Received: 25 March 2022 | Revised: 21 June 2022 | Accepted: 27 June 2022

DOI: 10.1111/jam.15703

Journal of Applied Microbiology

ORIGINAL ARTICLE

Effects of probiotic *Lactiplantibacillus plantarum* IMC 510 supplementation on metabolic factors in otherwise healthy overweight and obese individuals

Maria Magdalena Coman[1] | **Lorenzo Miorelli**[2] | **Maria Vittoria Micioni Di Bonaventura**[3] | **Carlo Cifani**[3] | **Chiara Salvesi**[4] | **Amedeo Amedei**[5] | **Stefania Silvi**[4] | **Maria Cristina Verdenelli**[1]

[1]Synbiotec S.r.l., Spin-off of UNICAM, Camerino, Italy

[2]Sacco System S.r.l., Zelo Buon Persico, Lodi, Italy

[3]Pharmacology Unit, School of Pharmacy, University of Camerino, Camerino, Italy

[4]School of Biosciences and Veterinary Medicine, University of Camerino, Camerino, Italy

[5]Department of Experimental and Clinical Medicine, University of Florence, Florence, Italy

Correspondence
Maria Magdalena Coman, Synbiotec S.r.l., Via Gentile III da Varano, Camerino 62032, Italy.
Email: magda.coman@unicam.it

Abstract

Aims: Probiotic supplementation approach offers the possibility to shape the gut microbiota (GM), enabling the development of innovative formulations able to improve intestinal well-being and consequently the related body weight modulation and energy metabolism. In the present clinical study, a new potential probiotic supplement based on *Lactiplantibacillus plantarum* IMC 510 was studied for weight management.

Methods and Results: Quantitative characterization by qPCR of representative bacterial groups of GM was used to determine the microbiota modulation at different supplementation periods. Furthermore, measurement of the endpoints linked to weight control (body mass index, body weight, waist circumference) was assessed. Specific questionnaires to evaluate the impact on psychological and physiological point of view were performed. Results showed that after 90 days, *Lact. plantarum* IMC 510 supplementation brought an improvement in endpoints linked to weight control and healthy status, although no significant changes in the microbiota composition were reported for analysed bacterial groups, except for *Lactobacillus* spp. and *Bifidobacterium* spp.

Conclusions: We concluded that *Lact. plantarum* IMC 510 supplementation could be an interesting tool for weight management. More studies are needed to understand the impact on GM, for example, evaluating the production of short-chain fatty acids, since their important role in dietary metabolism. Further research is necessary to better elucidate the relationship between GM and overweight and the mechanism of action by which *Lact. plantarum* IMC 510 modifies body weight.

Significance and Impact of the Study: However, these promising outcomes represent a clear advantage of probiotic supplementation and identify a new potential probiotic as a novel and safe therapeutic approach in the obesity prevention and management.

KEYWORDS

body weight, gut microbiota, *Lactiplantibacillus plantarum* IMC 510, obesity, overweight, probiotics, waist circumference, well-being

INTRODUCTION

Obesity is a multifactorial disease caused by socioeconomic, hormonal and neuronal mechanisms, unhealthy lifestyle, genetic and epigenetic factors. On the other hand, overweight and obesity are the results of a positive energy balance, meaning an increased energy intake and a decreased energy expenditure. Importantly, overweight and obesity positively correlate with other chronic diseases such as type 2 diabetes, cardiovascular diseases, some types of cancer, musculoskeletal disorders, infertility and others. The World Health Organization (WHO) defines obesity as an abnormal or excessive fat accumulation that may impair health, as result of an energy imbalance between caloric intake and energy expenditure and it is estimated that 39% of the worldwide adult population will become obese within 2035 (Schütz et al., 2021). The World Obesity Atlas (2022), published by the World Obesity Federation, predicts that 1 in 5 women and 1 in 7 men will be living with obesity by 2030, that correspond to one billion of people worldwide (World Obesity Atlas, 2022). Moreover, the global coronavirus disease 2019 pandemic and the consequent social restriction measures significantly changed individuals' body weight, physical activities, daily routines and eating habits (Ali Malik et al., 2022; Schneider et al., 2022), leading to an increase in eating disorders and obesity (Rodgers et al., 2020; Sideli et al., 2021; Urhan & Okut Aysin, 2022). The WHO criteria to diagnose obesity is the body mass index (BMI) that considers overweight a person with a BMI ≥ 25, while obese a person with a BMI ≥ 30 (World Health Organization, 2021). Therefore, BMI is a useful indicator of overall adiposity and different fat compartments are associated with differential metabolic risks. Thus, an evaluation of waist circumference represents a more accurate obesity classification: visceral/central waist circumference over 88 cm for women and over 102 cm for men, or subcutaneous obesity (Fox et al., 2007).

Many recent studies indicated that the human gut microbiota (GM) plays a fundamental role in rising of obesity affecting energy harvest and nutrients absorption. Moreover, it has been demonstrated that the intestinal microbiota differs between obese and lean individuals (John & Mullin, 2016). In fact, adult GM is characterized mainly by taxa belonging to two phyla, Firmicutes and Bacteroidetes, with a high ratio of Bacteroidetes to Firmicutes (B/F) in normal-weight individuals, whereas the alteration of this ratio is correlated to obesity status (Vallianou et al., 2020). It seems that a significant variation within Firmicutes may contribute to more efficient energy extraction from food sources and to the increase in energy storage in host fat tissue (Ejtahed et al., 2019), where a specific alteration of GM composition led to a proportional increase in Firmicutes and to a decrease in Bacteroidetes phylum. Other clinical studies revealed that subjects with low bacterial richness have higher leptin and C-protein levels, insulin resistance, dyslipidaemia, adiposity and inflammatory phenotype, gaining more weight respect to the individuals with high bacterial diversity (Schütz et al., 2021). Intestinal bacteria ferment indigestible carbohydrate residues, synthesize short-chain fatty acids (SCFA) and amino acids, possibly contributing to an increase in the amount of energy supplied to the host. On the other hand, by-products of bacterial fermentation can reduce appetite and increase satiety as well as a decrease in body weight and BMI. Consequently, the intestinal microbiota is a potentially modifiable factor associated with the prevention and treatment of excessive body weight.

The modulation of GM is involved in many benefits for weight control since evidences showed a role of GM in gastrointestinal protection, permeability degree of the mucosa, dietary polysaccharides metabolism by fermentation and absorption, higher catabolism and production of SCFA, highlighting its crucial role in the management of fat gathering and the consequent obesity. Knowledge of how the GM can modulate body weight and energy metabolism is essential to have insights overview on the role of intestinal bacteria. There are three primary GM mechanisms playing an important role: production of SCFA, control of bile acid metabolism, and induction and/or protection from metabolic endotoxaemia (Brusaferro et al., 2018).

In this scenario, in addition to nutrients and dietary patterns, probiotics have been investigated on the capacity to positively modulate the GM composition, to stimulate the immune system and to maintain the intestinal barrier integrity through competitive effects against certain pathogens. According to a joint FAO/WHO, F.A.O (2002), probiotics are defined as 'live microorganisms that confer a health benefit to the host when administered in adequate amounts'. Several studies demonstrated the positive effects of probiotics on obesity and related health complications, such as reduction of adipose tissue inflammation, endotoxemia, adiposity, body mass, leptin levels and energy intake (Guazzelli Marques et al., 2020).

Based on studies carried out in experimental animals, most of the formulations tested in clinical trials contained strains of the genera *Lactobacillus* and *Bifidobacterium* with the purpose to evaluate the impact of probiotics on the obesity status, especially on weight changes, while the use of other strains can be deleterious. Many studies are available and most of them reports positive results in both adults and children (Brusaferro et al., 2018). Recently, an interesting work on obese rabbits (Bouaziz et al., 2021), supplemented with these two probiotics strains, revealed their beneficial effect on

several biochemical and morphometric parameters related to obesity and metabolic syndrome. Moreover, the authors strongly emphasize that the anti-obesity effects of probiotics to the host are species specific and strain specific and dosage, and duration of administration dependent, in line with other studies (Daniali et al., 2020; Ejtahed et al., 2019).

