Using chlorhexidine varnish to prevent early childhood caries in American Indian children

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Abstract

Objectives: To test the efficacy of 10% chlorhexidine (CHX) dental varnish applied to the mothers' dentition in preventing caries in American Indian children.
Methods: This was a placebo-controlled, double-blind, randomized clinical trial. Mother–child pairs were enrolled when the child was 4.5-6.0 months. Mothers received 4 weekly applications of the study treatment (CHX or placebo) followed by single applications when her child was age 12 and 18 months. Children received caries examinations at enrollment, 12, 18 and 24 months. Analyses were limited to the intent-to-treat (ITT) group: children whose mothers received the first study treatment and who received at least one post-baseline exam. The outcome variable was the number of new carious surfaces (NNCS) at the child's last visit. Wilcoxon nonparametric and Fisher's exact tests were used to test differences between the active and placebo groups.
Results: We randomized 414 mother–child pairs, with 367 (88.6%) included in the ITT group (active = 188, placebo = 179). The proportion of children caries-free at their final exam was 51.1% and 50.8% for the active and placebo groups ($P > 0.99$). The mean NNCS for the active and placebo groups was 3.82 (standard deviation [SD] = 8.18) and 3.80 (SD = 6.08), respectively ($P = 0.54$). The proportion with NNCS > 6 was 18.1% for active children versus 27.9% for placebo (relative risk [RR] = 0.65, $P = 0.03$). The number needed to treat to shift one child from NNCS > 6 to a lower severity was 10.2.
Conclusions: In this population CHX varnish did not reduce the mean NNCS or proportion of children with caries, but did reduce the proportion with severe caries.

Introduction

Caries in the primary dentition (commonly referred to as early childhood caries, or ECC) is the most common pediatric chronic disease, with a prevalence of 28% in US children 2-5 years of age (1). Low-income and minority children are disproportionately affected by caries, with American Indian and Alaskan Native (AI/AN) children being one of the racial/ethnic groups at highest risk (2). Despite decades of ECC control efforts by AI/AN communities and Indian Health Service (IHS) – the federal agency whose mission is to provide health services to AI/AN – little progress has been made. The IHS ECC-prevention efforts have focused primarily on community water fluoridation, behavior modification for the mother regarding the child's diet and oral hygiene, and application of fluoride varnish (FV) in both clinical and community settings. However, the outcome of these efforts was recently described as "minor, transient victories, at best" (3).

Caries is a multifactorial disease, with the presence of cariogenic bacteria being a necessary but not sufficient component. The majority of the literature suggests that children acquire cariogenic bacteria by vertical transmission from the

Table 1 Treatment (for Mothers) and Exam (for Children) Timeline and Number Completed*

	Intent to treat	
	n = 367	
Interval between treatments	Active	Placebo
n/a	Treatment 1 and caries exam *n* = 188 mother–child dyads	Treatment 1 and caries exam *n* = 179 mother–child dyads
1 week (5-14 days)	Treatment 2 *n* = 182 mothers	Treatment 2 *n* = 173 mothers
1 week (5-14 days)	Treatment 3 *n* = 177 mothers	Treatment 3 *n* = 170 mothers
1 week (5-14 days)	Treatment 4 *n* = 174 mothers	Treatment 4 *n* = 169 mothers
6 months (± 14 days)	Treatment 5 and caries exam *n* = 157 mothers *n* = 177 children	Treatment 5 and caries exam *n* = 152 mothers *n* = 165 children
6 months (± 14 days)	Treatment 6 and caries exam *n* = 139 mothers *n* = 165 children	Treatment 6 and caries exam *n* = 129 mothers *n* = 152 children
6 months (± 14 days) (end of study)	Caries exam *n* = 164 children	Caries exam *n* = 156 children

* Per protocol visit interval; individuals who completed the visit outside of the protocol interval are also included in the counts.

mother (4,5), although horizontal transmission has also been identified (6). Once teeth are colonized, cariogenic bacteria produce acid when exposed to sugar, resulting in demineralization of the enamel if the acid is of sufficient quantity and duration (7). The newly emerged primary dentition is especially susceptible to caries because the hydroxyapatite of the enamel is highly susceptible to acid dissolution (8), and requires time to "mature." There are no published reports on the level of cariogenic bacteria among AI/AN children, although there is documentation that AI/AN have a much higher incidence and prevalence of other infectious diseases of childhood (9,10).

