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ORIGINAL ARTICLE

Airborne enteric micro-organisms and ammonia levels in diaper-changing rooms in kindergartens

P.E. Vaattovaara[1], M. Kivimäenpää[1], P. Vaattovaara[2], P. Pasanen[1] and H. Heinonen-Tanski[1]

1 Department of Environmental Science, University of Eastern Finland, Kuopio, Finland
2 Department of Applied Physics, University of Eastern Finland, Kuopio, Finland

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Correspondence
Piia E. Vaattovaara, Department of Environmental Science, University of Eastern Finland, 70211 Kuopio, Finland.
E-mail: piia.vaattovaara@uef.fi

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Abstract

Aims: We evaluated risks associated with diaper changing in Finnish kindergartens where children were using either modern disposable paper or reusable cloth diapers.
Methods and Results: We determined enteric micro-organisms and ammonia in diaper-changing rooms in four kindergartens in autumn and winter in the ambient air. No coliphages were detected in the air. The numbers of faecal coliforms and enterococci in air were typically low regardless of whether the children used either paper or cloth diapers. Ammonia concentrations increased over the background level because of diaper changing.
Conclusions: The numbers of bacteria or coliphages are not expected to pose any high air hygiene risks, and increased ammonia air concentrations are unlikely to impair the health of staff or children when diapers are changed in modern kindergartens. However, increased ammonia gas concentrations indicate that some other diaper-related gas-phase emissions should be studied to understand better diaper-related health risks.
Significance and Impact of the Study: Modern reusable cloth baby diapers and the modern paper baby diapers used in this study are equally safe with respect to risks from airborne virus, bacteria or ammonia.

Introduction

Humans can be exposed to airborne micro-organisms through many kinds of different contacts and also through vector-borne transmission. It is well known that influenza infections can spread by the aerosol route because the lower respiratory tract is a common site of initiation of infection (Knight 1980). Douwes *et al.* (2003) stated that exposure to environmental bioaerosols can be associated with a wide range of health effects, for example infectious diseases, respiratory diseases and even cancers. Several disease outbreaks have been reported in different environments such as hospital bathrooms where individuals can be exposed to micro-organisms (Kool *et al.* 1999). One aspect of this kind of health risk was reported by one of the biggest Finnish daily newspapers (Turun Sanomat, 9.10.2008), that is, health risks posed by baby diapers. It was speculated that cloth diapers would be sources of viruses by spreading them into the air. For example, this could occur when many children are using the same toilet and some children can be so-called healthy carriers spreading pathogenic micro-organisms. It is known that the bacterial numbers can be rather high, but the numbers of viruses may be lower in infections that cause diarrhoea (Knight 1980; Nicas *et al.* 2004). This concept is supported by a previous kindergarten study, where there were more faecal coliforms present on the surfaces of furnitures and toys if children used cloth diapers as compared to the situation where paper disposable diapers were in use (Van *et al.* 1991). On the other hand, Holaday *et al.* (1995) did not find any significant difference in the intensity of faecal contamination in kindergarten surfaces between children who used cloth or paper diapers. Today, however, it should be noted that there have been major improvements in cloth diapers (Zwane 2010), and it is important to conduct a trial investigating possible health risks and differences in emissions into air between modern-day paper and cloth diapers.

There has been intense debate about the differences between paper and cloth diapers from the perspective of sustainable development and possible health effects. It has been observed that the dirty paper (disposable) baby diapers represent a major load being dumped into solid waste collection system and landfills, for example in Finland 7% of all municipal solid waste consists of disposable paper diapers after source segregation of recyclable materials (YTV 2004). To reduce the amount of waste, many laws restrict the dumping of waste into landfills (EC 1999). Furthermore, the cost of disposable paper diapers can be rather high (Zwane 2010) especially to low-income, younger families with small children (Statistical Center, Finland, 2010). Consequently, many families have started to use reusable cloth diapers to reduce diaper waste and to save money. One consequence is that reusable cloth diapers have become more common in many kindergartens.

