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Dietary Supplementation with Lactobacilli and Bifidobacteria Is Well Tolerated and Not Associated with Adverse Events during Late Pregnancy and Early Infancy[1–3]

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Abstract

Lactic acid bacteria and bifidobacteria are increasingly being administered to pregnant women and infants with the intention of improving health. Although these organisms have a long record of safe use, it is important to identify any adverse effects in potentially vulnerable populations. In a randomized, double-blinded, placebo-controlled trial, we evaluated the safety of a bacterial dietary supplement for the prevention of atopy in infants. Two strains of lactobacilli (*Lactobacillus salivarius* CUL61 and *Lactobacillus paracasei* CUL08) and bifidobacteria (*Bifidobacterium animalis subsp. lactis* CUL34 and *Bifidobacterium bifidum* CUL20) with a total of 1×10^{10} colony-forming units were administered daily to women during the last month of pregnancy and to infants aged 0–6 mo. Adverse events (AE) were classified according to WHO International Statistical Classification of Diseases criteria. Common symptoms were recorded by regular questionnaires. Baseline characteristics of 220 mother-infant dyads in the treatment and 234 in the placebo group were similar. Compliance with the trial interventions, loss to follow-up, symptoms, drug usage, infant growth, method of feeding, visits to the doctor, and mothers' assessment of infant health were similar in the 2 groups. Fifteen (6.8%) mothers and 73 (33.2%) infants in the treatment group and 21 (9.0%) mothers and 75 (32.1%) infants in the placebo group reported AE ($P = 0.49$ and $P = 0.84$, respectively). Severe AE occurred in 18 mothers and 63 infants with a similar frequency in each group. None of the AE were attributed to the intervention. Our findings support the safe use of this consortium of organisms during pregnancy and early infancy. J. Nutr. 140: 483–488, 2010.

Introduction

A range of microbial supplements are consumed by various groups of people with the aim of improving health. The term probiotics, although defined as live microorganisms which, when administered in adequate amounts, confer health benefits on the host (1), is often applied inaccurately to organisms in which no evidence for a clear health benefit exists. Probiotic organisms include the lactic acid bacteria (principally *Lactobacillus* and *Streptococcus*) and bifidobacteria.

Lactic acid bacteria and bifidobacteria have a long record of safe use (2). Clinical studies have provided evidence for their use in acute and antibiotic-associated diarrhea, necrotizing enterocolitis, and the prevention of atopy (3), and the safety of probiotics in children has recently been reviewed (4). However, there are rare case reports of infection associated with lactic acid bacteria administration, primarily in compromised hosts (5). Other potential concerns include horizontal gene transfer of virulence factors or antimicrobial resistance genes between probiotic organisms and other intestinal or food-borne microorganisms, the production of harmful metabolites, and possible adverse immunologic effects (6). The report of possible adverse effects of probiotics in acute pancreatitis (7) highlights the need to assess the safety of specific probiotic organisms in specific at-risk populations. In view of these concerns, many researchers have emphasized the need for careful monitoring for potential adverse effects (3–6).

We undertook a prospective, randomized, controlled trial of a microbial dietary supplement in the prevention of atopy. Two strains of lactobacilli and 2 strains of bifidobacteria were

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[3] Supplemental Figure 1 is available with the online posting of this paper at http://jn.nutrition.org.

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administered to mothers during the last month of pregnancy and to infants during the first 6 mo of life. Here, we report common symptoms and adverse effects that occurred in the study according to intervention group.

Materials and Methods

The study was approved by the Swansea Local Research Ethics Committee in February 2004 (International Standard Randomized Controlled Trial, ISRCTN 26287422).

Participants. Women aged 16 y or older with a normal singleton pregnancy attending antenatal clinics in hospitals or general practice surgeries were eligible to join the study. Most women were carrying a fetus at increased risk of atopy, defined as a fetus with a first-degree relative with either asthma or eczema diagnosed by a health professional or allergic rhinitis treated by a doctor. Some women carrying a fetus not at increased risk of atopy were also recruited. Exclusion criteria were any known adverse condition affecting the woman, fetus, or the likely outcome of the pregnancy; if a member of the fetus's sibship or household was already recruited to the study; or if the woman was unwilling to discontinue use of other live bacterial dietary supplements. Information regarding demographic factors and other factors that may be associated with adverse events (AE),[8] such as smoking during pregnancy and housing conditions, was collected at recruitment on standard forms.

Randomization. A computer-generated, random allocation sequence without blocks allocated the mother-infant dyad at 36 wk of gestation to either the treatment or placebo arm of the study on a 1:1 basis. The allocation sequence was generated by the independent statistician (D.W.) and was also held in sealed, opaque envelopes at the trial site for emergency access but was not available to any member of the research team. Research staff allocated dyads sequentially to the next number in the sequence.

Intervention. Women during the last month of pregnancy and their infants from birth to age 6 mo received daily vegetarian capsules composed of hydroxypropyl methylcellulose containing either the treatment consisting of *Lactobacillus salivarius* CUL61 [National Collection of Industrial, Food and Marine Bacteria (NCIMB) 30211] 6.25×10^9 colony-forming units (cfu), *Lactobacillus paracasei* CUL08 (NCIMB 30154) 1.25×10^9 cfu, *Bifidobacterium animalis subsp. lactis* CUL34 (NCIMB 30172) 1.25×10^9 cfu, and *Bifidobacterium bifidum* CUL20 (NCIMB 30153) 1.25×10^9 cfu or identical placebo capsules containing maltodextrin. The identity of the organisms was confirmed at the species and strain level by 16S rRNA gene sequencing, rep PCR fingerprinting and cluster analysis, and Random Amplified Polymorphic DNA typing. The products were independently tested at a United Kingdom Accreditation Service-accredited laboratory prior to release for the study.

Mothers were provided with 2 bottles each containing 30 capsules at recruitment. The mother either took the capsule by mouth or sprinkled the contents of the capsule onto food. For infants, 6 bottles of capsules were provided and the contents of the capsule were mixed with formula or expressed breast milk or sprinkled directly into the baby's open mouth.

Follow-up for AE. AE were defined as any untoward medical occurrence in a participant in the trial, including events that were not necessarily caused by or related to the intervention (8). Parents/caregivers were issued with a study card with the contact details of a named research nurse and were encouraged to maintain close contact by phone, to report any AE occurring in the mother or infant, and to discuss any concerns regarding the study. Parents and caregivers were asked to report their participation in the trial to hospital staff in the event of either the mother or infant requiring admission to hospital. If mothers or infants were admitted to the hospital, staff were alerted to their participation in the trial by a sticker placed on the cover of both the mother and baby case notes and encouraged to notify the research team. Throughout the course of the study, the hospital microbiology laboratory results were checked to identify lactobacilli or bifidobacteria infections.

