MOTHERISK ROUNDS

Probiotic Safety in Pregnancy: A Systematic Review and Meta-analysis of Randomized Controlled Trials of *Lactobacillus, Bifidobacterium, and Saccharomyces spp.*

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Abstract

Our objective in this study was to review systematically the evidence for safety of *Lactobacillus, Bifidobacterium* and *Saccharomyces spp*. during pregnancy and to conduct a meta-analysis of randomized controlled trials (RCTs). Eleven databases were searched from inception to September 2007 for RCTs of probiotic use during pregnancy. Two independent reviewers searched databases. Random-effects models combined data. Eleven studies on *Lactobacillus* and/or *Bifidobacterium* examined 1505 patients for four outcomes with no data heterogeneity; no miscarriage data were reported. Five studies reported Caesarean section outcomes (OR 0.88; 95% CI 0.65 to 1.19). Six studies reported birth weight (weighted difference 45 g; 95% CI −181 to 271). Three studies reported gestational age (weighted difference 0.4 weeks; 95%CI −0.4 to 1.2). No malformations were reported in the probiotic group. No RCTs were available for Saccharomyces during pregnancy. *Lactobacillus* and *Bifidobacterium* had no effect on the incidence of Caesarean section, birth weight, or gestational age. The safety of Saccharomyces during pregnancy is unknown.

Résumé

Dans le cadre de cette étude, notre objectif était de procéder à l'analyse systématique des résultats soutenant l'innocuité de *Lactobacillus*, de *Bifidobacterium* et de *Saccharomyces spp.* au cours de la grossesse, et de mener une méta-analyse des essais comparatifs randomisés (ECR). Du début de l'étude jusqu'à septembre 2007, des recherches ont été menées dans 11 bases de données afin d'y repérer les ECR portant sur l'utilisation de probiotiques au cours de la grossesse. Deux évaluateurs indépendants ont procédé aux recherches dans les bases de données. Des modèles à effets aléatoires ont combiné les données. Onze études portant sur *Lactobacillus* et/ou *Bifidobacterium* ont examiné 1 505 patientes en fonction de quatre critères d'évaluation sans hétérogénéité des données; aucune donnée sur la fausse couche n'a été signalée. Cinq études ont signalé les issues de la césarienne (RC, 0,88; IC à 95 %, 0,65–1,19). Six études ont signalé le poids de naissance (différence pondérée : 45 g; IC à 95 % −181 à 271). Trois études ont signalé l'âge gestationnel (différence pondérée : 0,4 semaine; IC à 95 % −0,4 à 1,2). Aucune malformation n'a été signalée dans le groupe « probiotique ». Nous ne disposions d'aucun ECR portant sur *Saccharomyces* pendant la grossesse. *Lactobacillus* et *Bifidobacterium* n'exerçaient aucun effet sur l'incidence de la césarienne, le poids de naissance ou l'âge gestationnel. L'innocuité de *Saccharomyces* pendant la grossesse est inconnue.

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INTRODUCTION

The use of natural health products during pregnancy is quite common, the prevalence of herbal medicine use during pregnancy being between 7% and 55%.[1–4] A survey in the United States found that between 45% and 93% of midwives had prescribed some form of natural health product to women during their pregnancy.[5] A collection of 75 systematic reviews, published as a textbook on natural health products in pregnancy and lactation, concluded that evidence for their safety and efficacy during pregnancy and lactation was lacking.[6]

Probiotics, one group of natural health products, consist of live bacteria or non-pathogenic yeast that colonize the gastrointestinal tract and provide a health benefit to the host.[7]

Lactobacillus spp. and *Bifidobacterium spp.* were reported as the most commonly used probiotic strains.[8] *Saccharomyces spp.* have also been reported as being commonly used.[8]

Lactobacillus spp. refers to a group of lactic acid-producing, Gram-positive rods that are obligate and facultative anaerobes.[9] *Lactobacillus* species include *L. acidophilus, L. bulgaricus, L. casei rhamnosus, L. delbrueckii, L. fermentum, L. plantarum, L. reuteri, L. rhamnosus GG*, and *L. sporogenes.*[10] *Bifidobacterium spp.* are anaerobic, rod-shaped, Gram-positive bacteria that normally colonize the human colon.[11] They constitute a predominant part of the anaerobic flora of the human colon and are the predominant intestinal organisms of breast-fed infants.[11] *Bifidobacterium* species include *B. adolescentis, B. animalis, B. bifidum, B. breve, B. infantis, B. lactis, and B. longum.*[12] *Saccharomyces spp.* are a group of non-pathogenic probiotic yeast which includes *S. boulardii* and *S. cerevisiae.*[13,14]

