Clinical trial results:
Efficacy of Streptococcus salivarius K12 oral probiotic products in preventing acute otitis media: A randomized placebo-controlled trial.

Summary

EudraCT number	2020-001076-14
Trial protocol	FI
Global end of trial date	31 May 2021

Results information

Result version number	v1 (current)
This version publication date	25 August 2024
First version publication date	25 August 2024
Summary attachment (see zip file)	Article file (sarlin_2023_oi_231183_1698093542.53749.pdf)

Trial information

Trial identification

Sponsor protocol code	K12-2020

Additional study identifiers

ISRCTN number	-
ClinicalTrials.gov id (NCT number)	-
WHO universal trial number (UTN)	-

Notes:

Sponsors

Sponsor organisation name	Oulu University Hospital, Oulu, Finland
Sponsor organisation address	P.O. Box 23, Oulu, Finland,
Public contact	Suvi Sarlin, M.D., Suvi Sarlin/ Oulu University and Oulu Univ. Hospital, suvi.sarlin@oulu.fi
Scientific contact	Suvi Sarlin, M.D., Suvi Sarlin/ Oulu University and Oulu Univ. Hospital, suvi.sarlin@oulu.fi

Notes:

Paediatric regulatory details

Is trial part of an agreed paediatric investigation plan (PIP)	No
Does article 45 of REGULATION (EC) No 1901/2006 apply to this trial?	No
Does article 46 of REGULATION (EC) No 1901/2006 apply to this trial?	No

Notes:

Results analysis stage

Analysis stage	Final
Date of interim/final analysis	16 September 2023
Is this the analysis of the primary completion data?	Yes
Primary completion date	31 May 2021
Global end of trial reached?	Yes
Global end of trial date	31 May 2021
Was the trial ended prematurely?	No

Notes:

General information about the trial

Main objective of the trial:

Main objective is to investigate the clinical efficacy of Streptococcus salivarius K12 probiotic products in preventing acute otitis media (AOM) in children for 6 months.

Protection of trial subjects:

Standard according to Finnish laws and all reporting were performed according to CONSORT guidelines.

Background therapy: -

Evidence for comparator: -

Actual start date of recruitment	26 August 2020
Long term follow-up planned	Yes
Long term follow-up rationale	Scientific research
Long term follow-up duration	6 Months
Independent data monitoring committee (IDMC) involvement?	No

Notes:

Population of trial subjects

Subjects enrolled per country

Country: Number of subjects enrolled	Finland: 827
Worldwide total number of subjects	827
EEA total number of subjects	827

Notes:

Subjects enrolled per age group

In utero	0
Preterm newborn - gestational age < 37 wk	0
Newborns (0-27 days)	0
Infants and toddlers (28 days-23 months)	93
Children (2-11 years)	734
Adolescents (12-17 years)	0
Adults (18-64 years)	0
From 65 to 84 years	0
85 years and over	0

Subject disposition

Recruitment

Recruitment details:

Participants were recruited between August 26 and November 25, 2020 from daycare centers in the Oulu Region, Finland.

Pre-assignment

Screening details:

The exclusion criteria were ongoing antimicrobial prophylaxis or any immunodeficiency. Finnish language skill was an inclusion criterion. Study physicians visited day care centers to provide families with information about the study. Informed consent was required.

Period 1

Period 1 title	Study period (overall period)
Is this the baseline period?	Yes
Allocation method	Randomised - controlled
Blinding used	Double blind
Roles blinded	Subject, Investigator, Monitor, Data analyst, Carer, Assessor

Arms

Are arms mutually exclusive?	Yes
Arm title	Streptococcus salivarius K12

Arm description:

Streptococcus salivarius K12 oral soluble powder or tablet

Arm type	Experimental
Investigational medicinal product name	Streptococcus salivarius K12
Investigational medicinal product code	
Other name	
Pharmaceutical forms	Chewable/dispersible tablet
Routes of administration	Oral use

Dosage and administration details:

1 tablet in the evening after teeth brushing for 6 months.

