Method number:	W4002
Target concentration:	340 µg/100 cm^2
Procedure:	Samples are collected by using firm hand pressure to move the 4.75 in. × 4.75 in. Ghost Wipe across the surface of interest. Before sampling, the media are moistened with 0.5-mL 50:50 isopropanol:water. After sampling, immediately place the media in vials containing 5 mL of a derivatizing reagent solution. The samples are prepared, then analyzed by high performance liquid chromatography(HPLC) using fluorescence (FL) and ultraviolet (UV) detectors.
Reliable quantitation limit:	0.962 µg
Special Requirements:	The time interval, from beginning to collect the sample, until the sample is placed in the vial containing the derivatizing reagent, should not exceed three minutes.
Status of method:	Evaluated method. This method has been subjected to the established evaluation procedures of the Methods Development Team.

March 2002

Chemists: Richard Lawrence, Steven Thygerson

Applied Industrial Hygiene Chemistry Team
Program Support Division
OSHA Salt Lake Technical Center
Salt Lake City UT 84115-1802

1. General Discussion

1.1 Background

1.1.1 History

This wipe sampling method was developed to provide a uniform and practical means of taking wipe samples for 1,6-hexamethylene diisocyanate (HDI). The sampling of HDI on surfaces was performed using Ghost Wipes (Environmental Express). The Ghost Wipe was chosen for its durability and its use in surface sampling of other analytes. The samples are analyzed following the procedure in OSHA Method 42[1], using high performance liquid chromatography (HPLC). Glass plates were used as the ideal surface to check the surface sampler removal efficiency. The use of a surface that approximates an ideal surface (extremely smooth and non-porous) minimizes the effect of the surface on the evaluation of the media. No interactions between the glass plate and HDI were found. In this evaluation, the media are pre-wetted with a solution of 50:50 isopropanol:water. After sampling, the samples were immediately placed in vials containing 5 mL of a derivatizing reagent solution of 50:50 dimethyl sulfoxide (DMSO):ethyl acetate and 0.025 M 1-(2-pyridyl) piperazine (1-2PP). The derivatizing reagent (1-2PP) reacts with HDI by attaching a chromaphore to the molecule. This improves the analytical sensitivity of the method. Also, without the derivatizing reagent, the HDI will begin to hydrolyze or polymerize and the HDI on the sample will be lost. A derivatizing reagent solution of 0.025 M 1-2PP in acetonitrile was initially used. However, other isocyanates that were to be evaluated did not go into solution in this reagent. To make this method as uniform and practical as possible for other isocyanate sampling methods, the derivatizing reagent with DMSO and ethyl acetate as solvent was used. A study was conducted to investigate the reaction of HDI with the alcohol and the water in the wetting solution prior to being derivatized. It was determined that the time interval, from beginning collection of the sample, until the sample is placed in contact with the derivatizing reagent should not exceed three minutes. (Section 4.9)

1.1.2 Toxic effects (This section is for information only and should not be taken as the basis of OSHA policy.)

Many dermal exposure studies on animals describe the ability of HDI to cause contact sensitization or direct irritation to the skin of laboratory animals.[2] Respiratory hypersensitivity has been induced by exposure of diisocyanates to the skin of laboratory animals.[3, 4, 5, 6] Human skin exposure to diisocyanates has resulted in erythema, eczematous dermatitis, contact eczema characterized by follicular papules and dermal

---

[1] Diisocyanates, 1,6-Hexamethylene Diisocyanate (HDI), Toluene-2,6-Diisocyanate, Toluene-2,4-Diisocyanate, Method 42, 1989; http://www.osha-slc.gov/dts/sltc/methods/organic/org042/org042.html, (accessed May 2000).

[2] *Toxicological Profile for Hexamethylene Diisocyanate*, U.S. Department of Health and Human Services, Public Health Service, Agency for Toxic Substances and Disease Registry, Atlanta, GA, 1998, pp 59-67.

[3] Rattray, N., et al. Induction of Respiratory Hypersensitivity to Diphenylmethane-4,4' diisocyanate (MDI) in Guinea Pigs. Influence of Route of Exposure. *Toxicology* **1994,** *88*, pp 15-30.

