HHS Public Access

Author manuscript

Dermatitis. Author manuscript; available in PMC 2020 March 01.

Published in final edited form as:

Dermatitis. 2019 ; 30(2): 129–134. doi:10.1097/DER.0000000000000455.

Isothiazolinone Content of US Consumer Adhesives: Ultrahigh-Performance Liquid Chromatographic Mass Spectrometry Analysis

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Abstract

Background: There are limited data regarding the prevalence and concentration of isothiazolinone preservatives in consumer adhesives.

Objectives: The aim of this study was to determine the prevalence and concentration of 5 specific isothiazolinones (methylisothiazolinone [MI], methylchloroisothiazolinone [MCI], benzisothiazolinone [BIT], butyl BIT, and octylisothiazolinone) in US adhesives.

Methods: Thirty-eight consumer adhesives were analyzed using ultrahigh-performance liquid chromatographic–mass spectrometry. Fisher exact tests were used to test for isothiazolinone content and: 1) glue format (2) application purpose and 3) extraction method.

Results: Nineteen adhesives (50%) had at least 1 isothiazolinone, and 15 contained 2 isothiazolinones. Frequencies and concentrations were as follows:MI (44.7%; 4–133 ppm), MCI (31.6%; 7–27 ppm), BIT (15.8%; 10–86 ppm), and octylisothiazolinone (2.6%; 1 ppm). Butyl BIT was not detected in any of the adhesives. Format (stick vs liquid) was not statistically associated with isothiazolinone presence. At least half of adhesives in the following application purposes had at least 1 isothiazolinone: shoe, craft, fabric, and school. All-purpose glues had a statistically significant lower concentration of MI and MCI, whereas craft glues were associated with higher concentrations of MI and MCI. Compared with other glues, fabric adhesives were associated with a higher risk of containing BIT.

The authors have no conflicts of interest to declare.

No reprints available.

Conclusions: Half of the tested adhesives contained at least 1 isothiazolinone. Methylisothiazolinone and MCI were the most common. Consumers and dermatologists should be aware of adhesives as a source of isothiazolinones.

Isothiazolinone preservatives are commonly used in water-based consumer products. Several isothiazolinones have been reported to cause allergic contact dermatitis, including benzisothiazolinone (BIT), octylisothiazolinone (OIT), butyl BIT (BBIT), methylchloroisothiazolinone (MCI), and methylisothiazolinone (MI).[1–6] The epidemic of allergy to MCI and MI in Europe and North America has been extensively addressed elsewhere.[7]

Occupational contact dermatitis to adhesives is well recognized.[8–10] In a study of North American production workers, adhesives were the most common source of occupationally related allergens.[11] Isothiazolinones are newly recognized adhesive allergens; MI, MCI/MI, and BIT have been reported in industrial adhesives.[12–16] A study of 1745 Finnish patch test patients revealed 36 patients with occupationally related MCI and/or MI sensitivity including 1 case attributed to glue exposure in a carpenter.[17]

In addition to occupational exposures to isothiazolinones in glue, consumers are also at risk. Silva et al[18] described a 20-year-old patient with foot dermatitis and a positive patch test reaction to MI. A use test revealed a positive reaction to an adhesive-covered portion of her shoe; subsequent chemical analysis revealed the presence of MI. The Park Nicollet Contact Dermatitis Group recently reported a child with hand dermatitis and MI sensitivity; the source was traced to MI in school glue used to make slime, a homemade stretchy play material.[19] Since that case, we have had an additional 3 children with allergy to slime due to MI in consumer glue (data unpublished). Difficulty finding alternatives for these patients led to this study.

Very little information is available regarding the frequency and concentration of isothiazolinones within adhesive products. One Japanese study sought to characterize isothiazolinone presence (MCI, MI, BIT, and OIT) in wallpaper adhesives using gas chromatography–mass spectrometry. Of the 7 tested adhesives, only 1 was free of isothiazolinones.[20]

In the United States, avoidance of isothiazolinones in personal care products is possible through careful review of labeled ingredients and use of computerized programs such as the American Contact Dermatitis Society's nonprofit Contact Allergen Management Program.[21] However, other products such as glues, paints, and cleaning products are not required to be labeled with nonhazardous materials such as isothiazolinone preservatives.[22] The aim of this project was to identify the presence and concentration of MCI, MI, BIT, BBIT, and OIT in US consumer glues.