Although never studied in AI/AN populations, there are a number of reports of attempting to reduce the rate of ECC by interfering with the vertical transmission of cariogenic bacteria from mother to infant. Kohler (11) treated caries-active mothers with either what was considered the standard of care or with the combination of dental restorations, chlorhexidine (CHX) mouth rinse, FV, dietary counseling, and oral hygiene. Children of treated mothers exhibited delayed colonization by mutans streptococci (MS) and a significant reduction in caries that was still evident when the children were 7 years (12).

There is extensive literature on the use of CHX-containing products in different vehicles and concentrations to prevent caries, with results as varied as the methods. Recent reviews (13-15) found evidence that in selected populations at certain concentrations and frequencies, CHX may be an effective agent in reducing caries, but that overall, the data are inconclusive. Some of these studies included using CHX on the mother's dentition in an attempt to interrupt or delay transmission of cariogenic bacteria to the child, again with varying results. The purpose of this research was to test the efficacy of CHX varnish in reducing the prevalence and severity of ECC among high-risk AI/AN children through interrupting the transmission of cariogenic bacteria from mother to child by periodic treatment of the mother with a 10% CHX formulation found effective in reducing decay in adults with xerostomia (16).

Materials and methods

Design

This was a placebo-controlled (1:1), double-blind, parallel group randomized clinical trial of the efficacy of a 10% CHX varnish applied to the mothers' dentition in preventing ECC. Mother–child pairs were enrolled when the child was between 4.5 and 6.0 months of age. After enrollment, each mother received four weekly applications of the study treatment (CHX or a placebo) followed by single applications when her child was approximately 12 and 18 months of age for a total of six treatments (Table 1). At enrollment and again at the 12- and 18-month exams, the mothers completed questionnaires on their interim medical history, concurrent medications, and adverse effects from the study medication. The children received comprehensive surface-specific caries examinations at enrollment and when the child was approximately 12, 18, and 24 months old. Children received a FV treatment after the

12- and 18-month exams – the standard of care at the study sites. The FDA approved this Phase III clinical trial under IND #45,466.

Study sites

Participants were recruited from four different AI communities in Oregon, Washington, and Arizona – all with fluoridated water systems. The study was conducted at the local IHS or tribally operated community dental clinic. At three of the sites, the facility dentists were trained as examiners, while the fourth and largest site hired a dentist examiner specifically for the study.

Participants

Women were eligible to participate if they could provide informed consent, had at least 20 natural teeth, had unrestored caries or a previous child with documented ECC, and had a child between 4.5 and 6.0 months of age with or without teeth. Exclusion criteria included the presence of orthodontic appliances and pregnancy. Prior to enrollment, the mother's cavitated carious lesions were restored. The Institutional Review Board of record and the local tribal health authority for each community approved the protocol, and written informed consent was obtained from all participants.

Randomization and study treatments

Study sites received consecutively numbered boxes of the study product which were numbered by the research pharmacist prior to shipment. Each box contained separate vials for each study visit. As participants were enrolled, they were assigned the next numbered product box. Because the active and placebo study products were identical in color, smell, taste, and viscosity, neither the participants nor study staff knew whether the product was active or placebo. Study treatments were applied in a dental clinic setting by trained hygienists or dental assistants after a brief rubber cup prophylaxis. The study treatments were applied in two stages: Stage 1 contained either 10% CHX diacetate w/v suspended in a solution of Sumatra benzoin and alcohol (the active treatment) or only the Sumatra benzoin and alcohol solution (the placebo treatment); Stage 2 was a proprietary aqueous dispersion of inert methacrylate approved for use by the FDA under license K023671. The stage 2 coating is designed to prolong the contact time between the CHX and the tooth. For participants in the active treatment arm, the mean dose of CHX at each application visit was 37.4 mg ($\pm$14); the cumulative mean dose was 224 mg. The visits for the mother–child pair took 45-60 minutes each for the four visits in which the mothers filled out the interim history questionnaires in addition to getting a caries examination and study medication application, and about 15 minutes each for the four other visits.