[4] Karol, M. H., et al. Dermal Contact with Toluene Diisocyanate (TDI) Produces Respiratory Tract Hypersensitivity in Guinea Pigs, *Toxicology and Applied Pharmacology* **1981,** *58*, pp 221-230.

[5] Kimber, I. The Role of the Skin in the Development of Chemical Respiratory Hypersensitivity, *Toxicology Letters* **1996,** *86,* pp 89-92.

[6] Bickis, U. Investigation of Dermally Induced Airway Hyperreactivity to Toluene Diisocyanate in Guinea Pigs. Ph.D. thesis, Queen's University, Kingston, Canada, 1994.

sensitization.[7] A procedures for conducting urinalysis for 1,6-hexamethylene diamine as an indicator of the biological metabolite of HDI has been published.[8]

1.1.3 Workplace exposure

Occupations with the greatest potential for exposure to HDI are painters and paint spraying machine operators, aircraft engine mechanics, and aircraft machinists. Other occupations with potential for exposure to HDI include construction laborers, chemical technicians, mixing and blending machine operators in the chemical industry, plumbers, pipe fitters, steam fitters, metal plating machine operators, miscellaneous machine operators in the aircraft equipment industry, and production workers and supervisors in the fabricated structural metal industry.[9]

1.1.4 Physical properties and descriptive information[10]

CAS number:	822-06-0	vapor pressure (mmHg):	0.05
IMIS number:	1377[11]	molecular weight:	168.20
flash point (OC):	135°C	boiling point:	255°C
melting point:	-67°C	odor:	pungent
appearance:	Pale yellow liquid	synonyms:	1,6-diisocyanatohexane, HDI
molecular formula:	OCN-$(CH_2)_6$-NCO	solubility:	poorly soluble in water, reacts slowly with water
specific gravity:	1.04 g/mL		

$LD_{50,}$ rabbit, dermal: 570 µg/kg [12]

SD_{50} (sensitization dose) mouse, dermal: 0.088 mg/kg

LOAEL (Lowest observable adverse effects level), guinea pig, dermal: 0.1 mg

This method was evaluated according to the OSHA SLTC "Evaluation Guidelines for Surface Sampling Methods".[13] The Guidelines define analytical parameters, specify required laboratory tests, statistical calculations and acceptance criteria. The analyte surface concentrations throughout this method are based on the evaluated sampling area and analytical concentration parameters.

[7] *Criteria for a Recommended Standard... Occupational Exposure to Diisocyanates*; DHEW (NIOSH) Publ. No. 78- 215; U.S. Department of Health, Education and Welfare, Public Health Service, Center for Disease Control, National Institute for Occupational Safety and Health, U.S. Government Printing Office: Washington DC, 1978, pp 35-36.

[8] Maitre A., et. al. Urinary Hexane Diamine as an Indicator of Occupational Exposure to Hexamethylene Diisocyanate, *International Archives of Occupational and Environmental Health* **1996,** *69,* pp 65-68.

[9] *National Occupational Exposure Survey*; U.S. Department of Health, Education and Welfare, Public Health Service, Center for Disease Control, National Institute for Occupational Safety and Health, Cincinnati, OH, 1989.

[10] *Toxicological Profile for Hexamethylene Diisocyanate*, U.S. Department of Health and Human Services, Public Health Service, Agency for Toxic Substances and Disease Registry, Atlanta, GA, 1998, pp 59-67, 100-102.

[11] Chemical Sampling Information, Hexamethylene Diisocyanate, http://www.osha-slc.gov/dts/chemicalsampling/data/CH_245198.html, (accessed May 2001).

[12] RTECS 2000, 1,6-Diisocyanate Hexane, Registry of Toxic Effects of Chemical Substances, U.S. Department of Health and Human Services, National Institute for Occupational Safety and Health, Washington, DC., December 2000.

[13] Lawrence, R. *Evaluation Guidelines for Surface Sampling Methods*; OSHA Salt Lake Technical Center, U.S. Department of Labor: Salt Lake City, UT, 2001 (unpublished).