In addition to spontaneous reporting, regular questionnaires collected information on common symptoms, compliance with trial interventions, feeding method, visits to general practitioners, medicines received, hospital admissions, and the mother's or main caregiver's assessment of the baby's overall health. Questionnaires were scheduled at ages 6, 12, 18, and 24 wk. At 6 wk, questionnaires were completed during a home visit by a research nurse, at 12 and 18 wk by telephone interviews, and at 24 wk during a research clinic or, if the infant was unable to attend, by telephone interview. Research staff also made additional visits to participants' homes to complete follow-up if required. Unused trial preparations were collected at home visits and the 6-mo clinic visit as a further assessment of compliance.

Categorization and independent review of AE. All AE were entered into an electronic database and reviewed at regular intervals by independent safety monitors who also provided guidance on the attribution of serious unexpected AE according to Directive 2001/20/EC (8). Symptoms and signs related to atopy have not been included as these were the primary outcome measures of the trial and will be reported separately. All other AE recorded by spontaneous reporting and questionnaires were included in the analysis. AE were categorized independently by 2 pediatricians (S.J.A. and G.M.) using WHO International Statistical Classification of Diseases and Related Health Problems, 10th Revision (ICD10) criteria (9) and any discrepancies discussed. The severity of AE was graded according to Directive 2001/20/EC (8). All events where the participant was admitted overnight to hospital were classified as serious.

Data analysis. Once data collection and classification of AE was completed, the database was locked and sent for statistical analysis by intention to treat. Demographic variables and the frequency of AE in the 2 arms of the trial (treatment or placebo) were described. The number of spontaneously reported AE in infants was reported for the whole 6-mo period. Difficulties in contacting parents and caregivers, especially once many mothers had returned to work, often resulted in delayed or missed scheduled questionnaires. Information to the age of 8 wk was collated to assess the effects of exposure to the bacterial food supplement early in life and to ages 9–28 wk to assess effects of exposure later in infancy. Analysis was performed using SAS 9.1 (SAS Institute). Binary outcomes were compared with Fisher's exact test. Continuous variables at baseline are shown as mean and SD. Continuous outcomes tended to have skewed distributions, so were described using median values and ranges and analyzed by the Mann-Whitney U test. $P < 0.05$ was considered significant. Values in the text are medians (ranges) unless noted otherwise.

Results

From a total of 1419 pregnant women attending antenatal clinics who were assessed for eligibility to participate in the trial, 454 women were recruited, of whom 413 (91.0%) were carrying a fetus at increased risk of atopy. Two hundred and twenty women were allocated to the treatment group and 234 to the placebo group (**Supplemental Fig. 1**). Demographic variables and factors potentially associated with either AE or the reporting of AE were similar in the 2 arms of the study (**Table 1**). The number of respondents varied according to parents' and caregivers' ability to recall information.

Mothers. Median compliance with the trial intervention was 20 (range 0–60) d in 207 mothers in the treatment group and 20 (range 0–60) d in 213 in the placebo group ($P = 0.82$). Some

[8] Abbreviations used: AE, adverse event; CFU, colony-forming unit; ICD10, International Statistical Classification of Diseases and Related Health Problems, 10th Revision; NCIMB, National Collection of Industrial, Food and Marine Bacteria; SAE, serious adverse event.

TABLE 1 Baseline demographic and environmental factors that may be associated with AE or the reporting of AE

Variable	Treatment group, *n* = 220		Placebo group, *n* = 234	
	n		*n*	
Mother				
Age, *y*	220	29.0 ± 5.6	234	29.3 ± 6.0
Alcohol in pregnancy, *n (%)*	218	93 (42.7)	233	100 (42.9)
Current cigarette smoker, *n (%)*	220	33 (15.0)	234	40 (17.1)
Smoked cigarettes during pregnancy, *n (%)*	220	52 (23.6)	233	62 (26.6)
Education, *n (%)*				
University or college education	219	136 (62.1)	232	133 (57.3)
Other professional qualification	219	29 (13.2)	232	37 (15.9)
Currently working, *n (%)*	219	150 (68.5)	234	164 (70.1)
Father				
Education, *n (%)*				
University or college education	216	113 (52.3)	229	117 (51.1)
Other professional qualification	216	30 (13.9)	229	37 (16.2)
Currently working, *n (%)*	215	189 (87.9)	228	208 (91.2)
Housing				
Other children present, *n*	220	0.9 ± 0.9	234	1.0 ± 1.0
Adults present, *n*	220	2.1 ± 0.6	234	2.1 ± 0.6
House damp or condensation present, *n (%)*	218	57 (26.1)	232	56 (24.1)
House contains mold, *n (%)*	217	33 (15.2)	233	32 (13.7)
Cigarette smokers present, *n*	220	0.5 ± 0.7	233	0.6 ± 0.8

mothers mistakenly continued to take the trial interventions after delivery. In all, 36 mothers (7.9%) reported a total of 44 AE (**Table 2**). Eighteen were classified as serious AE (SAE), of which 13 SAE were categorized as ICD10 chapter XV (pregnancy, childbirth, and the puerperium) and consisted of pregnancy-induced hypertension (4 women), complications of Caesarean section (4), prolonged rupture of membranes (3), third-degree perineal tear (1), and pyrexia during delivery (1). There were 2 central nervous system SAE and both occurred in the treatment group. One woman developed acute inflammation of the central nervous system 6 d after delivery with persisting disability. Despite intensive investigation, no infectious or alternative cause was identified. One woman had transient demyelination of the pons occurring 16 mo after delivery with full recovery. There were 2 intrauterine deaths (ICD10 chapter XVI) and both occurred in the placebo group. One resulted from acute placental failure associated with severe pregnancy-induced hypertension and the other was unexplained but associated with adverse social circumstances. One mother in the placebo group collapsed 5 mo after delivery and made a full recovery; no cause was found (chapter XVIII). Categorized according to ICD10 chapters, the frequency of the AE was similar in the 2 groups (Table 2). No lactobacilli or bifidobacteria infections were identified and none of the AE were attributed to the trial interventions.

Infants. Birthweights did not differ ($P = 0.44$) and were 3.49 (2.1–4.9) kg in the treatment group ($n = 219$) and 3.55 (2.0–5.2) kg in the placebo group ($n = 233$). One infant in each group was delivered outside of the local National Health Service Trust and birthweight could not be ascertained. Loss to follow-up for infants in the 2 groups was similar at follow-up to 8 wk

TABLE 2 Number of mothers reporting AE classified according to ICD10 code chapter

ICD10 code chapter	Treatment group, *n* = 220	Placebo group, *n* = 234	*P*-value
	n (%)		
I: Certain infectious and parasitic diseases	0 (0.0)	1 (0.4)	0.61
III: Diseases of the blood and blood-forming organs and certain disorders involving the immune mechanism	0 (0.0)	1 (0.4)	1.00
VI: Diseases of the nervous system	2 (0.9)	0 (0.0)	0.23
X: Diseases of the respiratory system	1 (0.5)	0 (0.0)	0.49
XI: Diseases of the digestive system	2 (0.9)	2 (0.9)	1.00
XV: Pregnancy, childbirth and the puerperium	10 (4.5)	13 (5.6)	0.50
XVI: Certain conditions originating in the perinatal period	0 (0.0)	3 (1.3)	0.25
XVIII: Symptoms, signs and abnormal clinical and laboratory findings, not elsewhere classified	0 (0.0)	2 (0.9)	0.50
XXII: Codes for special purposes	1 (0.5)	0 (0.0)	0.49
Total mothers[1]	15 (6.8)	21 (9.0)	0.49

[1] 6 mothers reported 2 AE and 1 mother reported 3 AE.

