



## Review

# Overview of Probiotic Strains of *Weizmannia coagulans*, Previously Known as *Bacillus coagulans*, as Food Supplements and Their Use in Human Health

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**Abstract:** *Weizmannia coagulans*, previously known as *Bacillus coagulans* and before that as *Lactobacillus sporogenes*, is a spore-forming, lactic acid-producing, Gram-positive, bacillus-shaped bacterial species with several known probiotic strains, including GBI-30, 6086 Unique IS-2, MTCC 5856, LBSC (DSM 17654), TBC169, SNZ 1969, BC30, and T11. This review focusses on the health benefit of these strains. A total of 53 clinical trials were found to use various strains of *Weizmannia coagulans*. However, 19 of these clinical trials did not provide strain information. Clinical evidence has shown that supplementation with strains of *Weizmannia coagulans* resulted in statistically significant health effects in the probiotic groups compared to the placebo. Several health benefits of the *Weizmannia coagulans* strains were found including relieving symptoms of irritable bowel syndrome, constipation, diarrhoea, and other gastrointestinal symptoms, function recovery treatment of non-fatty liver disease, after surgery or in patients with rheumatoid arthritis, quality of life and glucose- and lipid-related biomarkers related to overweight or obese participants or diabetic patients, absorption of protein or muscle integrity and improvement of peri- and post-menopausal symptoms. The main mechanism of action is the modulation of the intestinal microbiota and host immunity. However, in terms of several clinical studies involving small patient populations, others did not provide strain information. Larger, well-designed clinical studies are warranted to support the health benefits of *Weizmannia coagulans* strains.

**Keywords:** spore-forming bacteria; probiotics; *Bacillus coagulans*; *Weizmannia coagulans*



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## 1. Introduction

A special group of beneficial microbes is probiotics, which are by definition ‘live microorganisms that, when administered in adequate amounts, confer a beneficial effect on the host’ [1]. The most common probiotics are strains of the following genera: *Lactis-eibacillus*, *Limosilactobacillus*, *Lactobacillus*, *Lactiplantibacillus*, *Latilactobacillus*, *Levilactobacillus*, *Limosilactobacillus*, *Bifidobacterium*, *Lactococcus*, *Leuconostoc*, *Enterococcus*, *Pediococcus*, *Streptococcus*, *Bacillus*, *Clostridium*, *Propionibacterium*, *Escherichia*, and *Saccharomyces* [2]. For a probiotic strain to confer a beneficial effect on the host, it must have certain characteristics including survival at relevant body sites, such as adhesion to mucus or intestinal epithelial cells, interaction with human immune cells, resistance to digestive enzymes, bile or acid, antibacterial activity via competitive exclusion or production of bacteriocins or hydrogen peroxide [3].

Spore-forming bacteria are a wide group of bacteria that contain many pathogens and also beneficial bacteria and belong to the phyla Bacillota (previously known as Firmicutes) [4]. The most common genera are *Bacillus* and *Clostridium*; both genera have been recently divided and renamed [5,6], with some new names of genera including *Weizmannia*,

*Geobacillus*, *Alkalihalobacillus*, *Priestia*, and *Clostridioides*. Endospores or shortly spores are robust, metabolically inert, and incredibly resilient structures that are resistant to environmental stresses, such as UV, desiccation, heat, many disinfectants, high temperatures, and antibiotics [7]. The robust properties of the spore are due to its multi-layered structure. The dense core in *Clostridioides difficile* (previously *Clostridium difficile*) contains the DNA that is surrounded by small proteins. Around this core is a thin layer of peptidoglycan with the same composition as in vegetative cells. This layer is surrounded by the cortex, which contains a much thicker layer of peptidoglycan and is followed by another membrane, derived from the mother cell and a lamellar coat that contain highly crosslinked proteins. The final outmost coat or exosporium of *Clostridioides difficile* contains an amorphous structure. Other spore formers also contain similar structures and a more hexameric organization of this outer membrane [7–10]. Once the environmental conditions improve, the spore germinates back to vegetative cells. Germination is initiated when chemical signals indicate the spore is in an environment that is conducive to vegetative cell survival and growth [7]. Due to their resistance to various conditions, many aerobic sporogenic bacteria are ubiquitous in the environment.