The aim of the present pilot study was to evaluate the potential modulatory effect of probiotics on weight management. The potential probiotic strain *Lactiplantibacillus plantarum* IMC 510, tested in the present pilot clinical trial, was previously isolated from healthy human subjects, identified by 16S rRNA gene sequence analysis, characterized as probiotic strain and tested for safety (resistance to low pH, bile salts and pancreatic juice, ability to adhere to intestinal cells and colonize the mucosa, antipathogenic activity against gram-positive and gram-negative bacteria and yeasts, non-transmissible antibiotic resistance genes', absence of plasmids) by Synbiotec S.r.l. (Coman et al., 2021; Coman & Cresci, 2014; Silvi et al., 2003). Furthermore, *Lact. plantarum* IMC 510 was evaluated in vivo in obese rats and it was able to decrease food intake, weight gain and, consequently, to induce relative beneficial effects, demonstrated by serological, biochemical and histological analyses, potentially through leptin control (Micioni Di Bonaventura et al., 2021). We hypothesized that daily consumption of one capsule of this probiotic strain could modulate the intestinal GM, thereby reducing body weight and fat, positively influencing several biomarkers of obesity-associated disorders including diabetes, hypertension and cardiovascular disease.

MATERIALS AND METHODS

Subjects and enrolment

This study involved 19 overweight/obese volunteers, consisting of 10 males and 9 females. Eligible participants were recruited mostly from the University of Camerino and Synbiotec Srl personnel via e-mail and word-of-mouth advertisement. The age of the subjects ranged between 26 and 73 years old. The inclusion criteria of the covered subjects were BMI equal to or greater than 25. Participants were asked to maintain their usual diet as well as their physical activity level. All the subjects gave their informed consent to participate and the followed procedures were in accordance with the ethical standards of the responsible committee on human experimentation and with the Helsinki Declaration of 1975, as revised in 1983.

There were several inclusion requirements for the feasibility study: men and women recruited had to be between 26 and 73 years, they had to be obese (BMI > 30, class I, II, III) or overweight (BMI 25÷29.9). There had to be willingness to collaborate during the study, providing informed consent and ability to follow guidelines, in particular the availability to complete questionnaires, records and journals associated with the study. Consumption of functional foods, dietary supplements with probiotics, laxatives and substances for body weight control had to be suspended. While the exclusion criteria prohibit the use of probiotics continuously, in the 4 weeks prior to treatment, it was also forbidden use of other treatments (drugs or nutritional programmes) that affect body weight, food intake and/or energy expenditure. Menopausal, pregnant or breastfeeding women cannot participate in the study as well as those who are enlist in another obesity treatment programme. Moreover, there were criteria for leaving the study, occurring one of the following conditions: non-continuity of administration, severe illness, continuous use of antibiotics and/or laxatives or other probiotics. Finally, the volunteers were divided into a probiotic supplemented group (seven females and five males) and a placebo group (two females and five males).

Probiotic strain supplementation

Probiotic strain was supplied by capsules. Each capsule contained 1.5×10^{10} bacterial cells of the human origin strain *Lact. plantarum* IMC 510, isolated and characterized by Synbiotec S.r.l., and deposited at the Deutsche Sammlung von Mikroorganismen und Zelkulturen (DSMZ) collection with the identification number DSM 32548 (Coman et al., 2021; Coman & Cresci, 2014; Silvi et al., 2003). Compared with the probiotic capsules, the placebo ones contained just the same carrier material and were similar in size, shape and taste.

Study design

A randomized, placebo-controlled study was performed with a 4-week run-in period followed by a 12-week intervention period. The subjects should not have antibiotic treatment in the previous month. During the run-in period, they were asked to discontinue the possible consumption of laxatives, dietary fibres and probiotics.

The intervention project involves three phases: Phase 1—evaluation period in which the recruited subjects respect the inclusion criteria to access the successive phase (1 month); Phase 2—period of 3 months of intervention in which the subjects had the dietary supplementation in the form of capsules containing probiotics with the indication to take one capsule a day; Phase 3—follow-up—1 month after the intervention period (Figure 1).

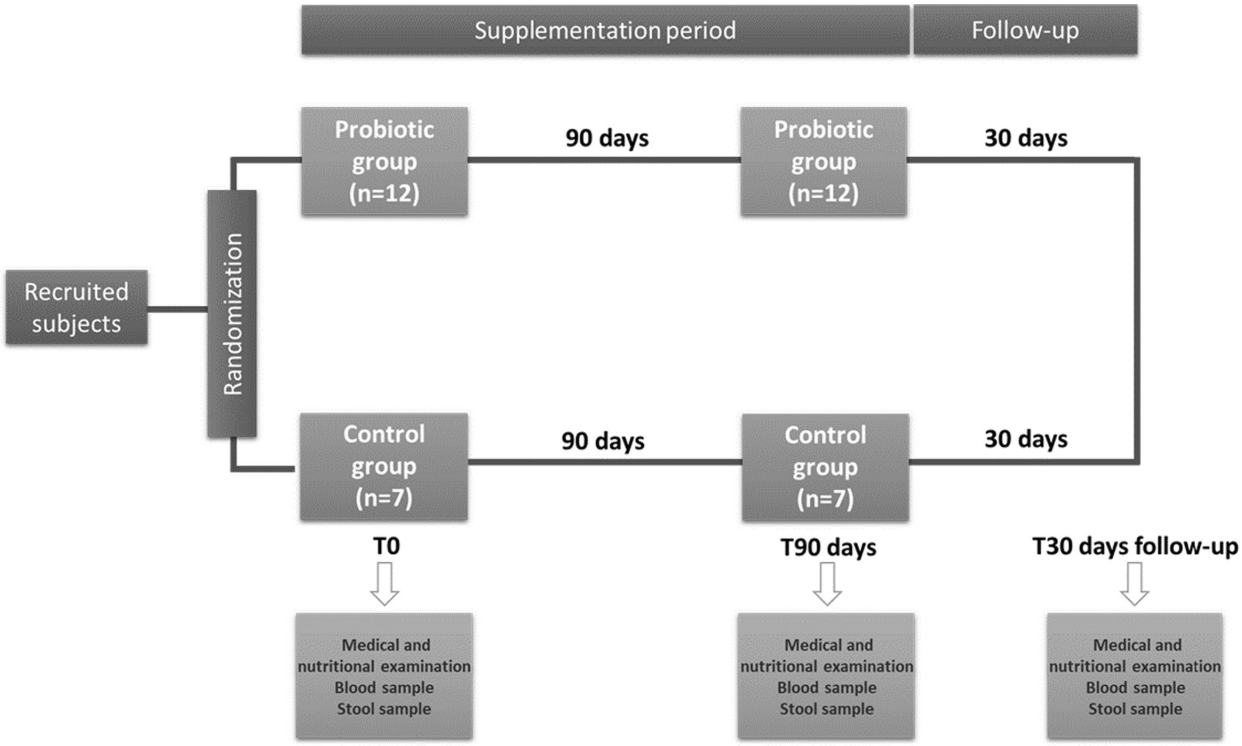

FIGURE 1 Clinical trial design

At the end of each phase, biological samples were collected and analysed. Participants were subjected, at each stage of study, to questionnaires on intestinal and psychological well-being and assessment of specific quality of life regarding obesity.

The safety evaluation of the new dietary supplement containing the selected potential probiotic strain (*Lact. plantarum* IMC 510) and the effect on the intestinal microflora and body weight reduction, keeping the same eating habits, was performed at the time of recruitment, after 3 months of probiotic treatment and after 1 month of follow-up, as first objectives. Moreover, the secondary objectives concerned the changes in BMI, waist circumference, blood parameters, such as glycemia, lipids levels, liver function (glutamic oxaloacetic transaminase and glutamic pyruvic transaminase in UI/l), intestinal well-being, quality of life, general well-being and distress level generated by overweight and obesity status.

