/s41598-022-27086-x.
84. Strandwitz P, Kim KH, Terekhova D, et al. GABA-Modulating bacteria of the human gut microbiota. *Nat Microbiol*. 2019;4(3):396–403. https://doi.org/10.1038/s41564-018-0307-3.
85. Jiang Y, Li K, Li X, Xu L, Yang Z. Sodium butyrate ameliorates the impairment of synaptic plasticity by inhibiting the neuroinflammation in 5XFAD mice. *Chem Biol Interact*. 2021;341:109452. https://doi.org/10.1016/j.cbi.2021.109452.
86. Frost G, Sleeth ML, Sahuri-Arisoylu M, et al. The short-chain fatty acid acetate reduces appetite via a central homeostatic mechanism. *Nat Commun*. 2014;5:3611. https://doi.org/10.1038/ncomms4611.
87. Kaelberer MM, Buchanan KL, Klein ME, et al. A gut-brain neural circuit for nutrient sensory transduction. *Science*. 2018;361(6408):eaat5236. https://doi.org/10.1126/science.aat5236.
88. Kaelberer MM, Rupprecht LE, Liu WW, et al. Neuropod cells: the emerging biology of gut-brain sensory transduction. *Annu Rev Neurosci*. 2020;43:337–353. https://doi.org/10.1146/annurev-neuro-091619-022657.