Several spore-forming bacteria are pathogens or opportunistic pathogens, including *Clostridioides difficile*, which is the most common cause of antibiotic-associated diarrhoea and pseudomembranous colitis [7], *Clostridium tetani* and *Clostridium botulinum* that cause tetanus and botulism, respectively [11], as well as *Bacillus anthracis* that causes anthrax [12], *Bacillus cereus* and *Clostridium perfringens* that cause food poisoning [13] and *Bacillus thuringiensis* that causes diarrhoeal illness [14].

On the other hand, some spore-forming bacteria are commensals. Some are important soil bacteria, plant and animal commensals that fix nitrogen, produce protective volatile organic compounds, and are used in bioremediation, such as *Bacillus subtilis*, *Bacillus thuringiensis*, *Bacillus amyloliquefaciens*, *Bacillus velezensis*, and *Bacillus pumilus* [15–21]. The thermophilic *Geobacillus stearothermophilus* (previously *Bacillus stearothermophilus*) is commonly used as a biological indicator of sterilization [22,23]. There are also some probiotic spore-forming strains such as *Weizmannia coagulans* (previously *Bacillus coagulans*) MTCC 5856 [24]; *Weizmannia coagulans* Unique IS-2 [25]; *Bacillus subtilis* BS50 [26]; *Bacillus subtilis* DE111 [27]; *Bacillus clausii* UBBC-07 [28]; *Clostridium butyricum* MIYAIRI [29]; and many others with health benefits, as confirmed by clinical trials.

As spores can survive the harsh conditions of the stomach with a very low pH due to gastric acid (hydrochloric acid) and bile acids (mainly derivatives of cholic acid) and, upon transit into the duodenum, begin to germinate back to vegetative cells that colonize and proliferate in the colon [7,30], they are often considered to have an important potential as a probiotic if a health benefit is found. A strain can be defined as a probiotic if it is supported by at least one positive human clinical trial with a statistically significant beneficial effect on health in the probiotic group compared to the placebo group.

In this review, we focussed on the health effects established from published randomised controlled clinical trials of the sporogenic probiotic strains of *Weizmannia coagulans*, as a comprehensive review of these strains has not been published and many health traits are strain specific.

## 2. Search Strategy

We used the search strategy: “probiotics” AND (*Bacillus coagulans* OR *Weizmannia coagulans* OR *Lactobacillus sporogenes*) in various databases (PubMed, ScienceDirect) and included placebo-controlled clinical trials, which investigated the health benefits of the sporogene probiotic. Clinical trials on animals, without the full text available or in languages other than English were excluded. A total of 53 clinical studies investigating the health benefit of the probiotic strains of *Bacillus coagulans* (*Weizmannia coagulans*) were found (up to 1 July 2023). Clinical studies without placebo control or strain information were not added in results in tables, but only in descriptive form in the results and the discussion.

### 3. Results

A total of 53 clinical studies on humans were found. Two cross-over studies [31,32], five studies that included various *Weizmannia coagulans* (previously *Bacillus coagulans*) strains as part of multi-strain probiotics [33–37] and twenty-five randomised controlled trials that included various *Weizmannia coagulans* (previously *Bacillus coagulans*) as single strain probiotics [24,25,38–60] are described in the results in tabular form. Then, 21 studies are described only descriptively; of these, 2 [61,62] were not placebo-controlled studies and 19 studies contained no strain information [63–81]. One study was excluded as it was in Italian [82]. The results of cross-over clinical trials and randomised controlled clinical trials that utilised *Weizmannia coagulans* (previously *Bacillus coagulans*) as single strains or part of multiple-strain probiotics are noted in separate tables.

Table 1 depicts two cross-over studies by Nyangale and co-authors [31,32]. The strain *Weizmannia coagulans* GBI-30, 608 was used in both cross-over clinical trials. The beneficial effect was connected to intestinal microbiota modulation. Five randomised controlled clinical trials [33–37] included various strains of *Weizmannia coagulans* (previously *Bacillus coagulans*) such as CGI314, Unique IS2, MY01, and SNZ 1969 as part of multi-strain probiotics. Safety and tolerance as well as the effect of examination stress on students, influence on functional dyspepsia symptoms, gastrointestinal discomfort, and overweight and obesity-related parameters were investigated and statistically significant health benefits were found in all clinical trials with multi-strain probiotics. These studies are described in Table 2.

