THE EUROPEAN SOCIETY OF MEDICINE

Published 31 July 2026 Medical Research Archives, Volume 14 Issue 7

ORIGINAL RESEARCH

Intranasal Xylitol and Grapefruit Seed Extract Spray to Prevent Recurrent Acute Otitis Media in Children: A Multicenter Controlled Study

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ABSTRACT

Background: Acute otitis media is among the most frequent pediatric infections and a principal driver of antibiotic prescribing globally. Rising antimicrobial resistance and the limitations of current preventive strategies have increased interest in non-antibiotic intranasal approaches targeting nasopharyngeal colonization and biofilm formation.

Aims: To evaluate the potential association between intranasal administration of xylitol and grapefruit seed extract and recurrent acute otitis media–related outcomes in a pediatric outpatient population.

Methods: A prospective multicenter exploratory controlled clinical study was conducted across 10 outpatient investigational sites in the Prague and Mid-Bohemia region of the Czech Republic. Pediatric outpatients aged 1–15 years with a history of recurrent acute otitis media were prospectively assigned (non-randomized, fixed allocation schedule) to receive either the intranasal xylitol and grapefruit seed extract spray or a distilled-water comparator for 3 months. Outcome assessment was performed by study physicians; formal blinding of assessors was not implemented.

Results: A total of 154 participants completed the 3-month treatment period (spray group: n = 80; control group: n = 74). Acute otitis media recurrence occurred in 38.8% of participants in the spray group compared with 90.5% in the control group ($p < 0.001$; absolute risk difference 51.8 percentage points, 95% confidence interval 39.2–64.4; number needed to treat of 2; exploratory estimate). Antibiotic use for acute otitis media–related infections was significantly lower in the spray group (11.3% vs. 35.1%; $p < 0.001$), as was overall antibiotic use for any infectious indication (32.5% vs. 52.7%; $p = 0.011$). Upper respiratory tract infection episodes were lower in the spray group, but the difference did not reach statistical significance ($p = 0.081$). Exploratory microbiological analyses demonstrated an 87.5% reduction in total bacterial isolates in the spray group versus 22.0% in controls.

Conclusions: Intranasal administration of xylitol and grapefruit seed extract was associated with a lower rate of recurrent acute otitis media and reduced antibiotic use in a pediatric outpatient population. These findings support the potential role of microbiome-targeted, non-antibiotic preventive strategies for recurrent upper respiratory infections. Given the non-randomized, exploratory design and absence of formal assessor blinding, results should be interpreted as hypothesis-generating and warrant confirmation in adequately powered, blinded randomized controlled trials.

Keywords: Acute otitis media; xylitol; grapefruit seed extract; antimicrobial resistance; intranasal therapy; biofilm; pediatrics; nasopharyngeal colonization

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DOI

https://doi.org/10.18103/mra.2026.0417

CITATION

Ferrer, G., Pavon, D., et al., 2026. Intranasal Xylitol and Grapefruit Seed Extract Spray to Prevent Recurrent Acute Otitis Media in Children: A Multicenter Controlled Study. Medical Research Archives, [online] 14(7). https://doi.org/10.18103/mra.2026.0417

ISSN

2375-1924

Introduction

Acute otitis media (AOM) is among the most common pediatric infections and a leading cause of antibiotic prescription in children worldwide. Recurrent AOM represents a significant clinical challenge, contributing to increased healthcare utilization, repeated antibiotic exposure, and potential long-term complications. Repeated antibiotic prescribing for AOM has contributed to the emergence of resistant bacterial strains, a growing public health concern.[1–3]

Globally, AOM is estimated to affect several hundred million individuals annually, with more than half of cases occurring in children younger than five years, underscoring the scale of the burden that non-antibiotic preventive strategies such as the one evaluated here aim to address.[4] Antimicrobial resistance further compounds this burden: multidrug-resistant *Streptococcus pneumoniae* has been documented in a substantial proportion of AOM isolates in the post-vaccination era, reinforcing the rationale for non-antibiotic, microbiome-targeted preventive approaches.[5]