METHODS

Adhesive Collection

A convenience sample of 37 adhesives was purchased from online distributors and local retailers in Minnesota between January and February 2017. Both liquid and stick adhesives

from 13 manufacturers were sampled from a variety of intended applications including all-purpose, school, wood, craft, fabric, eyelash, and shoe. One sample contained 2 liquid chambers; each was tested independently for a total of 38 samples. During acquisition, composition for most adhesives was not readily available on either packaging or product safety datasheets. Once all samples were collected, they were mailed via ground postage to the National Institute for Occupational Safety and Health (Morgantown, WV) at the Centers for Disease Control and Prevention for analysis.

Reagents

Methylisothiazolinone (≥98%, CAS# [Chemical Abstracts Service] 2682–20-4), BIT (96%, CAS# 2634–33-5), OIT (99.9%, CAS# 26530–20-1), atrazine-desethyl (99%, CAS# 6190–65-4, used as internal standard [IS]), and trichloroethylene were obtained from Sigma-Aldrich Inc (Millipore Sigma, St Louis, MO). Methylchloroisothiazolinone/MI (67.7% MCI, CAS# 26172–55-4) was obtained from Combi-Block, Inc (San Diego, CA), and BBIT (95%, CAS# 4299–07-4) was purchased from Creative Dynamics Inc (BOC Science, Shirley, NY). The MCI standard was a mixture of MCI and MI at a ratio of MCI/MI = 2/1 in H_2O. The mixture by ultrahigh-performance liquid chromatographic–mass spectrometry (UHPLC-MS) was found to contain only MCI and MI. The standard MI (≥98%) was used to determine the exact MI concentration in the mixture and then calculate the MCI concentration in that standard. Acetonitrile (LC/MS grade), methanol (high-performance liquid chromatographic grade), and formic acid (LC/MS grade) were purchased from Fisher Chemical Inc (Battle Ground, WA). Water was made by Milli-Q (Millipore Sigma, Burlington, MA).

Glues Sample Preparation: Extraction Method 1

Approximately100 mg of each glue was weighed in a 6-mL glass vial containing 3 mL of an ice-cold mixture-B consisting of 0.4% of formic acid and acetone at a ratio of 50/50 (vol/vol) containing 2.0 ng/μL of IS, immediately vortexed for 30 seconds, and then diluted 40-fold (wt/vol) with mixture-B. The sample was again vortexed for another 30 seconds and then sonicated for 20 minutes. Each sample was again briefly vortexed and then centrifuged for 20 minutes at 1360*g* at 4°C. The top phase was recovered and filtered through a 13-mm, 0.2-μm polytetrafluoroethylene syringe filter into 2 vials for UHPLC-MS analysis. Preliminary evaluations of 5-, 10-, 20-, 30-, 40-, and 50-fold dilutions of adhesives were conducted, and the 40-fold dilution was determined to be optimal for subsequent quantitative analyses. Extracts of samples with undetectable isothiazolinone levels at the 40-fold dilution were also evaluated without dilution.

Glues Sample Preparation: Extraction Method 2

For glues not solubilized by mixture-B, approximately100 mg of each adhesive was added to a 6-mL glass vial containing 1 mL of trichloroethylene and immediately vortexed for 30 seconds followed by addition of 3 mL of ice-cold mixture-B. After a 30-second vortex, the mixture was sonicated for 20 minutes and then centrifuged for 20 minutes at 1360*g* at 4°C. The top phase was filtered through a 13-mm, 0.2-μm polytetrafluoroethylene syringe filter into 2 vials for UHPLC-MS analysis.

Ultrahigh-Performance Liquid Chromatographic–Mass Spectrometry

All isothiazolinones in glues were analyzed by an ultrahigh-performance liquid chromatograph coupled to a quadrupole 3-dimensional ion trap mass spectrometer (Thermo Fisher Scientific, San Jose, CA) equipped with a heated electrospray ionization probe. The analytes were separated on an Accucore C18 high-performance liquid chromatography column (2.1 × 50 mm) by a mobile phase consisting of acetonitrile and 0.1% formic acid under a gradient elution program at a fixed 1-mL/min flow rate. The analytes were ionized by the heated electrospray ionization probe operated in positive mode. The MS detector was operated to select specific ions during different periods of each run, and the second-stage MS was set to zero collision energy. The protonated molecular ions (M + 1) monitored were 0.00 to 0.45 minutes for MI (116 m/z), 0.45 to 0.9 minutes for CMI (150 m/z), 0.9 to 1.4 minutes for BIT (152.3 m/z), 1.4 to 2.0 minutes for IS (188.3 m/z), 2.0 to 2.6 minutes for BBIT (208.3 m/z), and 2.6 to 3.5 minutes for OIT (214.4 m/z), with their respective individual tune method. Quantification of the analytes was done using response factors calculated from a 6-point calibration curve. All response factors were based on the ratio of peak area of each compound with IS. The concentrations of calibration samples were in picograms per microliter. The limits of quantification of this method at less than 20% relative SD were MI 150 pg/injection, MCI 200 pg/injection, BIT 230 pg/injection, BBIT 70 pg/injection, and OIT 75 pg/injection.