**Table 1.** Characteristics of two cross-over clinical trials using probiotic strains of the species *Weizmannia coagulans* (previously *Bacillus coagulans*), listed in descending chronological order.

Reference	Investigated Aim	Intention-to-Treat Population	Probiotics */Dosages, Cross-over Design	Main Findings after Consumption of Probiotics with <i>Weizmannia coagulans</i> and Other Strains
Nyangale 2015 [31]	Improvement of immune function	42 older people recruited; 36 completed study.	<i>Weizmannia coagulans</i> GBI-30, 6086, $1 \times 10^9$ cfu/day; 1 capsule per day for 28 days, followed by a 21-day washout period before switching to the other treatment.	Increase in beneficial groups of bacteria in the human gut and increase in production of anti-inflammatory cytokines.
Nyangale 2014 [32]	Modulating the gut microbiota	6 adults, randomised into 2 groups, 3 in each group.	<i>Weizmannia coagulans</i> GBI-30, 6086, $1 \times 10^7$ cfu/day; 1 capsule per day for 28 days, followed by a 21-day washout period before switching to the other treatment.	Modulation of the faecal microbiota using in vitro batch culture fermenters.

\* The new nomenclature *Weizmannia coagulans* is used for *Bacillus coagulans* (previously *Lactobacillus sporogenes*) [5,83].

**Table 2.** Characteristics of five randomised, placebo-controlled clinical trials using multi-strain probiotics, including *Weizmannia coagulans* (previously *Bacillus coagulans*) strains, listed in descending chronological order.

Reference	Investigated Aim	Intention-to-Treat Population	Multistrain Probiotic * Dosages (with <i>Weizmannia coagulans</i> (Previously <i>Bacillus coagulans</i> ) Strains)	Main Findings after Consumption of Multistrain Probiotic with <i>Weizmannia coagulans</i> Strain
Rea 2023 [33]	Safety, tolerance, and impact	98 study participants, 4 groups; 3 treatment groups (N = 24, 24, 25) and placebo (N = 25).	Group A: <i>Alkalihalobacillus clausii</i> CSI08, $1 \times 10^9$ cfu/day; Group B: <i>Priestia megaterium</i> MIT411, $1 \times 10^9$ cfu/day; Group C: probiotic cocktail ( <i>Bacillus subtilis</i> DE111, <i>Priestia megaterium</i> MIT411, <i>Weizmannia coagulans</i> CGI314, and <i>Alkalihalobacillus clausii</i> CSI08), $1 \times 10^9$ cfu/day for 45 days.	Decrease in incidence of loose stools. No adverse effects or safety concerns or changes in physiological symptoms and microbiota determination.

Table 2. Cont.