Clinically, meta-analyses and systematic reviews of clinical trials have demonstrated a significant benefit of *Lactobacillus spp.*, *Bifidobacterium spp.*, and *Saccharomyces spp.* alone or in combination for the prevention of acute diarrhea,[15] treatment of Clostridium difficile-associated diarrhea,[16] prevention of traveller's diarrhea,[17] treatment of yeast vaginitis and bacterial vaginosis,[18] treatment of acute diarrhea in children,[13,19] and treatment of antibiotic-associated diarrhea in children.[20] Many studies varying in methodological quality have addressed the safe use and benefits of *Lactobacillus spp., Bifidobacterium spp. or Saccharomyces spp.* throughout pregnancy.[21–28] For example, LGG was shown to be effective in the prevention of atopic disease in newborns when administered to pregnant women[21–27] and in the prevention of preterm labour.[28]

There is an outstanding need to determine the safety of administering probiotics during pregnancy. A number of clinical indications for probiotic use, such as diarrhea, yeast vaginitis, and bacterial vaginosis, have mostly been assessed in non-pregnant women. If deemed to be of minimal risk, probiotics could be of therapeutic benefit to pregnant women for these clinical indications.

Our objective was to conduct a systematic review of the scientific literature for randomized controlled trials on the administration of *Lactobacillus spp.*, *Bifidobacterium spp.*, and *Saccharomyces spp.* during pregnancy and to conduct a meta-analysis of these trials to provide more precise estimates of risk.

ABBREVIATIONS

BV	bacterial vaginosis
CFU	colony forming unit
LGG	*L. rhamnosus GG*
RCT	randomized controlled trial
TGF-β	transforming growth factor β

METHODS

For this systematic review, we included RCTs that assessed the effects of administering one or more of the three probiotic groups, i.e., *Lactobacillus spp.*, *Bifidobacterium spp.* or *Saccharomyces spp.*, or placebo to pregnant women. The probiotics administered could have been given at any time during pregnancy, at any dose, in any form (i.e., capsule, tablet, liquid, food [yogourt]), for any condition (e.g., atopic disease, vaccine antibodies), and given for at least one week. We included only studies with data on birth weight, gestational age, or incidence of the following: Caesarean section, malformations (major or minor) or miscarriage. Non-English journal articles were included, but studies related to probiotic administration after birth or during lactation were excluded.

Institutional Review Board approval is not required for systematic reviews and/or meta-analyses at the authors' institution.

In keeping with the principles of evidence-based practice, we endeavoured to identify and analyze the relevant RCTs that provided information on the safety of *Lactobacillus spp.*, *Bifidobacterium spp.*, and *Saccharomyces spp.* during pregnancy. Two independent reviewers systematically searched the following databases from their inception to September 2007: MEDLINE (1966–2007), OLDMEDLINE (1950–65), Cumulative Index to Nursing and Allied Health Literature (CINAHL) (1982–2007), Cochrane Database of Systematic Reviews, Cochrane Central Register of Controlled Trials, Database of Abstracts of Reviews of Effectiveness (DARE), Allied and Complementary Medicine (AMED) (1985–2007), EMBASE (1980–2007), AltHealthWatch. To ensure that no RCTs were overlooked, the following additional databases were consulted: Complete German Commission E Monographs by the American Botanical Council, Natural Database, and Natural Standard. We also hand-searched the bibliographies of relevant studies and contacted experts in the field to identify RCTs that may remain unpublished.

The databases were searched using the following MeSH terms, "lactobacillus," "bifidobacterium," "bifidobacteria," "saccharomyces," and each of these terms with "pregnancy." After we obtained the peer-reviewed publications, wherever possible, two reviewers independently assessed eligibility on the basis of the full text papers. Third-party arbitration was used in the case of any disagreements regarding inclusion.

Table 1. Randomized controlled trials of probiotics in pregnancy

Author (year)	n	Intervention	Dosage (daily)	Results	Statistically significant	Quality score (%)	Evidence grade (Table 2)	Comments
Abrahamsson et al. (2007)[21]	232	Oil droplets of *L. reuteri* ATCC 55730 or placebo	1×10^8 CFU	Treated infants had less IgE-associated eczema at 2 years of age than those from the placebo group	Yes	87.1	1a	Very strong scientific evidence
Gueimonde et al. (2006)[37]	53	LGG or placebo	NA	Infants whose mothers received LGG showed a significantly higher occurrence of *B. breve* and lower occurrence of *B. adolescentis* than those from the placebo group	Yes	45.2	1b	Strong scientific evidence
Kalliomäki et al. (2001)[22]	159	LGG or placebo	1×10^{10} CFU	At 2 years of age, the frequency of atopic eczema in the LGG group was half that of the placebo group. At 4 years of age, the frequency of atopic disease remained lower in the LGG group versus placebo	Yes	74.2	1a	Very strong scientific evidence
Kaplas et al. (2007)[38]	30	LGG or placebo	1×10^9 CFU	Dietary counselling with probiotics resulted in higher concentrations of linoleic and dihomo-c-linolenic acids compared with dietary counselling and placebo or with placebo alone	NA	51.6	1b	Strong scientific evidence
Kukkonen et al. (2006)[39]	61	Probiotics and galacto-oligosaccharides, or a placebo	5×10^9 CFU or 2×10^8 CFU or 2×10^9 CFU	Probiotics may improve response to Hib immunization	Yes	83.9	1a	Very strong scientific evidence
Kukkonen et al. (2007)[24]	925	Probiotics and galacto-oligosaccharides, or a placebo	5×10^9 CFU or 2×10^8 CFU or 2×10^9 CFU	Probiotic treatment showed no effect on the incidence of all allergic diseases by age 2 years but significantly prevented eczema and especially atopic eczema	Yes	87.1	1a	Very strong scientific evidence
Nishijima et al. (2005)[28]	24	Fermented milk containing *L. johnsonii*	10^9 CFU per ml	Oral administrations of probiotics can restore vaginal flora in pregnant women	Yes	29.0	1b	Strong scientific evidence. Small sample size
Rautava et al. (2002)[26]	62	LGG or placebo	2×10^{10} CFU	LGG increases the immunoprotective potential of breast milk when administered during pregnancy and lactation	Yes	51.6	1a	Sub-group analysis of data from Kalliomäki et al. (2001) Included because Kalliomäki et al. (2001) did not contain data on Caesarean section