Data Analysis

Data were collected and stored in Excel (Excel 2010; Microsoft Corporation, Redmond, WA). Statistical analyses were performed using SAS (version 9.2, Statistical Analysis System; SAS Institute Inc, Cary, NC). Fisher exact tests were used to test for the following associations with isothiazolinone content: (1) glue format (liquid vs stick), (2) application purpose (7 categories), and (3) extraction method (1 vs 2). Three categories of isothiazolinone concentrations were examined: high, low, and none. On the basis of overall average concentrations, an arbitrary cutoff of 25 ppm was used to define “high” versus “low” isothiazolinone content. $P \leq 0.05$ was considered statistically significant.

RESULTS

A total of 33 liquid and 5 stick adhesives were analyzed. Most were all-purpose (31.2%) or school (26.3%) glues (Table 1). Of the 38 adhesives tested, 19 (50%) were found to contain at least 1 isothiazolinone. Fifteen (39.5%) had 2 isothiazolinones. The most common isothiazolinones were MI (n = 17) and MCI (n = 12), followed by BIT (n = 6) and OIT (n = 1). Butyl BIT was not detected in any tested adhesives (Tables 2, 3). The concentration of isothiazolinones was highest for MI (4–133 ppm), followed by BIT (11–86 ppm), MCI (7–28 ppm), and OIT (1 ppm). Isothiazolinone presence by application purpose is listed in Table 4.

Format: Stick Versus Liquid

Five stick adhesives were tested; 3 were identified as “school glue,” and 2 were “all-purpose.” Two (1 school and 1 all-purpose) contained an isothiazolinone, BIT.

Thirty-three liquid adhesives were tested; 17 contained at least 1 isothiazolinone. Liquid glues contained the following isothiazolinones: MI (n = 17), MCI (n = 12), BIT (n = 4), and OIT (n = 1). There was no statistical association between format (liquid vs stick) and isothiazolinone presence (yes vs no; $P = 1.000$).

Methylisothiazolinone

Seventeen adhesives contained MI. There was no association of MI concentration (high vs low, excluding none; high vs low including none; any vs none) and application purpose (all-purpose vs other; school vs other; wood vs other; fabric vs other; craft vs other; all Ps > 0.3068) with the following exceptions. Compared with other glues, all-purpose adhesives were associated with a lower likelihood of containing MI ($P = 0.0336$). Compared with all other glues, craft glues were associated with a higher likelihood of containing MI at higher concentrations (high vs low excluding no isothiazolinone, approached significance at $P = 0.0987$) (high vs low including no isothiazolinone, $P = 0.0207$).

Methylchloroisothiazolinone

Twelve adhesives contained MCI. There was no association of MCI concentration (high vs low, excluding none; high vs low including none; any vs none) and application purpose (all-purpose vs other; school vs other; wood vs other; fabric vs other; craft vs other; $P > 0.2867$) with the following exceptions. Compared with other glues, all-purpose adhesives were associated with a lower likelihood of containing MCI (approached significance at $P = 0.0600$). Compared with all other glues, craft glues were associated with a higher likelihood of containing MCI ($P = 0.0272$) and at higher concentrations (high vs low excluding none, approached significance at $P = 0.0909$) (high vs low including none, $P = 0.0142$).

Benzisothiazolinone

Six adhesives (15.8%) contained BIT. There was no association of BIT concentration (high vs low, excluding none; high vs low including none; any vs none) and application purpose (all-purpose vs other; school vs other; wood vs other; fabric vs other; craft vs other; $P > 0.2639$) with 1 exception. Compared with other glues, fabric adhesives were associated with a higher likelihood of containing BIT ($P = 0.0045$).