Physiological measurements

Body weights of the volunteers were recorded using a calibrated scales after the removal of shoes and jackets. Waist circumference was measured two fingers below the umbilicus, while height was measured after shoes removal. For each participant, all the measurements were taken at the same time of the day, at each check time point (T0, T90 days and T30 days of follow-up).

Evaluation of the health status by questionnaires

Bowell well-being

This first questionnaire concerned bowel well-being and stool consistency, which is a basic parameter in the description of normal or altered intestinal habit (Silvi et al., 2014). These parameters were self-evaluated by subjects at the end of the intervention by questions on 'change in the numbers of times of defecation per day as stool frequency' and 'change in the number of eggs (large size) that correspond to the volume by visual estimation'. The people were asked to record if in the last 3 months and after 1-month follow-up their bowel well-being in terms of intestinal regularity and stool volume has remained the same, improved or worsened compared to the period before beginning the consumption of the probiotic products and also the degree of change on a combined scale leading to a 10-point Likert scale (−5, 0, +5). Secondary outcome measures were also investigated: ease at defecation, bloating, constipation, abdominal pain, intestinal cramps, feeling of incomplete defecation, incontinence and halitosis (also the

change of these individual bowel habits was assessed with the same combined Likert scale). Stool consistency was defined by the Bristol Stool Form Scale (Lewis & Heaton, 1997).

Health-related quality of life

Health-related quality of life (HRQoL) of subjects was assessed by self-administration of Psychological General Well-Being Index (PGWBI) (Dupuy, 1984) that is a general questionnaire measuring psychological well-being and distress and it is composed of 22 items, which constitute six dimensions (anxiety, depression, self-control, positive well-being, general health and vitality). Each scale includes 3–5 items. Questions allow multiple-choice answers with scores ranging from 0 to 5 (best score value). The PGWBI global score represents the sum of all items and ranges and higher scores (100—best) indicate greater psychological well-being (Silvi et al., 2014).

Obesity related well-being questionnaire

The Obesity Related Well-being (ORWELL) questionnaire is a psychometric test for assessing the specific quality of life regarding obesity. The areas investigated include both physical well-being and emotional state and relationship life. For each item, the participants were asked to score on a 4-point Likert scale (0, not at all; 1, just a little; 2, not so much; 3, much) related to the occurrence and/or severity of the symptom (occurrence) and the subjective relevance of the symptom-related impairment in one's own life (relevance). The score of the item is calculated as the product of occurrence and relevance. The total ORWELL 97 score is obtained as the sum of the scores of individual items (Mannucci et al., 1999). The sums of the scores related to occurrence (ORWELL 97-O) and relevance (ORWELL 97-R) of symptoms in individual items were also calculated. Higher ORWELL scores mean a lower quality of life.

GM characterization

Faecal samples

Faecal samples were collected from each subject of both probiotic and placebo groups at time 0 (T0), corresponding to the starting day of probiotic/placebo supplementation, after 90 days of probiotic/placebo supplementation (T90 days) and after 30 days of follow-up period (T30 days follow-up). The faeces were frozen at −80°C until performing the microbiota analysis (selected bacterial group enumeration by quantitative Real-Time PCR [qPCR] and bacterial probiotic strain colonization).

DNA extraction

DNA extraction from all faecal samples was performed using a Stool DNA Isolation Kit (NorgenBiotek Corp.) with a modified protocol following the manufacturer's instructions specific for the faecal samples. Quantity and purity of all extracted DNA were checked with a NanoDrop ND-1000 Spectrophotometer (Thermo Scientific) and then stored at −20°C until used for molecular analysis.

Quantitative Real-Time PCR

A qPCR procedure was used for the quantification of selected bacterial groups from probiotic and placebo subjects' faeces.

On DNA extracted from faecal samples, a SYBR Green Real-Time PCR amplification was performed using an iCycler iQ Real-Time Detection System (Stratagene) associated with MXP Software. All PCR experiments were carried out in triplicate with a reaction volume of 20.6 µl using iCycler IQ 96-well optical grade PCR tubes (Stratagene) covered with iCycler optical cap (Stratagene). The efficiency of PCR amplification was optimized with primers concentration of 500 ng/µl (pmol/µl) proved to be optimal for the amplifications of target sequences. The reaction mixtures contained 9 µl QuantiNova SYBR Green PCR Kit (Qiagen), 9.8 µl distilled sterile water and 0.4 µl of each primer (forward and reverse, DSMZ). Subsequently, 1 µl of faecal DNA (or water for negative control) was added to the reaction mixture. *Lactobacillus* spp., *Bifidobacterium* spp., Enterobacteriaceae, *Clostridium coccoides–Eubacterium rectale* group, *Staphylococcus* spp. and *Bacteroides–Prevotella-Porphyromonas* spp. were the bacterial groups of interest. For quantification of the above-mentioned target groups of bacteria, standard curves, previously generated for each of them as reported by Avella et al. (2010) and Nasuti et al. (2016), were used. They were previously obtained using the extracted DNA from reference strains for the quantification of the target species.

Recovery of *Lact. plantarum* IMC 510 from faeces after supplementation period

To confirm the presence of the tested strain in the intestine, faecal samples were analysed by enumeration of lactobacilli onto MRS agar (de Man, Rogosa and Sharpe agar; Oxoid) by 10-fold serial dilution method (Verdenelli

et al., 2009). After aerobic incubation at 37°C for 48–72 h, 10%–20% of the total colonies per sample randomly selected from countable agar plates were isolated and checked for purity. DNA extracted from the selected colonies using a modified benzyl chloride method (Zhu et al., 1993) was analysed by an RAPD technique (Verdenelli et al., 2009).

Blood parameters

Each volunteer went to the Analisi Biemme laboratory (Castelraimondo, MC, Italy), where blood samples were collected and then analysed. Blood samples were collected three times: at the beginning of the study, after 90 days of probiotic/placebo supplementation and after 30 days of follow-up the supplementation period.

Statistical analysis

The results are expressed as mean ± standard deviation or standard error. Statistical significance of the differences between the probiotic group and the placebo group was analysed using Student *t*-test. Significant differences between mean values were determined by Tukey's test after one-way analysis of variance using GraphPad PRISM® 5.1 program. A *p*-value less than 0.05 was considered statistically significant. For the bowel habits, where $p < 0.05$ reflects significant difference, a four multiple comparison test was used.

RESULTS

Recruitment and baseline characteristics of subjects

In all, 19 volunteers were recruited to the clinical study. There were no dropouts, exclusions or adverse events in either arm of the study and compliance to the supplementation was 100%. Baseline demographics of the participants who completed the study are shown in the Table 1.

Subjects were well balanced over both groups, probiotic and placebo one's, respect to the baseline characteristics, such as gender, height, waist circumference and weight. In fact, there were not significant differences within the supplemented and placebo group ($p > 0.05$, Student *t*-test), except for the age ($p < 0.05$).

Effects of *Lact. plantarum* IMC 510-dietary supplementation on body weight

Variations in body weight, BMI and waist circumference (mean values) are summarized in Figure 2. The probiotic group revealed significant decreases ($p < 0.05$) from T0 in the following parameters at the started time points. The control group (placebo), by contrast, showed no significant decrease in any parameter of the measured values at any time point.

As reported in Figure 3, at the end of 90 days of supplementation, the subjects from the probiotic group registered a significant reduction in terms of weight (−3.39 kg), waist circumference (−4.29 cm) and BMI (−1.15). It is important to highlight also the 'placebo effect', since it was recorded a very similar trend in the placebo group respect to probiotic group. The tendency to lose weight and to reduce the waist circumference was maintained also after 30 days of follow-up of supplementation period.