Table 2. Levels of evidence for harm[6]

Level	Evidence
1a	VERY STRONG SCIENTIFIC EVIDENCE
	Statistically significant evidence from one or more systematic reviews or RCTs.
1b	STRONG SCIENTIFIC EVIDENCE
	Statistically significant evidence from one or more outcome studies OR cohort studies OR case control studies.
1c	GOOD SCIENTIFIC EVIDENCE
	Evidence from one or more case series.
2	FAIR SCIENTIFIC EVIDENCE
	Evidence based on case reports.
3	*IN VITRO* SCIENTIFIC EVIDENCE
	Evidence based on scientific studies conducted on animals, insects or microorganisms OR laboratory studies on human cells.
4	INDIRECT EVIDENCE
	Evidence based on scientific theory OR expert opinion.
5	UNKNOWN
	No available information.

Two reviewers conducted data extraction independently and assessed the study quality of each publication using the checklist developed and validated by Downs and Black.[29] The checklist uses the following criteria to evaluate quality: reporting, external validity, internal validity (bias and confounding/selection bias), and power.[29] Reviewers were not blinded to authorship or journal name.

In cases where reviewer scores were within 10% of each other, reviewers discussed the differences in their quality evaluations and then established a consensus for the final scoring of the publication. In cases where the difference in reviewer scores exceeded 10%, third party arbitration was used to address the differences between their quality evaluations and then to establish a consensus for the final scoring of the publication. Once scored, each publication was referenced in our database and tabulated (Table 1). Additionally, studies were assigned an evidence level for harm (Table 2). The evidence levels used in Table 2 are standardized and described further in previous publications.[6,30–33]

When available, data on birth weight, gestational age and incidence of Caesarean section were extracted from the RCTs and tabulated. Data were combined using random effects meta-analytical models.[34] In the case of Caesarean section data, incidences were pooled across the strata of 2 × 2 tables, and the odds ratio was calculated using a random effects meta-analytic model.[34] In the case of birth weight and gestational age, the difference between treatment and placebo group means was calculated using a random effects model for continuous variables (i.e., weighted mean difference). For quality assurance purposes, data were examined using the chi-squared test[34] and the I^2 test for heterogeneity.[35] The statistical software used during this study was Review Manager (RevMan) Version 4.2 for Windows (Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration, 2003).