Reference	Investigated Aim	Intention-to-Treat Population	Multistrain Probiotic * Dosages (with <i>Weizmannia coagulans</i> (Previously <i>Bacillus coagulans</i> ) Strains)	Main Findings after Consumption of Multistrain Probiotic with <i>Weizmannia coagulans</i> Strain
Venkataran 2021 [34]	Examination stress	80 students, 2 groups; 40 in probiotic group (with glutamine) and 40 in placebo group.	<i>Weizmannia coagulans</i> Unique IS2, <i>Lactocaseibacillus rhamnosus</i> UBLR58, <i>Bifidobacterium lactis</i> UBBLA70, <i>Lactiplantibacillus plantarum</i> UBLP40; <i>Bifidobacterium breve</i> UBBR01, <i>Bifidobacterium infantis</i> UBBI01, $2 \times 10^{10}$ cfu/day for 28 days.	Reduction of level of stress and fasting serum cortisol levels.
Wauters 2021 [35]	Functional dyspepsia	68 patients with functional dyspepsia, 2 groups; 32 in probiotic group (with glutamine) and 36 in placebo group.	<i>Weizmannia coagulans</i> MY01, <i>Bacillus subtilis</i> MY02 $5 \times 10^9$ cfu/day for 8 weeks.	Efficacious and safe in the treatment of functional dyspepsia. Beneficial immune and microbial changes.
Soman 2019 [36]	Undiagnosed gastrointestinal (GI) discomfort.	60 adults with GI discomfort, 2 groups, 30 in each group.	<i>Weizmannia coagulans</i> SNZ 1969, <i>Alkalihalobacillus clausii</i> SNZ 1971, <i>Bacillus subtilis</i> SNZ 1972, $2 \times 10^9$ cfu/day for 30 days.	Improvement of several symptoms of GI discomfort.
Sudha 2019 [37]	Overweight/obesity-related parameters	90 overweight/obese subjects, 2 groups, 45 in each group.	<i>Ligilactobacillus salivarius</i> UBLS-22, <i>Lactocaseibacillus casei</i> UBLC-42, <i>Lactiplantibacillus plantarum</i> , UBLP-40, <i>Lactobacillus acidophilus</i> UBLA-34, <i>Bifidobacterium breve</i> UBBR-01, <i>Bacillus coagulans</i> Unique IS2 $6 \times 10^{10}$ cfu/day for 12 weeks.	Improvement of quality of life of overweight/obese participants.

\* New nomenclature of bacilli [5] and lactobacilli [84] is used, where relevant.

Twenty-five randomised controlled clinical trials [24,25,38–60] used single-strain *Weizmannia coagulans* probiotics. The most used strain (9 clinical trials) was GBI-30, 6086 [45,46,48,54,56–60], followed by 5 clinical trials with the strain Unique IS-2 [25,41,47,51,53], 5 clinical trials with MTCC 5856 [24,38,39,44,55], and 2 clinical trials with LBSC (DSM 17654) [42,52]. The remaining four clinical trials were each conducted with a different strain, namely TBC169 [40], SNZ 1969 [43], BC30 [49], and T11 (IBRC-M10791) [50]. The investigated beneficial effects of the probiotic strains included relieving symptoms of irritable bowel syndrome [42,46,51,53,55,58,59], constipation, diarrhoea, and other gastrointestinal symptoms [24,25,43,47,48,52,56,60], modulation of gut microbiota [38], function recovery treatment of non-fatty liver disease [45], after surgery [40] or in patients with rheumatoid arthritis [57], quality of life and glucose- and lipid-related biomarkers related to overweight or obese participants [44,49] or diabetic patients [50], absorption of protein or muscle integrity [41,54], and the skin anti-aging effect [39]. All clinical studies found at least one health benefit. Table 3 depicts detailed data of these 25 randomised controlled clinical trials that utilised single-strain *Weizmannia coagulans* probiotics.

**Table 3.** Characteristics of twenty-five randomised, placebo-controlled clinical trials using only a single strain probiotic of the species *Weizmannia coagulans* (previously *Bacillus coagulans*), listed in descending chronological order.

Reference	Investigated Aim	Intention-to-Treat Population	Strain/Dosage of Single Probiotic	Main Findings after Consumption of <i>Weizmannia coagulans</i> (Previously <i>Bacillus coagulans</i> ) Strain *
Majeed 2023a [38]	Modulation of healthy gut microbiota	30 healthy participants, 2 groups, 15 in each group.	MTCC 5856 $2 \times 10^9$ spores/day for 28 days.	Minor changes in few bacterial species in the intestinal microbiota of healthy individuals.

Table 3. Cont.

