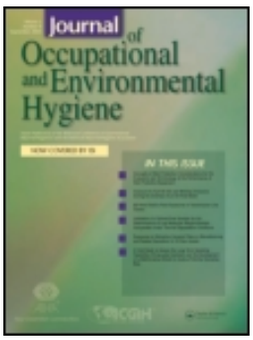

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Development of a Sampling Patch to Measure Dermal Exposures to Monomeric and Polymeric 1,6-Hexamethylene Diisocyanate: A Pilot Study

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The purpose of this study was to develop and evaluate a patch sampler to monitor dermal exposures to monomeric and polymeric 1,6-hexamethylene diisocyanate (HDI) in the automotive refinishing industry. Different patch materials were used to construct the patches, and patches impregnated with a derivatizing solution were compared with those that were not impregnated. We observed that impregnated felt patches measured significantly more HDI monomer ($p = 0.04$) than non-impregnated patches in a controlled experiment. Both impregnated and non-impregnated patches were compared with the tape-strip method by monitoring three spray painters' dermal exposure to monomeric and polymeric HDI. Isocyanurate was the predominant species measured by all three sampler types with detectable levels in >86% of samples. Overall, tape-strips of exposed skin measured lower levels of monomeric and polymeric HDI than impregnated patch samplers at the same sampling site on the skin. Unlike tape-strips, impregnated patches are not as prone to evaporative or reactive losses or losses due to rapid penetration into the skin. Further investigations are warranted to evaluate these and other methods to measure dermal exposure to workers under occupational conditions to better understand the relationship between dermal exposure and internal dose.

Keywords 1,6-hexamethylene diisocyanate (HDI), dermal exposure, dermal sampling, polyisocyanate, sampling method

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INTRODUCTION

Respiratory exposure to diisocyanates has long been considered the primary route of exposure, and thus, research, regulation, and prevention have focused almost exclusively on airborne isocyanate exposures.[1] Airborne isocyanate exposures have been reduced through improved controls and use of less volatile isocyanates; however, isocyanate asthma continues to be prevalent in workplaces where measured isocyanate inhalation exposures are very low or non-detectable but where there is a clear opportunity for dermal exposure.[2] Animal studies have linked respiratory sensitization with prior dermal exposure to isocyanates.[3,4] Respiratory sensitization was induced after epicutaneous exposure to 1,6-hexamethylene diisocyanate (HDI) in mice[5] and to toluene diisocyanate (TDI) in guinea pigs[6] and after intradermal and topical, but not inhalation, exposure to diphenylmethane diisocyanate (MDI) in guinea pigs.[4]

In spray painting operations, monomeric and polymeric HDI can be present in both aerosol and vapor forms. Most exposure assessments have only focused on the characterization of airborne exposures.[7–12] However, aerosol deposition on the skin and skin contact with contaminated surfaces and liquid product also constitute an important contact site for exposure. Isocyanates are commonly mixed with various solvents, polyols, and other substances such as catalysts and blowing agents, which may affect isocyanate reactivity, skin absorption, and health effects.[2] Urine and blood biomarkers of isocyanate exposure can potentially be used to assess internal dose but not to distinguish whether exposure is due to the dermal or respiratory route.[2] The higher the volatility of the isocyanate, the shorter its residence time is on the skin. Therefore, the less volatile polymeric isocyanates (HDI biuret and isocyanurate) may potentially have longer residence time on the skin and, thus, may have skin and systemic effects different from that of the monomer.

Fent et al.[13] postulated that differences between dermal exposures for polymeric HDI are likely due to different rates of skin absorption or chemical reactivity. Exposure of the skin to isocyanates could contribute to a significant part of the total body burden. For example, Bello et al.[2] estimated that 1% skin absorption of a small MDI droplet (10 mg) would result in a dose approximately 4.5-fold (450%) higher than a 15-min inhalation exposure to a concentration at the United Kingdom

Health and Safety Executive short-term exposure limit (70 μg NCO/m^3), assuming 100% lung retention and a ventilation rate of 7 L/min.