TABLE 3 Common symptoms, drug usage, and growth up to age 8 wk[1]

Variable	Treatment group, n = 220		Placebo group, n = 234		P-value
	n		n		
Age at follow-up, *d*	167	46 (26–60)	161	46 (33–62)	0.47
Common symptoms (yes), *n (%)*					
Colic	167	83 (49.7)	161	75 (46.6)	0.58
Regurgitation	167	123 (73.7)	161	128 (79.5)	0.24
Constipation	167	26 (15.6)	161	44 (27.3)	0.01
Diarrhea	167	23 (13.8)	161	21 (13.0)	0.87
High temperature	163	6 (3.7)	159	12 (7.5)	0.15
Drugs (yes), *n (%)*					
Antibiotic	166	31 (18.7)	161	25 (15.5)	0.47
Paracetamol	167	23 (13.8)	161	27 (16.8)	0.54
Weight,[2] *kg*	159	4.90 (3.6–7.1)	156	4.99 (3.6–6.9)	0.79
Weeks exclusively breast-fed, *n*	167	2 (0–8)	161	3 (0–8)	0.20
Visits to doctor, *n (%)*	166	1 (0–4)	161	1 (0–8)	1.00
Mother`s assessment of infant`s health, *n (%)*					1.00
Very healthy	166	145 (87.3)	160	139 (86.9)	
Occasionally unwell	166	20 (12.0)	160	20 (12.5)	
Nearly always unwell	166	1 (0.6)	160	1 (0.6)	

[1] Information collected either during home visit or by telephone interview. Data are medians (range) or *n* (%).
[2] Either weight measured during home visits or recent weight recorded by community midwife during telephone interviews

(P = 0.11) and between 9 and 28 wk (P = 0.29; Supplemental Fig. 1). The most common reason given for loss to follow-up was that parents and caregivers were too busy. Many parents and caregivers who could not be contacted before 8 wk did provide information at later follow-ups.

During the first 8 wk, compliance did not differ between groups (P = 0.99) and was 27 (0–60) d (n = 217) in the treatment group and 26 (0–61) d (n = 228) in the placebo group. Parents and caregivers in both groups reported that the trial interventions were easy to administer to infants: 109/142 (76.8%) in the treatment group and 110/139 (79.1%; P = 0.67) in the placebo group. Drug usage, infant weight, feeding practice, number of visits to the doctor, and mothers' assessments of their infants' health were also similar in the 2 groups (**Table 3**). Constipation occurred in significantly fewer infants in the treatment than the placebo group (Table 3). The frequency of other symptoms was similar in the 2 groups.

During 9–28 wk, compliance with the trial interventions was 106 (0–182) d (n = 206) in the treatment group and 105 (0–180) d (n = 216) in the placebo group (P = 0.82). There were no significant differences in the various factors between the 2 groups (**Table 4**).

In the first 6 mo, parents or caregivers reported AE in about 1 in 3 infants in each group (Table 5). Most AE were common illnesses in infancy and occurred at a similar frequency in each group. Overall, AE in 27 (12.3%) infants in the treatment group and 36 (15.4%) in the placebo group were classified as serious (P = 0.41). **Table 5** also shows specific common AE (reported in $\geq$*5* infants in either group). One infant living in a family with adverse social circumstances presented with a sudden unexpected death in infancy. This mother and infant had been allocated to the placebo arm of the study. A total of 14 infants were classified as having "unspecified acute lower respiratory infection" (ICD10 code J22) and this occurred more frequently in the treatment than the placebo group. Nine of these had a respiratory illness that was treated with an antibiotic by their General Practitioners. Five were admitted to the hospital and were therefore SAE (4 in the treatment and 1 in the placebo group; P = 0.20) and all recovered. A specific infectious agent was not identified in any of these infants and it is likely that some had viral infections.

Five infants reported vomiting and/or diarrhea, none classified as serious, all in the treatment group (ICD10 code P93). In 2 infants with regurgitation, the mother considered that the trial intervention caused the symptoms and discontinued adminis-

TABLE 4 Common symptoms and drug usage recorded between ages 9 and 28 wk[1]

Variable	Treatment group, n = 220		Placebo group, n = 234		P-value
Common symptoms (yes), *n (%)*	*n*		*n*		
Colic	191	65 (34.0)	195	63 (32.3)	0.75
Regurgitation	191	135 (70.7)	195	145 (74.4)	0.43
Constipation	191	55 (28.8)	195	62 (31.8)	0.58
Diarrhea	191	63 (33.0)	195	55 (28.2)	0.32
High temperature	191	51 (26.7)	195	53 (27.2)	1.00
Drugs (yes), *n (%)*					
Antibiotic	192	39 (20.3)	195	48 (24.6)	0.33
Paracetamol	192	172 (89.6)	195	177 (90.8)	0.73
Growth[2]					
Weight, *kg*	138	7.90 (5.8–10.2)	141	7.90 (5.4–10.4)	0.32
Length, *cm*	137	68.0 (44.1–75.3)	141	68.1 (46.5–83.7)	0.94
Head circumference, *cm*	131	43.5 (38.2–49.7)	139	44.0 (39.0–48.7)	0.26
Weeks received cereals, *n (%)*	146	4 (0–12)	150	5 (0–12)	0.17
Trial intervention was easy to administer (yes), *n (%)*	130	109 (83.8)	130	110 (84.6)	1.00
Visits to doctor, *n (%)*	192	1 (0–11)	195	1 (0–9)	0.62
Mother`s assessment of baby`s health,[1] *n (%)*					0.62
Very healthy	146	126 (86.3)	151	135 (89.4)	
Occasionally unwell	146	16 (11.0)	151	14 (9.3)	
Nearly always unwell	146	4 (2.7)	151	2 (1.3)	

[1] Data are medians (range) or *n* (%).
[2] Measured or assessed at the clinic visit scheduled for 24 wk.