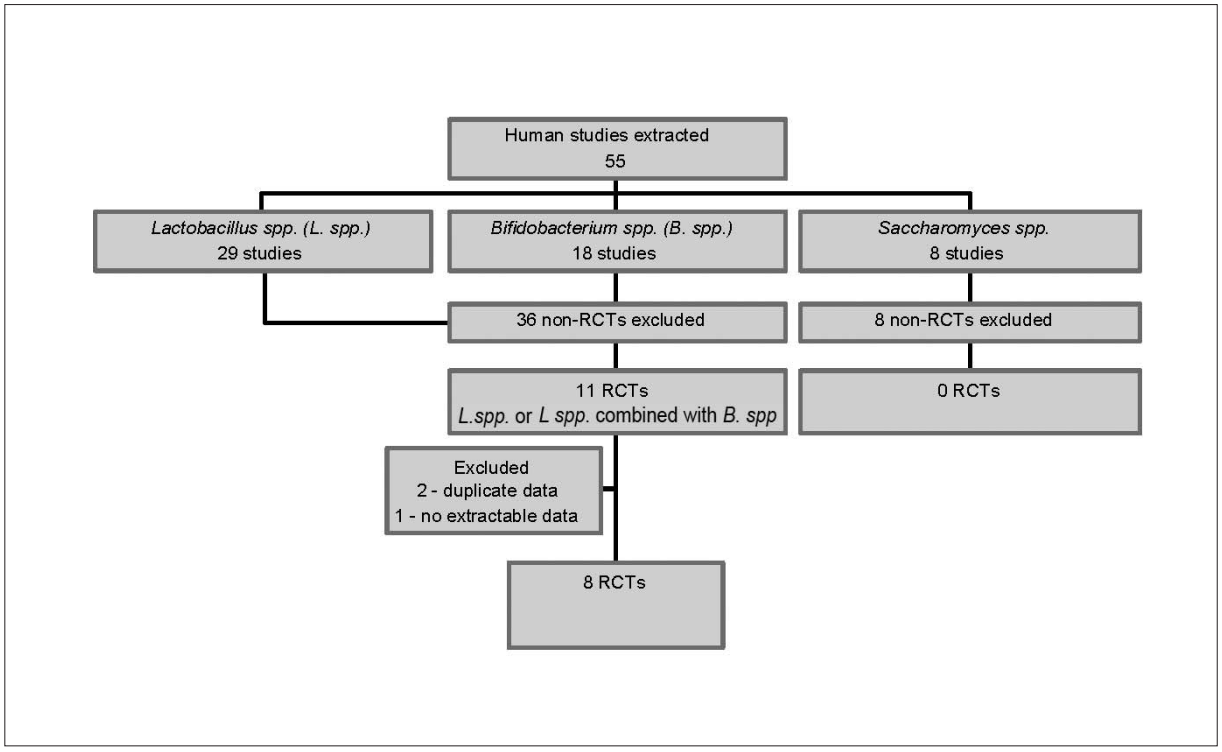
RESULTS

In total, 55 human studies were extracted from the scientific literature on probiotics during pregnancy, of which 11 were RCTs (Table 1 and Figure 1). Two studies were excluded (Rinne et al. 2005 and 2006) as they were sub-group analyses of the Kalliomäki et al. (2001) study. The Rautava et al. study[26] was also a sub-group analysis of the Kalliomäki et al. (2001) study[22]; however, it was included in the meta-analysis as it contained data on Caesarean section rates that were not presented in the Kalliomäki et al. (2001) study.[22] Lastly, the Neri et al.[36] study was excluded from meta-analysis as it did not contain any extractable data on pregnancy safety. No data on malformations and miscarriage incidence were available or suitable for meta-analysis.

Of the remaining eight RCTs included for meta-analysis, the probiotic intervention was of *Lactobacillus spp.* alone[21,22,26,28,37] or in combination with *Bifidobacterium spp.*[24,38,39] There were no RCTs on *Saccharomyces spp.* during pregnancy. A flow chart of extracted studies is presented in Figure 1.

Based on the reported Caesarean section outcomes, we found no evidence of publication bias using Begg-Mazumdar test (tau = 0.200, $P = 0.624$).[40] Nonetheless, we cannot completely rule out the possibility of

Figure 1. Probiotics in pregnancy flow chart



spp: species

publication bias because of the small number of studies (K = 5) in our search guidelines that reported the incidence of Caesarean section.

The RCTs are summarized in Table 1. The findings from the RCTs are discussed individually. Results of the meta-analysis are presented at the end of the Results section.

Systematic Review

Abrahamsson et al.[21] conducted a randomized controlled trial of 232 pregnant women with a family history of atopic disease.[21] The women received oil droplets of *L. reuteri* ATCC 55730 (1×10^8 CFU) or placebo daily from week 36 of gestation until expected delivery.[21] At delivery, 227 women delivered healthy infants with no reports of malformations (major or minor), low birth weight, or preterm delivery.[21] The infants were then directly administered *L. reuteri* or placebo for two years. At the end of the trial, the authors reported that infants treated with *L. reuteri* had significantly less IgE-associated eczema at two years of age and therefore may possibly have a reduced risk of developing later respiratory allergic disease.[21]

Gueimonde et al. conducted a randomized controlled trial in 53 pregnant women with a family history of atopic disease.[37] Pregnant women were administered an unreported daily dose of LGG or placebo from four weeks before the date of expected delivery until expected delivery.[37] The objective was to study the effects of LGG or placebo on mother–infant *Bifidobacteria* transfer at birth and the development of *Bifidobacteria* during the first weeks of life.[37] At delivery, 53 women delivered infants with no reports of malformations (major or minor), low birth weight, or preterm delivery.[37] At five days of age, infants whose mothers received LGG showed a significantly higher occurrence of *B. breve* and lower occurrence of *B. adolescentis* than those from the placebo group.[37] LGG intake increased the *Bifidobacterial* diversity in infants and reduced the *Bifidobacterium* species similarity between mother and infant.[37]