Methods for monitoring dermal exposures are less advanced than those for air sampling techniques. However, several groups have measured exposure of the skin to isocyanates using qualitative SWYPE colorimetric indicators (CLI, Des Plains, Ill.),[14,15] quantitative wipes,[16] and quantitative tape-strips.[13,17,18] These methods may underestimate exposures due to losses from absorption, chemical reactions, or poor removal efficiency.[2,19]

The objective of this study was to develop a sampling patch to quantify exposure to monomeric and polymeric HDI deposited on the skin in the spray painting environment and to compare the method with the dermal tape-strip method as described by Fent et al.[13,17] The tape-strip method has the ability to quantify monomeric and polymeric HDI that has penetrated into the stratum corneum and that covers the skin's surface. Comparison of the tape-strip and the patch methods allowed us to investigate the potential limitations of these sampling techniques and to improve our understanding of the relationships among dermal exposure, penetration, and the contribution of this route of exposure to the total body burden. Our ability to measure dermal isocyanate exposure accurately is critical for exposure and risk assessment in order to predict systemic exposure, develop sensitive and predictive models through multiple exposure routes, and ultimately protect the health of workers.

METHODS

Laboratory Studies

Three materials (polyester felt, wool, and 37-mm glass fiber filter) were tested to determine their suitability for use as a patch sampler. Different materials may have different absorption, retention, and repellency characteristics that can affect exposure assessment.[20] There are no standard guidelines for the type of material that patches should be made of, the only criterion being that they should be absorbent.[20] These materials were chosen based on their availability, they easily fit into the patch design, and their potential ability to absorb chemical substances. We evaluated reactivity of these materials with a derivatizing solution and for the recovery of HDI monomer.

Reactivity of Materials with Derivatizing Solution

A piece of either felt or wool (5 cm^2) was placed in a 20-mL glass jar (I-Chem, New Castle, Del.) with 5 mL of derivatizing solution, which was made by dissolving 2 g of 1-(2-methoxyphenyl)-piperazine (MPP; 192.3 g/mol) in 1:1 of 30% v/v solution of *N*,*N*-dimethylformamide (DMF; 73.09 g/mol) in acetonitrile (ACN; 41.05 g/mol). The materials were left at room temperature for 24 hr. Materials were visually assessed periodically throughout the 24 hr for signs of breakdown, discoloration, and disintegration.

Recovery of HDI Monomer with Felt, Wool, and GFF

Pieces of wool (N = 6) and felt (N = 6) (2.5 cm $\times$ 4 cm) along with 37-mm glass fiber filters (GFF, Type AE; SKC, Eighty Four, Pa.) (N = 3) were placed separately into 20-mL glass jars (I-Chem). Each material type was spiked with 40 μL of a mixture of HDI in toluene (1550 pmol/μL HDI in toluene [TOL; 92.14 g/mol]). After spiking, the lids of the jar were affixed and samples held at room temperature for 15 min. Reference samples were prepared by spiking the same amount of HDI/TOL onto the glass in an empty glass jar.

Following the 15-min period, 10 mL of derivatizing solution (2 g/L MPP in 30% DMF-ACN solution) was added to each glass jar. Acetic anhydride (200 μL) was added to acetylate residual MPP. After 15 min, the internal standard (2 pmol/μL urea derivative of 1,8-octamethylene diisocyanate; ODIU) was combined (1:1 v/v ratio) with aliquots of each sample to give an internal standard concentration of 1 pmol/μL. All samples were analyzed for HDI monomer using liquid chromatography-mass spectrometry (LC-MS) as described elsewhere.[13] Wool, felt, and GFF samples were compared with reference samples to determine the percentage recovery from each material type.

Recovery of HDI Monomer from Impregnated Felt

Pieces of felt (2.5 cm $\times$ 4 cm) were impregnated with a derivatizing solution (1.09 g/mL MPP in TOL) designed to collect and derivatize isocyanate vapors. The felt pieces were impregnated with 1200 μL of 6 g/L MPP in TOL and allowed to dry for 20 min. Each impregnated felt was placed into a 20-mL glass jar and spiked with 40 μL of HDI/TOL (1550 pmol/μL). After spiking, the lids of the jars were affixed and jars held at room temperature for 15 min. Reference samples were collected by spiking the same amount of HDI/TOL onto the glass in an empty glass jar. Following the 15-min period, 10 mL of derivatizing solution (2 g/L MPP in 30% DMF-ACN solution) was added to each glass jar. Sample processing and analysis were performed as described above. All samples were analyzed for HDI monomer using LC-MS as described elsewhere.[13] Felt patch samples were compared with reference samples to determine percentage recovery.