TABLE 5 Number of infants aged 0–6 mo with AE classified according to ICD10 chapter and specific common AE (≥5 in 1 group) by ICD10 code

ICD10 chapter or code	Treatment group, *n* = 220	Placebo group, *n* = 234	*P*-value
	n (%)		
I: Certain infectious and parasitic diseases	15 (6.8)	12 (5.1)	0.55
A09: Gastroenteritis	6 (2.7)	3 (0.9)	0.33
B34.9: Viral infection, unspecified	2 (0.9)	6 (2.1)	0.29
B37.0: Candidal stomatitis	5 (1.8)	2 (0.9)	0.27
IV: Endocrine, nutritional and metabolic diseases	0 (0.0)	1 (0.4)	1.00
VII: Diseases of the eye and adnexa	6 (2.7)	12 (5.1)	0.23
H10.0: Conjunctivitis	6 (2.7)	8 (3.4)	0.79
VIII: Diseases of the ear and mastoid process	3 (1.4)	3 (1.3)	1.00
X: Diseases of the respiratory system	24 (10.9)	16 (6.8)	0.14
J06: Upper respiratory tract infection	5 (2.3)	5 (2.1)	1.00
J21.9: Bronchiolitis	9 (3.6)	9 (3.8)	1.00
J22: Lower respiratory tract infection	11 (3.6)	3 (0.9)	0.03
XI: Diseases of the digestive system	8 (3.6)	14 (6.0)	0.28
K21: Gastro-esophageal reflux disease	4 (1.8)	6 (2.6)	0.75
XII: Diseases of the skin and subcutaneous tissue	12 (5.5)	12 (5.1)	1.00
L01.0: Impetigo	7 (3.2)	3 (1.3)	0.21
XIV: Diseases of the genitourinary system	0 (0.0)	1 (0.4)	1.00
XV: Pregnancy, childbirth and the puerperium	4 (1.8)	5 (2.1)	1.00
O98.9: Unspecified maternal infectious or parasitic disease complicating pregnancy, childbirth and the puerperium	4 (1.8)	5 (2.1)	1.00
XVI: Certain conditions originating in the perinatal period	10 (4.5)	18 (7.7)	0.18
P59.9: Neonatal jaundice, unspecified	0 (0.0)	6 (2.6)	0.03
P92.5: Neonatal difficulty in feeding at breast	3 (1.4)	7 (3.0)	0.34
P93: Reactions and intoxications due to drugs administered to fetus and newborn	5 (2.3)	0 (0.0)	0.026
XVII: Congenital malformations, deformations and chromosomal abnormalities	12 (5.5)	12 (5.1)	1.00
Q82.5: Congenital non-neoplastic naevus	1 (0.5)	5 (2.1)	0.22
XVIII: Symptoms, signs and abnormal clinical and laboratory findings, not elsewhere classified	7 (3.2)	4 (1.7)	0.37
XIX: Injury, poisoning and certain other consequences of external causes	2 (0.9)	2 (0.9)	1.00
XX: External causes of morbidity and mortality	0 (0.0)	2 (0.9)	0.50
Total number of infants reporting ≥1 AE	73 (33.2)	75 (32.1)	0.84

tration. One infant was receiving an antibiotic and this was considered the likely cause of the symptoms. In 2 infants, the relationship with the trial intervention was unclear. Six infants had physiological neonatal jaundice (ICD10 code P59.9) and all occurred in the placebo group. All 6 infants with a SAE classified as a disease of the digestive system (ICD10 chapter XI) were in the placebo group ($P = 0.03$). Two were surgical cases (obstructed inguinal hernia, umbilical hernia repair) and 4 were admitted with a variety of symptoms, including colic and constipation. With these exceptions, the frequency of SAE was similar in the 2 groups ($P > 0.05$), with SAE categorized either according to ICD10 chapter or specific code. Apart from the 2 possible adverse reactions listed above, no AE was considered to be associated with this supplement. In addition, no lactobacilli or bifidobacteria infections were identified. All life-threatening and fatal AE in both mothers and infants were reviewed carefully by the research team and the external safety monitors and none were considered to be attributable to the supplement.

Discussion

Administration of 2 strains of lactobacilli and 2 strains of bifidobacteria to mothers during the last month of pregnancy and infants during the first 6 mo of life, most of whom were at increased risk of atopic disease, was not associated with an increase in AE. Only a small proportion of women reported 1 or more AE, which were mostly events related to pregnancy, delivery, and the puerperium. It was not possible to identify a specific cause in the 2 women in the treatment group who developed neurological illnesses after delivery. However, we are not aware of any previous reports of similar illnesses attributed to ingestion of lactobacilli or bifidobacteria. In keeping with the findings in this study, many studies of probiotics administered during pregnancy have not identified adverse outcomes and a recent systematic review reported no adverse effects of lactobacilli and bifidobacteria during pregnancy (10).

In infants, lower respiratory tract infections were reported more frequently in the treatment than the placebo group. Although these infants received antibiotic treatment, it is likely that many were suffering from viral rather than bacterial infection. Also, the difference between the 2 groups may have arisen by chance given the large number of comparisons reported. Regurgitation may have been an adverse reaction to the dietary supplement in 2 infants, but this seems unlikely. The questionnaires identified regurgitation as the most common symptom and it occurred with similar frequency in each group. The supplement appeared to reduce the frequency of constipation in early infancy.

However, no effect on constipation was observed later in infancy and this early difference may have arisen by chance given the large numbers of comparisons reported. The greater frequency of physiological jaundice in the placebo than the treatment group was also likely to be a chance finding.

A strength of the current study was the identification of AE both by spontaneous reporting as well as regular questionnaire and clinical follow-up in a double-blind study. A weakness was loss to follow-up of ~15% overall. Many mothers were working and were too busy; domestic difficulties were common with some women living in a refuge. Although loss to follow-up was similar in both groups, the possibility that the bacterial supplement caused AE in some of these participants cannot be excluded.

Many different bacterial preparations have been administered to large numbers of healthy infants in research studies without reported adverse effects and some authors consider that there is no evidence that probiotics are harmful to children (11). However, the increased use of bacterial dietary supplements, their inclusion in infant formula milks, and the fact that AE may be strain specific (5) have prompted closer scrutiny for possible adverse effects. In reports focusing on safety, AE were not attributable to the supplemented organisms in infants administered *Lactobacillus rhamnosus* HN001 and *Bifidobacterium animalis* subsp. lactis HN019 (12), combinations of *Bifidobacterium longum* BL999, *Lactobacillus rhamnosus* LPR and *Lactobacillus paracasei* ST11 (13), and *Bifidobacterium lactis* BB-12 and *Lactobacillus reuteri* ATCC 55730 (14). Similarly, adverse outcomes have not been associated with infant formula milks containing 1 or more bacterial species with a prebiotic; organisms have included *Bifidobacterium longum* BL999 (15), *Lactobacillus rhamnosus* GG and LC705, *Bifidobacterium breve* Bb99 and *Propionibacterium freudenreichii* spp. *shermanii* (16), and *Lactobacillus paracasei* ssp. *paracasei* and *Bifidobacterium animalis* ssp. *lactis* (17). Finally, the administration of *Bifidobacterium lactis* with long-chain PUFA was not associated with detrimental effects (18).