Effects of *Lact. plantarum* IMC 510-dietary supplementation on GM

A qPCR procedure was used for the quantification of selected bacterial groups from volunteers' faeces. The bacterial groups of interest were *Bacteroides–Prevotella–Porphyromonas* spp., *Staphylococcus* spp., *Cl. coccoides–E. rectale* group, *Lactobacillus* spp., *Bifidobacterium* spp. and

TABLE 1 Demographic and baseline characteristics of the study population. Values are expressed as mean ± standard deviation

	Probiotic group ($n = 12$)	Placebo group ($n = 7$)	p^*
Age (years)	44.2 ± 11.8	59.3 ± 11.6	<0.05
Gender			
Males (*n* [%])	5 (41.7)	5 (71.4)	
Females (*n* [%])	7 (58.3)	2 (28.6)	
Body weight (kg)	84.7 ± 20.3	98.2 ± 16.4	>0.05
Height (m)	1.7 ± 0.1	1.7 ± 0.1	>0.05
Waist circumference (cm)	101.8 ± 20.0	115.1 ± 15.5	>0.05

*Significance level of $p < 0.05$ by Student *t*-test.

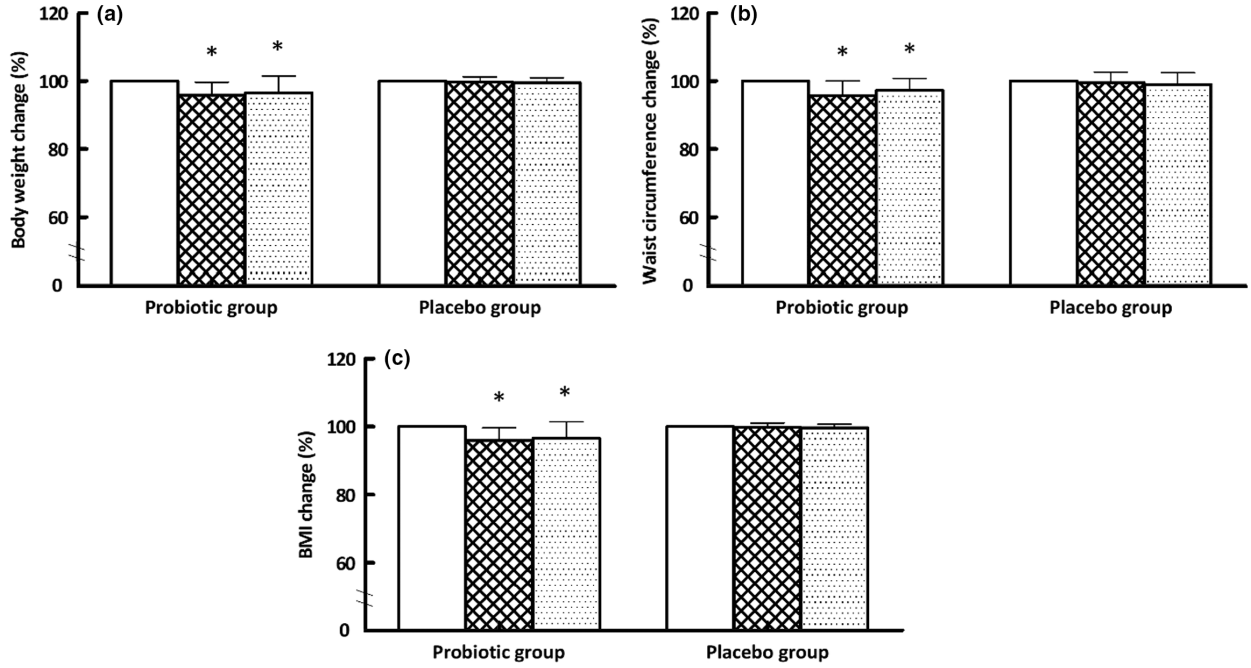

FIGURE 2 Changes (%) in body weight (a), waist circumference (b) and BMI (c) during different time points (□T0, ▩ T90 days, ▨ T30 days follow-up) relative to the two groups of volunteers (probiotic group and placebo group). *Significantly different from the starting point with supplementation (Student *t*-test, $p < 0.05$).

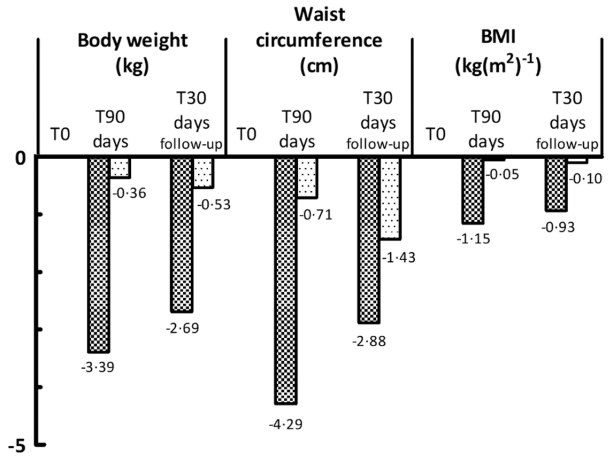

FIGURE 3 Reduction respect to starting point (T0) in body weight, waist circumference and body mass index (BMI), during the different time points (T90 and T30 days follow-up) relative to the probiotic (▩) and placebo (■) groups of volunteers.

Enterobacteriaceae. Figure 4 shows the changes of microbiota composition over the supplementation period and after 30 days of follow-up in both probiotic and placebo groups.

The probiotic supplementation significantly increased the concentration of *Lactobacillus* spp. (Figure 3, $p < 0.05$) after the 90 days of treatment respect to T0. In addition, also *Bifidobacterium* spp. level increased after 90 days of probiotic supplementation, even if not in a significant way. Any significant modification was found for the other bacterial groups (*Bacteroides–Prevotella–Porphyromonas* spp., *Staphylococcus* spp., *Cl. coccoides–E. rectale* group and Enterobacteriaceae) between the two groups during the supplementation period (Figure 4). However, the proportion of subjects with increased/decreased bifidobacterial concentration after supplementation period tended to be different between the two groups (chi square, $p = 0.01$). In particular, *Bifidobacterium* spp. level increased in 10 subjects out of 12 (83.3%) participants in the probiotic group. An opposite trend, with decreasing values, was observed in five subjects out of seven (71.4%) participants in the placebo group (data not shown).

Intestinal colonization of *Lact. plantarum* IMC 510

This test was carried out to confirm intestinal transit survival in human subjects. The recovery of *Lact. plantarum* IMC 510 from all the faecal samples (collected during the three different collection time points) demonstrated the presence of the strain during the consumption of the dietary supplement. Furthermore, the strain was also still identified in all the participants' faeces at the end of follow-up period. The strain was not detected in any of the faeces' samples collected before the consumption of the bacterial strain (Figure 5).

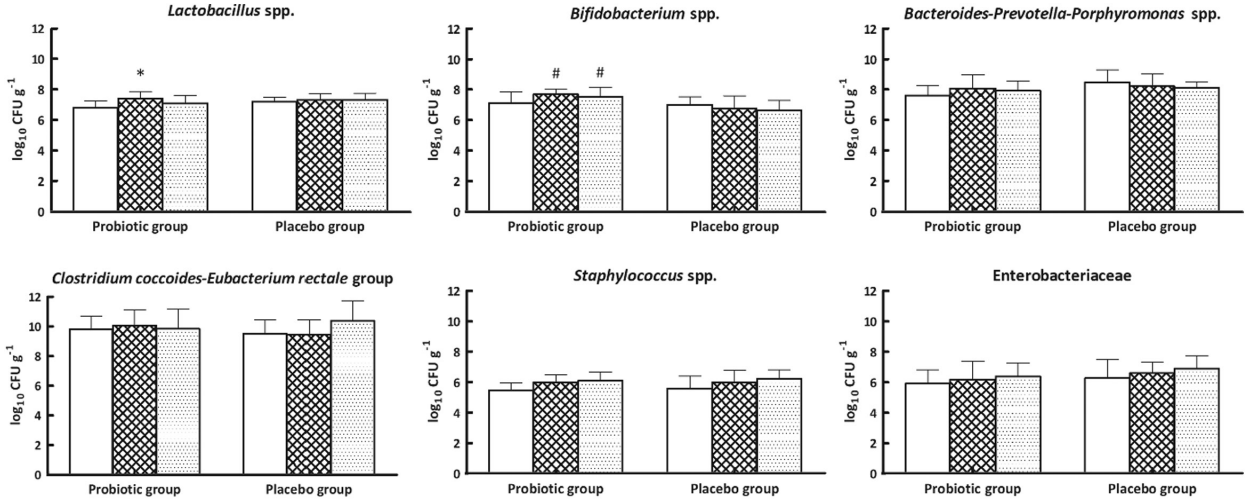

FIGURE 4 Faecal bacteria concentration ($\log_{10}$ CFU per gram ± standard deviation) of target bacterial groups during the different time points (☐ T0, ▩ T90 days, ⬚ T30 days follow-up) relative to the two groups of volunteers (probiotic group and placebo group). *Significantly different ($p < 0.05$) from T0 and #from the placebo group at the same time point by Tukey's test following one-way analysis of variance.