Field Studies

Comparison of Impregnated and Non-Impregnated Patches

Clearcoat Spiking. Felt patches (2.5 cm $\times$ 4 cm) were impregnated as described above. Both impregnated and non-impregnated felt patches were placed separately in 20-mL glass jars. A mixed clearcoat (15 μL; BASF, Münster, Germany), used by an automotive spray painter, was spiked on each patch using a 20-μL pipette. After spiking, the open jars with samples were allowed to sit at room temperature for 15 min. Leaving the jars open mimicked more closely the conditions of field sampling and allowed us to investigate evaporation. The pipette tips were ejected into a separate glass vial containing 15 mL of derivatizing solution (2 g/L MPP in 30% DMF/ACN) to account for total mass.

Following the 15-min period, glass jars were filled with 15 mL of derivatizing solution (2 g/L MPP in 30% DMF/ACN). A reference sample of a mixed clearcoat (15 μL) was drawn into a 20-μL pipette and dispensed into a glass vial (I-Chem) filled with 15 mL of derivatizing solution (2 g/L MPP in 30% DMF/ACN). The pipette tip was also ejected into the solution to eliminate sidewall losses due to the viscosity of the clearcoat. All samples were then placed into a cooler ($\sim$4°C) and returned to UNC laboratory for storage at –40°C until analyzed.

Sample processing and analysis were performed as described above. A total of nine impregnated and nine non-impregnated felt patches and respective pipette tips were analyzed for HDI monomer, biuret, and isocyanurate using LC-MS as described elsewhere.[13] The percentage recovery for each patch sample was calculated by adding the sum of mass measured for each patch sample and respective pipette tip. This mass was then compared with a specific reference sample to calculate the percentage recovery.

Side-by-Side Spray Painting. Felt patches (2.5 cm × 4 cm) were impregnated with 2000 μL of impregnating solution (6 g/L MPP in TOL) as described above. Both impregnated (N = 4) and non-impregnated felt patches (N = 4) were lined up alternating on a cardboard backing. A spray painter sprayed the patches with a Deltron (Strongsville, Ohio) clearcoat mixture. After spraying, the patches were allowed to remain in the spray booth for 12 min, which is the approximate time it takes to apply one coat of clearcoat to an automobile. After this time, patches were placed into glass jars containing 10 mL of derivatizing solution (2 g/L MPP in 30% DMF/ACN) and capped. All samples were then placed into a cooler ($\sim$4°C) and returned to UNC laboratory for storage at –40°C until analyzed. Sample processing and analysis were performed as described above. Samples were analyzed for HDI monomer, biuret, and isocyanurate using LC-MS as described elsewhere[13] and the results of the impregnated and non-impregnated felt patches compared.

Comparison of Patches and Tape-Strips During Application of Clearcoat

Patch sampling was performed on three workers during 11 spray painting tasks in central North Carolina. These painters were enrolled in our exposure assessment study described elsewhere.[21] Patches (size 5.5 cm × 3.5 cm with the sample collection surface of 2.5 cm × 4 cm) were constructed with felt, aluminum foil, and Cover-Roll adhesive tape (Beiersdorf AG, Hamburg, Germany). The felt was impregnated with 2000 μL of impregnating solution (6 g/L MPP in TOL) as described above. The impregnated felt was backed with two layers of aluminum foil, and the foil was folded around the edges to prevent any potential run-off of the gel-like MPP and contact with the skin. The adhesive tape was attached to the back of the aluminum foil for easy placement of the patch to the skin of the worker.

Sample Collection. Both impregnated and non-impregnated patches were placed on the right and left volar forearm of workers during 11 spray painting tasks (22 samples sets). As depicted in Figure 1, a space in between the patches was left for the collection of tape-strip samples after the paint task. The locations of the impregnated and non-impregnated patches on the worker's arm were randomized. Painters in this study did not wear coveralls (as part of their normal work practice) and hence their arms were exposed to spray paint mist.