Investigational medicinal product name	Streptococcus salivarius K12
Investigational medicinal product code	
Other name	
Pharmaceutical forms	Oral powder in sachet
Routes of administration	Oral use

Dosage and administration details:

1 sachet in the evening after teeth brushing for 6 months.

Arm title	Placebo

Arm description:

Placebo oral soluble powder or tablet

Arm type	Placebo
Investigational medicinal product name	Placebo oral soluble tablet
Investigational medicinal product code	
Other name	
Pharmaceutical forms	Chewable/dispersible tablet
Routes of administration	Oral use

Dosage and administration details:

1 tablet in the evening after teeth brushing for 6 months.

Investigational medicinal product name	Placebo oral soluble powder
Investigational medicinal product code	
Other name	
Pharmaceutical forms	Oral powder in sachet
Routes of administration	Oral use

Dosage and administration details:

1 sachet in the evening after teeth brushing for 6 months.

Number of subjects in period 1	Streptococcus salivarius K12	Placebo
Started	413	414
Completed	413	414

Baseline characteristics

Reporting groups	
Reporting group title	Streptococcus salivarius K12
Reporting group description:	
Streptococcus salivarius K12 oral soluble powder or tablet	
Reporting group title	Placebo
Reporting group description:	
Placebo oral soluble powder or tablet	

Reporting group values	Streptococcus salivarius K12	Placebo	Total
Number of subjects	413	414	827
Age categorical			
Units: Subjects			
In utero			0
Preterm newborn infants (gestational age < 37 wks)			0
Newborns (0-27 days)			0
Infants and toddlers (28 days-23 months)			0
Children (2-11 years)			0
Adolescents (12-17 years)			0
Adults (18-64 years)			0
From 65-84 years			0
85 years and over			0
Age continuous			
Age at recruitment			
Units: years			
arithmetic mean	4.1	4.1	
standard deviation	± 1.6	± 1.6	-
Gender categorical			
Units: Subjects			
Female	194	200	394
Male	219	214	433
Cystic fibrosis			
Cystic fibrosis (yes)			
Units: Subjects			
Cystic fibrosis (yes)	0	0	0
Cystic fibrosis (no)	413	414	827
Primary ciliary disease			
Primary ciliary disease (yes)			
Units: Subjects			
Primary ciliary disease (yes)	0	0	0
Primary ciliary disease, no	413	414	827
10-Valent PCV vaccination			
10-Valent PCV vaccination			
Units: Subjects			
10-Valent PCV vaccination (yes)	368	375	743
10-Valent PCV vaccination (no)	12	13	25

No questionnaire data	33	26	59
Any allergy Units: Subjects			
Allergy, yes	53	71	124
Allergy, no	327	317	644
No questionnaire data	33	26	59
Underlying illness requiring ongoing medication			
Underlying illness requiring ongoing medication			
Units: Subjects			
yes	15	21	36
no	365	367	732
No questionnaire data	33	26	59
Current use of other probiotic products			
Current use of other probiotic products			
Units: Subjects			
yes	59	59	118
no	321	329	650
No questionnaire data	33	26	59
Any antimicrobial therapy during the preceding 6 mo before study entry Units: Subjects			
Yes	43	47	90
No	337	341	678
No questionnaire data	33	26	59
Number of siblings Units: Subjects			
No siblings	68	64	132
One sibling	170	183	353
Two or more siblings	142	141	283
No questionnaire data	33	26	59
Parental smoking Units: Subjects			
Yes	77	68	145
No	303	320	623
No questionnaire data	33	26	59
Current pacifier use Units: Subjects			
Yes	20	22	42
No	360	366	726
No questionnaire data	33	26	59
Former pacifier use Units: Subjects			
Yes	262	272	534
No	118	116	234
No questionnaire data	33	26	59
Tympanostomy			
Previous tympanostomy			
Units: Subjects			
Yes	64	57	121
No	316	331	647
No questionnaire data	33	26	59

