

# Unsafe Child Feces Disposal is Associated with Environmental Enteropathy and Impaired Growth

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**Objective** To investigate the relationship between unsafe child feces disposal, environmental enteropathy, and impaired growth, we conducted a prospective cohort study of 216 young children in rural Bangladesh.

**Study design** Using a prospective cohort study design in rural Bangladesh, unsafe child feces disposal, using the Joint Monitoring Program definition, was assessed using 5-hour structured observation by trained study personnel as well as caregiver reports. Anthropometric measurements were collected at baseline and at a 9-month follow-up. Stool was analyzed for fecal markers of environmental enteropathy: alpha-1-antitrypsin, myeloperoxidase, neopterin (combined to form an environmental enteropathy disease activity score), and calprotectin.

**Findings** Among 216 households with young children, 84% had an unsafe child feces disposal event during structured observation and 75% had caregiver reported events. There was no significant difference in observed unsafe child feces disposal events for households with or without an improved sanitation option (82% vs 85%,  $P = .72$ ) or by child's age ( $P = .96$ ). Children in households where caregivers reported unsafe child feces disposal had significantly higher environmental enteropathy scores (0.82-point difference, 95% CI 0.11-1.53), and significantly greater odds of being wasted (weight-for-height z score  $< -2$  SDs) (9% vs 0%,  $P = .024$ ). In addition, children in households with observed unsafe feces disposal had significantly reduced change in weight-for-age z-score ( $-0.34$  [95% CI  $-0.68, -0.01$ ] and weight-for-height z score ( $-0.52$  [95% CI  $-0.98, -0.06$ ]).

**Conclusion** Unsafe child feces disposal was significantly associated with environmental enteropathy and impaired growth in a pediatric population in rural Bangladesh. Interventions are needed to reduce this high-risk behavior to protect the health of susceptible pediatric populations. (*J Pediatr* 2016; ■: ■-■).

Undernutrition is estimated to be the underlying cause of death for more than one-half of young children globally and is associated with an increased risk of cognitive delays, susceptibility to infections, and lower economic productivity.<sup>1-4</sup> There is a growing body of literature demonstrating an association between environmental enteropathy and undernutrition in susceptible pediatric populations.<sup>5-9</sup> Environmental enteropathy is defined by abnormal intestinal morphology, reduced intestinal barrier function, and increased intestinal inflammation resulting in malabsorption of nutrients and growth faltering in children.<sup>10-18</sup> This disorder is thought to occur from unsanitary environmental conditions, leading to repeated exposures to enteric pathogens.<sup>12-18</sup>

Sanitation interventions implemented in the water, sanitation, and hygiene field typically focus on construction of improved sanitation options targeted at ambulatory populations.<sup>19-21</sup> There is little attention given to open defecation events by young children, despite this practice being common among children in many low-income countries.<sup>22-24</sup> Unsafe disposal of child feces through practices such as disposal in open areas increases exposures to fecal pathogens in susceptible pediatric populations by allowing direct contact with human feces and contaminated soil during play behavior and through vectors such as flies spreading fecal pathogens to food.<sup>25,26</sup> Child feces not being disposed of in a latrine has been associated with an increased risk of diarrhea in young children.<sup>22,27-31</sup> In a meta-analysis of studies on unsafe child feces disposal, this practice was associated with a 23% increased risk of diarrheal diseases.<sup>32</sup>

Furthermore, the health impacts of unsafe feces disposal can extend beyond diarrheal disease. A study in rural Bangladesh found that unsafe child feces disposal was associated with an increased risk of soil-transmitted helminth infections in children younger than 2 years of age.<sup>33</sup> Most recently a cohort study conducted in Mirzapur, Bangladesh, found that young children mouthing soil during play in households with visible feces on their compound had an increased risk of environmental enteropathy and stunting.<sup>6</sup>

CFU	Colony-forming units
HAZ	Height-for-age z scores
icddr,b	International Centre for Diarrhoeal Disease Research, Bangladesh
WAZ	Weight-for-age z scores
WHZ	Weight-for-height z scores

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Our objective in conducting this prospective cohort study was to assess child feces disposal practices in rural Bangladesh and to determine the relationship between this behavior, exposure to enteric pathogens in soil, environmental enteropathy, and growth in a pediatric population. We hypothesized that unsafe disposal of child feces was associated with impaired growth in children through increased exposure to enteric pathogens leading to environmental enteropathy.