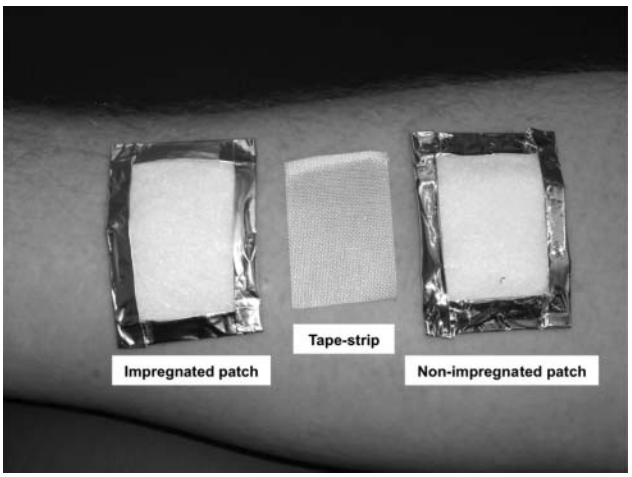

FIGURE 1. Right volar forearm of a worker depicting the location of the patches during spraying and the tape-strip after spraying. The exposed patch surfaces and the tape-strips were each 2.5 cm × 4 cm.

The painters in this study used BASF, Dupont Nason (Wilmington, Del.), and Deltron products. After each paint task, patches were immediately placed in 10 mL of derivatizing solution (2 g/L MPP in 30% DMF/ACN). In addition, five successive tape-strips (Cover-Roll, 2.5 cm × 4 cm) were collected adjacent to each patch sampler site and tapes placed in 5 mL of derivatizing solution (2 g/L MPP in 30% DMF/ACN) as described elsewhere.[13] Each tape-strip represents approximately one cell layer; thus, we removed approximately five cell layers. A bulk sample of mixed basecoat or clearcoat (10 μL) was drawn into a 20-μL pipette and dispensed into a glass vial (I-Chem) filled with 15 mL of derivatizing solution (2 g/L MPP in 30% DMF/ACN) to confirm the presence of isocyanates. The pipette tip was also ejected into the solution to eliminate sidewall losses due to the viscosity of the clearcoat. All samples were then placed into a cooler ($\sim$4°C) and returned to UNC laboratory for storage at –40°C until analyzed.

Sample Analysis. For analysis, samples were returned to room temperature, and acetic anhydride was added (200 μL for patch and bulk; 100 μL for tape-strips) to acetylate residual MPP. After 15 min, an internal standard (2 pmol/μL ODIU) was combined (1:1 v/v ratio) with aliquots of each bulk paint and patch sample to give an internal standard concentration of 1 pmol/μL. For tape-strips, after 15 min internal standard (52 pmol/μL ODIU) was added (100 μL) to give an internal standard concentration of 1 pmol/μL. Samples were analyzed for HDI monomer, biuret, and isocyanurate using LC-MS as

described elsewhere.[13] Comparisons were made between impregnated patch, non-impregnated patch, and tape-strip results.

RESULTS

Laboratory Studies

Based on visual inspection for reactivity of the three patch materials (felt, wool, and GFF) with derivatizing solution, we did not observe the material types to be reactive with the derivatization solution. The average percentage recovery (sample/reference × 100%) for each patch type compared with the reference samples for HDI monomer for all material types was >94%. The average percentage recoveries ± standard deviations for the felt, wool, and GFF were 104 ± 5, 99 ± 4, and 94 ± 8%, respectively. We also observed that HDI did not react with this material type (recoveries ∼100%), and therefore, the recovery of HDI monomer with impregnated felt was determined. The average percentage recovery and standard deviation for HDI monomer with the impregnated felt was 105 ± 9%. The laboratory results provided evidence of sufficient recovery with the impregnated felt. Therefore, we proceeded to test the felt patch design in an occupational field setting.

Field Studies

Comparison of Impregnated and Non-Impregnated Felt Patches

The average percentage recovery and standard deviation for each analyte when either felt patch type was spiked with the clearcoat are presented in Table I. Although not significant (two-sample means *t*-test, α-level of 0.05), the impregnated patches measured more monomeric and polymeric HDI compared with non-impregnated patches. We also observed that monomeric and polymeric HDI did not react with the felt as the results showed recoveries ∼100%.

Table II provides the results from the side-by-side spray-painting experiment for each analyte reported as ratios of impregnated patches to non-impregnated patches. Paired *t*-test (α-level of 0.05) indicated that the impregnated patches measured significantly more HDI monomer ($p = 0.04$) than non-impregnated patches. Although not significant, the impregnated patches generally measured more HDI biuret and isocyanurate than non-impregnated patches.