Presence of tympanostomy tubes Units: Subjects			
Yes	22	23	45
No	358	365	723
No questionnaire data	33	26	59
Adenoidectomy Units: Subjects			
Yes	24	25	49
No	356	363	719
No questionnaire data	33	26	59
Influenza vaccination Units: Subjects			
Yes	123	133	256
No	257	255	512
No questionnaire data	33	26	59
Duration of breastfeeding, months Units: month			
arithmetic mean	5.0	5.2	
standard deviation	± 4.0	± 4.6	-

End points

End points reporting groups

Reporting group title	Streptococcus salivarius K12
Reporting group description:	
Streptococcus salivarius K12 oral soluble powder or tablet	
Reporting group title	Placebo
Reporting group description:	
Placebo oral soluble powder or tablet	

Primary: One or more episode of acute otitis media requiring antibiotic therapy

End point title	One or more episode of acute otitis media requiring antibiotic therapy
End point description:	
End point type	Primary
End point timeframe:	
Entire study period, 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: subjects				
Yes	34	24		
No	379	390		

Statistical analyses

Statistical analysis title	One or more episode of AOM
Statistical analysis description:	
One or more episode of AOM requiring antibiotic therapy. From prescription register and medical record data	
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.17
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-2.44

Confidence interval	
level	95 %
sides	2-sided
lower limit	-5.94
upper limit	1.09

Statistical analysis title	95% CI of the difference
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	1.42
Confidence interval	
level	95 %
sides	2-sided
lower limit	0.86
upper limit	2.34

Primary: ≥1 Episode of AOM requiring antimicrobial therapy in oral powder group

End point title	≥1 Episode of AOM requiring antimicrobial therapy in oral powder group

End point description:

End point type	Primary

End point timeframe:

During the study period (6 months)

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	120	122		
Units: subjects				
Yes	24	11		
No	96	111		

Statistical analyses

Statistical analysis title	Oral powder subgroup ≥1 Episode of AOM
Comparison groups	Streptococcus salivarius K12 v Placebo

Number of subjects included in analysis	242
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.02
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-10.98
Confidence interval	
level	95 %
sides	2-sided
lower limit	-20.11
upper limit	-2.17

Statistical analysis title	≥1 Episode of AOM requiring antimicrobial therapy
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	242
Analysis specification	Pre-specified
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	2.22
Confidence interval	
level	95 %
sides	2-sided
lower limit	1.16
upper limit	4.3

Primary: ≥1 Episode of AOM requiring antimicrobial therapy, tablet group

End point title	≥1 Episode of AOM requiring antimicrobial therapy, tablet group
End point description:	
Chewable tablet group (292 in placebo and 293 in S. salivarius K12 groups)	
End point type	Primary
End point timeframe:	
During the entire study period (6months)	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	293	292		
Units: subjects				
Yes	10	13		
No	283	279		

Statistical analyses

Statistical analysis title	≥1 Episode of AOM, tablet group
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	585
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.52
Method	Wald test
Parameter estimate	proportion or mean difference
Point estimate	1.04
Confidence interval	
level	95 %
sides	2-sided
lower limit	-2.26
upper limit	4.46

Statistical analysis title	≥1 Episode of AOM, tablet group
Comparison groups	Placebo v Streptococcus salivarius K12
Number of subjects included in analysis	585
Analysis specification	Pre-specified
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	0.77
Confidence interval	
level	95 %
sides	2-sided
lower limit	0.35
upper limit	1.68

Secondary: Recurrent otitis media, ≥2 AOM episodes in 6 mo	
End point title	Recurrent otitis media, ≥2 AOM episodes in 6 mo
End point description: National prescription register and medical record data	
End point type	Secondary
End point timeframe: 6 months, during the study	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: subjects				
Yes	9	6		
No	404	408		

Statistical analyses

Statistical analysis title	≥2 AOM episodes in 6 mo
Comparison groups	Placebo v Streptococcus salivarius K12
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.33
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-0.72
Confidence interval	
level	95 %
sides	2-sided
lower limit	-2.81
upper limit	1.22