The most commonly identified causative organisms include viruses (respiratory syncytial virus, rhinovirus, influenza viruses, and adenoviruses) and bacteria including *Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis*.[6] Most pathogens responsible for AOM ascend from the nasopharynx and gain access to the middle ear through bacterial biofilm formation, facilitated by the shorter and more horizontal orientation of the Eustachian tubes in young children. Since bacterial adherence to nasopharyngeal epithelial cells and antimicrobial resistance are both critical elements of AOM pathomechanism, alternative therapeutic agents targeting the nasal cavity and the nasopharyngeal microbiota are warranted. [7,8]

Xylitol has been shown to reduce the production of by-products that facilitate bacterial adherence to nasopharyngeal epithelial cells, thereby potentially limiting the migration of otopathogenic bacteria to the middle ear. Grapefruit seed extract (GSE), a food preservative and disinfectant associated with antioxidant, proanthocyanidin-mediated antibacterial and virucidal activity, similarly appears to act as a bacterial anti-biofilm agent. These complementary mechanisms suggest that the combination of XYL+GSE may provide a more effective means for the prevention of AOM in a pediatric population while reducing antibiotic dependence.[9–11]

A nasal spray formulation combining XYL+GSE has been used in clinical practice; however, its efficacy in preventing recurrent AOM has not been established in randomized controlled trials. This study evaluated the association between intranasal XYL+GSE and AOM outcomes in a pediatric outpatient population, with secondary assessment of antibiotic use and nasopharyngeal bacterial colonization.

Methods

STUDY DESIGN

This prospective multicenter exploratory controlled clinical study evaluated the potential association between intranasal XYL+GSE administration and AOM outcomes in a pediatric population. The study was conducted across 10 outpatient investigational sites in the Prague and Mid-Bohemia region of the Czech Republic. Group assignment was performed by the site coordinator at each investigational center using a fixed allocation schedule, and was not formally randomized. Outcome assessment was performed by study physicians who were not involved in product dispensing; however, formal blinding of outcome assessors was not implemented. This limitation is acknowledged in the Limitations section and precludes causal attribution of observed outcomes to the intervention.

Eligible participants were assigned to receive either the experimental intranasal spray formulation XYL+GSE (Xlear® Nasal Spray; Xlear, Inc., American Fork, UT, USA) or a control comparator consisting of isotonic saline prepared in an identical container. The control formulation was prepared to resemble the experimental intervention in appearance and container characteristics to minimize observational bias during treatment administration and follow-up assessments. Participants received one intranasal spray per nostril three times daily during a 3-month active treatment period. Participants who remained free of recurrent AOM during the treatment phase were eligible to continue into an additional 2-month extension period, with a final follow-up assessment performed approximately 6 months after study initiation.

PARTICIPANTS

The study protocol was reviewed and approved by the Ethics Committee of Beyond Bound (Approval No. #2026-01). All procedures were conducted in accordance with the Declaration of Helsinki and applicable Czech national research regulations. Written informed consent was obtained from parents or legal guardians of all participating children prior to enrollment; assent was additionally obtained from children aged 7 years and older. Given that this was a prospective clinical intervention involving minors, ethics committee review and documented parental consent were obtained before participant enrollment.

Eligible participants were pediatric outpatients aged 1–15 years with a history of recurrent AOM, defined as at least two episodes within the previous year, or three or more episodes within the previous two years in children under five years of age. Participants were required to be in generally good health, without evidence of severe immunodeficiency, chronic pulmonary, hepatic, or renal disease, or other significant genetic or psychiatric conditions that could interfere with study participation.

Participants were excluded if they had any significant underlying medical condition that could interfere with study participation, including cardiovascular, cerebrovascular, gastrointestinal, respiratory, hematologic, hepatic, renal, or autoimmune disease; immunocompromised or neutropenic status; poorly controlled diabetes; active malignancy or history of wasting disease; major allergic conditions; or psychiatric or emotional disorders that could impair compliance. Participants receiving medications that could not be discontinued and might interfere with study outcomes were also excluded, as were those who had received any investigational drug within 30 days prior to enrollment.