Kalliomäki et al.[22] conducted a randomized controlled trial in 159 pregnant women with a family history of atopic disease. The women received either two capsules daily of LGG (1×10^{10} CFU) or placebo from two to four weeks before the expected date of delivery until actual delivery.[22] At delivery, 155 women delivered healthy infants with no reports of malformations (major or minor), low birth weight, or preterm delivery.[22] After delivery, the infants received LGG or placebo directly for six months and were monitored to four years of age.[22,23] At two years of age, the frequency of atopic eczema in the LGG group was half that of the placebo group (RR = 0.51; 95% CI 0.32–0.84).[22] At four years of age, the frequency of atopic disease remained lower in

the LGG group than in the placebo group (14/53 vs. 25/54; RR = 0.57; 95% CI 0.33–0.97).[23]

Kaplas et al.[38] conducted a small randomized controlled trial in a group of pregnant women participating in a cohort study[41] on nutritional intake during pregnancy. At a mean of 13.8 ± 1.4 weeks' gestation, pregnant women were randomized to received two capsules daily of LGG and *B. lactis* (1×10^9 CFU per capsule) or placebo until the end of their pregnancy.[38] The groups were further divided into three: group 1 received the probiotic and dietary counselling, group 2 received the placebo and dietary counselling, and group 3 received the placebo.[38] The pregnancies were uncomplicated and the infants were delivered at term; there were no reports of malformations (major or minor), miscarriages, low birth weight, or preterm delivery.[38] Concentrations of linoleic (18:2n-6) and dihomo-c-linolenic acids (20:3n-6) in placental and umbilical cord samples taken at birth and up to 24 hours later were significantly higher ($P < 0.05$) in women given dietary counselling with probiotics than in women given dietary counselling and placebo or placebo alone.[38]

Kukkonen et al. (2006) [39] conducted a randomized controlled trial in a sub-group of 87 pregnant women already enrolled in the Kukkonen et al. (2007) [24] atopic disease prevention trial previously discussed. The purpose of this study was to observe the effects, if any, of probiotic intake on vaccine antibody response in newborns.[39] The pregnant women received the same dosing regimen pre- and post-pregnancy as discussed above in the Kukkonen et al. study.[24,39] Infants followed Finland's routine vaccination schedule to six months of age.[39] At six months of age, there was no difference in antibody responses to diphtheria, tetanus or *Haemophilus influenzae* type b vaccination between probiotic and placebo groups.[39]

Kukkonen et al. conducted a randomized controlled trial of 1223 pregnant woman with a family history of atopic disease.[24] At two to four weeks before expected delivery, pregnant woman received one capsule twice daily of either placebo or a probiotic combination containing four strains: *LGG* (5×10^9 CFU), *L. rhamnosus* LC705 (5×10^9 CFU), *B. breve* (2×10^8 CFU) and *Propionibacterium freudenreichii spp. Shermanii* (2×10^9 CFU).[24] At delivery, there were no reports of malformations (major or minor) in the probiotic group, but three reports in the placebo group (0/610 and 3/613, respectively).[24] There were 27 reports of preterm delivery in the probiotic group and 18 reports in the placebo group (27/610 and 18/613, respectively), and no reports of low birth weight.[24] After delivery, infants received either an opened capsule of the probiotics administered to their mother and 20 drops of sugar syrup (0.8 g of galacto-oligosaccharides) or an opened capsule of placebo and 20 drops of sugar syrup without galacto-oligosaccharides daily for the first six months of life.[24] Infants were monitored until two years of age.[24] At three and six months of age, fecal analysis indicated that *Lactobacillus spp.* and *Bifidobacterium spp.* more frequently colonized the guts of probiotic-supplemented infants,[24] At two years of age, probiotic treatment was associated with a significant reduction in eczema and atopic eczema in treated infants compared with the placebo group.[24]

Neri et al. conducted an open randomized trial of 64 women with bacterial vaginosis in their first trimester of pregnancy.[36] Pregnant women were divided into three groups: group 1 received a 10 to 15 mL vaginal douche (inserted with a syringe) of commercially available yogourt containing *L. acidophilus* ($> 1 \times 10^8$ CFU per mL) with a pH < 4.5, group 2 received a large vaginal tampon soaked with 10 to 15 mL of 5% acetic acid before insertion, and group 3 received no treatment (control).[36] Treatments were administered twice daily for seven days and then repeated one week later. The effect of the treatment was evaluated at four and eight weeks after completion of treatment by monitoring the presence or absence of BV criteria and patients' subjective feelings.[36] At four and eight weeks post treatment, the probiotic group had a significant absence of BV in comparison to both the acetic acid and control groups.[36] The authors concluded that the continuous correction of both the vaginal pH and *Lactobacillus spp.* flora was crucial for normal vaginal ecology, and was responsible for the high treatment response rate.[36]