Child caries examination and primary outcome

Caries were diagnosed visually by calibrated examiners in a dental clinic setting using a mirror, periodontal probe, dental light, and air. We used the diagnostic nomenclature of Pitts and Fyffe (17): non-cavitated lesions (d1); lesions where the cavitation extends into, but not through, the enamel (d2); and cavitated lesions that involve the dentine (d3). At the onset of the study, an examiner calibration session was held with a gold standard examiner,[1] followed by recalibrations at approximately 6-12-month intervals. Because of staff turnover at the sites that used facility dentists as examiners, there were a total of 17 examiners among the four sites. Each new examiner received the same calibration training from the same trainer before examining study participants. The inter-examiner reliability kappa scores for examiners compared to the standard ranged from 0.54 to 0.77, and intra-examiner kappas were 0.77-1.00. The primary outcome variable was the child's number of new carious surfaces (NNCS) at the last post-baseline visit.

Definition of the intent-to-treat population evaluated

For sample size calculation, we estimated a mean baseline NNCS of 4.0 (standard deviation [SD] 3.0), with a minimal clinical improvement of 20%, resulting in a target completion of 221 children in each treatment group (total 442). The intent-to-treat (ITT) population consisted of children of randomized mothers who received the first study medication application and who returned with their child for at least one of the three post-baseline exams. For children who did not complete all visits according to study protocol, the NNCS score from the last post-baseline examination was carried forward (last observation carried forward).

Statistical analysis

The children's NNCS scores at their last post-baseline visit were analyzed as a continuous measure for the mean NNCS – the primary outcome variable. They were also analyzed as a categorical variable based on two post-randomization sets of cutoffs. Categorization #1 (NNCS = 0, NNCS = 1-5, NNCS $\geq$6) was defined as a proxy for clinical severity based on the expert opinion of the facility dentists participating in our

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study that in their specific communities any child at 24 months of age having ≥6 cavitated tooth surfaces would likely require in-hospital treatment under general anesthesia (GA). Categorization #2 (NNCS = 1-3, NNCS = 4-7, NNCS ≥ 8) is the tertile distribution of the NNCS scores for placebo children who had >0 NNCS scores at their last exam. These categorizations were used to determine whether, irrespective of the mean NNCS, there was any shift in the distribution of caries severity as a result of the treatment intervention. To determine the effect of FV applications to the children, we analyzed the frequency distribution of the number of FV applications and used this for an adjusted analysis. The Wilcoxon nonparametric test was used to test the differences between the active treatment and placebo groups for continuous measures. The nonparametric test was used because it is more robust to departures from normality and outliers, and in the dataset being evaluated, there were several large outliers. The Fisher's exact test was used to test for differences in the proportion of study participants in the two different categorization schemes between the active and placebo groups. All analyses were conducted using SAS/STAT software Version 9.2 (18).

Results

Study population

We randomized a total of 414 AI/AN mother–child pairs, of whom 367 (the ITT group) returned for at least one post-baseline visit (Figure 1). At enrollment, these 367 mothers ranged in age from 14.1 to 43.5 years (mean = 26.8, SD = 6.4). At enrollment, children were 4.5-6 months old (mean = 5.26, SD = 0.64), and at the end of study were 22.0-30.3 months old (mean = 24.3, SD = 1.4). A total of 188 mother–child pairs were in the active treatment group, while 179 were in the placebo group. There was no difference between the active and placebo group in the children's mean age at enrollment or age at their last exam (Table 3). There were no serious study-related adverse events to either mothers or children.

Protocol compliance

There was no difference ($P = 0.9$) between active versus placebo in protocol compliance: 104 mother–child pairs (active $n = 54$, placebo $n = 50$) completed all activities per protocol; 119 (active $n = 61$, placebo $n = 58$) completed six of the study medication applications but not per protocol timeline; 99 (active $n = 49$, placebo $n = 50$) had at least five of the six applications; and 45 (active $n = 24$, placebo $n = 21$) had less than five applications (Table 2).

Caries increment

The mean NNCS did not differ between the active and placebo groups: 3.82 (SD = 8.18) versus 3.80 (SD = 6.08), respectively ($P = 0.54$) (Table 3). The intraquartile range was 0-4 for the active group and 0-6 in the placebo group, indicating that 75% of the individuals in the active group had NNCS ≤ 4 compared to 75% who had NNCS ≤ 6 in the placebo group. The proportion of children caries-free at their final exam was the same for active (51.1%) versus placebo (50.8%) ($P > 0.99$; Table 4).