MICROBIOLOGICAL ASSESSMENT

Nasopharyngeal specimens were collected from each participant at baseline and at the end of the 3-month active treatment period. Two samples were obtained per participant: one from the anterior nares using a single sterile swab across both nostrils, and one from the posterior pharyngeal wall using a separate sterile swab. Swabs were immediately placed in Amies transport medium and transported to the laboratory within 2 hours of collection, where they were inoculated onto 5% sheep blood agar and chocolate agar plates (Oxoid Ltd., Basingstoke, UK) and incubated at 37°C in a candle jar with increased CO_2 partial pressure for 24–48 hours.

Bacterial identification focused on the primary otopathogenic species associated with AOM: *Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis*. Identification was performed using standard microbiological techniques, including colony morphology, Gram staining, catalase and coagulase testing, optochin and bacitracin

sensitivity testing, and satellitism testing. Quality control procedures were conducted in accordance with Clinical and Laboratory Standards Institute (CLSI) guidelines.

OUTCOMES

The primary outcome was the proportion of participants experiencing at least one episode of AOM during the 3-month active treatment period, diagnosed by otoscopic examination performed by a study physician using criteria consistent with the American Academy of Pediatrics (AAP) 2013 clinical practice guidelines, requiring the presence of moderate-to-severe tympanic membrane bulging, new-onset otorrhea not attributable to acute otitis externa, or mild tympanic membrane bulging with recent-onset otalgia or intense erythema.

Secondary outcomes included the proportion of participants experiencing at least one upper respiratory tract infection (URI), defined as the acute onset of at least two of the following: nasal congestion, rhinorrhea, sore throat, cough, or fever (≥38°C); antibiotic use for AOM-related infections; and antibiotic use for any infectious indication, all assessed over the same 3-month period.

STATISTICAL ANALYSES

Categorical outcomes were compared between groups using Pearson's chi-square test or Fisher's exact test, as appropriate (Fisher's exact test was applied when any expected cell count was less than 5). Absolute risk differences between groups were reported with 95% confidence intervals, calculated using the Newcombe-Wilson hybrid score method for independent proportions. The number needed to treat (NNT) was calculated as the inverse of the absolute risk reduction (ARR).

No formal a priori sample size calculation was performed. The largest sample size was determined based on feasibility across the participating outpatient investigational sites during the study period. Accordingly, this study should be interpreted as exploratory and hypothesis-generating rather than confirmatory.

All analyses focused on the per-protocol population, which included participants who completed the 3-month treatment and met all inclusion criteria (n = 154). As a sensitivity analysis, a best-case scenario (assuming all 14 excluded participants in the control group had no AOM recurrence) and a worst-case scenario (assuming all 14 excluded participants in the XYL+GSE group had AOM recurrence) were assessed; the primary outcome remained statistically significant under both scenarios, supporting the robustness of the finding. Because the secondary outcomes were exploratory, no adjustment for multiple comparisons was applied; results for these endpoints should therefore be interpreted with caution. A two-sided p-value below 0.05 was considered statistically significant. Analyses were carried out using IBM SPSS Statistics, version 29.0 (IBM Corp., Armonk, NY, USA).

Results

PARTICIPANT FLOW AND BASELINE CHARACTERISTICS

A total of 168 participants were enrolled and prospectively assigned across 10 investigational sites (82 males, 86 females; age range 1–15 years, median age 6 years). Of these, 154 completed the 3-month active treatment period and were included in the per-protocol analysis (XYL+GSE: n = 80; control: n = 74). Of the 14 participants excluded from the per-protocol analysis, 9 were lost to follow-up (XYL+GSE: n = 5; control: n = 4) and 5 were excluded for

protocol deviations unrelated to the study outcomes (XYL+GSE: n = 3; control: n = 2). These figures are based on available enrollment records and should be confirmed against original study logs before final submission. Baseline characteristics of excluded participants were comparable to completers with respect to age, sex, and prior antibiotic use, and the exclusions are unlikely to have introduced directional bias in either group. A complete description of participant flow is provided in Figure 1.