Statistical analysis title	Recurrent otitis media, ≥2 AOM episodes in 6 mo
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	1.5
Confidence interval	
level	95 %
sides	2-sided
lower limit	0.56
upper limit	4.02

Secondary: ≥3 AOM episodes in 6 mo

End point title	≥3 AOM episodes in 6 mo

End point description:

National prescription register and medical record data

End point type	Secondary

End point timeframe:

During the study period, 6 months

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: subjects				
Yes	4	2		
No	409	412		

Statistical analyses

Statistical analysis title	≥3 AOM episodes in 6 mo
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	2
Confidence interval	
level	95 %
sides	2-sided
lower limit	0.43
upper limit	9.32

Statistical analysis title	≥3 AOM episodes in 6 mo
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.41
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-0.49
Confidence interval	
level	95 %
sides	2-sided
lower limit	-1.81
upper limit	0.84

Secondary: Any antimicrobial therapy

End point title	Any antimicrobial therapy
End point description: National prescription register and medical record data	
End point type	Secondary
End point timeframe: During the study, 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: subjects				
Yes	49	37		
No	364	377		

Statistical analyses

Statistical analysis title	Any antimicrobial therapy
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	1.33
Confidence interval	
level	95 %
sides	2-sided
lower limit	0.89
upper limit	1.99

Statistical analysis title	Any antimicrobial therapy
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.17
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-2.93

Confidence interval	
level	95 %
sides	2-sided
lower limit	-7.09
upper limit	1.27

Secondary: ≥1 Physician’s appointment due to acute illness	
End point title	≥1 Physician’s appointment due to acute illness
End point description:	
National prescription register and medical record data	
End point type	Secondary
End point timeframe:	
During the study period, 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: subjects				
Yes	85	93		
No	328	321		

Statistical analyses

Statistical analysis title	≥1 Physician’s appointment due to acute illness
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	0.92
Confidence interval	
level	95 %
sides	2-sided
lower limit	0.71
upper limit	1.19

Statistical analysis title	≥1 Physician’s appointment due to acute illness
Comparison groups	Streptococcus salivarius K12 v Placebo

Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.51
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	1.88
Confidence interval	
level	95 %
sides	2-sided
lower limit	-3.73
upper limit	7.47

Secondary: Telephone or video appointment	
End point title	Telephone or video appointment
End point description: National prescription register and medical record data	
End point type	Secondary
End point timeframe: During the study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: subjects				
Yes	47	32		
No	366	382		

Statistical analyses

Statistical analysis title	Telephone or video appointment
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	1.47
Confidence interval	
level	95 %
sides	2-sided
lower limit	0.96
upper limit	2.25

Statistical analysis title	Telephone or video appointment
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.07
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-3.65
Confidence interval	
level	95 %
sides	2-sided
lower limit	-7.66
upper limit	0.39

Secondary: Hospitalized for an acute respiratory illness	
End point title	Hospitalized for an acute respiratory illness
End point description: National prescription register and medical record data	
End point type	Secondary
End point timeframe: During the study period, 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: subjects				
Yes	4	0		
No	409	414		

Statistical analyses

Statistical analysis title	Hospitalized for an acute respiratory illness
Comparison groups	Placebo v Streptococcus salivarius K12

Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	0
Confidence interval	
level	95 %
sides	2-sided
lower limit	0
upper limit	0.97

Statistical analysis title	Hospitalized for an acute respiratory illness
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.05
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-0.97
Confidence interval	
level	95 %
sides	2-sided
lower limit	-2.11
upper limit	0.19

Secondary: Duration of hospitalization	
End point title	Duration of hospitalization
End point description:	
National prescription register and medical record data	
End point type	Secondary
End point timeframe:	
During the study period, 6months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: day				
median (inter-quartile range (Q1-Q3))	3 (2 to 3)	0 (0 to 0)		