TABLE I. Recovery of HDI Monomer, Biuret, and Isocyanurate from Impregnated and Non-Impregnated Felt Patches (N = 9) After Application of 15 μL of Clearcoat

	Average Percentage Recovery[A] ± Standard Deviation		
Analyte	Impregnated Patch	Non-Impregnated Patch	*p*-value
HDI monomer	117 ± 12	108 ± 15	0.18
Biuret	90 ± 13	82 ± 16	0.26
Isocyanurate	92 ± 14	82 ± 17	0.67

Note: Following application of clearcoat, the patches were allowed to sit for 15 min at room temperature before adding derivatizing solution.
[A]Sample amount/reference amount × 100.

Comparison of Felt Patches and Tape-Strips During Application of Clearcoat

Results of the felt patch and tape-strip sampling performed on the three workers during 11 spray painting tasks are presented in Table III. The percentage recovery of tape-strips compared with the patches was calculated for each of the 22 sample sets collected by summing the mass of analyte measured by the five consecutive tape-strips and comparing it with the masses measured in the patch samplers. Results of the five successive tape-strips show a decreasing trend in analyte mass, thus indicating penetration into the stratum corneum (data not shown). Bulk paint sample analysis confirmed the presence of HDI (187 ± 172 mg/L), biuret (3331 ± 7274 mg/L), and isocyanurate (48,482 ± 45,250 mg/L) in all spray painting tasks.

Overall, the impregnated patches measured more monomeric and polymeric HDI than non-impregnated patches or the tape-strip samples. Impregnated patches detected HDI monomer in 63% of the samples, while non-impregnated patches and tape-strips measured detectable levels of monomer in 9% and 36% of the samples, respectively. At most, the tape-strips recovered 35% of HDI monomer measured by impregnated patches. Biuret was detectable in 18% of impregnated and non-impregnated patch samples and 14% of tape-strip samples. The amount of biuret measured by the tape-strip samples varied from 11 to 60% of the amount measured by the impregnated patches.

Isocyanurate was the predominant species measured with all samplers. Tape-strip samples detected isocyanurate in 100% of the samples, while the impregnated and non-impregnated patches measured detectable levels of isocyanurate in 91% and 86% of the samples, respectively. Due to the much greater detection of isocyanurate than all other species, paired *t*-test (α-level of 0.05), in which samples below the limit of detection (LOD) or limit of quantitation (LOQ) were assigned values by dividing the respective limits by $\sqrt{2}$,[22] was performed. Values were natural log-transformed to satisfy the normality assumption. Shapiro-Wilks tests for normality indicated that isocyanurate levels measured with impregnated patches ($W = 0.93$), non-impregnated patches ($W = 0.93$), and tape-strips ($W = 0.81$) were approximately log-normally distributed. Geometric mean (GM) levels for each sampler type were calculated. Paired *t*-test indicated that significantly greater amounts of isocyanurate were measured using impregnated patches (GM = 1.4 μg; $p < 0.01$) and non-impregnated patches (GM = 1.0 μg; $p = 0.01$) than tape-strips (GM = 0.43 μg). However, for two tasks (Worker 3, Tasks 2 and 3), the tape-strips measured greater levels of isocyanurate than the impregnated patches. Although only borderline significant

TABLE II. Comparison of Measured Isocyanates from Clearcoat Sprayed onto Impregnated and Non-Impregnated Felt Patches

Analyte	Sample Number	Impregnated Patch (μg)	Non-Impregnated Patch (μg)	Ratio Impregnated/Non-Impregnated	*p*-value
HDI monomer	1	0.86	0.48	1.8	0.04
	2	0.58	0.41	1.4	
	3	1.35	0.65	2.1	
	4	1.55	0.78	2.0	
Biuret	1	2.23	2.19	1.0	0.23
	2	1.92	2.24	0.9	
	3	5.36	3.34	1.6	
	4	6.64	3.99	1.7	
Isocyanurate	1	239.65	221.69	1.1	0.29
	2	177.87	214.07	0.8	
	3	327.69	248.48	1.3	
	4	359.94	282.37	1.3	

($p = 0.07$), the impregnated patches collected more isocyanurate than non-impregnated patches.

DISCUSSION

In this study, we developed and evaluated a patch sampler to quantitatively measure dermal exposure to monomeric and polymeric HDI in the spray painting environment. Impregnated felt patches collected more than non-impregnated felt patches and tape-strips for the majority of the analytes and experiments. When clearcoat was directly sprayed onto the samplers, impregnated felt patches collected significantly more HDI monomer than non-impregnated patches. The comparison of samplers worn by painters during spray painting provided additional evidence that the impregnated patches may be more efficient at measuring isocyanates than the tape-strips or non-impregnated patches.