Nishijima et al. conducted a randomized controlled trial in 24 pregnant women near full term (35 weeks of gestation).[28] Pregnant women received either 120g/day of fermented milk containing 1×10^9 CFU of *L. johnsonii* or a placebo fermented milk for two weeks.[28] Vaginal fluid samples were collected before and after administration of the fermented milk. In the probiotic group, pathogenic bacteria such as *Gardnerella vaginalis* and *Corynebacterium spp.* were detected in four of the 12 subjects, but were undetectable after two weeks of probiotic administration.[28] In the placebo group, pathogenic bacteria were detected in three of the 12 subjects and remained present after two weeks of placebo treatment.[28] The authors concluded that oral administration of probiotics can restore the vaginal flora in pregnant women.[28] Since this study was published as a letter to the editor and was very brief, the authors did not provide any information on the birth outcomes in either group or report any major or minor malformations.[28]

Rautava et al.[26] conducted a sub-group analysis on 62 mother-infant pairs receiving either LGG or placebo as part of the Kalliomaki et al.[22] study. Breast milk samples were analyzed for TGF-β concentration. Elevated levels of

TGF-β are believed to enhance the ability of infants to produce specific IgA antibodies against dietary antigens and therefore, prevent atopic disease in breastfed infants.[42] The amount of breast milk TGF-β in mothers receiving LGG was significantly superior to placebo, thereby suggesting that LGG increases the immunoprotective potential of breast milk when administered during pregnancy and lactation.[26]

Meta-analysis

The primary pregnancy outcomes were Caesarean section rate, birth weight, and gestational age. Only one study reported the incidence of malformations; no malformations were reported in the probiotic group, while three cases with malformations were reported in the placebo group.[24] In six of the eight RCTs, the probiotic interventions occurred at 32–34 weeks' gestation, so data on miscarriage and malformation incidences were unlikely. Two studies were conducted in the first trimester and could therefore have provided data on miscarriage incidence; however, the authors did not report any incidences of miscarriage.[36,38] The studies that contained data on Caesarean section, birth weight, and gestational age are presented in Tables 3, 4, and 5, respectively.

For the outcome of Caesarean section, the odds ratio (OR) was 0.88 (95% CI 0.65–1.19) (Figure 2). There was no evidence of heterogeneity in the data ($c^2 = 0.97$, $P = 0.915$; $I^2 = 0$, $P = 0.24$).

For the outcome of birth weight, the meta-analytic means were 3.609 kg (± 0.146 kg) in the probiotic group and 3.587 kg (± 0.122 kg) in the placebo group. Compared with placebo, there was a non-significant increase in birth weight of 45 g ($P = 0.699$) associated with taking probiotics during pregnancy. The data did not display heterogeneity of effects ($c^2 = 0.46$, $P = 0.994$; $I^2 = 0$, $P = 0.334$).

For the outcome of gestational age, the meta-analytic means were 39.8 weeks (± 0.4 weeks) in the probiotic group and 39.2 weeks (± 0.5 weeks) in the placebo group. Compared with placebo, there was a non-significant increase in gestational age of 0.4 weeks ($P = 0.336$) associated with taking probiotics during pregnancy. The data did not display heterogeneity of effects ($c^2 = 0.79$, $P = 0.673$; $I^2 = 0$, $P = 0.804$).

DISCUSSION

Of the studies systematically reviewed, the majority rated well according to the quality checklist by Downs and Black[29] (Table 1). Some methodological weaknesses were identified; for example, Gueimonde et al. did not report the dose of probiotics administered.[37]

Given the poor quality of evidence typically found when researching natural health products during pregnancy, as observed in previous publications,[6] the identification of 11 RCTs on use of probiotics during pregnancy is impressive. The studies were related to topics on atopic disease prevention, mother-to-infant microflora transfer, BV, placental transfer of fatty acids, and immunization.

Before a unifying statement can be made that probiotics are either safe or effective during pregnancy, it must be emphasized that probiotics constitute a group of microorganisms, and there may be differences between individual species. Three genera were reviewed in this study: *Lactobacillus, Bifidobacterium*, and *Saccharomyces*. The studies extracted in this review and meta-analysis relate only to the three specific probiotic strains studied. For example, since LGG was the most commonly studied *Lactobacillus sp*. intervention in pregnancy, it cannot be concluded that all *Lactobacillus spp*. will have the same clinical effect on atopic disease prevention or safety profile.

Another consideration when evaluating safety is the timing of the probiotic intervention during pregnancy. The majority of the RCTs were conducted between weeks 32 to 36 and delivery. A probiotic intervention at 32 to 36 weeks of gestation is unlikely to increase miscarriage risk or affect organogenesis. For example, it is difficult to conclude that the *L. reuteri* administered in the study of Abrahamsson et al.[21] had any effect on gestational age: the intervention commenced one week before the beginning of what is considered full-term, i.e., the intervention was at week 36, and the normal full term for pregnancy is between weeks 37 and 42. Only Kaplas et al.[38] and Neri et al.[36] conducted studies in which probiotics were administered in the first trimester of pregnancy. Despite there being no reports of miscarriages or malformations in these two studies, it is not possible to conclude that probiotics have no effect on miscarriage or malformations incidence until more RCTs are available for meta-analysis.