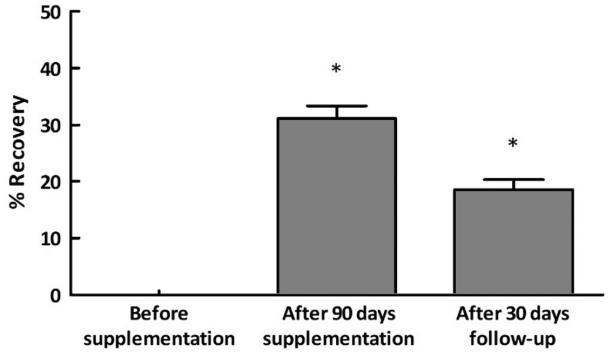

FIGURE 5 Recovery percentage of *Lactiplantibacillus plantarum* IMC 510 (■) respect to the total *Lactobacillus* spp., assessed by an RAPD method in faecal samples of subjects under probiotic supplementation. *$p < 0.05$ (using Student *t*-test).

The average value of recovery percentages of the probiotic *Lact. plantarum* IMC 510 (Figure 5) reached the 31.12% of the total *Lactobacillus* spp. at the end of the administration period and remained high, 18.50% for *Lact. plantarum* IMC 510, after 30 days of follow-up with respect to the total *Lactobacillus* spp. recovery.

Blood parameters

No significant changes in physiological, metabolic and inflammatory markers of the metabolic syndrome were observed in the probiotic group respect to the initial status of the volunteers (Table 2).

TABLE 2 Blood parameters levels during the clinical trial

	Probiotic group			Placebo group		
Parameters	**T0**	**T90 days**	**T30 days follow-up**	**T0**	**T90 days**	**T30 days follow-up**
Glycaemia	96.11 ± 18.25	93.33 ± 17.04	94.50 ± 18.58	116.00 ± 28.28	119.50 ± 33.23	110.50 ± 20.51
GOT	25.50 ± 6.89	20.75 ± 4.65*	20.50 ± 5.58*	21.50 ± 4.95	21.00 ± 4.24	22.00 ± 5.66
GPT	27.00 ± 10.01	22.25 ± 8.88	21.50 ± 8.94	20.50 ± 3.54	21.00 ± 2.83	22.50 ± 0.71
Cholesterol	194.11 ± 34.11	195.56 ± 41.99	189.88 ± 47.69	170.00 ± 53.74	178.50 ± 41.72	168.50 ± 55.86
HDL	62.67 ± 15.63	58.11 ± 9.84	55.38 ± 10.70	47.00 ± 4.24	46.00 ± 5.66	44.50 ± 7.78
LDL	113.71 ± 31.46	121.96 ± 35.51	122.20 ± 42.15	104.50 ± 64.35	95.00 ± 77.78	101.50 ± 68.59
Triglycerides	112.89 ± 48.84	104.44 ± 42.45	105.75 ± 54.92	116.50 ± 43.13	160.00 ± 104.65	136.50 ± 71.42

Note. Data expressed as mg/dl, are the mean ± standard deviation.

Abbreviations: GOT, serum glutamic-oxaloacetic transaminase; GPT, serum glutamic pyruvic transaminase; HDL, high-density lipoprotein; LDL, low-density lipoprotein.

*Significantly different from T0, $p < 0.05$ (Student *t*-test).

Bowel habits

The significant changes in bowel habits, considered as primary outcomes from the bowel well-being questionnaire, are listed in Table 3. Generally, the subjects using probiotics formulation presented significantly higher values ($p < 0.05$) in the positive scale scores related to the bowel habits. HRQoL was estimated as PGWBI global score and the probiotic group, after the 90 days of supplementation and also the 30 days of follow-up, showed values statistically higher ($p < 0.05$) respect to index values registered at the beginning of the probiotic consumption.

Quality of life

To assess obesity-related quality of life, self-reported measurement through the ORWELL 97 questionnaire was attempted to evaluate the subjective relevance and intensity of physical and psychological distress.

The total ORWELL 97 score is obtained as the sum of the scores of individual items; furthermore, to understand the results shown in Figure 6, it is important to keep in mind that higher ORWELL 97 scores are related to a lower quality of life, analysed using Student *t*-test with significance level lower than 0.05.

The value of the total score, after taking probiotics and also at 30 days of follow-up, has a decreasing trend, even if not significant ($p > 0.05$), which translates into a slight increase in the quality of life.

DISCUSSION

In the present study, probiotic *Lact. plantarum* IMC 510 supplementation significantly reduced body weight, BMI and waist circumferences in the overweight/obese subjects. The reduction in waist circumference is important because it is involved in a useful measure of fat distribution and it is closely correlated with atherogenic lipid profiles (Kadooka et al., 2010). Meanwhile, we noticed neither significant alteration nor physiological abnormality in lipid metabolism-related parameters such as triglycerides, total, low-density lipoprotein- or high-density

TABLE 3 Mean absolute changes in bowel habits frequency score, stool consistency and global score of psychological general well-being index (PGWBI) questionnaire over the 3 months of probiotic/placebo supplementation period and after 1 month of follow-up period

	Probiotic group (mean ± confidence limits)		Placebo group (mean ± confidence limits)	
Bowel habits[a]	**T90 days**	**T30 days follow-up**	**T90 days**	**T30 days follow-up**
Intestinal regularity	1.8 ± 0.3[d,e]	1.1 ± 0.3[d]	−0.1 ± 0.3	0.0 ± 0.4
Stool volume	1.8 ± 0.4[d,e]	1.3 ± 0.3[d]	−0.1 ± 0.4	0.1 ± 0.5
Stool consistency	2.2 ± 0.3[d,e]	1.8 ± 0.3[d,f]	0.0 ± 0.1	0.0 ± 0.1
Ease of defecation	1.8 ± 0.3[d,e]	1.3 ± 0.3[d]	0.0 ± 0.5	-0.1 ± 0.5
Rumbling of the stomach	1.8 ± 0.3[d]	1.4 ± 0.3	−0.3 ± 0.4	-0.1 ± 0.5
Swelling	1.6 ± 0.5[d]	1.3 ± 0.3[d]	−0.3 ± 0.1	-0.3 ± 0.1
Flatulence	1.2 ± 0.3[d]	1.3 ± 0.4	0.0 ± 0.1	0.0 ± 0.1
Constipation	0.9 ± 0.4	0.0 ± 0.2	0.0 ± ±0.5	0.0 ± 0.5
Diarrhoea	0.2 ± 0.1[e]	0.0±0.1[f]	−0.1 ± 0.5	0.0 ± 0.6
Dermatitis	0.7 ± 0.2	0.4 ± 0.1	−0.1 ± 0.4	0.0 ± 0.5
Food intolerance	0.8 ± 0.1	0.6 ± 0.1	0.0 ± 0.5	0.0 ± 0.5
Stool consistency[b]				
Type	3 ± 0.2[d]	3 ± 0.2[d]	3 ± 0.5	4 ± 0.5
PGWBI[c]				
Global Score	76.2 ± 3.2[d]	76.3 ± 3.8[d]	73.7 ± 3.3	72.0 ± 2.8

[a]Changes in bowel habits were assessed with a 10-combined Likert scale (−5, 0, 5).