## Methods

This prospective cohort study of 216 randomly selected children 6-30 months of age was conducted in Mirzapur upazila in the Tangail district of Bangladesh at the site of the Global Enteric Multicenter Study demographic surveillance system. This study was nested within a larger investigation of the association between geophagy (mouthing of soil), environmental enteropathy, and stunting. The sample size was based on the number of study participants who could be recruited from February to April 2014. Study participants 6-30 months of age were selected to target children most susceptible to growth faltering.<sup>34</sup> A 9-month follow-up was conducted in study households between November and December 2014. A stool sample was collected from each child at baseline, and research assistants trained in standardized anthropometry measured the child's weight once and height 3 times. These measurements were used to calculate z scores according to the World Health Organization child growth standards.<sup>35</sup> Two soil samples also were collected in the outdoor courtyard area where the enrolled child was observed playing in a subset of 128 randomly selected households.

Informed consent was obtained from all study participants, and study procedures were approved by the research ethics committees of the International Centre for Diarrhoeal Disease Research, Bangladesh (icddr,b); an exemption was obtained from the ethical review board at the Johns Hopkins Bloomberg School of Public Health.

A 5-hour structured observation session was conducted by a trained research assistant between 8:00 a.m. and 1:00 p.m. from February to April 2014 in the household of each enrolled child. A structured observation tool was used to collect information on whether the child had a defecation event and how the caregiver disposed of the child's feces. Defecation events were divided into the following categories: open defecation event; child toileting event (if the child used a toilet or latrine); and a child potty event. We had the following categories for child feces disposal: (1) feces disposed of in a toilet or latrine; (2) feces scattered in yard or compound; (3) feces disposed of in an open space adjacent to the household compound; (4) feces buried; (5) feces thrown in a location designated for household waste (eg paper, wrappers); (6) no feces disposal; (7) other; and (8) did not observe. We used the Joint Monitoring Program definition of "safe feces disposal," which was defined as feces disposal in a latrine/toilet or buried. Any other method of feces disposal was defined as "unsafe feces disposal."<sup>36</sup> Using

the aforementioned categories, we also asked caregivers how they disposed of their child's feces.

We also observed child hand washing practices after a defecation event during the structured observation period. We defined hand washing behavior in the following categories: (1) no hand washing; (2) hand washing with one hand; (3) hand washing with 2 hands; and (4) could not observe. The cleansing agent used during the child hand washing event was recorded as follows: water only; bar soap and water; and did not observe.

All stool samples collected were transported in cooler boxes to the Enteric Microbiology Laboratory at icddr,b in Dhaka, Bangladesh and stored at  $-80^{\circ}\text{C}$  until analysis. Alpha-1-antitrypsin (Biovendor, Asheville, North Carolina), Neopterin (Genway, San Diego, California), and Calprotectin (ALPCO, Salem, New Hampshire) enzyme-linked immunosorbent assay kits were run for sample analysis according to the package insert. Myeloperoxidase enzyme-linked immunosorbent assay kits also were run according to the manufacturer specified instruction, except for a 1:500 dilution used for initial runs (ALPCO). The environmental enteropathy disease activity score was calculated by the use of fecal myeloperoxidase, alpha-1-antitrypsin, and neopterin, according to previously published methods.<sup>10</sup>

Soil samples were stored in cooler boxes and transported to the Enteric Microbiology Laboratory at icddr,b in Dhaka, Bangladesh, where total *Escherichia coli* counts and diarrheagenic *E coli* were detected according to previously published methods.<sup>6,37,38</sup> The complete soil findings were published previously elsewhere.<sup>6</sup> The objective of the current analysis was to stratify the soil findings by child feces disposal practices.

## Statistical Analyses

Our primary objective in conducting this study was to determine whether unsafe child feces disposal was significantly associated with elevated markers of environmental enteropathy and impaired growth in young children. Therefore, our primary study outcomes are calprotectin, environmental enteropathy disease activity score, and height-for-age z scores (HAZ), weight-for-age z scores (WAZ), and weight-for-height z scores (WHZ). A z score less than  $-2$  was classified as stunted for HAZ, underweight for WAZ, and wasted for WHZ.<sup>39,40</sup> Our measurements of unsafe child feces disposal were based on the observed behavior during 5-hour structured observation and caregiver reported behavior. To assess the association between unsafe child feces disposal and the selected fecal markers of environmental enteropathy, linear regression models were used with calprotectin and environmental enteropathy disease activity score as the outcomes and our measures of unsafe child feces disposal as predictors.