Of the 11 RCTs identified, only eight contained pregnancy outcome data for meta-analysis: Caesarean section rate, birth weight, or gestational age. [21,22,24,26,28,37–39]Administering LGG alone or in combination with *L. rhamnosus*, *B. breve and P. freudenreichii spp. Shermanii* did not affect the incidence of Caesarean section (OR = 0.88; 95% CI 0.65–1.19). With respect to birth weight and gestational age, only mean values, and not incidences of low birth weight or preterm infants, were available. Administering *L. reuteri* alone, LGG alone or in combination with *B. lactis, L. rhamnosus, B. breve* and *P. freudenreichii spp. Shermanii* did not affect birth weight. Administering LGG alone or in combination with *B. lactis* did not appear to have affected gestational age. Taking these probiotics appeared to have been related to a small and non-significant increase in birth weight of 45 g and a non-significant increase in gestational age of approximately

Table 3. Results from studies reporting on the relationship between use of probiotics and Caesarean section

	Probiotic group		Placebo group		
Studies	Caesarean section	Vaginal	Caesarean section	Vaginal	*P*
Abrahamsson et al. (2007)[21]	11	103	15	98	0.517
Gueimonde et al. (2006)[37]	3	26	4	20	0.787
Kukkonen et al. (2007)[24]	75	376	79	374	0.814
Kukkonen et al. (2006)[39]	5	42	6	34	0.776
Rautava et al. (2002)[26]	4	26	4	28	0.780
Overall	98	573	108	554	0.413

Table 4. Results from studies reporting on the relationship between use of probiotics and birth weight in kilograms

	Probiotic group			Placebo group			
Studies	Mean	SD	n	Mean	SD	n	*P*
Abrahamsson et al. (2007)[21]	3.66	0.42	114	3.60	0.47	113	0.312
Kalliomäki et al. (2001)[22]	3.63	0.48	77	3.61	0.47	82	0.791
Kaplas et al. (2007)[38]	3.80	0.14	10	3.71	0.16	20	0.142
Kukkonen et al. (2007)[24]	3.60	0.48	461	3.59	0.48	464	0.752
Kukkonen et al. (2006)[39]	3.58	0.42	47	3.57	0.47	40	0.917
Nishijima et al. (2005)[28]	2.80	0.39	12	2.97	0.34	12	0.267
Overall	3.51	0.39	721	3.51	0.4	731	1.0

SD: standard deviation

Table 5. Results from studies reporting on the relationship between use of probiotics and gestational age

	Probiotic group			Placebo group			
Studies	Mean	SD	n	Mean	SD	n	P
Kalliomäki et al. (2001)[22]	39.0	1.3	77	39.0	1.4	82	0.064
Kaplas et al. (2007)[38]	40.0	0.4	10	39.4	0.6	20	0.008
Nishijima et al. (2005)[28]	38.5	1.1	12	38.9	1.1	12	0.383
Overall	39.2	0.9	99	39.1	1.0	114	0.447

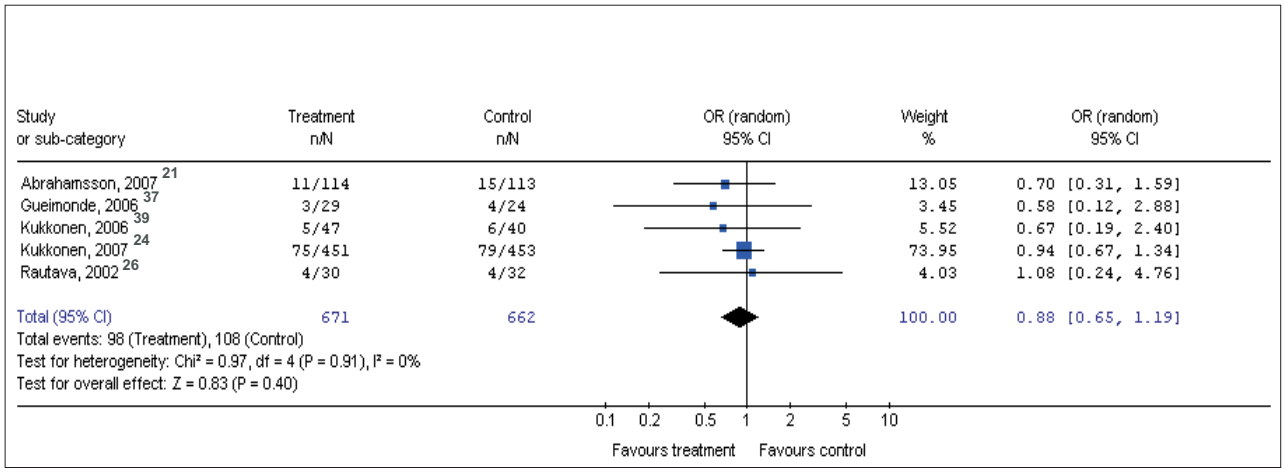
SD: standard deviation

three days (0.4 weeks). Even if these results were statistically significant, it is unlikely that these results would be judged as clinically relevant.