When worn by painters, impregnated patches detected more HDI monomer (63%) than the non-impregnated patches (9%) and tape-strips (36%). For HDI monomer, it is no surprise that the impregnated patch performed better than the non-impregnated patch and tape-strips because HDI monomer is more volatile than the polymeric forms and, thus, may evaporate more rapidly. The impregnated patches should quickly derivatize isocyanates into more stable molecules thereby minimizing evaporative or reactive losses. Tape-strip samples, on the other hand, can exhibit losses due to evaporation, polymerization, reactivity, and penetration into the skin (beyond the layers of the stratum corneum sampled). Because the felt used to construct the patches did not react with HDI monomer, the non-impregnated patches should exhibit only losses due to evaporation/polymerization. Therefore, the differences between the non-impregnated patch and the tape-strip are likely due to chemical reactions that are occurring on the skin's surface or due to penetration into the skin.

Our results indicate uptake and penetration into the stratum corneum as we measured HDI in successive tape-strip samples as also indicated in our previous publication.[17] Bello et al.[23] observed that octyl isocyanate, a volatile low-molecular weight monoisocyanate used as a surrogate for low-molecular weight diisocyanates such as HDI, disappeared from the skin surface relatively fast (~70% of the applied dose in 10 min). Their data indicated that the losses due to evaporation were minimal, thus suggesting rapid penetration of this low molecular weight isocyanate.

When the patches were sprayed directly with clearcoat, we observed less than a 2-fold difference between the impregnated and non-impregnated patches for the HDI polymers. Greater differences were observed in a few instances when the patches were worn by the painters. For Worker 1, Task 1, the impregnated patches measured 2.3 and 9.4 times more biuret and 2.6 and 4.2 times more isocyanurate than the non-impregnated patches. However, for the other workers and tasks, the levels of biuret and isocyanurate were similar for impregnated and non-impregnated patches. It is possible that the results observed for Worker 1 may be due to unusual spatial variability in clearcoat overspray, accidental touching of the impregnated sampler with contaminated hands, or some other sampling error/malfunction.

Biuret was detected in 18% of the patch samples and 14% of the tape-strip samples. The impregnated-patch samplers measured 40–89% more biuret than the tape-strips. The tape-strips measured levels of biuret similar to the non-impregnated patch samplers (77–100%). In a previous study, we found evidence of high reactivity and sampling challenges for biuret.[24] Biuret is likely more difficult to measure than HDI monomer and isocyanurate. We measured biuret the least, despite the confirmation of its presence in the bulk paint samples. Similarly, we previously measured detectable levels of biuret in 83% of the bulk paint samples but in only 9% of the tape-strips collected from the lower arms of painters who

TABLE III. Summary of Felt Patch and Tape-Strip Measurements Obtained from Adjacent Sample Areas from Three Workers During Different Spray Painting Tasks