Reference	Investigated Aim	Intention-to-Treat Population	Strain/Dosage of Single Probiotic	Main Findings after Consumption of <i>Weizmannia coagulans</i> (Previously <i>Bacillus coagulans</i> ) Strain *
Majeed 2023b [39]	Anti-skin aging	56 female adults, 2 groups, 28 in each group.	MTCC 5856 $2 \times 10^9$ spores/day for 10 weeks.	Reduction of visibility of wrinkles and fine lines and improvement in skin elasticity.
Majeed 2023c [24]	Functional gas and bloating.	70 adults with gastrointestinal symptoms, 2 groups; 35 in each group.	MTCC 5856 $2 \times 10^9$ spores/day for 4 weeks.	Reduction of gastrointestinal symptoms in adults with abdominal gas and distension.
Venkataraman 2023 [25]	Functional constipation.	150 adults with functional constipation, 3 groups; 50 in probiotic with lactulose group, 50 in lactulose, 50 in placebo group.	Unique IS2 $2 \times 10^9$ spores/day for 4 weeks.	Relief of symptoms of constipation in a shorter period.
Li 2022 [40]	Function recovery after gynaecological laparoscopic surgery.	130 women undergoing gynaecological laparoscopic surgery, 3 groups; 44 in high-dose probiotic group, 43 in conventional dose probiotic group, and 43 in placebo group.	TBC169 $1.75 \times 10^8$ or $1.05 \times 10^8$ cfu/day until first flatus.	Intestinal function recovery and reduction of postoperative adverse events in patients after gynaecological laparoscopic surgery.
Tarik 2022 [41]	Absorption and utilisation of protein during training.	70 resistance-trained males, 2 groups; 35 in probiotic group (with whey protein) and 35 in placebo group.	Unique IS-2 $5 \times 10^9$ cfu/day for 60 days.	Increase in absorption of branched-chain amino acids and improvement of muscle power in the lower body.
Gupta 2021 [42]	Irritable bowelsyndrome (IBS).	40 adults with diagnosed IBS, 2 groups, 20 in each group.	LBSC (DSM 17654) $6 \times 10^9$ sores/day for 80 days.	Effective in alleviation of overall symptoms of IBS.
Kang 2021 [43]	Mild intermittent constipation.	80 healthy subjects with mild intermittent constipation, 2 groups, 40 in each placebo group.	SNZ 1969 $1.0 \times 10^9$ for 8 weeks.	Amelioration of intestinal motility and gut microbiota composition in adults with mild constipation.
Kazzi 2021 [44]	Clinical outcome after laparoscopic sleeve gastrectomy (LSG).	60 obese patients undergoing LSG, 2 groups; 30 in probiotic group (with galactomannans) and 30 in placebo group.	MTCC 5856 $4.5 \times 10^9$ cfu/day for 3 months post surgery.	Improvement of clinical outcomes of morbidly obese patients undergoing LSG (triglycerides, LDL, weight loss and AST).
Abhari 2020a [45]	Non-alcoholic fatty liver disease (NAFLD).	53 patients with NAFLD, 2 groups; 27 in probiotic group (with inulin) and 27 in placebo group.	GBI-30 $10^9$ spores/day for 12 weeks.	Effective for treatment of NAFLD and its related inflammation without any significant effects on related cardiovascular risk factors.
Abhari 2020b [46]	Irritable bowelsyndrome (IBS).	50 IBS patients, 2 groups, 25 in each group	GBI-30, 6086 $10^9$ spores/day for 8 weeks.	Reduction of severity score of IBS symptoms.
Madempudi 2020 [47]	Functional constipation.	100 adults diagnosed with functional constipation, 2 groups, 50 in each placebo group.	Unique IS-2 $5 \times 10^9$ cfu/day for 4 weeks.	Decrease in symptoms of constipation.
Anaya-Loyal 2019 [48]	Upper respiratory tract infections (URTI), gastrointestinal tract infections.	80 healthy school-aged children, 2 groups, 30 in each group.	GBI-30, 6086 $10^9$ cfu/day for 3 months.	Reduction of URTI symptoms (nasal congestion, bloody nasal mucus, itchy nose, hoarseness) and gastrointestinal tract infections (GITI) symptoms (flatulence rate).
Angelino 2019 [49]	Glucose- and lipid-related biomarkers.	46 healthy sedentary overweight and obese participants, 2 groups; 23 in probiotic group (with barley $\beta$ -glucans) and 23 in placebo group.	BC30 $10^7$ cfu/day for 12 weeks.	Improvement of glycemia- and lipid-related markers and resistin in a subgroup of healthy obese or hyperglycemic volunteers.
Arani 2019 [50]	Diabetic nephropathy (DN).	60 patients with DN, 2 groups; 30 in probiotic group (with honey) and 30 in placebo group.	T11 (IBRC-M10791) $10^8$ cfu/g for 12 weeks.	Improvement of insulin metabolism, total cholesterol, HDL cholesterol, serum hs-CRP, and plasma MDA levels.
Madempudi 2019 [51]	Irritable bowelsyndrome (IBS).	136 adults with IBS, 2 groups, 68 in each group.	Unique IS-2 $2 \times 10^9$ cfu/day for 8 weeks.	Reduction of abdominal pain intensity and increasing bowel movements.