This study was initiated because almost nothing was known about the effect of faecal and urinary soiling of different modern diaper types on microbial numbers and the ammonia concentration in the ambient air. We therefore wanted to provide information for users about the possible risks to air hygiene of modern cloth and paper diapers. Another aspect was to determine whether there were any differences between modern cloth and paper diapers during different seasons and whether the nursery personnel would be at an increased risk for occupational problems if children used either cloth or paper diapers.

Materials and methods

The kindergartens and the diapers

Four kindergartens in Kuopio (62°53′32″N; 27°40′42″E), Finland, were included in this study. These kindergartens were selected owing to the high number of diaper-using children, that is, there were five to ten children in each group. Experiments were carried out in autumn (no snow) and in winter (snow cover) when the indoor air was drier because of more intensive heating. The parents of children and the nursery personnel were asked to provide consent to participate in the experiment. The children were 11–32 months old during the experiments. All nursery personnel in each kindergarten and *c.* 80% of all diaper-using children in each kindergarten participated in the study.

The cloth diaper type used here was a commercial pocket diaper, which is often used in kindergartens and at home. Pocket diapers consist of an outer layer of a polyester interlock knit fabric that has been laminated to a thin film of polyurethane (waterproof material) and an inner layer of Coolmax (100% polyester) that allows the child to feel dry. A removable, 30 × 30 cm foldable insert cloth made of 1–2 layers and of urine absorbing bamboo and hemp is located inside the diaper pocket. The cloth diapers used were produced by Myllymuksut Oy, Juupajoki, Finland. The paper diapers examined were also a common commercial diaper type with a frontal tape system common in both kindergartens and homes. The paper diapers were produced by Kesko Oyj, Helsinki, Finland. All the cloth diapers were washed using PIRKKA sensitive washing liquid (Kesko, Finland) in a washing machine at 60°C before each use.

There were 2–8-week measurement periods in the study: the first one in the autumn of 2009 from the end of September up to middle of November and the second one in the winter of 2010 from late January up to late March. Each of four kindergartens was studied four times per season for two nonconsecutive days per week during two consecutive weeks. On each trial week, the children used cloth diapers on 1 day and paper diapers on the other study day. The order of diaper types was changed between the 2 weeks. The diapers were changed by the kindergarten caregivers. They followed the normal hand washing practices and washing of the baby skin contaminated by faeces. The used, dirty diapers were processed following normal practices in the kindergartens, that is, the paper diapers were put into a plastic bag in a closed waste basket in the bathroom, and the cloth diapers were put into separate closed plastic bags, which then were collected by a research worker. Any faeces present in the diaper were left there after a diaper change. The children who did not participate in the study used another toilet during the trial.

Sampling and the laboratory analyses

Coliphages, bacteria and ammonia concentrations in ambient air were sampled around noon during the change of diapers in the bathrooms. The changing of diapers was done continuously without pause one child after one. The air sampling time lasted until 10 min after the last child had had their diaper changed, so that a maximum of 30 min sampling time was used. For example, the sampling lasted 26 min, if the changing took 16 min.

The sampling height was about 1·1–1·3 m above the floor, which is the average breathing height for children and caregivers when they are changing diapers. All samplers were placed as close to the exact changing location at times when children would be wearing diapers or having them changed. In addition, the number of diapers used, the number of urine and faeces in each diaper and the number of dry diapers were calculated. Air relative humidity (RH) and the temperature were measured by Testo 635 (Testo Inc., NJ, USA).