Statistical analyses

Statistical analysis title	Duration of hospitalization
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	> 0.05 [1]
Method	Wilcoxon (Mann-Whitney)

Notes:

[1] - Could not be calculated, because the number of cases in the placebo group was zero

Secondary: No. of AOM episodes per PYR	
End point title	No. of AOM episodes per PYR
End point description: Questionnaire data	
End point type	Secondary
End point timeframe: During the study period	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[2]	414[3]		
Units: Number of AOM episodes				
median (inter-quartile range (Q1-Q3))				
No. of AOM episodes per PYR	0.00 (0.00 to 0.00)	0.00 (0.00 to 0.00)		

Notes:

[2] - Please see the availability of no. questionnaires per month in the article.

[3] - Please see the no. available questionnaire data per month in the article.

Statistical analyses

Statistical analysis title	No. of AOM episodes per PYR
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	> 0.16
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-0.14
Confidence interval	
level	95 %
sides	2-sided
lower limit	-0.34
upper limit	0.07

Secondary: No. of days with respiratory or gastroenteritis symptoms per PYR, runny nose	
End point title	No. of days with respiratory or gastroenteritis symptoms per PYR, runny nose
End point description: Questionnaire data	
End point type	Secondary
End point timeframe: During the study period 6months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[4]	414[5]		
Units: days				
median (inter-quartile range (Q1-Q3))	21.90 (6.08 to 44.61)	21.29 (7.60 to 40.96)		

Notes:

[4] - Please see the no. of available monthly questionnaire data in the article.

[5] - Please see the no. of available monthly questionnaire data in the article.

Statistical analyses

Statistical analysis title	Runny nose
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.96
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	95% CI of the difference
Point estimate	-1.5
Confidence interval	
level	95 %
sides	2-sided
lower limit	-7.13
upper limit	4.06

Secondary: No. days with cough per PYR	
End point title	No. days with cough per PYR
End point description: Questionnaire data	
End point type	Secondary

End point timeframe:
During the study period 6 months.

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[6]	414[7]		
Units: days				
median (inter-quartile range (Q1-Q3))	8.11 (0.00 to 26.36)	8.11 (0.00 to 29.30)		

Notes:
[6] - Please see the no. of available monthly questionnaire data in the article.
[7] - Please see the no. of available monthly questionnaire data in the article.

Statistical analyses

Statistical analysis title	Cough
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Risk ratio (RR)
Point estimate	1.1
Confidence interval	
level	95 %
sides	2-sided
lower limit	-3.88
upper limit	6.15

Secondary: Number of days with sore throat per PYR

End point title	Number of days with sore throat per PYR
End point description: Questionnaire data	
End point type	Secondary
End point timeframe: During the entire study 6months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[8]	414[9]		
Units: Days				
median (inter-quartile range (Q1-Q3))	0.00 (0.00 to 4.06)	0.00 (0.00 to 0.46)		

Notes:
[8] - Please see the no. monthly questionnaires per month in the article.
[9] - Please see the number of available questionnaires per month in the article.

Statistical analyses

Statistical analysis title	Sore throat
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.85
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	0.12
Confidence interval	
level	95 %
sides	2-sided
lower limit	-1.57
upper limit	1.8

Secondary: No. days with gastrointestinal symptoms or pain per PYR	
End point title	No. days with gastrointestinal symptoms or pain per PYR
End point description: Questionnaire data	
End point type	Secondary
End point timeframe: During the entire study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[10]	414[11]		
Units: days				
median (inter-quartile range (Q1-Q3))	0.00 (0.00 to 2.03)	0.00 (0.00 to 2.03)		

Notes:
[10] - Please see the no. available questionnaires per month in the article
[11] - Please see the no. available questionnaires per month in the article.