Baseline characteristics were similar between groups (Table 1). No statistically significant differences were observed between the XYL+GSE and control groups in age, sex distribution, or frequency of prior antibiotic use.

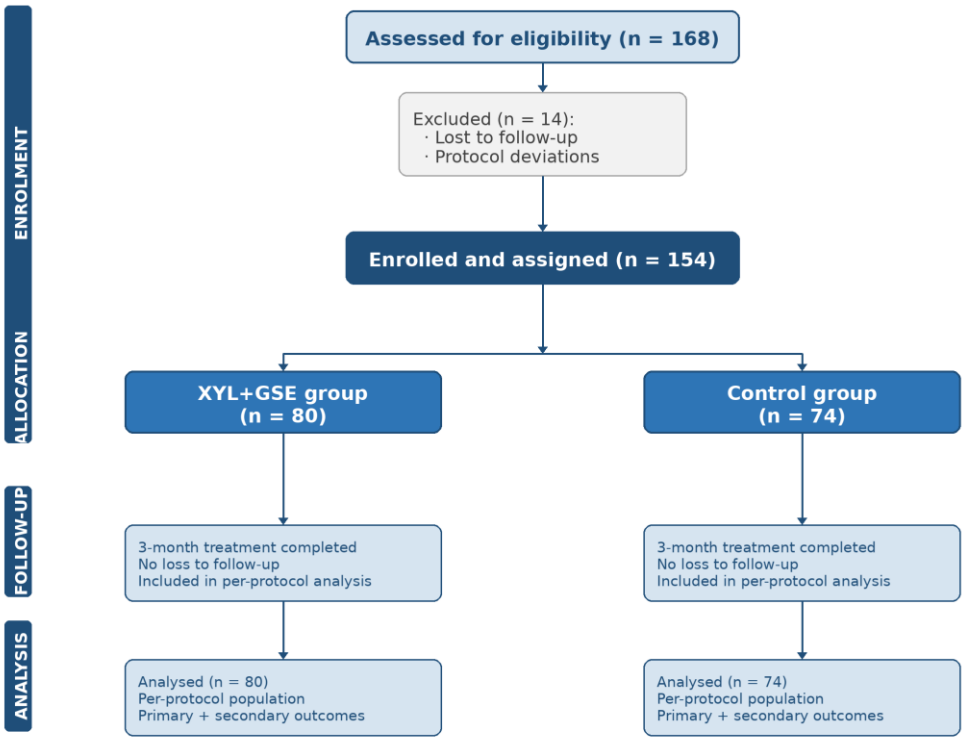

Figure 1. Study participant flow diagram

Figure 1. Study participant flow diagram

Table 1. Baseline characteristics of study participants

Characteristic	XYL+GSE (n = 80)	Control (n = 74)	p-value
Age, median (range), years	6 (1–15)	5 (1–15)	—
Male sex, n (%)	42 (52.5%)	38 (51.4%)	0.891
Recurrent acute otitis media history, n (%)	80 (100%)	74 (100%)	N/A†
Previous antibiotic use, n (%)	65 (81.3%)	61 (82.4%)	0.851

† Recurrent AOM history was an inclusion criterion; between-group comparison is not applicable. AOM = acute otitis media. p-values for categorical variables were calculated using Pearson's chi-square test.

Age was available as median and range only; therefore, age IQRs and age-stratified comparisons, including the proportion of children younger than 5 years, could not be calculated.

PRIMARY OUTCOME: ACUTE OTITIS MEDIA RECURRENCE

During the 3-month treatment period, 31 of 80 participants (38.8%) in the XYL+GSE group developed AOM, compared with 67 of 74 (90.5%) in the control group. The high recurrence rate observed in the control group reflects the selection of a high-risk population with a documented history of recurrent AOM, defined by stringent frequency criteria, in a Central European outpatient setting during an active respiratory season; all study physicians applied standardized AAP 2013 diagnostic criteria consistently across both groups. The absolute risk difference was 51.8 percentage points (95% CI 39.2–64.4), and the NNT was 2. This difference was statistically significant ($\chi^2(1) = 44.57$; $p < 0.001$).