			HDI (μg)			Biuret (μg)			Isocyanurate (μg)		
Worker	Task	Arm Location	Impregnated Patch[A]	Non-Impregnated Patch[A]	Tape[B]	Impregnated Patch[A]	Non-Impregnated Patch[A]	Tape[B]	Impregnated Patch[A]	Non-Impregnated Patch[A]	Tape[B]
1	1	Left	4.72 (35%)	1.75 (95%)	1.66	60.6 (34%)	26.95 (77%)	20.8	972 (34%)	369 (91%)	334.1
		Right	0.22	0.03	<LOQ	2.35 (11%)	0.25 (100%)	0.25	40.5 (6%)	9.7 (24%)	2.32
	2	Left	<LOQ	ND	ND	<LOQ	ND	ND	2.71 (39%)	2.32 (46%)	1.06
		Right	<LOQ	ND	ND	ND	<LOQ	ND	4.71 (17%)	3.65 (22%)	0.79
2	1	Left	ND	ND	ND	ND	ND	ND	2.73 (16%)	1.31 (34%)	0.44
		Right	ND	ND	<LOQ	ND	ND	ND	0.76 (54%)	0.89 (46%)	0.41
	2	Left	0.09	ND	<LOQ	ND	ND	ND	1.74 (27%)	2.32 (20%)	0.47
		Right	0.12 (8%)	ND	0.01	ND	ND	ND	4.71 (14%)	3.65 (18%)	0.65
	3	Left	0.03	ND	ND	ND	ND	ND	8.53 (12%)	7.37 (14%)	1.06
		Right	0.02	ND	ND	ND	ND	ND	8.81 (18%)	7.15 (23%)	1.62
3	1	Left	ND	ND	ND	ND	ND	ND	ND	ND	<LOQ
		Right	ND	ND	<LOQ	ND	ND	ND	ND	ND	0.06
	2	Left	ND	ND	ND	ND	ND	ND	0.62 (16%)	0.38 (26%)	0.1
		Right	ND	ND	ND	ND	ND	ND	0.30 (274%)	nd	0.82
	3	Left	ND	ND	ND	ND	ND	ND	0.74 (46%)	0.60 (56%)	0.34
		Right	ND	ND	ND	ND	ND	ND	0.44 (205%)	0.32 (286%)	0.9
	4	Left	0.32	ND	ND	ND	ND	ND	0.54	<LOQ	<LOQ
		Right	0.12	ND	<LOQ	ND	ND	ND	<LOQ	0.35	<LOQ
	5	Left	0.33	ND	<LOQ	ND	ND	ND	1.07 (27%)	0.7 (41%)	0.29
		Right	0.2	ND	ND	ND	ND	ND	1.23 (59%)	0.89 (82%)	0.73
	6	Left	<LOQ	ND	ND	ND	ND	ND	1.40	1.40	<LOQ
		Right	0.02	ND	ND	0.25 (60%)	0.19 (79%)	0.15	1.50 (11%)	1.80 (9%)	0.16
LOD			0.003	0.003	0.002	0.04	0.04	0.02	0.04	0.04	0.02
LOQ			0.008	0.008	0.004	0.1	0.1	0.05	0.1	0.1	0.05

[A]Percentage measured by five consecutive tape-strips compared with the respective patch sampler provided in parentheses.
[B]Summation of five consecutive tape-strips.

did not wear protective clothing in a larger study of automotive spray painters.[18]

A borderline significant difference ($p = 0.07$) was observed between the impregnated and non-impregnated patches and, thus, may suggest some polymerization. Evaporation is possible, but unlikely, given the low vapor pressure (5.3×10^{-9} mmHg) of isocyanurate. Compared with the patches, on average, the tape-strips recovered half the amount of isocyanurate ($p < 0.01$). These results indicate that isocyanurate is either reacting with the skin or rapidly penetrating into the deeper layers of the skin, thus leaving less of the compound for sampling with the tape-strips.

Bello et al.[23] investigated dermal penetration of polymeric HDI, polymeric isophorone diisocyanate (IPDI), and MDI and found that chemical reaction was minimal, thus suggesting permeation into the deeper layers of the skin. Our results confirm penetration into the stratum corneum, as we measured isocyanurate in the successive tape-strip samples. Another possibility is that the tape-strip is not efficient at removing isocyanurate from the skin's surface. However, this seems unlikely because we have previously observed that the tape-strip technique removes >95% of a high molecular weight compound.[25] In addition, if the tape-strips had poor collection efficiency, we would expect to see similar results with biuret; yet, 77–100% of biuret measured with the non-impregnated patches was also measured with tape-strips.

Soutar et al.[20] provided a thorough discussion regarding the advantages and disadvantages of patch and whole-body sampling. A limitation of the tape-strip and patch sampling techniques is that they only provide an estimate on the amount of a substance deposited on a particular area (the sample location). These methods assume that contamination is uniformly distributed over the sample area. However, the sample area represent only a small portion of a particular region, and thus, regional dermal estimates could lead to either overestimation or underestimation of exposure. During spraying, if the droplets miss the sampler, exposure could be underestimated, and conversely, if a splash lands on the sampler, exposure could be overestimated. The main limitation of the patch is that it represents only the amount of isocyanate deposited on the skin. The tape-strip technique, however, is capable of measuring uptake of isocyantes in the skin through successive tape-strips.