An analysis of the children's end of study NNCS scores using the post-randomization clinical severity Categorization #1 (NNCS = 0, 1-5 or ≥ 6) resulted in a difference ($P = 0.03$) in the distribution of caries scores between the active and placebo groups, with fewer active children in the most severe category (NNCS ≥ 6) (Table 4). The relative risk for having NNCS ≥ 6 was reduced by 35.2% among the active group. Severity Categorization #2 compared only the 187 children with NNCS > 0 at their final examination using the tertile distribution of caries from the placebo group (NNCS = 1-3, 4-7, and ≥8). Using this distribution resulted in a nonsignificant ($P = 0.11$) trend for a greater proportion of the active children to be in less severe tertiles (Table 5).

We used the frequency distribution of FV applications for adjusted analysis. Because of an error in printing the paper Case Record Forms for the study, FV status was not reliably recorded at two of the four study sites, so we evaluated the effects on the children ($n = 295$) at the two other sites. Fisher's exact test resulted in a nonsignificant trend ($P = 0.16$) for a greater proportion of active (74.2%) than placebo (66.2%) children to have received 2 versus 0-1 FV applications. However, the mean NNCS was the same (4.2) for placebo children who received 2 ($n = 98$) versus 0-1 ($n = 50$) FV. Adjusted analysis controlling for the frequency of FV application did not change the result of the distribution for severity Categorization #1 or #2 as the parameter estimates of treatment effect adjusted and unadjusted were essentially the same.

Number needed to treat

The number needed to treat (NNT) metric is a useful way to assess the practicality of implementing a specific preventive intervention (19). Using severity Categorization #1, the NNT was 10.2 [1/(27.9%–18.1%)] to shift one child from the most severe caries category (NNCS ≥ 6) to a less severe outcome.

Discussion

This study is the first completed randomized controlled clinical trial intended to prevent ECC among AI/AN children by interrupting the vertical transmission of cariogenic bacteria – primarily MS, but not excluding others – from mother to child by treating only the mother. This study was initiated in the context of a very high prevalence, severity, and morbidity from ECC in many AI/AN communities.