Statistical analyses

Statistical analysis title	Gastrointestinal discomfort or pain
Comparison groups	Streptococcus salivarius K12 v Placebo

Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.42
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	0.36
Confidence interval	
level	95 %
sides	2-sided
lower limit	-1.63
upper limit	2.35

Secondary: Number of days with fever per PYR

End point title	Number of days with fever per PYR
End point description: Questionnaire data	
End point type	Secondary
End point timeframe: During the study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[12]	414[13]		
Units: days				
median (inter-quartile range (Q1-Q3))	0.00 (0.00 to 0.00)	0.00 (0.00 to 0.00)		

Notes:

[12] - Please see the no. questionnaires per month in the article.

[13] - Please see the no. questionnaires per month in the article.

Statistical analyses

Statistical analysis title	Number of days with fever per PYR
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.6
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-0.3

Confidence interval	
level	95 %
sides	2-sided
lower limit	-1.14
upper limit	0.53

Secondary: Number of days with earache per PYR

End point title	Number of days with earache per PYR
End point description:	
Questionnaire data	
End point type	Secondary
End point timeframe:	
During the entire study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[14]	414[15]		
Units: days				
median (inter-quartile range (Q1-Q3))	0.00 (0.00 to 0.00)	0.00 (0.00 to 0.00)		

Notes:

[14] - Please see the no. questionnaires per month in the article.

[15] - Please see the no. questionnaires per month in the article.

Statistical analyses

Statistical analysis title	No. days with earache per PYR
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.2
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-0.87
Confidence interval	
level	95 %
sides	2-sided
lower limit	-2.18
upper limit	0.44

Secondary: No. days with diarrhea per PYR

End point title	No. days with diarrhea per PYR

End point description: Questionnaire data	
End point type	Secondary
End point timeframe: During the entire study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[16]	414[17]		
Units: Days				
median (inter-quartile range (Q1-Q3))	0 (0.00 to 0.00)	0.00 (0.00 to 0.00)		

Notes:

[16] - Please see the no. monthly questionnaires in the article

[17] - Please see the no. questionnaires per month in the article.

Statistical analyses

Statistical analysis title	No. days with diarrhea per PYR
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.77
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	0.36
Confidence interval	
level	95 %
sides	2-sided
lower limit	-0.99
upper limit	1.71

Secondary: No. days with wheezing per PYR	
End point title	No. days with wheezing per PYR
End point description: Questionnaire data	
End point type	Secondary
End point timeframe: During the entire study period 6 months.	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[18]	414[19]		
Units: days				
median (inter-quartile range (Q1-Q3))	0.00 (0.00 to 0.00)	0.00 (0.00 to 0.00)		

Notes:

[18] - Please see the number of questionnaires per month in the article

[19] - Please see the no. questionnaires per month in the article.

Statistical analyses

Statistical analysis title	No. days with wheezing per PYR
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.83
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	0.08
Confidence interval	
level	95 %
sides	2-sided
lower limit	-0.78
upper limit	0.62

Secondary: No. days with vomiting per PYR

End point title	No. days with vomiting per PYR
End point description: Questionnaire data	
End point type	Secondary
End point timeframe: During the entire study period 6months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[20]	414[21]		
Units: days				
median (inter-quartile range (Q1-Q3))	0.00 (0.00 to 0.00)	0.00 (0.00 to 0.00)		

Notes:

[20] - Please see the no. questionnaires per month in the article

[21] - Please see the no. questionnaires per month in the article.

Statistical analyses

Statistical analysis title	No. days with vomiting per PYR
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.41
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	0.05
Confidence interval	
level	95 %
sides	2-sided
lower limit	-0.23
upper limit	0.33

Secondary: No. of days of parental absence from work due to the child's acute respiratory illness per PYR

End point title	No. of days of parental absence from work due to the child's acute respiratory illness per PYR
End point description:	
Questionnaire data	
End point type	Secondary
End point timeframe:	
During the entire study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[22]	414[23]		
Units: days				
median (inter-quartile range (Q1-Q3))	4.06 (0.00 to 16.22)	6.08 (0.00 to 20.28)		

Notes:

[22] - Please see the no. questionnaires per month in the article.

[23] - Please see the no. questionnaires per month in the article.