To assess the association between unsafe child feces disposal and growth, linear regression models were used with the change in HAZ, WAZ, and WHZ as outcomes and logistic regression models with being underweight, stunted, or wasted as outcomes and unsafe child feces disposal as the predictor. For our adjusted models, covariates were selected if their association with the outcome had significance

less than 0.2. To assess the relationship between *E coli* in soil and unsafe child feces disposal practices a logistic regression model was conducted with the presence of diarrheagenic *E coli* as the outcome and a linear regression model with *E coli* counts as the outcome and unsafe child feces disposal as the predictor. For individual level variables, a Fisher exact test was used for categorical variables.

## Results

The demographic characteristics of the study population are presented in **Table I**. The median age of study children was 17 months, and 54% of children were female. Of 216 children, 36% had a defecation event during the 5-hour structured observation period, with 85 child defecation events total (**Table II**). Ninety-one percent (71/78) of children had a single defecation event, and 9% (7/78) had 2 defecation events. Of the 78 children with an observed defecation event, 89% had an open defecation event, 10% had a child potty event, and 1 child had a caregiver-assisted toilet event. There were no significant differences in open defecation events by age category ( $P = .45$ ; 6-12 months: 31%; 12-18 months: 31%; 18-24 months: 31%; and 24-30 months: 36%). Only 1 child was reported to be wearing a diaper during the structured observation period (0.5%). All households reported having a sanitation facility.

Eighty-four percent (65/77) of households with a child defecation event had an observed unsafe feces disposal event during the structured observation period. Seventy-eight percent (66/85) of child feces were disposed of in an open

space adjacent to the household compound, and 14% (12/85) were disposed of in a toilet or latrine. Of the 8 child potty defecation events, 4 had feces disposal events in a toilet or latrine, and 4 had child feces disposal events in an open space adjacent to the household compound. There was no significant difference in the proportion of observed unsafe child feces disposal events between those households with or without an improved sanitation option (82% vs 85%,  $P = .72$ ) or by age category (6-12 months: 90%; 12-18 months: 83%; 18-24 months: 84%; and 24-30 months: 83%,  $P = .96$ ).

Seventy-five percent (163/216) of children had caregivers who reported unsafe disposal of child feces. Ninety-one percent (59/65) of observed unsafe feces disposal events also were caregiver reported. Twenty-five percent (53/216) of caregivers reported disposing of child feces in a toilet or latrine, 71% (154/216) in an open space adjacent to the household compound, and 4% (9/216) in a location designated for household waste.

Only 8% of 77 children were observed hand washing with soap after defecation, and 26% had any hand washing event (soap or water). All hand washing events (except for one) involved both hands and assistance from a caregiver. There was no significant difference in hand washing by age category (6-12 months: 20%; 12-18 months: 33%; 18-24 months: 26%; and 24-30 months: 17%;  $P = .67$ ).

Children in households where caregivers reported practicing unsafe feces disposal had significantly greater environmental enteropathy disease activity scores (0.82-point difference, 95% CI 0.11-1.53), after adjustment for age, age squared, caregiver educational level, and family size in the fully adjusted models (**Table III**). There were no other significant associations found between child feces disposal practices and fecal markers of environmental enteropathy.

We were able to locate 92% (71/77) of children with baseline observed child feces disposal events and 95% (205/216) with caregiver reported child feces disposal events at our 9-month follow-up. The odds of wasting (WHZ  $< -2$  SDs) at follow-up was significantly higher for children in households where caregivers reported practicing unsafe child feces disposal at baseline (9% unsafe feces disposal vs 0% safe feces disposal,  $P = .024$ ) in the adjusted model (**Table IV**). Furthermore, children in households with observed unsafe feces disposal events at baseline had significantly reduced changes in WAZ (coefficient:  $-0.34$  [95% CI:  $-0.68, -0.01$ ]) and WHZ ( $-0.52$  [95% CI  $-0.98, -0.06$ ]) in the adjusted models. There were no significant associations between child feces disposal practices and other anthropometric measurements.