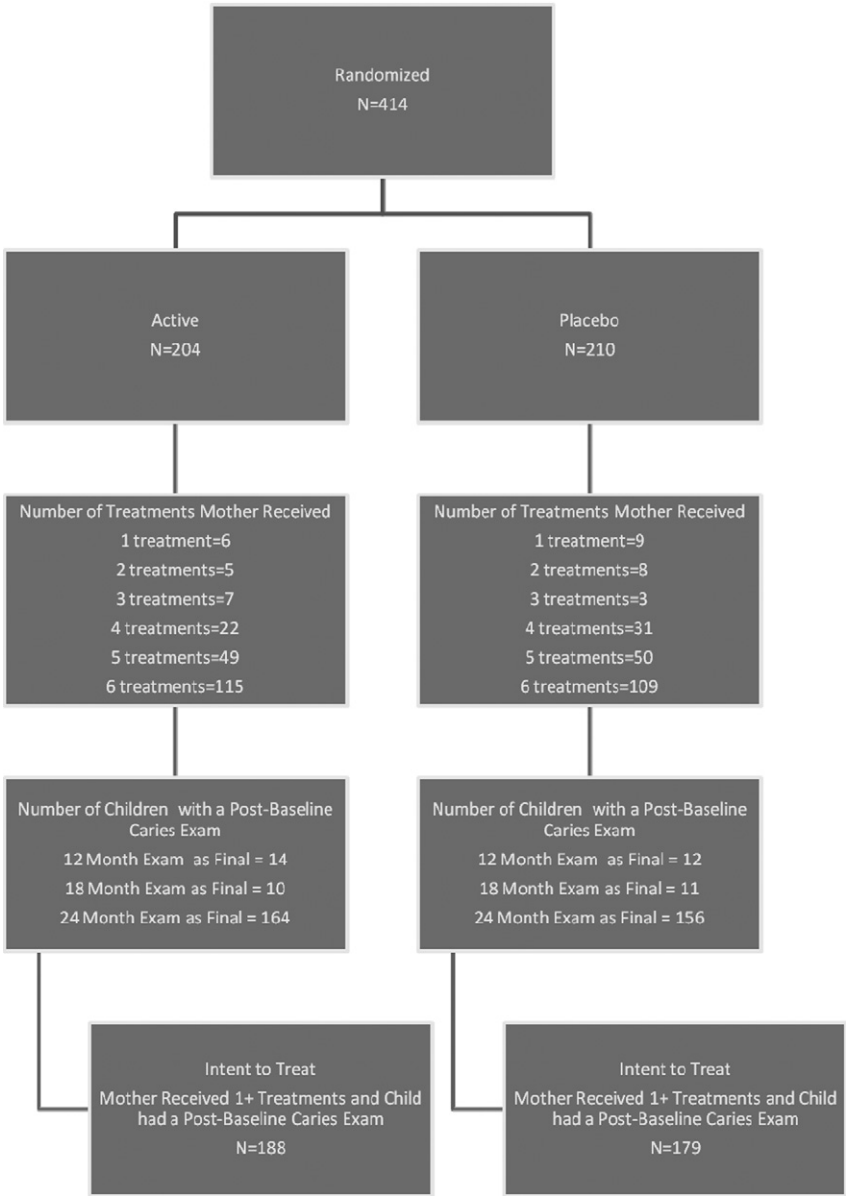

Figure 1 Number of study participants, by treatment group and protocol compliance.

There were acknowledged limitations of the study – foremost being the absence of a microbiological assessment for either mothers or children. Because transmission of cariogenic bacteria was the issue of concern, DNA fingerprinting would have been required, which would have greatly exceeded the budgetary and logistic capability of the study. There were ongoing recruitment and retention problems at two sites; thus we did not achieve our goal of 442 completed children for analysis. Given that the study was conducted using the community dental facility dentists as examiners, we used multiple examiners at each site, although all were trained by the same individual. We did not control for

Table 2 Protocol Compliance for Intent-to-Treat Population, by Treatment

Protocol compliance	Active *n* = 188	Placebo *n* = 179	Total *n* = 367
6 treatments per protocol	54	50	104
6 treatments but not per protocol	61	58	119
5 of 6 treatments	49	50	99
4 of 6 treatments	14	14	28
3 of 6 treatments	6	3	9
2 of 6 treatments	4	4	8
1 of 6 treatments	0	0	0

Overall *P*-value = 0.9 (Fisher's exact test).

Table 3 Number of New Carious Surfaces (NNCS) and Age at the Final Exam – Intent-to-Treat Population

	Treatment group		
Variable	Active $n = 188$	Placebo $n = 179$	*P*-value*
NNCS			
Mean (SD)	3.82 (8.18)	3.80 (6.08)	0.540
Median (IQR)	0 (0-4)	0 (0-6)	
Range	0-67	0-27	
Child's age (months)			
Mean (SD)	23.0 (3.6)	23.2 (3.7)	0.699
Range	10.9-30.5	10.8-30.1	

* Wilcoxon nonparametric test.

potential confounding variables such as antibiotic treatment for infectious disease, although the randomized placebo design should minimize any effect from these. Last, a cost–benefit analysis was beyond the scope of the study.

We found no difference between active versus placebo in either the mean NNCS for the children or proportion of caries-free children. Despite this, in each of two analyses based on post-randomization categorization by clinical severity, there was a trend among active children toward reduced caries severity (Table 4 and 5). This suggests that for children at the highest risk, mean NNCS and the proportion of caries-free children may not adequately describe changes in disease severity. The need for a new metric for caries in the primary dentition that correlates well with morbidity was emphasized in the 2010 Symposium on ECC among AI/AN children sponsored by the American Dental Association (http://www.ada.org/5154.aspx).

For very high-risk populations, the most important issue is whether morbidity from severe ECC, such as pain, infection, and hospitalization, can be prevented through an intervention. A good model for this construct found in the pediatric infectious disease literature may be the varicella (chickenpox) vaccine. A review found that the vaccine was 84.5% effective (range 44%–100%) in preventing *all* cases of varicella, but 100% effective in preventing *severe* varicella (20). This does not imply that varicella and caries are equivalent in all attributes, but rather provides a documented example that interventions may achieve reductions in the proportion of individuals with severe disease while not eliminating all cases. Similarly, our primary goal for ECC prevention among AI/AN children is to reduce the prevalence of severe cases, especially those requiring restorative care under GA (21,22). The cost of a full mouth GA restoration in a child has been estimated at $2,000-10,000 per case. Thus, both the human and financial costs are extremely high (23). Anecdotally, in many AI/AN communities, >25% of all children require restorations under GA for severe ECC prior to entry into the first grade. This compares to published data from two administrative databases showing a rate of about 0.2% of non-AI/AN children (24).