Several other randomized trials have evaluated probiotics in the prevention of atopy in infants at increased risk of atopy. Most have shown favorable outcomes, but Taylor et al. (19) reported increased allergen sensitization in infants who received *Lactobacillus acidophilus* (LAVRI-A1). Symptoms and signs of atopy are the primary clinical endpoints in this current study and these outcomes will be reported.

In conclusion, this study assessed the safety of a dietary supplement of *L. salivarius* CUL61, *L. paracasei* CUL08, *B. animalis subsp. lactis* CUL34, and *B. bifidum* CUL20 in mothers during the last month of pregnancy and infants during the first 6 mo of life. There was no evidence that these strains were associated with adverse effects in these women or their infants.

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Probiotics in the prevention of eczema: a randomised controlled trial

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ABSTRACT

Objective To evaluate a multistrain, high-dose probiotic in the prevention of eczema.

Design A randomised, double-blind, placebo-controlled, parallel group trial.

Settings Antenatal clinics, research clinic, children at home.

Patients Pregnant women and their infants.

Interventions Women from 36 weeks gestation and their infants to age 6 months received daily either the probiotic (*Lactobacillus salivarius* CUL61, *Lactobacillus paracasei* CUL08, *Bifidobacterium animalis subspecies lactis* CUL34 and *Bifidobacterium bifidum* CUL20; total of 10^{10} organisms/day) or matching placebo.

Main outcome measure Diagnosed eczema at age 2 years. Infants were followed up by questionnaire. Clinical examination and skin prick tests to common allergens were done at 6 months and 2 years.

Results The cumulative frequency of diagnosed eczema at 2 years was similar in the probiotic (73/214, 34.1%) and placebo arms (72/222, 32.4%; OR 1.07, 95% CI 0.72 to 1.6). Among the secondary outcomes, the cumulative frequency of skin prick sensitivity at 2 years was reduced in the probiotic (18/171; 10.5%) compared with the placebo arm (32/173; 18.5%; OR 0.52, 95% CI 0.28 to 0.98). The statistically significant differences between the arms were mainly in sensitisation to cow's milk and hen's egg proteins at 6 months. Atopic eczema occurred in 9/171 (5.3%) children in the probiotic arm and 21/173 (12.1%) in the placebo arm (OR 0.40, 95% CI 0.18 to 0.91).

Conclusions The study did not provide evidence that the probiotic either prevented eczema during the study or reduced its severity. However, the probiotic seemed to prevent atopic sensitisation to common food allergens and so reduce the incidence of atopic eczema in early childhood.

Trial registration Number ISRCTN26287422.

What is already known on this topic?

- Altered exposure to microbial organisms in early life might influence the development of atopy.
- Meta-analysis suggests that probiotics may be effective in the primary prevention of atopy.

What this study adds?

- Our data do not support use of the study probiotic for the prevention of eczema in early childhood.
- Skin prick sensitivity to common food allergens and atopic eczema were reduced among children receiving the probiotic.

INTRODUCTION

The major atopic disorders, eczema, allergic rhinitis and asthma, cause significant disease burdens worldwide. Symptoms of atopy were reported to occur in 15–40% of children aged 13–14 years living in the UK in the mid-1990s[1] and prevalence has increased in many countries in recent years.[2–4]

Despite the high atopic disease burden, pathogenic mechanisms remain poorly understood. Atopic disorders are heterogeneous resulting from complex interactions between environmental factors, including exposure to microbes, and host genes modulating innate and acquired immune responses, and mucosal and skin integrity.[5] A critical issue is the role of atopy defined as a genetic propensity to develop immunoglobulin E antibodies following exposure to allergens[6] and assessed by skin prick tests (SPTs) or measurement of specific IgE in serum. For eczema[7] and asthma,[8] atopic sensitisation early in life has been associated with persistent disease.

The hygiene hypothesis, based on the observation of increased atopy in smaller families,[9] postulates that reduced exposure to infections in early childhood results in aberrant immunological responses to allergens.[10] Intestinal microbiota provide a critical source of early immune stimulation and variations in early gut colonisation have been associated with the development of atopic disease.[11] Probiotics are defined as live microorganisms which, when administered in adequate amounts, confer health benefits on the host.[12] However, the term 'probiotic' is commonly used for microorganisms that are undergoing evaluation for possible health benefits and it is this sense that is used here. Clinical trials suggest a role for probiotics in the primary prevention of atopic eczema.[13] The mechanisms whereby probiotics may prevent atopy are unclear but might involve reduced exposure to allergens through improved epithelial barrier function and modulation of the developing immune system to prevent IgE sensitisation.[14]

We evaluated the effect of a probiotic comprising two strains of lactobacilli and two strains of bifidobacteria, or an inert placebo, administered to

mother-infant dyads in the perinatal period on clinical and laboratory manifestations of atopy. Here, we report the clinical manifestations of atopy at age 0–2 years.

METHODS

The safety profile of the probiotic[15] and factors determining compliance with the trial procedures[16] have been published.

Participants

Women aged 16 years or more, with a normal singleton pregnancy and intending to give birth at Singleton Hospital, Swansea were invited to join the study. Details of the recruitment process are described elsewhere.[16] Women with known adverse conditions likely to affect them, the fetus, or the outcome of the pregnancy, were excluded. Women were informed of the trial, by letter, at booking, and provided with further information when attending for antenatal care. Those returning an expression of interest were contacted by researchers. Signed, informed consent was obtained by a researcher at 35–36 weeks gestation.

We had intended to recruit infants at 'high-risk' of developing atopy defined as those with a first degree relative with either asthma or eczema diagnosed by a health professional or allergic rhinitis treated by a doctor.[17] However, sufficient details of atopy in first degree relatives were often difficult to obtain at recruitment. Therefore, in practice, infants with and without an increased risk of atopy were recruited. To preserve the integrity of the random allocation sequence, all infants are included in this report. Key findings relating to those at increased risk of atopy are presented in Webtable 1.

Randomisation

Pregnant women were allocated on a 1:1 basis at 36 weeks gestation to either the active or placebo arm of the study according to a computer-generated, random sequence without blocks. The random sequence was generated by the commercial partners and held independently of the research team. Women were allocated consecutively to the next number in the randomisation sequence and provided with the corresponding, preprepared intervention pack.

Intervention

We selected a multispecies probiotic preparation on the basis of greater efficacy than either single species or single organism preparations in the prevention or treatment of diseases in animal models and clinical trials.[18] Species that had been evaluated in previous clinical trials, including the prevention of eczema were selected.[17] Organisms were selected on the basis of activity in promoting responses in vitro consistent with protection against allergy (personal communication; Dr S. Plummer 2003).