Overall, 14% (18/128) of households sampled had soil with detectable diarrheagenic *E coli*; when stratified by type of child feces disposal, this was 4% (1/28) for households with safe feces disposal compared with 17% (17/100) for households with unsafe feces disposal (OR 5.5 [95% CI 0.70, 43.52]). The overall median for *E coli* counts in soil was 6250 colony-forming units (CFU)/g, with median 4237 CFU/g for safe feces disposal and 6488 CFU/g for unsafe feces disposal practices (105 825 CFU/g point difference [95% CI  $-287\ 294, 498\ 944$ ]).

**Table I. Study population and household characteristics**

Number of children	216
Female	54%
Age, mo, median $\pm$ SD (min-max)	17 $\pm$ 5.8 (18-30)
Baseline anthropometric measurements	
Proportion WAZ $< -2$	22%
Proportion HAZ $< -2$	26%
Proportion WHZ $< -2$	7%
Number of individuals living in household, median $\pm$ SD (min-max)	5 $\pm$ 1.9 (1-12)
Age of caregiver, y, median $\pm$ SD (min-max)	25 $\pm$ 6.2 (17-52)
Caregiver educational level	
No formal education	10%
Primary school education	26%
Secondary education or greater	64%
Floor type in sleeping room	
Earth	76%
Concrete	23%
Other	1%
Unimproved sanitation option*	16%
Fecal calprotectin, $\mu$ g/g	402.67 (193.37-822.30)
EE score	5 (3, 7)
Fecal alpha-1-antitrypsin, mg/g	0.26 (0.16-0.51)
Fecal myeloperoxidase, ng/mL	3576.75 (1969.50-5998)
Fecal neopterin, nmol/L	1505.50 (572.00-3011)

WAZ, weight-for-age z-score; HAZ, height-for-age z scores; WHZ, weight-for-height z scores; EE, environmental enteropathy.

Unless otherwise noted, 95% CIs are shown in parentheses.

\*Unimproved sanitation (defined as no sanitation option, open pit latrine, latrine with broken slab, bucket, or hanging toilet).

**Table II.** Characteristics of child defecation and child feces disposal events during structured observation

	%	n	Total
Children with defecation events	36%	78	216
Total number of child defecation events	-	85	-
Number of defecation events per child			
1	91%	71	78
2	9%	7	78
Children with an open defecation event during structured observation	89%	69	78
Defecation event type			
Open defecation	89%	76	85
Caregiver assisted child toileting event	1%	1	85
Child potty event	9%	8	85
Children with a unsafe feces disposal event during structured observation	84%	65	77
Type of disposal of child feces			
Feces disposed of in a toilet or latrine	14%	12	85
Feces scatter in yard or compound	1%	1	85
Feces disposed in an open space adjacent to compound	78%	66	85
Feces were buried	0%	0	85
Feces disposed of in a location designated for household waste (eg, paper, wrappers)	2%	2	85
No feces disposal	2%	2	85
Other	1%	1	85
Did not observe	1%	1	85
Child handwashing during defecation event			
No	74%	63	85
Yes, 1 hand	1%	1	85
Yes, 2 hands	24%	20	85
Could not observe	1%	1	85
Cleansing agents used during child handwashing events			
Water only	64%	14	22
Bar soap and water	32%	7	22
Did not observe	5%	1	22

## Discussion

This is the first prospective cohort study to our knowledge to investigate the association between unsafe child feces disposal practices, fecal markers of environmental enteropathy, and impaired growth in young children. We found a significant association between caregiver-reported unsafe child feces disposal and elevated environmental enteropathy disease activity scores. Furthermore, both observed and caregiver-reported unsafe feces disposal was associated with impaired growth at our 9-month follow-up. These findings suggest that this practice puts susceptible pediatric populations at risk of environmental enteropathy and growth faltering and supports the hypothesis that unsanitary environmental conditions lead to impaired growth through increased exposure to fecal pathogens causing environmental enteropathy.