Viruses

The total viable enteric coliphages were sampled from ambient air in the diaper-changing room by a BioSampler liquid impinger (SKC, Inc., Eighty Four, PA, USA) at flow 12·5 l min^{-1} into 20 ml phosphate buffer as described by Heinonen-Tanski *et al.* (2009). Coliphages were determined with the method developed by Grabow and Coubrough (1986). A 100 ml portion of melted phage agar was mixed with 10 ml sampling solution and 5 ml of 1% triphenyltetrazolium chloride (Merck, Darmstadt, Germany) solution in ethanol (Etax A, Altia, Finland) and 90 ml of sterile, distilled and deionized water and 7 ml of 2- to 3-h-old host bacterium cultures were carefully mixed to avoid air bubbles. *Escherichia coli* strains ATCC 13706 and ATCC 15597 were used as hosts for somatic and male-specific coliphages. After overnight incubation at 37°C, the phage plaques were counted and the concentration expressed as plaque-forming units in 1 m^3 of air (PFU m^{-3}). The lowest detection limit was four PFU m^{-3} (20 min sample).

Bacteria

The faecal coliforms and enterococci were sampled from the air in the diaper-changing room by a 6-stage Andersen impactor at flow rate 28·3 l min^{-1} (Andersen 1958). Chromocult agar (Merck) was used for the faecal coliforms. For the enterococci, we used Slanetz–Bartley agar (Lab M, Lancashire, UK). Following regular microbiological practices, the faecal coliforms were incubated at 37°C for 24 and 48 h and the enterococci at 37°C for 48 and 72 h. The colonies were counted after incubation, and the counts were corrected by the positive-hole method (Andersen 1958). The results were expressed as numbers of colony-forming units in 1 m^3 of air (CFU m^{-3}). The lowest detection limits for both bacteria were 1·2–2·7 CFU m^{-3}.

Ammonia

Ammonia was sampled by the liquid impinger (model 7540-10; ACE Glass, Vineland, NJ, USA) at flow rate 2·2 l min^{-1} during the autumn measurement period. To ensure that the flow rates are sufficient also in the case when just few diapers are changed, the flow rates were increased to 2·7 l min^{-1} during winter measurement period. To avoid vaporization of ammonia from the collected sample, 10 ml of 0·05 mol l^{-1} H_2SO_4 solution was located at the bottom of the impinger. The ammonia was sampled two times per measurement session. The first sample was taken for background concentration of ammonia before any diaper changing or some other use of that room. The second sample for assaying the ammonia level was taken during diaper changing. The voltages (mV) owing to the samples were measured with an ammonia electrode (Ammonia Electrode Model 95-10; Orion Research Incorporated, Cambridge, MA, USA) and ion analyzer (Orion Research Expandable Ion Analyzer EA920). Standard ammonia concentrations were used to analyse ammonia concentrations in kindergarten air. The ammonia concentrations of the background and changing time samples, respectively, were estimated from a standard curve (voltage of ion analyzer vs. ammonia concentration).

Statistics

The average numbers of coliforms, enterococci and the concentration of ammonia were calculated per diaper type (cloth or paper) for each season separately (autumn or winter; the number of statistical replicates was the number of measurements, that is, $n = 8$) and for both seasons together (autumn and winter; $n = 16$). The geometric mean (GM) was used for micro-organisms (Robertson 1932) and the arithmetic mean for ammonia. General linear models for repeated-measures ANOVA for seasons as a within-subject factor and diaper type as a between-subject factor were used to study whether the parameters were affected by diaper type or season. This allowed a determination of whether the numbers of airborne contamination organisms were attributable to the use of different diaper types as well as the effect of the season. The number of statistical replicates was the number of kindergartens ($n = 4$). The preliminary investigation found no association between air concentrations of emitted ammonia or micro-organisms and other background variables (e.g. time or number of diapers worn by children). Thus, no weighting of the data was performed. When necessary, data were logarithmically transformed to fulfil requirements of the ANOVA.

Results

Viruses

In this study, no coliphage viruses were detected; thus, all the numbers of both coliphages were less than the detection limit (four PFU m^{-3}) in all air samples regardless of whether paper or cloth diapers were in use or whether the season was autumn or winter.