[b]The assessment has been obtained following the Bristol Stool Chart.

[c]The global score ranged from 0 to 100 (best).

[d]Significantly different from the starting point of the supplementation T0.

[e]Significantly different from Placebo T90 days.

[f]Significantly different from Placebo T30 days follow-up (Student *t*-test, $p < 0.05$).

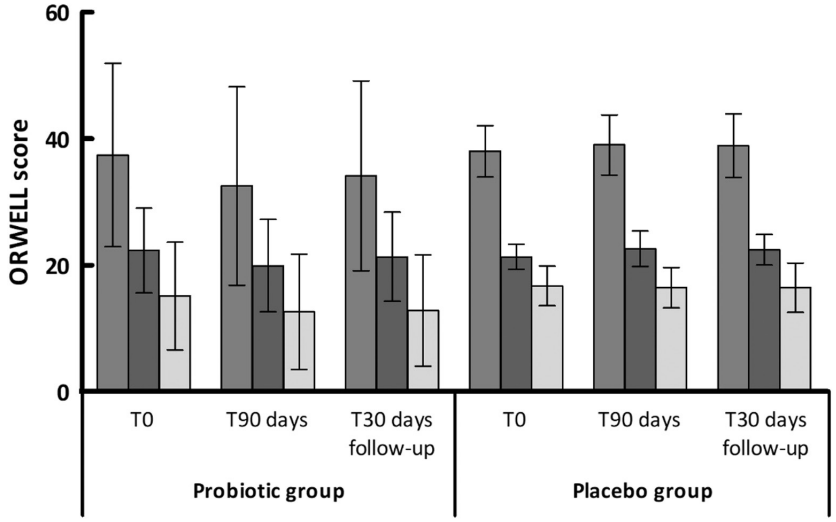

FIGURE 6 Obesity Related Well-being (ORWELL) 97 score (■-ORWELL Total score, ■-ORWELL relevance score, ■-ORWELL occurrence score) expressed as mean ± standard deviation during the different time points (T0—beginning of the study, T90 days of supplementation and T30 days follow-up the supplementation) relative to the two groups of volunteers (probiotic group and placebo group).

lipoprotein-cholesterols. No major changes in the faecal microbiota were detected in response to the probiotic supplementation like other researchers, who observed no changes in the GM composition in healthy adults after probiotic administration (Khalesi et al., 2019).

An increased abundance of lactobacilli was found in faecal samples from the supplemented group during the intervention period, suggesting the successful administration and compliance. Finally, no clinically problematic findings were observed throughout the study in any subject, confirming the safety of the probiotic tested.

The absence of changes in the plasma metabolic and inflammatory markers may be explained by the fact that the inclusion and exclusion criteria used in the present study prevented the recruitment of subjects displaying an unhealthy metabolic profile. Thus, these findings do not exclude that *Lact. plantarum* IMC 510 can improve the blood parameters and suggest that its benefits are possibly limited in subjects showing haematologic alterations. Notably, a large number of studies showed that certain probiotics may be able to lower glycaemia and serum lipids, particularly in diabetic or hypercholesterolemic subjects in clinical (Jones et al., 2012; Khalili et al., 2022; Kocsis et al., 2020; Wang et al., 2018; Wu et al., 2017; Zhang et al., 2022) and in pre-clinical research (Cabello-Olmo et al., 2022; Marques et al., 2020; Ooi & Liong, 2010; Reamtong et al., 2021; Wang et al., 2012). Notably, last year, three probiotics were tested in a healthy rabbit (see details in Kadja et al., 2021) and improved the biochemical and haematological profiles. These results are very promising and the choice of the rabbit as obesity and metabolic model is important, because they are sensitive to high-fat diets and consequently they develop hyperlipidaemia more quickly and easily (Fan & Watanabe, 2000; Waqar et al., 2010) than rats and mice, that are more resistant to develop hypercholesterolaemia (Andreadou et al., 2020; Giudetti et al., 2020; Martinelli et al., 2020; Micioni Di Bonaventura et al., 2017, 2020; O'Neal & Still, 1962).

Two hypotheses are plausible to speculate: the primary one is that an integrated diet with probiotics leads to reduction of weight and body circumference and to the regularization of appetite with consequent reduction in food intake. It has recently emerged that intestinal microbiota has an important role in body weight regulation, influencing the energy metabolism (Kadooka et al., 2010). For a decrease in body weight, it is necessarily a shift in the balance between available energy utilization and food intake, leading to a net energy deficit. Moreover, the probiotic supplementation confers intestinal well-being and improves the balance of the GM bringing general well-being to the organism. The secondary hypothesis, taking into consideration the strengthening of the immune system, supports the usefulness of the probiotics to improve the quality of life. It is very well known that obesity and overweight have a huge impact on the quality of life resulting from factors such as less ability to perform activities, early fatigue, increased anxiety, depression and low self-esteem (Michael et al., 2020). Using several validated questionnaires, the results of our study indicated significant improvements in participant scores for general wellness, state of health/energy/mood, intestinal well-being and quality of life after 3 months of probiotic supplementation.

Evidence so far demonstrates that the bacteria, commonly found in the human gastrointestinal tract, affect nutrient acquisition and energy regulation. This suggests an important role played by GM in the development of

obesity. The focus of many recent studies has been given to the role of these micro-organisms in the management of metabolic diseases and obesity, due to the beneficial effect of probiotics in adjusting the gut dysbiosis to rebalance intestinal microbiota (Ejtahed et al., 2019).

Changes in the GM composition caused by external factors may promote the development of metabolic diseases. The proposed mechanisms of action for probiotic-mediated weight loss include the modulation of GM composition and the production of SCFAs, the regulation of energy homeostasis and/or satiety, the improvement of the gut barrier function and the interruption of bile acid metabolism in the host (Cerdó et al., 2019).

Several findings indicated that the probiotic effect on body weight and metabolism is strain specific and that only *Lactobacillus* and *Bifidobacterium* genera are effective, whereas the use of other strains can be deleterious (Brusaferro et al., 2018). The two most detected bacterial phyla in humans are the Firmicutes and the Bacteroidetes, with a high ratio of B/F in normal-weight subjects, while a low B/F ratio is found after obesity development, highlighting the impacting role of GM in the fat metabolism regulation (Cheng & Liu, 2020). The present study reveals an increase in relative abundance of Firmicutes and higher Firmicutes/Bacteroidetes (F/B) ratio in overweight and obese placebo group, while in the supplemented group the probiotic consumption maintain lower the F/B ratio (data not shown), confirming the potentiality of *Lact. plantarum* IMC 510 to contrast obesity through GM modulation and reinforcing the probiotics use in the obesity management.

Obviously, probiotic action in body weight is not only species specific, but also strain specific and additional research is needed to clarify further the mechanisms underlying the effects observed in the present study.

Anyway, this study shows a positive translational effect of *Lact. plantarum* IMC 510 on body weight from a preclinical rats model of obesity (Micioni Di Bonaventura et al., 2021) to a human intervention study in otherwise healthy overweight and obese individuals. In fact, it was demonstrated a clear reduction in food intake and accordingly also in body weight gain, BMI, liver and white adipose tissue weight, hepatic lipid accumulation, adipocyte size and other parameters in obese rats after 84 days *Lact. plantarum* IMC 510 supplementation (Micioni Di Bonaventura et al., 2021). This highlights the promising potential of *Lact. plantarum* IMC 510 to be developed as a valuable supplement in decreasing specific obesity markers.

However, the identification of strains that are potentially associated with a beneficial effect is not enough to suggest their systematic use in the treatment of obesity and related metabolic disturbances. The correct dosage, duration of administration and long-term effects of the administration of the different strains are not known yet. Further studies are needed before probiotics can be rationally prescribed for the prevention or treatment of obesity. Control of the diet, environmental and lifestyle factors that promote the obesity development remain the best solution to problems related to weight gain.