The findings from our structured observation indicated that very few young children were wearing diapers in this setting (<1%) and that open defecation events were very common (89%). Our observed rates of unsafe child feces disposal (84%) are consistent with a country report that estimated that 78% of child feces was disposed of in an open space.<sup>41</sup> In addition, a recent study in Orissa, India, found that 81% of caregivers reported unsafe disposal of child feces.<sup>24</sup> There were no significant differences in unsafe child feces disposal practices by child age or by the presence of an improved sanitation option in the household. Furthermore, we observed that caregivers gave fairly accurate reports of this behavior when events during structured observation were compared. This finding suggests that caregiver-reported child feces disposal may be a low-cost alternative to more intensive structured observation as a measure of assessing this behavior. Furthermore, we observed that only 8%

**Table III.** Association between unsafe child feces disposal and fecal environmental enteropathy markers\*

Outcome	Caregiver unsafe feces disposal <sup>†</sup> coefficient (95% CI)			Observed unsafe feces disposal <sup>‡</sup> coefficient (95% CI)		
	Total N	Age adjusted	Fully adjusted <sup>§</sup>	Total N	Age adjusted	Fully adjusted <sup>§</sup>
EE score	216	0.85 (0.14, 1.55) <sup>¶</sup>	0.82 (0.11, 1.53) <sup>¶</sup>	77	0.48 (-1.01, 1.98)	0.45 (-1.07, 1.96)
Calprotectin, $\mu\text{g/g}$	216	-128.71 (-354.09, 96.67)	-146.53 (-372.33, 79.26)	77	3.76 (-350.13, 357.65)	23.25 (-377.51, 331.01)

\*Safe feces disposal was defined by the Joint Monitoring Program definition of feces disposal in a latrine/toilet or buried. Any other method of feces disposal was defined as "unsafe feces disposal."

<sup>†</sup>Children in households with a caregiver that reports unsafe child feces disposal practices at baseline.

<sup>‡</sup>Children in households where caregiver was observed practicing unsafe disposal of child feces.

<sup>§</sup>Fully adjusted models adjust for age, age squared, caregiver educational level, and family size.

<sup>¶</sup>P value less than .05.

Table IV. Association between unsafe child feces disposal and growth measurements\*

Outcome	Caregiver-reported unsafe child feces disposal OR (95% CI)				Observed unsafe child feces disposal OR (95% CI)					
	N	Safe feces disposal	Unsafe feces disposal	Unadjusted	Fully adjusted†	N	Safe feces disposal	Unsafe feces disposal	Unadjusted	Fully adjusted†
Follow-up proportion WAZ < -2	216	14%	26%	2.29 (0.92, 5.30)	2.28 (0.94, 5.51)	71	0%	21%	inf (0.52, inf)	0.97 (0.20, 4.81)
Follow-up proportion HAZ < -2	216	26%	33%	1.40 (0.68, 2.85)	1.33 (0.64, 2.74)	71	30%	28%	0.92 (0.21, 4.04)	0.97 (0.20, 4.81)
Follow-up proportion WHZ < -2	216	0%	9%	inf (1.122, inf)§	1.33 (0.64, 2.74) ‡	71	0%	8%	inf (0.14, inf)	0.97 (0.20, 4.81) ‡

  

Outcome‡	Caregiver-reported unsafe child feces disposal coefficient (95% CI)				Observed unsafe child feces disposal coefficient (95% CI)				
	N	Safe feces disposal †	Unsafe feces disposal	Unadjusted	Fully adjusted**	N	Safe feces disposal**	Unsafe feces disposal**	Fully adjusted†
Change in WAZ	216	-0.10	-0.18	-0.010 (-0.33, 0.13)	-0.06 (-0.28, 0.16)	71	0.15	-0.12	-0.34 (-0.68, -0.01) §
Change in HAZ	216	-0.30	-0.31	0.054 (-0.14, 0.25)	0.12 (-0.071, 0.30)	71	-0.06	-0.31	-0.028 (-0.50, 0.44)
Change in WHZ	216	0.24	-0.003	-0.18 (-0.49, 0.13)	-0.13 (-0.43, 0.17)	71	0.65	0.13	-0.52 (-0.98, -0.06) §

inf, infinity.

\*Safe feces disposal was defined by the Joint Monitoring Program definition of feces disposal in a latrine/toilet or buried. Any other method of feces disposal was defined as "unsafe feces disposal."

†Fully adjusted models adjust for caregiver educational level, and family size.