The active intervention comprised a vegetarian capsule containing *Lactobacillus salivarius* CUL61 (National Collection of Industrial, Food and Marine Bacteria (NCIMB) 30211) 6.25×10^9 colony forming units (CFUs), *Lactobacillus paracasei* CUL08 (NCIMB 30154) 1.25×10^9 CFUs, *Bifidobacterium animalis* subspecies *lactis* CUL34 (NCIMB 30172) 1.25×10^9 CFUs and *Bifidobacterium bifidum* CUL20 (NCIMB 30153) 1.25×10^9 CFUs as a freeze-dried powder. Organism identity was confirmed at the species and strain levels by 16S rRNA gene sequencing, rep PCR fingerprinting and cluster analysis, and Random Amplified Polymorphic DNA typing. Women in the placebo arm received capsules of identical appearance containing maltodextrin powder. The dose in both arms was one capsule daily from 36 weeks gestation until delivery. Infants received the same capsules as their mother once daily from birth to age 6 months. Women were asked to store the capsules in the fridge and either took the capsule by mouth or sprinkled the contents onto food. The contents of the capsule were sprinkled directly into the infant's open mouth or mixed with expressed breast milk or formula feed. Mothers were asked not to consume any commercially available probiotics or live yoghurts or administer these to their infants.

Unused capsules returned by participants from intervention and placebo groups for compliance monitoring were tested by an independent laboratory on an opportunistic basis. No live bacteria were identified in the placebo capsules, conforming with the random allocation sequence. Viability of the bacteria in the active preparation was consistent with the storage conditions confirming no significant deterioration in the product.

Data collection and allergen testing

Demographic information and possible risk factors for atopy were recorded at recruitment. Follow-up questionnaires were scheduled every 6 weeks to age 6 months and at 1 year and 2 years. A research assistant completed questionnaires with the parent/carer during a home visit at age 6 weeks, during research clinics at 6 months and 2 years and by telephone interview at other times. Where a follow-up was missed, parents/carers were asked to provide information for the period since the last questionnaire was completed. Questionnaires recorded any atopic disorders diagnosed by health professionals, treatments received and the occurrence and duration of common skin, respiratory and gastrointestinal symptoms.

At research clinics, infants were examined by either a clinician or a trained researcher. If present, the severity of eczema was evaluated by the scoring atopic dermatitis index.[19] SPTs using common food allergens (cow's milk, hen's egg), aeroallergens (house dust mite, cat dander, grass pollen) and positive (histamine) and negative controls were performed. The response to an allergen was considered positive if there was a wheal diameter ≥3 mm.[20]

Outcome measures

The primary outcome was the cumulative frequency of diagnosed eczema at 2 year follow-up (reported by parents/carers to have been diagnosed by a health professional or diagnosed during a research clinic). Eczema was defined as an itchy rash affecting the face, scalp or extensor surfaces of the limbs in infants and flexures in older children and of duration ≥4 weeks and with ≥1 exacerbation by age 24 months[21] based on the information from questionnaires. Secondary outcomes were responses to SPTs, atopic eczema defined as eczema with one or more positive SPTs, eczema of any duration and whether or not diagnosed by a health professional, the severity of eczema, treatment with topical steroid preparations, respiratory symptoms with asthma and allergic rhinitis and reported food allergy. We had intended to report outcomes up to age 2 years. However, scheduled follow-up assessments were often delayed particularly when mothers had returned to work. Therefore, we have included all information for children aged up to, but not including, 3 years.

Data management and analysis

Strict blinding of the clinical research teams regarding participant allocation was maintained until after databases were locked following completion of data collection. The period of follow-up, defined as the time from birth to the age of the last follow-up questionnaire, was calculated for each infant.

Table 1 Demographic characteristics and possible risk factors for atopy at baseline

Variable	Probiotic arm*	Placebo arm*
Socioeconomic status (Townsend rank; N; median, IQR)	220; 949 (333–1514)	234; 864 (330–1558)
Mother smoked during pregnancy	41/205 (20.0%)	47/218 (21.6%)
Vaginal delivery	152/216 (70.4%)	157/232 (67.7%)
House		
Cat, dog, rodent or bird kept indoors	112/220 (50.9%)	120/234 (51.3%)
Damp and/or mould as reported by participants	59/217 (27.2%)	56/232 (24.1%)
No. households with		
1 adult	10/220 (4.6%)	9/234 (3.9%)
≥3 adults	20/220 (9.1%)	25/234 (10.7%)
no other children	81/220 (36.8%)	93/234 (39.7%)
≥3 other children	16/220 (7.3%)	18/234 (7.7%)
No. infants at increased risk of atopy†	197/220 (89.6%)	205/234 (87.6%)

This table gives the number of participants (%) unless otherwise stated.
*Denominator varies according to information provided by parents/carers and compliance with follow-up.
†Defined as first degree relative with either asthma or eczema diagnosed by a health professional or allergic rhinitis treated by a doctor.

Demographic variables, possible risk factors for atopy, and primary and secondary outcomes were analysed by treatment allocated. Findings for binary outcomes were expressed in ORs with 95% CIs. A χ^2 test or Fisher's exact test was used to compare proportions. Continuous outcome variables had skewed distributions and were summarised using median values and IQR and compared by the Mann-Whitney U test.

We performed adjusted analyses to investigate whether the trends in atopic sensitivity or atopic eczema were modified by baseline demographic variables (table 1), possible risk factors for atopy (table 2) and mother and infant compliance. For compliance, participants were classified as no compliance and compliance levels 1–4 representing the quartiles of the total number of days that trial interventions were taken. We used a set of logistic regression models, including the study treatment in each case and adding the other variables in a stepwise manner, retaining them in the model if they resulted in a significant improvement in model fit (at the 5% level). Analysis was performed using R/SPSS V.16.0 (IBM, USA)/SAS V.9·2 (SAS Institute, USA).

Sample size

We aimed to recruit sufficient mother/infant dyads to observe a 50% reduction in the frequency of asthma by age 5 years from 20% to 10%.[22] For the purposes of this paper, we expected that 40% of infants at increased risk of atopy (defined as having a first degree relative with an atopic condition) in the placebo arm would have developed eczema by age 2 years.[17 23] A total of 308 infants (154 in each arm) would be sufficient to detect a 50% reduction in eczema frequency from 40% in the control arm to 20% in the probiotic arm with 90% power at the 1% significance level.

Table 2 Potential modulating factors for atopy during follow-up

Variable	Probiotic arm	Placebo arm
Breast fed (full or partial)—any duration	49/191 (25.7%)	47/205 (22.9%)
Age last breast fed in months (N; median, (IQR))	185; 1 [0–7]	190; 1 [0–5]
Attended child-minder in first 12 months	10/163 (6.1%)	16/172 (9.3%)
Attended nursery in first 12 months	65/162 (40.1%)	61/170 (35.9%)
Any oral/systemic antibiotics	155/214 (72.4%)	154/225 (68.4%)

This table gives the number of participants (%) unless otherwise stated.