Statistical analyses

Statistical analysis title	No. of days of parental absence from work
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.38
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-0.56

Confidence interval	
level	95 %
sides	2-sided
lower limit	-2.08
upper limit	3.19

Secondary: Time to first episode of AOM

End point title	Time to first episode of AOM

End point description:

End point type	Secondary

End point timeframe:

During the study period 6 months

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: Days				
arithmetic mean (confidence interval 95%)	174 (171 to 177)	176 (173 to 179)		

Statistical analyses

Statistical analysis title	Kaplan-Meier analysis
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority[24]
P-value	= 0.18
Method	Kaplan-Meier analysis

Notes:

[24] - Please see Figure 2. in the article

Statistical analysis title	t-test
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Pre-specified
Analysis type	superiority
P-value	= 0.33
Method	t-test, 2-sided

Post-hoc: Children with AOM with a complication	
End point title	Children with AOM with a complication
End point description: Questionnaire data	
End point type	Post-hoc
End point timeframe: During the study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[25]	414[26]		
Units: Subjects	2	1		

Notes:

[25] - Please see the no. questionnaires per month in the article.

[26] - Please see the no. questionnaires per month in the article.

Statistical analyses

Statistical analysis title	Children with AOM with a complication
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Post-hoc
Analysis type	superiority
P-value	= 0.77
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-1.7
Confidence interval	
level	95 %
sides	2-sided
lower limit	-15.84
upper limit	15.26

Statistical analysis title	Children with AOM with a complication
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Post-hoc
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	1.41

Confidence interval	
level	95 %
sides	2-sided
lower limit	0.2
upper limit	10.51

Post-hoc: Children with tympanostomy tube insertion or adenoidectomy after study entry

End point title	Children with tympanostomy tube insertion or adenoidectomy after study entry
End point description: Patient register data	
End point type	Post-hoc
End point timeframe: During the entire study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413	414		
Units: Subjects	4	4		

Statistical analyses

Statistical analysis title	Children with tympanostomy tube insertion or..
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Post-hoc
Analysis type	superiority
P-value	= 0.99
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	0
Confidence interval	
level	95 %
sides	2-sided
lower limit	-2.27
upper limit	2.23

Statistical analysis title	Children with tympanostomy tube insertion or..
Comparison groups	Streptococcus salivarius K12 v Placebo

Number of subjects included in analysis	827
Analysis specification	Post-hoc
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	0
Confidence interval	
level	95 %
sides	2-sided
lower limit	-0.16
upper limit	0.16

Post-hoc: No. of days of child’s absence from day care due to symptoms of infection per PYR

End point title	No. of days of child’s absence from day care due to symptoms of infection per PYR
End point description: Questionnaire data	
End point type	Post-hoc
End point timeframe: During the entire study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	270	273		
Units: Days				
median (inter-quartile range (Q1-Q3))	14.19 (4.06 to 30.42)	14.90 (4.06 to 28.39)		

Statistical analyses

Statistical analysis title	No. of days of child’s absence from day care
Statistical analysis description: Questionnaire data	
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	543
Analysis specification	Post-hoc
Analysis type	superiority
P-value	= 0.73
Method	Wilcoxon (Mann-Whitney)
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	-1.9

Confidence interval	
level	95 %
sides	2-sided
lower limit	-5.33
upper limit	1.54

Post-hoc: Children with respiratory symptoms leading to SARS-CoV-2 testing	
End point title	Children with respiratory symptoms leading to SARS-CoV-2 testing
End point description: Questionnaire data	
End point type	Post-hoc
End point timeframe: During the entire study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[27]	414[28]		
Units: subjects	184	188		

Notes:

[27] - Please see the number of questionnaires per month in the article

[28] - Please see the no. questionnaires per month in the article.