Table 3. Cont.

Reference	Investigated Aim	Intention-to-Treat Population	Strain/Dosage of Single Probiotic	Main Findings after Consumption of <i>Weizmannia coagulans</i> (Previously <i>Bacillus coagulans</i> ) Strain *
Maity 2019 [52]	Acute diarrhoea with abdominal discomfort.	60 adults, 2 groups, 30 in each group.	LBSC (DSM 17654) $2 \times 10^9$ cfu/day for 7 days.	Improvement of pathophysiological conditions related to acute diarrhoea and abdominal discomfort.
Sudha 2018 [53]	Irritable bowel syndrome (IBS).	141 children aged 4–12 years, 2 groups. 72 in probiotic group (chewable tablets) and 69 in placebo group.	Unique IS2 $2 \times 10^9$ cfu/day for 8 weeks.	Reduction of IBS symptoms (pain, abdominal discomfort, bloating) in children.
Gepner 2017 [54]	Muscle integrity and cytokine response during intense military training.	26 male soldiers, 3 groups; 9 in probiotic group with $\beta$ -hydroxy- $\beta$ -methyl-butyrate (HMB), 9 in HMB group, and 8 in placebo group.	GBI-30, 6086 $10^9$ cfu/day for 40 days.	Attenuation of inflammatory cytokine markers during highly intense military training.
Majeed 2016 [55]	Irritable bowelsyndrome (IBS).	36 diarrhoea-predominant IBS patients, 2 groups; 18 in each group.	MTCC 5856 $2 \times 10^9$ cfu/day for 90 days.	Management of diarrhoea-predominant IBS symptoms.
Yang 2014 [56]	Residual immune activation in chronic treated HIV infection	24 participants with HIV, 2 groups, 12 in each group.	GBI-30, 6086 $2 \times 10^9$ cfu/day for 90 days.	Improvement of gastrointestinal symptoms and increase in CD4 <sup>+</sup> T cells.
Mandel 2010 [57]	Functional capacities in patients with rheumatoid arthritis (RA).	45 adults with symptoms of RA, 2 groups; 23 in probiotic group and 22 in placebo group.	GBI-30, 6086 $2 \times 10^9$ cfu/day for 60 days.	Effective for patients suffering from RA. Reduction of pain, self-assessed disability, CRP. Improvement in participation in daily activities.
Dolin 2009 [58]	Irritable bowelsyndrome (IBS).	55 IBS patients, 2 groups; 28 in probiotic group and 27 in placebo group.	GBI-30, 6086 $2 \times 10^9$ cfu/day for 8 weeks.	Reduction of average number of bowel movements.
Hun 2009 [59]	Irritable bowelsyndrome (IBS).	50 subjects with diarrhoea-prominent IBS, 2 groups. Study was completed by 22 in probiotic group and 22 in placebo group. Initial data not shown.	GBI-30, 6086 $2 \times 10^9$ cfu/day for 8 weeks.	Relief of abdominal pain and bloating for patients with IBS.
Kalman 2009 [60]	Functional gastrointestinal (GI) symptoms.	61 adults, 2 groups, 30 in each group.	GBI-30, 6086 $2 \times 10^9$ cfu/day for 4 weeks.	Improvement of quality of life and reduction of GI symptoms in adults with post-prandial intestinal gas-related symptoms.

\* The new nomenclature *Weizmannia coagulans* is used for *Bacillus coagulans* (previously *Lactobacillus sporogenes*) [5,83].

Two of the 53 studies [61,62] were not placebo-controlled studies; however, a positive effect was found for an increased immune response to the viral challenge, where each participant's baseline results served as control [61] and utilisation of *B. coagulans* Unique IS-2 strain was efficient and safe to treat the patients with acute diarrhoea [62].