Extraction Method

Eleven adhesives were not solubilized by methanol, acetonitrile, acetone, ethanol, 1-propanol, or 2-propanol (extraction method 1), and thus the nonpolar solvent tetrachloroethylene (extraction method 2) was required (Table 3). Adhesives analyzed by method 1 were significantly more likely to have isothiazolinones (none vs any) as compared with those analyzed by method 2 (Fisher exact test, $P = 0.003$; data not shown).

DISCUSSION

This analysis of 38 US consumer adhesives has several important findings. First, half of the tested glues were found to contain at least 1 isothiazolinone. The most common were MI and MCI, followed by BIT and OIT. Butyl BIT was not detected in any tested adhesive. Second, format (stick vs liquid) was not statistically associated with isothiazolinone

presence. Third, at least half of adhesives in the following application purposes had at least 1 isothiazolinone: shoe, craft, fabric, and school. All-purpose glues had a statistically significant lower concentration of MI and MCI, whereas craft glues were associated with higher concentrations of MI and MCI. Compared with other glues, fabric adhesives were associated with a higher likelihood of containing BIT.

Isothiazolinone Presence

The presence of isothiazolinones in adhesives is not surprising as this has been documented in case reports. The MI concentration of the shoe glue in our study (7.4 ppm) was similar to that in a European shoe glue (9.8 ppm).[23] Rosero-Moreano et al[24] analyzed 3 food packaging adhesives for MCI and MI using high-performance liquid chromatography/MS. Methylchloroisothiazolinone was present in all 3 (8.8–10.6 ppm), and MI was present in 2 adhesives (46.2 and 55.2 ppm). Similarly, a study of 7 Japanese wallpaper adhesives found that MI and MCI were present in 85.6% (6/7) of the adhesives and OIT was present in 42.9% (3/7). Concentrations ranged from 4.4 to 10.4 ppm for MCI, 13.7 to 26.5 ppm for MI, and 5.9 to 133.0 ppm for OIT. Benzisothiazolinone was not present in any of the tested materials.[20] These studies highlight the prevalence and potential ubiquitous nature of these antimicrobials within adhesive products.

It is not surprising that adhesives not solubilized by methanol, acetonitrile, acetone, ethanol, 1-propanol, or 2-propanol (extraction method 1) and requiring the nonpolar solvent tetrachloroethylene (extraction method 2) had lower isothiazolinone content. These glues are less water soluble and therefore likely to have less need for preservatives such as isothiazolinones.

Format/Application of Adhesive

There are many formulas for making adhesives.[25] Water is an important component. Thus, preservatives are necessary to prevent bacterial growth. Therefore, the presence of isothiazolinones in liquid and semisolid adhesives, such as glue sticks, is altogether not surprising. We had surmised that stick adhesives may be less likely to contain isothiazolinones than liquid formulations; however, this was not the case. Of the 5 stick formulations in our study, 2 contained BIT. If more analyses of stick formulations verify that MI and MCI are not commonly used, stick glues may be a useful substitute for individuals with sensitivity to MCI/MI but not BIT. It is estimated that only 10% of individuals with MCI/MI allergy cross-react to BIT.[26]

At least half of the following types of glues contained isothiazolinones: school, craft, shoe, and fabric. The 1 eyelash glue tested did not have an isothiazolinone. More studies are needed to verify that all-purpose glues and wood glues are truly “safer” for patients sensitive to isothiazolinones.

Clinical Implications

There is a high degree of cross-reactivity between MCI and MI. Cross-reactivity of MCI/MI and OIT and BIT is variable, however. A study of 3938 patients tested to MI, BIT, and OIT found that fewer than 10% of individuals with MI allergy also reacted to BIT and OIT. In

contrast, individuals sensitized to BIT and OIT had higher proportions of positive reactions (20.5% and 50%, respectively) to MI.[26] Therefore, patch testing to all isothiazolinones may be useful in specific patients. For example, if a patient reacts only to MCI/MI but not BIT and OIT, stick glue formulations may be a good option based on our preliminary results.

More studies are also needed to establish the levels of isothiazolinones needed to elicit allergic contact dermatitis in sensitized individuals. In a double-blind, placebo-controlled study, Zachariae and colleagues[23] tested 25 individuals with confirmed MCI/MI allergy and 10 controls in a repeat open application test over 4 weeks. That study found that the elicitation threshold of MCI/MI is expected to be in the proximity of 0.025 μg/cm^2.