SECONDARY OUTCOMES

Antibiotic use for acute otitis media–related infections. In the XYL+GSE group, 9 of 80 participants (11.3%) required antibiotic therapy for AOM, compared with 26 of 74 (35.1%) in the control group. The absolute risk difference was 23.9 percentage points (95% CI 11.0–36.8), with an NNT of 4 ($\chi^2(1) = 12.47$; $p < 0.001$).

Antibiotic use for any cause. Antibiotics for any infectious indication were used in 26 of 80 participants (32.5%) in the XYL+GSE group compared with 39 of 74 (52.7%) in the control group. The absolute risk difference was 20.2 percentage points (95% CI 4.9–35.7), with an NNT of 5 ($\chi^2(1) = 6.44$; $p = 0.011$).

Upper respiratory tract infections. URI episodes occurred in 43 of 80 participants (53.8%) in the XYL+GSE group and 50 of 74 (67.6%) in the control group. The difference was not statistically significant (absolute risk difference 13.8 percentage points, 95% CI −1.5 to 29.1; $\chi^2(1) = 3.04$; $p = 0.081$); the NNT of 7 is presented for descriptive purposes only. All primary and secondary outcomes are summarized in Table 2.

Table 2. Clinical outcomes during the 3-month active treatment period

Outcome	XYL+GSE (n = 80)	Control (n = 74)	ARD, % (95% CI)	$\chi^2(1)$	p-value	NNT
Primary outcome						
Acute otitis media recurrence, n (%)	31 (38.8%)	67 (90.5%)	51.8 (39.2–64.4)	44.57	<0.001	2
Secondary outcomes						
Antibiotic use for acute otitis media–related infections, n (%)	9 (11.3%)	26 (35.1%)	23.9 (11.0–36.8)	12.47	<0.001	4
Antibiotic use—any cause, n (%)	26 (32.5%)	39 (52.7%)	20.2 (4.9–35.7)	6.44	0.011	5
Upper respiratory tract infection episodes, n (%)	43 (53.8%)	50 (67.6%)	13.8 (−1.5 to 29.1)	3.04	0.081	7‡

‡ NNT for URI is presented for descriptive purposes; the between-group difference was not statistically significant. AOM = acute otitis media; URI = upper respiratory tract infection; NNT = number needed to treat; ARD = absolute risk difference; CI = confidence interval. Absolute risk differences and 95% CIs were calculated using the Newcombe-Wilson hybrid score method. Pearson's chi-square test was used for all comparisons. A two-sided p-value <0.05 was considered statistically significant.

BACTERIAL COLONIZATION

Nasopharyngeal bacterial colonization was assessed by nasal and throat swab cultures at baseline and at the end of the 3-month treatment period (Table 3). Baseline colonization patterns were comparable between groups. *Streptococcus*

pneumoniae was the most prevalent pathogen at baseline, with similar counts across both groups (XYL+GSE: 19 total isolates; control: 15 total isolates). By end of treatment, *S. pneumoniae* was undetectable in both groups, suggesting natural resolution of colonization independent of treatment.

A clinically notable finding was the differential behavior of *Staphylococcus aureus*. In the XYL+GSE group, *S. aureus* nasal isolates decreased from 5 to 2, whereas the control group showed a marked increase, rising from 5 to 12 nasal isolates and from 2 to 8 throat isolates (combined increase from 7 to 20 isolates). Due to the small absolute numbers, this finding was not formally analyzed statistically. Notably, the control group received substantially more antibiotics during the study period, which may have selected for Staphylococcus aureus via competitive release following depletion of antibiotic-susceptible flora; this confound precludes attributing the S. aureus increase directly to the absence of XYL+GSE treatment. Overall, total bacterial isolates (nasal and throat combined) decreased by 87.5% in the XYL+GSE group (from 40 to 5 isolates), compared with a 22.0% reduction in the control group (from 41 to 32 isolates). No individual between-group difference in colonization by species reached statistical significance ($p > 0.05$ for all comparisons), likely reflecting insufficient power for this exploratory endpoint.