1.2 Limit defining parameters

1.2.1 Detection limit of the overall procedure

The detection limit of the overall procedure is 0.29 µg per sample. This is the smallest amount of HDI spiked on the wipe sampler that will give a detector response that is significantly different from the response of the wipe sampler blank. (Section 4.1)

1.2.2 Reliable quantitation limit

The reliable quantitation limit is 0.96 µg per sample. This is the amount of HDI spiked on the wipe sampler that will give a detector response that is considered the lower limit for a precise quantitative measurement. (Section 4.1)

1.2.3 Recovery effects of storage

The recovery of HDI from spiked samples used in a 15-day storage test remained above 88.6% when the samples were stored at 22°C. (Section 4.2)

1.2.4 Surface sampler removal efficiency

The removal efficiency of Ghost Wipe moistened with 0.5 mL of wetting reagent for HDI spiked at the target concentration of 340.0 µg/100cm^2 is 68.3%. This was the percentage of HDI that was removed from a glass plate surface, spiked at the target concentration. (Section 4.3)

1.2.5 Sampling reproducibility and analytical reproducibility

Six glass plate surfaces were spiked at the target concentration. Chemists, other than the one developing the method, conducted sampling on the glass plate surfaces as described in Section 2. The test was repeated with a second chemist performing the sampling. The first chemist was able to achieve a removal efficiency of 60.7%. The second chemist was able to achieve a removal efficiency of 55.5%. (Section 4.5.1)

Six samples spiked at the target concentration by liquid injection were submitted for analysis by the OSHA Salt Lake Technical Center. The samples were analyzed according to a draft copy of this procedure after 22 days of storage at 22 °C. The average analytical result was 111% of theoretical. (Section 4.5.2)

2. Sampling Procedure

All safety practices that apply to the work area being sampled should be followed. The sampling should be conducted in such a manner that it will not interfere with work performance or safety. The derivatizing reagent solution contains DMSO. DMSO is readily absorbed by the skin and is an excellent vehicle to transfer contaminants dissolved in it through the skin barrier. Skin contact with the derivatizing solution containing DMSO should be avoided.

2.1 Apparatus

2.1.1 Chemical resistant gloves; samples are collected using firm hand pressure to wipe the sampling medium across a surface. Wear a clean pair of gloves for each sample. The gloves selected are to be resistant to penetration of the chemical being sampled and any other chemicals expected to be present. Nitrile gloves were used for sampling 1,6-hexamethylene diisocyanate based on a review of glove manufacturer's chemical resistivity and degradation information.

2.1.2 Labeled 20-mL scintillation vials with PTFE lined caps, one for each sample, each containing 5 mL of derivatizing reagent solution.

2.1.3 Ghost Wipes, dry, catalog number SC4050, Environmental Express, Mt. Pleasant, S.C.

2.1.4 Disposable pipette and bulb, capable of delivering 0.5 mL. Non-sterile graduated pipette, catalog no. 13-711-9A, Fisher Scientific, Fair Lawn, NJ.

2.1.5 Optional steel or plastic measuring tape or disposable Sampling Template 10 × 10 cm, catalog number 1010, Environmental Express, Mt. Pleasant, S.C.

2.2 Reagents

2.2.1 Water, HPLC grade. A Millipore Milli-Q system was used to prepare the deionized water (DI water) for this evaluation.

2.2.2 Isopropanol, HPLC grade, Lot No. 001456, Fisher Scientific, Fair Lawn, NJ.

2.2.3 Ethyl acetate, HPLC grade, Lot No. 924366, Fisher Scientific, Fair Lawn, NJ.

2.2.4 Dimethyl sulfoxide (DMSO), HPLC grade, Lot No. BB965, Baxter, Muskegon, MI.

2.2.5 1-(2-Pyridyl) piperazine (1-2PP), Lot No. 09914JU, Aldrich, Milwaukee, WI.

2.2.6 Wetting reagent, 50:50 isopropanol:deionized water (DI) water.

2.2.7 Derivatizing solution, 50:50 ethyl acetate:DMSO with 0.025 M 1-2PP.

2.3 Shipping media and reagents to the sampling site and reagent preparation

The media are received from the supplier in packages containing 100 wipes per package. Wear clean gloves and remove a sufficient number of wipes to perform the sampling. Remember to include extra wipes to serve as blanks. Place the wipes in a 5 × 7 in. recloseable polyethylene zipper bag. Place the remaining wipes in a large recloseable polyethylene zipper bag. The selected wipes can now be shipped or taken to the workplace for sampling.