A total of 19 studies did not contain strain information [63–81]. The studies named the probiotic *Bacillus coagulans* or *Lactobacillus sporogenes*. A combination of undefined *B. coagulans* and *Clostridium butyricum* strains [63] or an *L. sporogenes* strain [68] were found to be effective as adjuvant therapy in *Helicobacter pylori* irradiation to reduce the burden of antibiotic resistance. Another clinical trial found that a *B. coagulans* strain together with prebiotics improved symptoms of irritable bowel syndrome [70]. Certain strains of *B. coagulans* slightly decreased salivary streptococci mutans counts [65,79]. Certain *B. coagulans* or *L. sporogenes* strains as part of synbiotic foods improved some metabolic parameters of patients with type 2 diabetes mellitus [66,67,69,72,73]. *L. sporogenes* strains alone or as part of synbiotics have also been investigated for women's health and reduced certain metabolic parameters in pregnant women [71], and improved certain peri- or post-menopausal symptoms [74,77,81]. An undefined *L. sporogenes* strain as part of synbiotics also improved constipation symptoms in children [75]. Supplementation

with an undefined *B. coagulans* strain appeared to be beneficial for maintaining power and short-term speed performance while attenuating the inflammatory response during intense training [64]. On the other hand, undefined *B. coagulans* strains had no therapeutic effect on acute diarrhoea in children [78], did not improve spontaneous bacterial peritonitis among consecutive cirrhotic patients [76], and were not effective in reducing the incidence of death or necrotizing enterocolitis in low-birth-weight infants [80].

#### 4. Discussion

*Weizmannia coagulans*, previously known as *Bacillus coagulans* and before that as *Lactobacillus sporogenes* or *Bacillus sporogenes* is a Gram-positive, spore-forming, lactic acid producing, rod-shaped, motile, catalase-positive and oxidase-negative bacterial species with several known probiotic strains, including GBI-30, 6086 Unique IS-2, MTCC 5856, LBSC (DSM 17654), TBC169, SNZ 1969, BC30 and T11, as shown in the clinical trials described in this review [24,25,31,32,38–82]. The species was isolated and described by Horowitz-Wlassowa and Nowtelnow in 1933. It is widely used in medicine and the food industry [83,85,86]. As it has characteristics of both the bacilli and lactobacilli group, the classification of *Weizmannia coagulans* has undergone several changes, from being part of the lactobacilli group, followed by the bacilli group, and recently it has been reclassified into the *Weizmannia* genus [5,83]. Most of the clinical trials found in our review utilised various single-strain *Weizmannia coagulans* probiotics (named also *Bacillus coagulans* or *Lactobacillus sporogenes*) [24,25,31,32,40–62,64–81], only a few studies investigated multiple strain probiotics with *Weizmannia coagulans* strains [33–37,63].

Randomised controlled trials (RCT) are the most common studies to evaluate health-care interventions. They consist of participants randomly allocated to either a treatment or intervention group or a control group. A control group allows for the most effective comparison of two groups that are initially similar. Randomisation prevents bias as it allows allocation blinding and statistical control. It is important that the design, conducting, and reporting are without selection, allocation, performance, attribution, or experimental bias. The reporting quality of an RCT should be transparent and clear, allowing readers to appropriately analyse and understand the study design and results that may change clinical practice [87–89]. In a crossover clinical trial, participants are randomly allocated to receive treatment one first, followed by a washout period, when no treatment is received and then allocated to the other treatment and vice versa. Removing patient variation in this way makes crossover trials potentially more efficient than similar-sized, parallel-group trials in which each subject is exposed to only one treatment. And the treatment effects can be estimated with greater precision given the same number of subjects. However, long washout periods to prevent carryover can cause the study to lose effect [90].