Bacteria

For the cloth diapers, the observed highest number of airborne coliforms was 24·2 CFU m^{-3}, whereas the lowest number was below the detection limit (1·2 CFU m^{-3}). Similarly, the highest numbers of enterococci were

72·1 CFU m^{-3}, and again the lowest numbers were below the detection limit (1·2 CFU m^{-3}). For paper diapers, the numbers of coliform bacteria varied from under the detection limit (1·4 CFU m^{-3}) up to 155·2 CFU m^{-3} and those for enterococci were below the detection limit (1·2 CFU m^{-3}) up to 30·7 CFU m^{-3} (Table 1).

No main effects of the season and diaper type were statistically significant for the number of bacteria (Table 1). However, the interactions between the season and diaper type were significant for the numbers of bacteria in the air (Table 1). The numbers of coliforms increased from autumn to winter, and the increase was higher when cloth diapers were being used. However, the numbers of coliforms were higher when paper diapers were in use. In contrast to the situation with coliforms, the numbers of enterococci declined from autumn to winter, and this decrease was more prominent when the paper diapers were being used. Furthermore, the numbers of enterococci were higher when cloth diapers were being used (Table 1). However, it must be noted that the observed numbers and the actual differences in the numbers of bacteria between the diaper types or between seasons were low.

In the winter, the numbers of coliforms included one exceptionally high value in the paper diapers, which differed remarkably from the other values. One possible reason for such a high value could be an infected child using a paper diaper. Nonetheless, there were no more illnesses than were normally recorded in that day care centre before or after that specimen.

Ammonia

The averaged background concentrations during the days when cloth diapers were in use were 13·7 μg m^{-3} in autumn and 9·5 μg m^{-3} in winter. Correspondingly, for paper diapers, the averaged background concentrations were 16·1 μg m^{-3} in autumn and 11·3 μg m^{-3} in winter (Table 1).

The ammonia levels in air varied for cloth diapers between 22·9–32·1 μg m^{-3} in autumn and 31·5–49·9 μg m^{-3} in winter during changing. Correspondingly, for paper diapers, the ammonia levels varied from 41·2 to 64·0 μg m^{-3} in autumn and from 24·7 to 31·7 μg m^{-3} in winter. There were no statistically significant differences in ammonia air concentrations between cloth and paper diapers, between seasons or due to combined season × diaper type (Table 1).

Based on the recommendations of the Finnish Ministry of Social Affairs and Health (2003), the detected ammo-

Table 1 The geometric means (GM) values ± standard error of mean (SEM) for indicator bacteria and the change in arithmetic averages ± SEM for ammonia concentrations measured in indoor air of four kindergartens with either cloth diapers or disposable paper diapers in autumn and winter with statistical significances (*P*-values) of main effects and interactions from general linear model for repeated measures (n = 4; the number of kindergartens) and ammonia air concentration distributions (%) divided into three levels (based on the Finnish Ministry of Social Affairs and Health recommendation, 2003)

Diaper type; season; *n*	Coliforms (CFU m^{-3})	Enterococci (CFU m^{-3})	Ammonia change (μg m^{-3})
Cloth; autumn; *n* = 8	3·2 ± 1·0	6·4 ± 0·6	13·8 ± 3·0
Cloth; winter; *n* = 8	7·6 ± 0·4	5·8 ± 0·5	31·6 ± 6·5
Cloth; both seasons; *n* = 16	4·9 ± 0·6	6·1 ± 0·8	22·7 ± 4·2
Paper; autumn; *n* = 8	5·4 ± 0·6	5·2 ± 0·5	36·5 ± 7·3
Paper; winter; *n* = 8	8·2 ± 0·6	2·9 ± 0·5	16·9 ± 2·8
Paper; both seasons; *n* = 16	6·6 ± 0·3	3·9 ± 0·3	26·7 ± 4·6
Statistical significance	*P*-values		
Diaper type	0·861	0·560	0·460
Season	0·310	0·094	0·812
Season × diaper type	0·040*	0·048*	0·059
	The distribution percentage (%) of ammonia levels in ambient air		
Diaper type; season; *n*	The normal level (<20 μg m^{-3})	Increased (20–40 μg m^{-3})	Remarkably increased (>40 μg m^{-3})
Cloth; autumn; *n* = 8	37·5	25	37·5
Cloth; winter; *n* = 8	0	62·5	37·5
Paper; autumn *n* = 8	0	37·7	62·5
Paper; winter; *n* = 8	12·5	75	12·5
Background; *n* = 16	100	0	0