The small sample size is the limitation of our analysis to be consider; however, the present study is one of the few to report the probiotic impact on weight loss in response to 3-month supplementation in a cohort of healthy, overweight and obese, free-living (without dietary or lifestyle restrictions) subjects. A subsequent and larger study is already planned and will be performed.

In conclusion, the potential probiotic strain *Lact. plantarum* IMC 510 showed lowering effects on body weight and other measures of subjects with obese tendencies, suggesting its beneficial influence on metabolic disorders.

Knowing that (i) the control of the diet and environmental and lifestyle factors that favour obesity development remain the best solution to problems related to weight gain and (ii) the dosage, duration of administration and long-term effects of the administration of the different strains, further studies are needed before probiotics can be rationally prescribed for the prevention or treatment of obesity.

CONFLICT OF INTEREST

No conflict of interest declared.

ORCID

Maria Magdalena Coman https://orcid.org/0000-0003-1594-4613
Maria Vittoria Micioni Di Bonaventura https://orcid.org/0000-0002-8044-1206
Maria Cristina Verdenelli https://orcid.org/0000-0001-5441-0361

REFERENCES

Ali Malik, G.M., Faheem, S. & Muhammad, F. (2022) Rise of eating disorders and obesity in young adults during the COVID-19 pandemic. *The Journal of the Pakistan Medical Association*, 72(2), 399.

Andreadou, I., Schulz, R., Badimon, L., Adameová, A., Kleinbongard, P., Lecour, S. et al. (2020) Hyperlipidaemia and cardioprotection: animal models for translational studies. *British Journal of Pharmacology*, 177, 5287–5311.

Avella, M.A., Olivotto, I., Silvi, S., Place, A.R. & Carnevali, O. (2010) Effect of dietary probiotics on clownfish: a molecular approach to define how lactic acid bacteria modulate development in a marine fish. *American Journal of Physiology. Regulatory, Integrative and Comparative Physiology*, 298, R359–R371.

Bouaziz, A., Dib, A.L., Lakhdara, N., Kadja, L., Espigares, E., Moreno, E. et al. (2021) Study of probiotic effects of *Bifidobacterium*

animalis subsp. lactis BB-12 and *Lactobacillus plantarum* 299v strains on biochemical and morphometric parameters of rabbits after obesity induction. *Biology*, 10, 131.

Brusaferro, A., Cozzali, R., Orabona, C., Biscarini, A., Farinelli, E., Cavalli, E. et al. (2018) Is it time to use probiotics to prevent or treat obesity? *Nutrients*, 10, 1613.

Cabello-Olmo, M., Oneca, M., Pajares, M.J., Jiménez, M., Ayo, J., Encío, I.J. et al. (2022) Antidiabetic effects of *Pediococcus acidilactici* pA1c on HFD-induced mice. *Nutrients*, 14, 692.

Cerdó, T., García-Santos, J.A., Bermúdez, M.G. & Campoy, C. (2019) The role of probiotics and prebiotics in the prevention and treatment of obesity. *Nutrients*, 11, E635.

Cheng, Y.-C. & Liu, J.-R. (2020) Effect of *Lactobacillus rhamnosus* GG on energy metabolism, leptin resistance, and gut microbiota in mice with diet-induced obesity. *Nutrients*, 12, 2557.

Coman, M.M. & Cresci, A. (2014) *Probiotics and prebiotics for human health: innovations and new trends*. Saarbrucken: Edizioni Accademiche Italiane.

Coman, M.M., Verdenelli, M.C. & Silvi, S. (2021) *ONE HEALTH: research, development and application of novel probiotics and prebiotics in human and animal health*. Wilington, DE: Generis Publishing.

Daniali, M., Nikfar, S. & Abdollahi, M. (2020) A brief overview on the use of probiotics to treat overweight and obese patients. *Expert Review of Endocrinology and Metabolism*, 15, 1–4.

Dupuy, H. (1984) The psychological general well-being (PGWB) index. In: Wenger, N.K., Mattson, M.E., Furberg, C.D., Elinson, J. (Eds.) *Assessment of quality of life in clinical trials of cardiovascular therapies*. Chap: Le Jacq Publishing, pp. 170–183.

Ejtahed, H.S., Angoorani, P., Soroush, A.R., Atlasi, R., Hasani-Ranjbar, S., Mortazavian, A.M. et al. (2019) Probiotics supplementation for the obesity management; a systematic review of animal studies and clinical trials. *Journal of Functional Foods*, 52, 228–242.

Fan, J. & Watanabe, T. (2000) Cholesterol-fed and transgenic rabbit models for the study of atherosclerosis. *Journal of Atherosclerosis and Thrombosis*, 7, 26–32.

FAO/WHO, F.A.O. (2002) *WHO working group report on drafting guidelines for the evaluation of probiotics in food*. London: Ontario, Canada, p. 30.

Fox, C.S., Massaro, J.M., Hoffmann, U., Pou, K.M., Maurovich-Horvat, P., Liu, C.-Y. et al. (2007) Abdominal visceral and subcutaneous adipose tissue compartments: association with metabolic risk factors in the Framingham heart study. *Circulation*, 116, 39–48.

Giudetti, A.M., Micioni Di Bonaventura, M.V., Ferramosca, A., Longo, S., Micioni Di Bonaventura, E., Friuli, M. et al. (2020) Brief daily access to cafeteria-style diet impairs hepatic metabolism even in the absence of excessive body weight gain in rats. *The FASEB Journal*, 34, 9358–9371.

Guazzelli Marques, C., de Piano Ganen, A., Zaccaro de Barros, A., Thomatieli dos Santos, R.V. & dos Santos Quaresma, M.V.L. (2020) Weight loss probiotic supplementation effect in overweight and obesity subjects: a review. *Clinical Nutrition*, 39, 694–704.

John, G.K. & Mullin, G.E. (2016) The gut microbiome and obesity. *Current Oncology Reports*, 18, 45.

Jones, M.L., Martoni, C.J. & Prakash, S. (2012) Cholesterol lowering and inhibition of sterol absorption by *lactobacillus reuteri* NCIMB 30242: a randomized controlled trial. *European Journal of Clinical Nutrition*, 66, 1234–1241.

Kadja, L., Dib, A.L., Lakhdara, N., Bouaziz, A., Espigares, E. & Gagaoua, M. (2021) Influence of three probiotics strains, *lactobacillus rhamnosus* GG, *Bifidobacterium animalis* subsp. *lactis* BB-12 and *Saccharomyces boulardii* CNCM I-745 on the biochemical and Haematological profiles and body weight of healthy rabbits. *Biology*, 10, 1194.

Kadooka, Y., Sato, M., Imaizumi, K., Ogawa, A., Ikuyama, K., Akai, Y. et al. (2010) Regulation of abdominal adiposity by probiotics (*Lactobacillus gasseri* SBT2055) in adults with obese tendencies in a randomized controlled trial. *European Journal of Clinical Nutrition*, 64, 636–643.

Khalesi, S., Bellissimo, N., Vandelanotte, C., Williams, S., Stanley, D. & Irwin, C. (2019) A review of probiotic supplementation in healthy adults: helpful or hype? *European Journal of Clinical Nutrition*, 73, 24–37.

Khalili, L., Eslamnezhad, K., Barzegar, A., Dehghani, A., Zakeri, N. & Mehrabi, E. (2022) Role of probiotics in diabetes control. In: Sayyed, R.Z. & Khan, M. (Eds.) *Microbiome-gut-brain Axis: implications on health*. Singapore: Springer Nature, pp. 337–353.

Kocsis, T., Molnár, B., Németh, D., Hegyi, P., Szakács, Z., Bálint, A. et al. (2020) Probiotics have beneficial metabolic effects in patients with type 2 diabetes mellitus: a meta-analysis of randomized clinical trials. *Scientific Reports*, 10, 11787.

Lewis, S.J. & Heaton, K.W. (1997) Stool form scale as a useful guide to intestinal transit time. *Scandinavian Journal of Gastroenterology*, 32, 920–924.