We previously used tape-strip sampling to measure dermal exposure to HDI-based polyisocyanates in 47 automotive spray painters performing a total of 296 paint tasks.[18] The results of tape-strip sampling performed on the lower arms of painters who did not wear protective clothing was directly comparable with the tape-strip sampling results presented here. The distribution of isocyanurate we collected with tape-strips (GM = 0.43 μg, GSD = 7.3, N = 22) was comparable to the distribution measured in our previous study[18] (GM = 1.5 μg, GSD = 8.1, N = 332). Although we cannot reliably calculate the distributions for HDI monomer and biuret due to the large number of non-detects and a small sample size, our detection rates (36, 14, and 100% for HDI monomer, biuret, and isocyanurate, respectively) were similar to the respective detection rates (44, 9, 96%, respectively) in our previous study.[18] Hence, our measurements may be representative of the automotive refinishing industry.

Previously, we compared our dermal tape-strip measurements obtained from spray painters who did not wear protective clothing and who wore coveralls and gloves[18] with the wipe sampling results reported by Bello et al.[16] who quantified dermal exposure (ng/cm^2) to TRIG in spray painters who did not wear protective clothing and who wore coveralls and gloves. Individual polyisocyanates measured with tape-strips[18] were converted into estimates of TRIG, and the results indicated that the tape-strips measured higher levels of polyisocyanates in the skin of painters who did not wear protective clothing and in the skin of painters who wore coveralls and gloves compared with the levels measured with wipe sampling. In our study, the impregnated patch samplers measured higher levels of polyisocyanates than the tape-strips. Therefore, we would expect the impregnated patch samplers to measure higher levels of polyisocyanates than the wipe samples used by Bello et al.[16] The specificity of our patch and tape-strip methods allows us to investigate individual monomeric and polymeric HDI concentrations on and in the skin.

Patch samplers are advantageous to use when investigating the effectiveness of protective clothing. Patches placed beneath protective clothing can indicate any permeation of isocyanates through the material. The tape-strip may not be as an effective indicator of permeation because of potential losses due to rapid penetration into the skin and due to evaporation. However, an advantage of the tape-strip method is that it is able to quantitatively measure polyisocyanate species in the non-viable skin layer, thus providing an estimate of the absorbed dose. To fully understand the relationships between dermal exposure, uptake, and internal dose, a study in which biological and dermal surface sampling of monomeric and polymeric HDI that excluded inhalation exposures would be necessary. This type of study would allow us to assess how to interpret dermal loading in relation to received internal dose.

Although the tape-strip is capable of measuring monomeric and polymeric HDI on the skin surface and that has penetrated into the skin, it appears that penetration into deeper layers of the skin may be occurring. Knowledge obtained on the penetration and absorption of monomeric and polymeric HDI into human skin is required to further our understanding on the effect of dermal exposure to internal dose received. Studies investigating urinary biomarkers of isocyanate exposure have provided indirect evidence of dermal uptake. The HDI hydrolysis product 1,6-hexamethylene diamine (HDA) has been measured in both blood plasma[26] and urine[27] of workers using respiratory protection, and elevated levels of urinary biomarkers have been detected in workers where isocyanate inhalation exposures were very low or non-detectable.[28,29]

Until we can establish dermal uptake, penetration patterns, and the fate of isocyanates in the skin, it seems prudent to measure dermal exposures with both impregnated patches

and tape-strips. The tape-strip samples seem to underestimate dermal exposure due to the rapid penetration, while non-impregnated patches suffer from losses due to evaporation/polymerization. Our ability to measure dermal isocyanate exposure accurately is critical in understanding the contribution of dermal exposure to the internal dose received and, thus, to the potential related health effects.

We acknowledge that this study measured exposure only to three workers. Future studies involving a larger population are required to further evaluate these and other methods (e.g., wipe sampling method) to measure dermal exposure as well as to assess dermal penetration of isocyanates. In addition, modification of the patch sampler design is necessary. The medical tape attached to the aluminum foil backing of the patch did not provide a sufficient hold on the workers' arms and resulted in the movement or loss of patches. In future studies, we will refine the design so that we can use Velcro and stick the patches to arm bands worn by the worker.

CONCLUSIONS

We developed and evaluated a dermal patch designed to measure monomeric and polymeric HDI during spray painting in the automotive refinishing industry. Although this study is limited in size, we demonstrate the potential use for the dermal patch sampling in the occupational setting. Overall, we measured greater levels of monomeric and polymeric HDI with impregnated patches compared with tape-strips. Further investigation comparing tape-strips with patch sampling as well as studies designed to further our understanding on dermal penetration and absorption patterns of monomeric and polymeric HDI in human skin are warranted.

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