We have no data on the subsequent caries experience of our study children to determine how many GA cases were prevented by the intervention used, and there is no national standard for assessing disease burden from ECC. Therefore, we evaluated the study results by two proxies for disease morbidity (severity categorizations 1 and 2). The expert

Table 4 Distribution of Caries by Categorization #1 (NNCS = 0, 1-5, ≥6) at Final Exam – Intent-to-Treat Population

		Treatment group			Overall
Variable	Statistic	Active	Placebo	RR	*P*-value*
Severity (categorization #1)	N	188	179		0.029
	NNCS = 0	96 (51.1%)	91 (50.8%)	1.01	
	NNCS = 1-5	58 (30.9%)	38 (21.2%)	1.45	
	NNCS ≥6	34 (18.1%)	50 (27.9%)	0.65	

* Fisher's exact test.

Table 5 Distribution of Caries by Categorization #2 (NNCS = 1-3, 4-7, ≥ 8) Among Caries-Active Children at the Final Exam – Intent-to-Treat Population

		Treatment group			Overall
Variable	Statistic	Active	Placebo	RR	*P*-value*
Severity (categorization #2)	N	92	88		
	NNCS = 1-3	42 (45.7%)	27 (30.7%)	1.49	0.113
	NNCS = 4-7	24 (26.1%)	31 (35.2%)	0.74	
	NNCS ≥ 8	26 (28.3%)	30 (34.1%)	0.83	

* Fisher's exact test.

opinion of the facility dentists participating in our study was that a caries dmfs (decayed, missing or filled surfaces) score ≥6 for 24 months old children would likely require in-hospital GA treatment. Based on this, we initially compared the active versus placebo groups for the distribution of caries when categorized into three groups: NNCS = 0, 1-5, ≥6. This resulted in a statistically significant change in the distribution of active versus placebo children, with 32% fewer children in the active group being in the ≥6 category compared to the placebo group ($P = 0.03$).

Given the absence of data showing the proportion of children who with any given dmfs score ultimately require treatment under GA, this categorization of children into these groups was admittedly a somewhat arbitrary attempt to correlate dmfs with morbidity. Thus, for further analysis, we next evaluated *only the caries-active children* by a "natural" distribution of tertiles (NNCS = 1-3, 4-7, ≥8) derived from the NNCS in our placebo group. This analysis resulted in a favorable but nonsignificant ($P = 0.11$) trend for a higher proportion of placebo children to be in the more severe two tertiles compared to the active children.

A remarkable incidental finding of our study was that by age 24 months, 49.2% of placebo children had cavitated caries (mean NNCS = 3.8) despite the study protocol specifying that children receive FV at 12 and 18 months. This contrasts with the report by Weintraub (25) that found caries prevention efficacy of even one FV annually. However, although purported to be a study of high-risk children ("low income, underserved populations"), this study featured the methodological curiosity of excluding from enrollment all the highest-risk children – namely those who had extant caries at the screening exam (26). Our finding of very limited preventive effectiveness of FV in our study population is consistent with the report by Holve (27) in which the "successful" group of AI/AN children who had ≥4 applications of FV at ages 9, 12, 15, 18, and 24 months nonetheless had a mean dmfs of 15.6 at age approximately 52 months.

There are a number of plausible reasons for the failure to achieve a positive treatment effect on the mean NNCS. The schedule of CHX varnish for the mothers may not have been adequate to inhibit transmission of all their cariogenic bacteria. Also, some of the children may have acquired their cariogenic bacteria from someone other than the mother; a recent report by Mitchell (28) found that 74% of children scheduled for full mouth restoration under GA had *Streptococcus mutans* subtypes that did not match their mothers' *S. mutans*. There are no published reports of the proportion of AI/AN children who acquire *S. mutans* from sources other than the mother, nor of correlation of caries severity with the source of transmission. Given the early and severe ECC that many AI/AN children develop, the microbiological paradigms that apply in lower-risk communities may not be applicable to AI/AN mothers and children.

Future research needed

Considering that almost 50% of the children in our placebo group had cavitated caries at age 24 months, clearly additional research is need to further quantify, understand, and control ECC in AI/AN children. Future research in this area should address transmission of cariogenic bacteria in conjunction with measures of reduction of incidence and morbidity from caries.

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