It could be argued that mean values of birth weight and gestational age were not ideal for evaluating pregnancy safety. The best basis for determining if a probiotic intervention is a risk factor for low birth weight or preterm delivery would have been the incidences of low birth weight infants and preterm deliveries in the probiotic and placebo groups. None of these data were available in the manuscripts extracted for meta-analysis. The corresponding authors were contacted to obtain these data, and, according to the authors who replied, the data were either not available or it was against the policy of their institutions to release any patient information (even anonymous information) outside their institution. We hope that these authors will publish their results in the future.

With respect to the results from the RCTs, there were a number of important clinical findings. LGG and *L. reuteri* appear to play a role in decreasing the incidence of atopic

Figure 2. Forest plot of Caesarean section outcomes for probiotic use during pregnancy



disease in infants up to four years of age. Since LGG and *L. reuteri* were also administered to the infants, it is not possible to conclude whether the observed prevention of atopic disease was due to the probiotics administered during pregnancy, those administered during infancy, or both. The most dramatic results with respect to atopic disease prevention were association with the administration of LGG. The less favourable results reported in the *L. reuteri* study may be due to methodological weaknesses in their study. Abrahamsson et al. used *L. reuteri* in a suspension of coconut (3/4) and peanut oil (1/4).[21] Although the authors reported that the peanut oil contained < 0.005% peanut protein and unknown amounts of coconut protein, it is plausible that administering known allergens and hyper-sensitizing agents such as peanuts and coconut may have exacerbated the atopic condition tested.[21] In fact, the intervention of *L. reuteri* would have to offset the familial susceptibility to atopic disease along with a repeated daily intake of an allergen and hyper-sensitizing agent. In the future, it would be more prudent for clinical studies of atopic disease to avoid using carrier solutions that are known allergens or hyper-sensitizing agents such as peanuts and coconut.

BV is a risk factor for prematurity and postpartum complications.[36] *L. acidophilus* administered as a yogourt suppository appears to be a safe and effective treatment of BV in pregnancy.[36] The continuous correction of both the vaginal pH and *Lactobacillus spp.* flora via the suppositories was believed by Neri et al. to be crucial for normal vaginal ecology and responsible for the high treatment rate.[41]

LGG appears to affect the mother–infant bifidobacteria transfer at birth and bifidobacteria development later in life. Gueimonde et al. reported that LGG given at four weeks pre-delivery decreased *B. adolescentis* and increased *B. breve*.[38] *B. adolescentis* is more frequently isolated from allergic infants than from healthy infants,[43,44] and *B. breve* may prevent atopic disease in infants.[45]

LGG and *B. lactis* may affect fatty acid transfer in the placenta, leading to higher placental concentrations of linoleic and dihomo-c-linolenic acids.[38] The administration of LGG, *L. rhamnosus, B. breve*, and *P. freudenreichii spp. Shermanii* to pregnant mothers did not affect antibody responses to diphtheria, tetanus, or *Haemophilus influenzae* type b vaccination.[39]

Lastly, the use of *Saccharomyces spp.* may be of concern during pregnancy because of reports about the implication of *S. boulardii* as an etiologic agent of invasive infection.[46] A comprehensive review of the scientific literature reported 92 cases of *Saccharomyces* invasive infection.[46] Clinically, *Saccharomyces* invasive infection was indistinguishable from an invasive candidiasis and had similar predisposing factors, such as intravascular catheter placement and antibiotic therapy.[46] The review also reported that treatment with intravenous amphotericin B and fluconazole were effective therapeutic options.[46] In our systematic review, no studies addressed *Saccharomyces spp.* as an intervention for pregnant women. Although the reported cases of *Saccharomyces* invasive infection were treatable, it is not prudent for *Saccharomyces spp.* to be administered to pregnant women, nor is it recommended.

CONCLUSIONS

The 11 studies of probiotic use in pregnancy that were reviewed were generally of good methodological quality and provided good evidence with respect to pregnancy. The meta-analysis did not indicate that administering certain strains of *Lactobacillus spp.* and *Bifidobacterium spp.* had any effect on Caesarean section rate, birth weight, or gestational age. Certain strains of *Lactobacillus sp.* and *Bifidobacterium sp.*

may prevent atopic disease in infants when administered to their mothers in pregnancy. *L. acidophilus* yogourt suppositories are a safe and effective treatment of BV in pregnancy. LGG and *B. lactis* increased fatty acid transfer to the placenta. LGG, *L. rhamnosus, B. breve*, and *P. freudenreichii ssp. Shermanii* administered to pregnant women did not affect antibody responses to diphtheria, tetanus or *Haemophilus influenzae* type b vaccination. At present, the safety of *Saccharomyces spp.* during pregnancy is unknown.

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