Role of the funding source

Cultech UK advised on study design, provided the trial interventions and generated the random allocation sequence but was not involved in data collection, analysis or interpretation of the findings.

Ethics approval

The study was approved by the local research ethics committee in February 2004 and registered with Current Controlled Trials (ISRCTN26287422).

RESULTS

Participants

Recruitment began in May 2005 and the last scheduled 2 year contact was in November 2009. Of 1419 women attending antenatal clinics who had expressed an interest in the study, 454 were recruited and randomised (figure 1; Trial profile).

Demographic variables and possible risk factors for atopy were similar in the 220 women randomised to the probiotic

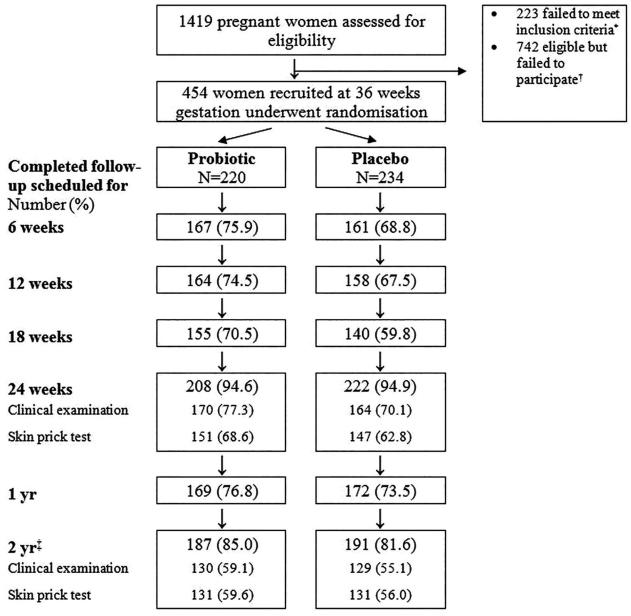

Figure 1 Trial profile *Reasons for exclusion: pregnancy complication (n=110); presented after 36 weeks gestation or at the end of the recruitment period and before 36 weeks gestation (83); multiple pregnancy (14); contacted again after recruitment terminated (9); previous infant participated in trial (6); unwilling to stop current probiotic (1). †Reasons for failure to participate: lost contact with research team (288); not sufficiently interested or disliked tests (184); concern that trial was "too much to take on" (129); unwillingness to take the investigation products (85); not prepared to be allocated to the placebo arm (21); developed a medical condition (16); language difficulties (10); not willing for this child to be treated differently from previous child (7); bereavement (2). ‡Follow-up was often delayed; includes children seen up to, but not including, age 3 years.

arm and 234 to the placebo arm (table 1). Maternal smoking during pregnancy, keeping pets and houses affected by damp or mould were common in both arms. Three hundred and seventy-four women were carrying an infant at high risk of developing atopy with a similar proportion in each study arm (table 1). Potential modifying factors for atopy during follow-up, such as feeding practice, exposure to other people and to antibiotics, were also similar in the two arms (table 2).

Completion of questionnaires and attendance at research clinics was similar in the two arms (figure 1). Median (IQR) age at follow-up was 2.11 years (2.01–2.28 years) in the probiotic arm and 2.09 years (2.01–2.24 years) in the placebo arm (p=0.28). We have reported previously that follow-up was more complete among less deprived families and mothers who were non-smokers.

The trial arm was not associated with retention or clinic attendance at 24 weeks and 2 years.[16]

Primary outcome measure

The cumulative frequency of diagnosed eczema (a composite of carers' reports and research clinic findings) at 2 years was similar in the probiotic (73/214; 34.1%) and placebo arms (72/222, 32.4%; OR 1.07, 95% CI 0.72 to 1.6; p=0.71).

Secondary outcome measures

Positive SPTs to one or more common allergens at either age 6 months or 2 years were significantly less frequent in the probiotic arm (OR 0.52, 0.28–0.98; p=0.036, table 3). The number needed to treat to prevent one infant becoming sensitised was 13 (95% CI 7 to 173). The reduced skin prick responses in the probiotic arm were mainly to food allergens (cow's milk and egg proteins) and statistically significant differences were already apparent at age 6 months. In contrast, sensitisation to aeroallergens (house dust mite, cat dander and grass pollen) occurred mainly after 6 months and was similar in the two arms (table 3).

Atopic eczema at 2 years was significantly less frequent in the probiotic arm (OR 0.40, 0.18–0.91; p=0.024) and was apparent by age 6 months (table 3). Differences between the two groups in non-SPT positive eczema were not statistically significant. At age 6 months, non-SPT positive eczema occurred in 37/147 (25.2%) children in the probiotic and 24/143 (16.8%) in the placebo arm (χ^2=3.06; p=0.08). At age 2 years, non-SPT positive eczema occurred in 57/171 (33.3%) children in the probiotic and 46/173 (26.6%) in the placebo arm (χ^2=1.86; p=0.17). The cumulative frequency of all eczema to age 2 years reported by parents and carers, whether or not diagnosed by a