Table 3. Nasopharyngeal bacterial colonization before and after 3 months of treatment

Bacterial Species	XYL+GSE Before (Nasal / Throat)	XYL+GSE After (Nasal / Throat)	Control Before (Nasal / Throat)	Control After (Nasal / Throat)
Streptococcus pneumoniae	10/9	0/0	9/6	0/0
Staphylococcus aureus	5/0	2/0	5/2	12/8
	2/6	0/1	2/4	2/2
Haemophilus influenzae	1/2	0/0	4/1	3/0
	2/0	0/0	2/3	0/0
	2/0	0/0	1/0	0/0
Escherichia coli	0/1	0/0	0/0	0/0
	0/0	1/1	2/0	2/3
Total isolates	22/18	3/2	25/16	19/13

§ *Branhamella catarrhalis* entries consolidated under *Moraxella catarrhalis* per current taxonomy. All between-group differences in bacterial colonization were non-significant ($p > 0.05$). Values represent number of culture-positive specimens per site per group.

Discussion

This exploratory controlled study evaluated the potential association between intranasal XYL+GSE administration and AOM incidence and antibiotic use in a pediatric outpatient population. These data suggest that intranasal XYL+GSE administration is associated with a statistically significant and potentially clinically meaningful reduction in recurrent AOM incidence and antibiotic use relative to a distilled-water comparator, with the caveat that the non-randomized design and absence of assessor blinding limit causal inference.

Two clinical trials established oral xylitol's efficacy in AOM prevention: xylitol administered at 8.4–10 g/day in five divided doses reduced AOM incidence by 40% with chewing gum and 30% with syrup over 2–3 months, with a concurrent decrease in antibiotic use. These findings, derived from oral xylitol regimens, contrast with the intranasal route used in the present study. The high AOM recurrence rate observed in the distilled-water control group (90.5%) reflects the enrollment of a high-risk, pre-selected population with documented recurrent AOM history and likely heightened susceptibility relative to general pediatric populations enrolled in previous xylitol trials; this rate is acknowledged as an outlier relative to published comparators and limits direct cross-study comparisons. However, xylitol failed to decrease AOM incidence in AOM-susceptible healthy children when administered three times daily, and was also ineffective in preventing AOM in children with tympanostomy tubes receiving the syrup formulation. Notably, none of those prior studies administered xylitol via the intranasal route, which may explain the divergent outcomes by targeting the nasopharyngeal site of pathogen colonization more directly.[12–14]

Komura et al. demonstrated that GSE at a 1:1,000 dilution rendered bacterial counts undetectable within 5 seconds under protein-free conditions and achieved a 3-log reduction within 5 seconds in the presence of 5% fetal bovine serum. Unlike hypochlorous acid (HOCl) — whose antimicrobial efficacy is markedly reduced when applied at a 30 cm distance, as demonstrated in in vitro spray models — the virucidal and bactericidal activity of GSE was not attenuated by spray distance, supporting its suitability as an intranasal antimicrobial agent.[10]

Xylitol inhibits bacterial growth and reduces pneumococcal adherence to nasopharyngeal epithelial cells, thereby limiting ascending colonization of the middle ear. Kontiokari et al. demonstrated that xylitol reduced the adherence of *Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis* to nasopharyngeal epithelial cells in vitro. The present study observed a corresponding reduction in colonization by AOM-associated pathogens in the XYL+GSE group.[9,12,13]

These findings are consistent with other non-antibiotic, nasal-spray-based strategies targeting the nasopharyngeal microbiota for AOM prevention. In a real-life clinical experience, children who received a nasal spray containing non-pathogenic bacterial strains as preventive bacteriotherapy after an initial AOM episode experienced significantly fewer recurrences than children managed with standard care alone, lending broader support to the rationale that topical, non-antibiotic modulation of nasopharyngeal colonization can reduce recurrent AOM.[15]