‡Adjusted ORs and CIs could not be calculated because there were no events in the safe feces disposal group.

§P value (&lt;.05).

¶Median.

\*\*Change is follow-up subtracted from baseline.

of children performed handwashing with soap event after defecation. This finding demonstrates a need for promotion of handwashing with soap in this age group because this a key time for habit formation.<sup>42</sup>

The significant associations between unsafe child feces disposal and elevated environmental enteropathy scores and faltering growth is consistent with the growing body of literature demonstrating that unsanitary environmental conditions put children at an increased risk of environmental enteropathy and impaired growth.<sup>5-7</sup> In our previous studies among children in the cohort, we have found that poor caregiver hand hygiene, presence of animals in the child's sleeping room, and children mouthing dirt during play behavior were all significantly associated with elevated fecal markers of environmental enteropathy.<sup>6,7</sup> In addition, an earlier study in rural Bangladesh found that children in "contaminated" households defined by water quality, unhygienic handwashing conditions, and unimproved sanitation had lower lactulose:mannitol, a measure of intestinal absorptive capacity, and impaired growth.<sup>5</sup> Furthermore, these findings build on earlier work demonstrating that unsafe child feces disposal increases the risk of enteric infections and diarrheal disease in pediatric populations.<sup>22,27-30,33</sup>

The lack of an association between unsafe child feces disposal practices and fecal calprotectin is likely attributed to the environmental enteropathy score representing a more comprehensive measure of intestinal inflammation. We suspect the lack of an association between observed unsafe child feces disposal and environmental enteropathy score is at least in part due to our small sample size. This association needs to be further investigated in a larger study population.

We found that households with unsafe child feces disposal practices had a more than 5 times greater odds of having diarrheagenic *E coli* in the soil in areas where study children were observed playing. This finding provides evidence to support the hypothesis that unsafe child feces disposal puts young children at a greater risk of exposure to enteric pathogens. Furthermore, this finding is consistent with the emerging body of literature demonstrating that contact with contaminated soils is an exposure route to enteric pathogens.<sup>43-46</sup> In rural Kenya, geophagy, defined as the consumption of soil, dirt, or mud, was common in young children and a risk factor for diarrhea and soil transmitted helminth infections.<sup>44,46</sup> In our cohort, geophagy was associated with environmental enteropathy and impaired growth.<sup>6</sup> Therefore interventions are urgently needed to reduce unsafe disposal of child feces that leads to exposure to enteric pathogen in young children.

An intervention study that promoted safe disposal of child feces in urban Bangladesh resulted in a significant 26% reduction in pediatric diarrhea.<sup>47</sup> This finding is consistent with 2 earlier intervention studies in rural Bangladesh that found disposing of child feces in a latrine and no visible feces being present on the household compound was associated with a 27% to 30% reduction in pediatric diarrhea.<sup>48,49</sup> In addition, a recent study in rural Bangladesh found that disposal of child feces in an enclosed space such as a latrine

lead to a 35% reduction in helminthiasis in children younger than 2 years of age.<sup>33</sup> These studies further demonstrate the impact of safe child feces disposal on child health.

This study has several limitations. First is the low number of child defecation and safe feces disposal events during the structured observation period. Future studies should observe child feces disposal practices using a larger sample size and for a longer duration. Second, we did not record how feces were transported for disposal in study households. Previous studies have found that dry leaves or straw or a digging hoe is used in this setting in rural Bangladesh.<sup>23</sup> Future studies should collect information on how child feces are transported for disposal because this could also be an exposure route to fecal pathogens. Third, we did not analyze the stool of study children for enteric pathogens. This would have added to our understanding of the relationship between unsafe feces disposal practices and enteric pathogens in susceptible pediatric populations. Fourth, our study was conducted in rural Mirzapur Bangladesh, and may not be representative of other rural areas in Bangladesh. Fifth, we only observed unsafe child feces disposal events and conducted stool collection at 1 time point. Future studies should assess this behavior and collect stool samples at multiple time points. Finally, we did not assess the caloric intake of study participants. This should be included in future studies.

The results of our study provide preliminary evidence to support the hypothesis that unsafe feces disposal practices leads to impaired growth through increased exposure to enteric pathogens causing environmental enteropathy. Interventions are needed to reduce this high-risk behavior in order to protect the health of susceptible children. ■

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