Table 3 Secondary outcomes according to intervention group

Variable	Probiotic arm	Placebo arm	OR (95% CI)	p Value*
SPT† positive at 6 m	6/151 (3.97%)	16/147 (10.88%)	0.34 (0.13 to 0.89)	0.023
▶ cow's milk	0/148 (0.0%)	5/147 (3.40%)	–	0.030*
▶ egg	5/148 (3.4%)	14/147 (9.5%)	0.33 (0.11 to 0.95)	0.032
▶ house dust mite	1/151 (0.66%)	0/147 (0.0%)	–	0.51*
▶ cat	0/151 (0.0%)	2/145 (1.4%)	–	0.24*
▶ grass	1/150 (0.67%)	0/147 (0.0%)	–	0.49*
SPT† positive at either 6 m or 2 yrs	18/171 (10.5%)	32/173 (18.5%)	0.52 (0.28 to 0.98)	0.036
▶ cow's milk	1/171 (0.6%)	6/173 (3.5%)	0.16 (0.02 to 1.4)	0.12*
▶ egg	9/171 (5.3%)	19/173 (11.0%)	0.45 (0.2 to 1.0)	0.052
▶ house dust mite	9/171 (5.3%)	11/173 (6.4%)	0.82 (0.3 to 2.0)	0.66
▶ cat	3/171 (1.8%)	7/173 (4.0%)	0.42 (0.1 to 1.7)	0.20
▶ grass	2/171 (1.2%)	2/173 (1.2%)	1.0 (0.14 to 7.2)	0.99*
Skin				
▶ Atopic eczema at 6 m	4/151 (2.7%)	13/147 (8.8%)	0.28 (0.089 to 0.88)	0.021
▶ Severity of eczema at 6 m clinic‡; median, (IQR)	14.3 (7.5–17.9)	14.4 (10.6–24.9)	–	0.46
▶ Atopic eczema at 2 yrs	9/171 (5.3%)	21/173 (12.1%)	0.40 (0.18 to 0.91)	0.024
▶ Severity of eczema at 2 yr clinic‡; median (IQR)	11.1 (7.2–20.1)	14.2 (7.2–14.2)	–	0.85
▶ All reported eczema§,¶	119/214 (55.6%)	132/226 (58.4%)	0.90 (0.61 to 1.3)	0.55
▶ Received topical steroid preparation	30/214 (14.0%)	40/226 (17.7%)	0.76 (0.45 to 1.2)	0.29
Respiratory				
▶ All reported asthma¶	23/171 (11.9%)	20/179 (10.0%)	1.2 (0.63 to 2.3)	0.57
▶ Night-time cough	156/214 (72.9%)	164/226 (72.6%)	1.0 (0.67 to 1.5)	0.93
▶ Night-time or daytime cough	178/214 (83.2%)	188/226 (83.2%)	1.0 (0.6 to 1.6)	0.99
▶ Wheezing without symptoms of a virus infection	50/214 (23.4%)	55/171 (24.3%)	0.95 (0.61 to 1.47)	0.81
▶ Inhaled bronchodilator or steroid	26/214 (12.1%)	27/226 (11.9%)	1.0 (0.57 to 1.8)	0.94
▶ Allergic rhinitis¶	10/190 (5.3%)	10/201 (5.0%)	1.1 (0.43 to 2.6)	0.90
▶ Sneezing and/or snuffling	207/214 (96.7%)	212/226 (93.8%)	1.95 (0.77 to 4.93)	0.15
Any reported food allergy	22/200 (11.0%)	31/204 (15.2%)	0.69 (0.38 to 1.2)	0.21

This table gives the number of participants (%) unless otherwise stated.
χ^2 test for contingency tables or Fisher's exact test where expected values in cells were <5.
†SPT valid if diameter of wheal for the positive control was ≥1 mm than diameter for the negative control; SPT positive if diameter of wheal for antigen was ≥3 mm than diameter for negative control.
‡SCORAD index[19].
§Of any duration.
¶Whether or not diagnosed by a health professional.
SCORAD, scoring atopic dermatitis; SPT, skin prick test.

health professional, its severity, the duration of rash and the use of topical steroids were similar in the two arms. The cumulative frequency of asthma and allergic rhinitis and symptoms and treatments consistent with these conditions and of reported food allergy were also similar in both arms (table 3).

The differences between the two arms in the risk of atopic eczema and SPT positivity among infants with one or more relatives with atopy were very similar to those in the whole cohort (webtable 1). However, with the exception of reduced sensitivity to cow's milk protein at 6 months, differences in the smaller cohort did not reach statistical significance. Median (IQR) compliance with the trial interventions in pregnant women was 20 (12–28) days in the probiotic arm and 20 (12–28) days in the placebo arm (p=0.98, Mann-Whitney U test). In infants, the corresponding values were 106 (30–141) days and 103 (11–151) days (p=0.97, Mann-Whitney U test). In logistic regression analyses, in almost all cases, additional variables (table 1) were not significantly related to clinical outcomes and did not change the estimated OR for the effect of the probiotic. The exception was the presence of a smoker in the household; this was a significant risk factor for atopic eczema and atopic sensitivity (OR at 2 years: 2.48, 95% CI 1.08 to 5.70; p=0.033 and OR 2.25, 95% CI 1.16 to 4.37; p=0.017 respectively). This effect was independent of the treatment and did not significantly modify the univariate OR for the treatment effect. There was no evidence for an interaction effect between smoking and treatment allocation.

We have reported previously that probiotic administration was not associated with adverse effects in either mothers or their infants.[15]

DISCUSSION

Our findings do not support the administration of a multistrain, high-dose probiotic to mothers during late pregnancy and their infants from birth to age 6 months for the prevention of eczema in early childhood. However, probiotic administration was associated with a reduced frequency in sensitivity to food antigens and atopic eczema.

The strengths of our study are the relatively large number of mother-infant dyads recruited, the pragmatic nature of the trial, and confirmation of identity and viability of the probiotic organisms.

The study had several weaknesses. We had intended to evaluate clinical outcomes in infants at increased risk of atopy identified by having one or more first degree relatives with atopy. In practice, this was difficult to determine accurately at recruitment. However, the frequency of atopy and effects of the probiotic were similar in infants with and without a first degree relative with atopy. We expected that administering a novel intervention to healthy young infants would be difficult and, although compliance with the trial interventions in mothers was good, compliance was lower in infants. Finally, many mothers returned to work and this resulted in missed or delayed follow-up questionnaires and study clinic attendances. Although we compensated for missed follow-ups by subsequently asking parents and carers for information since the last assessment was done, this may have resulted in some loss of accuracy of information.

Our finding that the study probiotic did not affect the frequency of eczema is inconsistent with a meta-analysis of 13 randomised, placebo-controlled trials (3092 infants/children), where probiotic administration was associated with modest reductions in the frequency of eczema (fixed effects analysis; RR 0.79; 95% CI 0.71 to 0.88).[24] Despite agreement between studies in the meta-analysis (I^2=24.0%),[24] variability in outcomes between studies are likely to result from differences in the probiotics used,[25] the contribution of atopic and other pathogenic factors including variations in host mucosal and skin integrity,[5 26] the diagnostic criteria, and outcome measures used for eczema.[27]

Our observations of reduced frequency of skin prick sensitivity to food allergens and eczema in children with demonstrated allergic sensitivity in the probiotic arm are consistent with those of a meta-analysis (20 cohorts; 4031 participants, including our findings) where probiotic administration during infancy and childhood was associated with reduced serum IgE and atopic sensitisation to food or inhalant allergens in random effects analysis with limited heterogeneity between studies and without evidence of publication bias.[28]

In our study, in contrast to food allergens, sensitivity to aeroallergens was similar in the two arms. This tends to develop after infancy[29] and, with the exception of two studies,[30 31] most researchers have also reported that probiotic administration during early infancy did not reduce aeroallergen sensitivity.[25 32–39] Our observation that probiotic administration did not reduce the frequency of asthma or wheeze in young children is consistent with other studies.[28]

CONCLUSION

Administration of probiotics in early life may have a role in the prevention of atopic sensitisation. However, a better understanding of strain-specific probiotic effects[25 40] and how these address underlying atopic mechanisms is needed to guide the selection of strains for evaluation in clinical trials. Long-term follow-up to determine the effect of probiotic administration on the allergic march is also a priority.[41]

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