Several found clinical studies [42,46,51,53,55,58,59,70] investigated the effectiveness of strains of *Weizmannia coagulans* probiotics in relieving the symptoms of irritable bowel syndrome in adults and children and found that all investigated strains including DSM 17654, GBI-30, 6086, Unique IS-2 and MTCC 5856 were effective. Therefore, this seems to be one of the core benefits of all *Weizmannia coagulans* strains [1]. General gastrointestinal health and microbiota also seem to be a core benefit of supplementation with *Weizmannia coagulans* strains as constipation, diarrhoea, bloating, and other gastrointestinal symptoms were improved by the strains Unique IS-2, MTCC 5856, SNZ 1969, GBI-30, and 6086 [24,25,31,32,38,43,47,48,52,56,60,75].

Other investigated effects of supplementation with *Weizmannia coagulans* strains included function recovery treatment of non-fatty liver disease [45], after surgery [40], for maintaining power and short-term speed performance, while attenuating the inflammatory response during intense training [64], or in patients with rheumatoid arthritis [57]. *Weizmannia coagulans* strains (previously *B. coagulans* or *L. sporogenes*) together with prebiotics were also effective in improving glucose- and lipid-related biomarkers related to overweight or obese participants or people with type 2 diabetes mellitus some metabolic parameters after supplementation [44,49,50,66,67,69,71–73]. Even certain peri- or post-menopausal symp-

toms improved [74,77,81]. The main mechanisms of action were modulation of the gut microbiota and host immunity.

Multistrain probiotics were investigated in some clinical trials [33–37] with various strains of *Weizmannia coagulans* (previously *Bacillus coagulans*) including CGI314, Unique IS2, MY01 and SNZ 1969, and statistically significant health benefits were found in all clinical trials with multi-strain probiotics.

Nineteen studies did not contain information on strains [63–81]. One of the studies without strain information [63] investigated the influence of *Clostridium butyricum* and *Bacillus coagulans* and another study [76] investigated a multistrain probiotic without information on any of the strains. The rest of the studies without strain information utilised only *Bacillus coagulans* (or *Lactobacillus sporogenes*). The study by Hoffman and co-authors [64] also noted that an inactivated (or non-viable) probiotic was used. This is not in line with the beginning of the definition of probiotics, as they are ‘live microorganisms’ [1]. Conflicting information by the authors was also the concentration of the daily serving as  $10^9$  cfu, as colony-forming units are produced by multiplication and metabolic activity of live active and viable microbes. Although this study may have used spores of *Bacillus coagulans* or postbiotics, this was not clear from the information. Also, the method of the inactivation of microbes was not evident. Authors of clinical studies on probiotics need to be more accurate when describing their clinical trials as, although several benefits are core benefits due to shared mechanisms [1,91], the action of probiotics is often strain-specific, especially when more specific ailments are researched [3]. When authors do not accurately note the specific probiotic strain used in their clinical study, it is like generalising dog breeds and saying it does not matter which dog breed is used as all are efficient working dogs, when it is general knowledge that a German shepherd dog is probably a more efficient working dog than, for example, a pug.

Many sporogenic bacteria are also beneficial animal probiotics, including *Bacillus amyloliquefaciens* TOA5001, *Bacillus subtilis*, *Clostridium butyricum* [92–94] and others for broiler chickens; *Bacillus licheniformis* for egg-laying hens [95]; *Bacillus subtilis* strains for ducks [96] and turkeys [97]; *Bacillus toyonensis*, *Bacillus cereus* var. Toyoi and other *Bacillus*-strains for piglets [98–100]; and *Bacillus subtilis* natto and other *Bacillus* strains for dairy cows [101,102].

Although this review focussed on sporogenic probiotic strains of *Weizmannia coagulans* (previously *Bacillus coagulans* or *Lactobacillus sporogenes*), there are many effective probiotic strains with or without spores as supported by a vast number of robust, well-designed clinical studies. Even if the survival of viable probiotic strains on their passage into the intestine is difficult, there are many novel applications, such as the manufacturing of double-coated capsules, that enable the survival of probiotic strains to the far parts of intestine without spores, where they can colonize the intestine, produce organic acids, bacteriocins, and other metabolites and modulate the intestinal microbiota to achieve eubiosis and modulate host immunity by regulating the functions of mucosal immune cells and intestinal epithelial cells. However, it is important to emphasize that the quality of clinical studies can be improved and that more large well-designed clinical trials are needed to support the many health claims of probiotics.

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