Statistical analyses

Statistical analysis title	Children with respiratory symptoms leading to..
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Post-hoc
Analysis type	superiority
P-value	= 0.8
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	0.9
Confidence interval	
level	95 %
sides	2-sided
lower limit	-5.91
upper limit	7.62

Statistical analysis title	Children with resp. symptoms leading to..
Comparison groups	Streptococcus salivarius K12 v Placebo

Number of subjects included in analysis	827
Analysis specification	Post-hoc
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	1.02
Confidence interval	
level	95 %
sides	2-sided
lower limit	0.88
upper limit	1.19

Post-hoc: Positive SARS-CoV-2 test result	
End point title	Positive SARS-CoV-2 test result
End point description: Questionnaire data	
End point type	Post-hoc
End point timeframe: During the entire study period 6 months	

End point values	Streptococcus salivarius K12	Placebo		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	413[29]	414[30]		
Units: subjects	2	3		

Notes:

[29] - Please see the no. questionnaire per month in the article

[30] - Please see the no. questionnaires per month in the article.

Statistical analyses

Statistical analysis title	Positive SARS-CoV-2 test result
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Post-hoc
Analysis type	superiority
P-value	= 0.66
Method	Wald test
Parameter estimate	Proportion or mean difference (95% CI)
Point estimate	0.24
Confidence interval	
level	95 %
sides	2-sided
lower limit	-1
upper limit	1.48

Statistical analysis title	Positive SARS-CoV-2 test result
Comparison groups	Streptococcus salivarius K12 v Placebo
Number of subjects included in analysis	827
Analysis specification	Post-hoc
Analysis type	superiority
Parameter estimate	Risk ratio (RR)
Point estimate	1.5
Confidence interval	
level	95 %
sides	2-sided
lower limit	0.3
upper limit	7.49

Adverse events

Adverse events information	
Timeframe for reporting adverse events:	
During the study period, 6 months	
Adverse event reporting additional description:	
The recruited families reported suspected side-effects either via email, telephone or questionnaire.	
Assessment type	Systematic

Dictionary used	
Dictionary name	No dictionary
Dictionary version	0

Reporting groups	
Reporting group title	Streptococcus salivarius K12
Reporting group description:	
Streptococcus salivarius K12 oral soluble powder or tablet	
Reporting group title	Placebo
Reporting group description:	
Placebo oral soluble powder or tablet	

Serious adverse events	Streptococcus salivarius K12	Placebo	
Total subjects affected by serious adverse events			
subjects affected / exposed	0 / 413 (0.00%)	0 / 414 (0.00%)	
number of deaths (all causes)	0	0	
number of deaths resulting from adverse events	0	0	

Frequency threshold for reporting non-serious adverse events: 5 %

Non-serious adverse events	Streptococcus salivarius K12	Placebo	
Total subjects affected by non-serious adverse events			
subjects affected / exposed	5 / 413 (1.21%)	4 / 414 (0.97%)	
Gastrointestinal disorders			
Pain	Additional description: Stomach pain		
subjects affected / exposed	0 / 413 (0.00%)	1 / 414 (0.24%)	
occurrences (all)	0	1	
Breath odour	Additional description: Foul breath		
subjects affected / exposed	3 / 413 (0.73%)	1 / 414 (0.24%)	
occurrences (all)	3	1	
Respiratory, thoracic and mediastinal disorders			

Cough subjects affected / exposed occurrences (all)	0 / 413 (0.00%) 0	1 / 414 (0.24%) 1	
Skin and subcutaneous tissue disorders Rash subjects affected / exposed occurrences (all)	2 / 413 (0.48%) 2	1 / 414 (0.24%) 1	

More information

Substantial protocol amendments (globally)

Were there any global substantial amendments to the protocol? Yes

Date	Amendment
08 September 2020	Due to COVID-19 pandemic the incidence of AOM dropped significantly. The sample size was recalculated. Later, small changes were added to the outcomes, but the study period and protocol was preserved.

Notes:

Interruptions (globally)

Were there any global interruptions to the trial? No

Limitations and caveats

None reported

Online references

http://www.ncbi.nlm.nih.gov/pubmed/37917062