LIMITATIONS

This study has several limitations. First, a convenience sampling methodology was used for gathering samples rather than a computer-generated randomization scheme. Second, adhesives were purchased in Minnesota. Although it is unlikely that ingredients of major brands vary by region, our findings may not be generalizable to other countries. Third, adhesives analyzed by method 2 were less likely to have isothiazolinones than those analyzed by method 1; although this is most likely due to the lower water content (thus less need for preservative), it is possible that the chemical analysis method affected detection. Finally, only consumer-based adhesives were tested, so results may not be reflective of concentration or prevalence of isothiazolinones in occupational settings.

CONCLUSIONS

Isothiazolinones were present in half of the commercial adhesives tested. Methylisothiazolinone and MCI were the most commonly found isothiazolinone compounds, whereas BBIT was not detected in any of the samples. Isothiazolinones were common in craft, fabric, and school glues.

No. Containing IC MI MCI BIT OIT BBIT

Acknowledgments

Funding was provided by an American Contact Dermatitis Society Research Grant.

This study involved the use of resources and facilities at the Minneapolis Veterans Affairs Medical Center as well as the National Institute for Occupational Safety and Health of the Centers for Disease Control and Prevention. The contents do not represent the views of the US Department of Veterans Affairs or the US Government. The findings and conclusions of this report are those of the authors and do not necessarily represent the official position of the National Institute for Occupational Safety and Health, Centers for Disease Control and Prevention. Mention of any company or product does not constitute endorsement by the National Institute for Occupational Safety and Health, Centers for Disease Control and Prevention.

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TABLE 1.

Characteristics of Adhesives

Adhesive Property	n (N = 38*)	%
Format		
Liquid	33*	86.8
Stick	5	13.2
Application purpose		
All-purpose	12*	31.2
School	10	26.3
Wood	5	13.2
Craft	5	13.2
Fabric	4	10.5
Eyelash	1	2.6
Shoe	1	2.6

* Thirty-seven adhesives were tested; 1 adhesive contained 2 separate chambers, both of which were tested.

TABLE 2.

Isothiazolinone Content and Concentration

Isothiazolinone	Samples, n (%)	Range, ppm	Average (SD), ppm
MI	17 (44.7)	4–133	25 (30)
MCI	12 (31.6)	7–27	17 (7)
BIT	6 (15.8)	11–86	57 (33)
OIT	1 (2.6)	1.52	—
BBIT	0 (0.0)	—	—

BBIT indicates butyl benzisothiazolinone; BIT, benzisothiazolinone; MCI, methylchloroisothiazolinone; MI, methylisothiazolinone; OIT, octylisothiazolinone.

TABLE 3.

Isothiazolinone Content of Adhesives

Manufacturer	Product Name	Application Purpose	Extraction Method	Format	MI, ppm	MCI, ppm	BIT, ppm	BBIT, ppm	OIT, ppm
3M	Scotch Maximum Strength Adhesive	All-purpose	2	Liquid	ND	ND	ND	ND	ND
	Scotch Permanent Glue Stick	All-purpose	2	Stick	ND	ND	ND	ND	ND
American International Industries	DUO Striplash Adhesive	Eyelash	2	Liquid	ND	ND	ND	ND	ND
Beacon Adhesives Inc	527 Multi-Use Glue	All-purpose	2	Liquid	ND	ND	ND	ND	ND
Crayola LLC	Washable Glue	School	1	Liquid	6.6	10.1	ND	ND	ND
	Washable Glue Sticks	School	1	Stick	ND	ND	ND	ND	ND
Duncan Enterprise Company	Aleene’s Clear School Tacky Glue	School	1	Liquid	4.3	8.3	ND	ND	ND
	Aleene’s Fabric Fusion	Fabric	1	Liquid	133.4	ND	83.2	ND	ND
	Aleene’s Fast Grab Tacky Glue	Craft	1	Liquid	34.77	27.59	ND	ND	ND
	Aleene’s Flexible Stretchable Fabric Glue	Fabric	1	Liquid	10.9	ND	73.8	ND	ND
	Aleene’s Original Tacky Glue	Craft	1	Liquid	38.0	11.0	ND	ND	ND
	Aleene’s Quick Dry Tacky Glue	Craft	1	Liquid	34.2	26.9	ND	ND	ND
	Aleene’s School Tacky Glue	School	1	Liquid	32.5	8.3	ND	ND	ND
	Aleene’s Stop Fraying	Fabric	1	Liquid	9.6	ND	63.87	ND	ND
Eclectic Products, Inc	Amazing Goop	All-purpose	2	Liquid	ND	ND	ND	ND	ND
	E6000	All-purpose	2	Liquid	ND	ND	ND	ND	ND
	E6000 Jewelry & Bead	Craft	2	Liquid	ND	ND	ND	ND	ND
	Original Shoe Goo Boots & Gloves		2	Liquid	7.4	ND	ND	ND	ND
Elmer’s Products Inc	Carpenter’s Wood Glue	Wood	1	Liquid	11.3	23.5	ND	ND	1.5
	Glue-All	All-purpose	1	Liquid	26.5	18.7	ND	ND	ND
	Krazy Glue	All-purpose	2	Liquid	ND	ND	ND	ND	ND
	Rubber Cement	School	1	Liquid	ND	ND	ND	ND	ND
	School Glue	School	1	Liquid	ND	ND	ND	ND	ND
	X-TREME School Glue	School	1	Liquid	12.8	19.1	ND	ND	ND
	All-Purpose Gluestick	All-purpose	1	Stick	ND	ND	86.5	ND	ND
	School Glue Naturals	School	1	Stick	ND	ND	ND	ND	ND
Franklin International	Titebond II Premium Wood Glue	Wood	1	Liquid	ND	ND	ND	ND	ND