*Statistically significant difference.

nia indoor air concentrations have been divided into three levels: below the background level (<20 μg m^{-3}), elevated level (20–40 μg m^{-3}) and considerably elevated level (>40 μg m^{-3}) subdivided according to whether both cloth and paper diaperings were being used and for both seasons (Table 1). The amount of ammonia was either elevated or considerably elevated in the air in the diaper-changing room.

Temperature and RH

The temperatures increased in the diaper-changing rooms during the measurements during both seasons. The average temperature increased from 22·6 to 24·3°C at the end of measurement for cloth diapers and from 22·9 to 24·2°C for paper diapers in autumn. Correspondingly, the average temperatures increased from 22·9 to 24·2°C for cloth diapers and from 22·7 to 24·1°C for paper diapers in winter.

The averaged RH decreased in the air in the diaper-changing room during the measurements in the autumn but increased in the winter. The averaged RH decreased from 25·5 to 24·1% for cloth diapers and from 26·3 to 25·6% for paper diapers in the autumn. The averaged RH increased from 11·7 to 13·8% for cloth diapers and from 11·9 to 14·3% for paper diapers in winter.

Discussion

This study investigated the hygiene-related properties of modern diapers. The most interesting result was the fact that no coliphages were found in any of air samples of kindergarten bathrooms regardless of whether the air sampling was done when children were using modern reusable cloth diapers or disposable paper diapers or whether it was autumn or winter. This result was not due to the analytical method, because coliphages were detected in the air of wastewater treatment plants using a similar sampling and determination protocol (Heinonen-Tanski *et al.* 2009).

Importantly for air hygiene, the numbers of enteric airborne bacteria were generally low. Therefore, changing diapers is not expected to produce any significant air-related microbe risks in the typical diaper-changing room conditions in Finnish kindergartens.

The ambient indoor bathroom air ammonia concentrations in background air and during changing diapers were clearly lower than usually regulated occupational atmospheric exposure maximum limits (18–40 mg m^{-3}; WHO 1990), which means that they should not cause any health effects in workers. In spite of the fact that the indoor air concentrations of ammonia were typically elevated or considerably elevated during diaper changing, it is unlikely that they would be responsible for any acute or chronic health disorders when the babies' diapers are being changed in bathrooms where the ventilation is working properly. The results of this study also show that there are no statistically significant differences between paper and cloth diapers in terms of the amounts of ammonia gas in air in bathrooms of the kindergartens. There were no statistical differences between the seasons. However, the ammonia gas concentrations that accumulate when diapers are being changed suggest that also other possible diaper-related gas-phase emissions need to be studied to understand better the air hygiene risks associated with different diaper types.

In this study, paper diapers and cloth diapers proved to be equally safe to use in terms of viruses, bacteria or ammonia levels. It should also be noted that whatever the diaper type is chosen, the diaper should not be left on the child for long periods to avoid discomfort, skin damage and infections.

Generally, the results of this study can partly be applied into modern sheltered housing for the elderly and hospitals and other environments where diapers are worn. Furthermore, the research and development of diapers and diaper-changing conditions needs to continue and provide information about good diapering practices, which should not be overlooked.

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