AOM continues to represent a substantial public health and economic burden despite widespread antibiotic use and pneumococcal conjugate vaccination. Estimates indicate that otitis media accounts for millions of pediatric healthcare visits annually in the United States, generating direct healthcare costs approaching USD 4.8 billion per year in children under five years of age, with a considerable proportion attributable to recurrent disease, hearing complications, and tympanostomy tube placement.[16–19]

Current management of recurrent AOM poses several challenges. Repeated antibiotic courses contribute to antimicrobial resistance and cumulative antibiotic exposure during early childhood. Pneumococcal conjugate vaccines have reduced part of the disease burden; however, recurrent AOM remains prevalent in the post-PCV era due to persistent non-vaccine pneumococcal serotypes, non-typeable *H. influenzae*, viral pathogens, and polymicrobial biofilm-associated disease.

Tympanostomy tube placement is associated with procedural costs and complications, including otorrhea, and offers variable long-term benefit on recurrence prevention.[19–21]

These results support the feasibility of a non-antibiotic, nasopharynx-targeted strategy for recurrent AOM prevention. By addressing bacterial colonization and biofilm formation at the site of pathogen ascent, intranasal XYL+GSE may interrupt a proximal step in AOM pathogenesis, rather than treating established infection. The European Position Paper on Rhinosinusitis and Nasal Polyps 2020 acknowledges intranasal xylitol among agents with potential benefit in sinonasal conditions, supporting the plausibility of this mechanistic pathway. The observed reductions in both AOM recurrence and antibiotic use, if confirmed in larger randomized trials, could support antimicrobial stewardship efforts in pediatric primary care.[22] A prospective cohort study similarly reported fewer AOM episodes and reduced antibiotic use during intranasal xylitol treatment in young children with recurrent AOM.[23]

The observed 87.5% reduction in total bacterial isolates in the XYL+GSE group versus 22.0% in controls is of particular interest and aligns with the proposed dual mechanism of action. The differential behavior of *S. aureus* colonization — decreasing in the XYL+GSE group but markedly increasing in controls — warrants further investigation in adequately controlled studies. This observation should not be interpreted as direct evidence of a protective effect of XYL+GSE on *S. aureus* nasopharyngeal expansion; differential antibiotic exposure between groups is a plausible confound, as the control group received substantially more antibiotics, which may have selected for S. aureus via competitive release following depletion of antibiotic-susceptible flora.

Limitations

Several limitations must be acknowledged. First, the exploratory, non-randomized design and modest sample size restrict external validity and limit statistical power for secondary and microbiological endpoints. Second, allocation was performed by site coordinators using a fixed schedule without formal randomization, and outcome assessors were not formally blinded, introducing potential selection and ascertainment bias; given that the primary endpoint (AOM diagnosis) requires clinical judgment, this constitutes the principal threat to internal validity and should be addressed in future confirmatory trials through allocation concealment and blinded endpoint adjudication. Third, microbiological analyses relied on culture-based methods with low absolute isolate counts, precluding species-level between-group inference. Fourth, the 3-month active treatment period precludes assessment of long-term recurrence or post-treatment microbiome dynamics. Finally, as a non-randomized exploratory study, causal inference is not warranted. Confirmatory randomized controlled trials with adequate statistical power, longer follow-up, and standardized microbiological methods are needed.

Conclusions

In this exploratory study, intranasal XYL+GSE administration was associated with a lower rate of recurrent AOM and reduced antibiotic use in a pediatric outpatient population. These findings support the potential role of microbiome-targeted, non-antibiotic preventive strategies for recurrent upper respiratory infections. Given the exploratory, non-randomized design and the absence of confirmed outcome-assessor blinding, these results should be interpreted as hypothesis-generating and warrant confirmation in adequately powered, blinded randomized controlled trials with pre-specified allocation concealment.

Conflicts of interest

The authors have no conflicts of interest to declare.

Funding

This study received no external funding.

Acknowledgments

The authors thank the staff of the 10 participating investigational sites in the Prague and Mid-Bohemia region for their contributions to participant recruitment and data collection.

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