Mannucci, E., Ricca, V., Barciulli, E., Di Bernardo, M., Travaglini, R., Cabras, P.L. et al. (1999) Quality of life and overweight: the obesity related well-being (Orwell 97) questionnaire. *Addictive Behaviors*, 24, 345–357.

Marques, A.M., Sarandy, M.M., Novaes, R.D., Gonçalves, R.V. & Freitas, M.B. (2020) Preclinical relevance of probiotics in type 2 diabetes: a systematic review. *International Journal of Experimental Pathology*, 101, 68–79.

Martinelli, I., Micioni Di Bonaventura, M.V., Moruzzi, M., Amantini, C., Maggi, F., Gabrielli, M.G. et al. (2020) Effects of *Prunus cerasus* L. seeds and juice on liver steatosis in an animal model of diet-induced obesity. *Nutrients*, 12, 1308.

Michael, D.R., Jack, A.A., Masetti, G., Davies, T.S., Loxley, K.E., Kerry-Smith, J. et al. (2020) A randomised controlled study shows supplementation of overweight and obese adults with lactobacilli and bifidobacteria reduces bodyweight and improves well-being. *Scientific Reports*, 10, 4183.

Micioni Di Bonaventura, M.V., Cecchini, C., Vila-Donat, P., Caprioli, G., Cifani, C., Coman, M.M. et al. (2017) Evaluation of the hypocholesterolemic effect and prebiotic activity of a lentil (*Lens culinaris* Medik) extract. *Molecular Nutrition & Food Research*, 61, 1700403.

Micioni Di Bonaventura, M.V., Coman, M.M., Tomassoni, D., Micioni Di Bonaventura, E., Botticelli, L., Gabrielli, M.G. et al. (2021) Supplementation with *Lactiplantibacillus plantarum* IMC 510 modifies microbiota composition and prevents body weight gain induced by cafeteria diet in rats. *International Journal of Molecular Sciences*, 22, 11171.

Micioni Di Bonaventura, M.V., Martinelli, I., Moruzzi, M., Micioni Di Bonaventura, E., Giusepponi, M.E., Polidori, C. et al. (2020) Brain alterations in high fat diet induced obesity: effects of tart cherry seeds and juice. *Nutrients*, 12, 623.

Nasuti, C., Coman, M.M., Olek, R.A., Fiorini, D., Verdenelli, M.C., Cecchini, C. et al. (2016) Changes on fecal microbiota in rats exposed to permethrin during postnatal development. *Environmental Science and Pollution Research*, 23, 10930–10937.

O'Neal, R.M. & Still, W.J. (1962) Pathogenesis of atherosclerosis. *Federation Proceedings*, 21(4)Pt 2, 12–14.

Ooi, L.G. & Liong, M.T. (2010) Cholesterol-lowering effects of probiotics and prebiotics: a review of in vivo and in vitro findings. *International Journal of Molecular Science*, 11, 2499–2522.

Reamtong, O., Thiangtrongjit, T., Kosoltanapiwat, N., Panbangred, W. & Prangthip, P. (2021) Potential benefits of *L. acidophilus* in dyslipidemic rats. *Scientific Reports*, 11, 6115.

Rodgers, R.F., Lombardo, C., Cerolini, S., Franko, D.L., Omori, M., Fuller-Tyszkiewicz, M. et al. (2020) The impact of the COVID-19 pandemic on eating disorder risk and symptoms. *The International Journal of Eating Disorders*, 53(7), 1166–1170.

Schneider, J., Pegram, G., Gibson, B., Talamonti, D., Tinoco, A., Craddock, N. et al. (2022) A mixed-studies systematic review of the experiences of body image, disordered eating, and eating disorders during the COVID-19 pandemic. *The International Journal of Eating Disorders*. https://doi.org/10.1002/eat.23706

Schütz, F., Figueiredo-Braga, M., Barata, P. & Cruz-Martins, N. (2021) Obesity and gut microbiome: review of potential role of probiotics. *Porto Biomedical Journal*, 6, e111.

Sideli, L., Lo Coco, G., Bonfanti, R.C., Borsarini, B., Fortunato, L., Sechi, C. et al. (2021) Effects of COVID-19 lockdown on eating disorders and obesity: a systematic review and meta-analysis. *European Eating Disorders Review*, 29(6), 826–841.

Silvi, S., Verdenelli, M.C., Cecchini, C., Coman, M.M., Bernabei, M.S., Rosati, J. et al. (2014) Probiotic-enriched foods and dietary supplement containing SYNBIO positively affects bowel habits in healthy adults: an assessment using standard statistical analysis and support vector machines. *International Journal of Food Science and Nutrition*, 65, 994–1002.

Silvi, S., Verdenelli, M.C., Orpianesi, C. & Cresci, A. (2003) EU project Crownalife: functional foods, gut microflora and healthy ageing isolation and identification of *lactobacillus* and *Bifidobacterium* strains from faecal samples of elderly subjects for a possible probiotic use in functional foods. *Journal of Food Engineering*, 6, 195–200.

Urhan, M. & Okut Aysin, E. (2022) Nutritional and health behaviour predictors of the weight gain during the COVID-19 pandemic. *European Journal of Nutrition*. https://doi.org/10.1007/s00394-022-02862-7

Vallianou, N., Stratigou, T., Christodoulatos, G.S., Tsigalou, C. & Dalamaga, M. (2020) Probiotics, prebiotics, Synbiotics, postbiotics, and obesity: current evidence, controversies, and perspectives. *Current Obesity Reports*, 9, 179–192.

Verdenelli, M.C., Ghelfi, F., Silvi, S., Orpianesi, C., Cecchini, C. & Cresci, A. (2009) Probiotic properties of *lactobacillus rhamnosus* and *lactobacillus paracasei* isolated from human faeces. *European Journal of Nutrition*, 48, 355–363.

Wang, J., Zhang, H., Chen, X., Chen, Y., Menghebilige & Bao, Q. (2012) Selection of potential probiotic lactobacilli for cholesterol-lowering properties and their effect on cholesterol metabolism in rats fed a high-lipid diet. *Journal of Dairy Science*, 95, 1645–1654. https://doi.org/10.3168/jds.2011-4768

Wang, L., Guo, M.J., Gao, Q., Yang, J.F., Yang, L., Pang, X.L. et al. (2018) The effects of probiotics on total cholesterol. *Medicine (Baltimore)*, 97, e9679.

Waqar, A.B., Koike, T., Yu, Y., Inoue, T., Aoki, T., Liu, E. et al. (2010) High-fat diet without excess calories induces metabolic disorders and enhances atherosclerosis in rabbits. *Atherosclerosis*, 213, 148–155. https://doi.org/10.1016/j.atherosclerosis.2010.07.051

World Health Organization. (2021) *Obesity and overweight* [WWW document]. https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight

World Obesity Atlas 2022. (2022) World Obesity Federation [WWW document]. https://www.worldobesity.org/resources/resource-library/world-obesity-atlas-2022

Wu, Y., Zhang, Q., Ren, Y. & Ruan, Z. (2017) Effect of probiotic *Lactobacillus* on lipid profile: a systematic review and meta-analysis of randomized, controlled trials. *PLoS One*, 12, e0178868. https://doi.org/10.1371/journal.pone.0178868

Zhang, C., jiang, J., Wang, C., Li, S., Yu, L., Tian, F. et al. (2022) Meta-analysis of randomized controlled trials of the effects of probiotics on type 2 diabetes in adults. *Clinical Nutrition*, 41, 365–373.

Zhu, H., Qu, F. & Zhu, L.H. (1993) Isolation of genomic DNAs from plants, fungi and bacteria using benzyl chloride. *Nucleic Acids Research*, 21, 5279–5280.

How to cite this article: Coman, M.M., Miorelli, L., Micioni Di Bonaventura, M.V., Cifani, C., Salvesi, C. & Amedei, A. et al. (2022) Effects of probiotic *Lactiplantibacillus plantarum* IMC 510 supplementation on metabolic factors in otherwise healthy overweight and obese individuals. *Journal of Applied Microbiology*, 00, 1–13. Available from: https://doi.org/10.1111/jam.15703