Manufacturer	Product Name	Application Purpose	Extraction Method	Format	MI, ppm	MCI, ppm	BIT, ppm	BBIT, ppm	OIT, ppm
	Titebond III Ultimate Wood Glue	Wood	1	Liquid	ND	ND	ND	ND	ND
	Titebond Original Wood Glue	Wood	1	Liquid	ND	ND	ND	ND	ND
Gorilla Glue Inc	Epoxy Clear (Tube 1)	All-purpose	1	Liquid	ND	ND	ND	ND	ND
	Epoxy Clear (Tube 2)	All-purpose	1	Liquid	ND	ND	ND	ND	ND
	Original Gorilla Glue	All-purpose	2	Liquid	ND	ND	ND	ND	ND
	Wood Glue	Wood	1	Liquid	10.5	7.3	ND	ND	ND
Henkel Corporation	Loctite Vinyl, Fabric & Plastic Flexible Adhesive	Fabric	2	Liquid	ND	ND	ND	ND	ND
Plaid Enterprises Inc	Mod Podge Gloss	Craft	1	Liquid	20.5	20.7	ND	ND	ND
Target Brands Inc	up&up Washable School Glue	School	1	Liquid	8.1	18.4	ND	ND	ND
	up&up Disappearing purple glue sticks	School	1	Stick	ND	ND	10.8	ND	ND
Weldbond Adhesives Products	Universal Adhesive	All-purpose	1	Liquid	16.5	ND	20.7	ND	ND

BBIT indicates butyl benzisothiazolinone; BIT, benzisothiazolinone; MCI, methylchloroisothiazolinone; MI, methylisothiazolinone; ND, not detected; OIT, octylisothiazolinone.

TABLE 4.

Isothiazolinone Content Based on Application Purpose

		No. Containing IC	MI	MCI	BIT	OIT	BBIT
Category Use	**n (N = 38)**	**n (%)**	**n (Range, ppm)**	**n (Range, ppm)**	**n (Range, ppm)**	**n (Range, ppm)**	**n (Range, ppm)**
All-purpose *	12	3 (25)	2 (16.5–26.5)	1 (18.7)	2 (20.7–86.5)	—	—
School	10	6 (60)	5 (4.3–32.5)	5 (8.3–19.1)	1 (10.8)	—	—
Craft	5	4 (80)	4 (20.5–38.0)	4 (11.0–27.6)	—	—	—
Wood	5	2 (40)	2 (10.5–11.3)	2 (7.3–23.5)	—	1 (1.5)	—
Fabric	4	3 (75)	3 (9.6–133.4)	—	3 (63.9–83.2)	—	—
Shoe	1	1 (100)	1 (7.4)	—	—	—	—
Eyelash	1	0 (0.0)	—	—	—	—	—

* One sample contained 2 separate adhesive chambers, both of which were tested.

BBIT indicates butyl benzisothiazolinone; BIT, benzisothiazolinone; IC, isothiazolinone; MCI, methylchloroisothiazolinone; MI, methylisothiazolinone; OIT, octylisothiazolinone