Place a disposable pipette and bulb, capable of delivering 0.5 mL in a small recloseable polyethylene zipper bag and ship with the wipe media.

Prepare the 50:50 isopropanol:DI water wetting reagent that is needed to moisten the wipe prior to sampling. Place 50.0 mL of the wetting reagent in a labeled bottle and cap securely.

Prepare the derivatizing solution that is combined with the wipe sample after sampling. The derivatizing solution consists of 50:50 ethyl acetate:DMSO with 0.025 M 1-2PP.

Place 5.0 mL of derivatizing solution in each scintillation vial, one for each sample. Cap the vials securely.

Following current Department of Transportation (DOT) regulations. Pack the bottle containing the wetting reagent and the vials containing the derivatizing solution in a shipping container and label it.

2.4 Sampling technique

Label a sufficient number of vials containing the derivatizing solution with a unique number, for the projected sampling needs. Wear clean unpowered gloves when handling the media.

Prepare a diagram of the area or rooms to be wipe sampled along with the locations of key surfaces.

Wear a new pair of clean gloves for each sample to prevent contamination of future samples as well as oneself. The gloves selected are to be resistant to penetration of the chemical being sampled and any other chemicals expected to be present. Nitrile gloves are recommended for sampling 1,6-hexamethylene diisocyanate based on a review of glove manufacturer's chemical resistivity and degradation information.

Record the sample vial number and the location where the sample is taken. Withdraw a Ghost Wipe from the zipper bag with your gloved fingers or clean tweezers. Use the disposable pipette to moisten the medium with 0.5 mL of the wetting reagent.

Depending on the purpose of the sample, it may be useful to determine the surface loading of the contamination (e.g., in micrograms of analyte per area). For these samples, it is necessary to determine the area of the surface wiped (e.g., 100 cm^2). To make this measurement, hold the measuring tape above the surface (without touching) or place the disposable sampling template on the surface before sampling. This would not be necessary for samples taken to simply show the presence of the contaminant.

The amount of time, from beginning to collect the sample, until the sample is placed in the vial containing the derivatizing reagent, should not exceed three minutes. (Section 4.9)

Firm pressure should be applied when wiping. Start at the outside edge and progress toward the center making concentric squares of decreasing size. Fold the medium with the contaminant side inward and repeat.

Without allowing the medium to come into contact with any other surface, fold the medium with the exposed side inward. Place the medium in a sample vial containing the derivatizing solution, cap and shake vigorously for one minute. Place a number corresponding the sample to the location on the diagram. Include notes with the sketch giving any further description that may prove useful when evaluating the sample results (e.g., a description of the surface sampled, such as: pencil, doorknob, safety glasses, lunch table, inside respirator, employee's names, etc.).

Submit at least one blank wipe medium, treated in the same fashion as the wipe samples, but without wiping.

Record sample location, employees names, surface area (if pertinent), work description, PPE, and any other necessary information, along with any potential interferences on the OSHA-91A form.

Submit the samples to the OSHA Salt Lake Technical Center together with OSHA-91A forms as soon as possible after sampling. Ship any bulk samples separate from the surface samples. Package, label, and ship samples according to current DOT regulations.

2.5 Extraction efficiency (Section 4.4)

It is the responsibility of each analytical laboratory to determine the surface sample extraction efficiency because the wipe sampling media, reagents and laboratory techniques may be different than the those listed in this evaluation and influence the results.

2.5.1 The mean extraction efficiency for 1,6-hexamethylene diisocyanate from Ghost Wipes over the range of RQL to 10 times the target concentration (0.99 to 3400 µg per sample) was 96.8%.

2.5.2 Extracted samples remain stable for at least 24 h.

2.6 Interferences, sampling

Suspected interferences should be reported to the laboratory with submitted samples.

Blank wipe sampling media was moistened with the wetting reagent, placed in vials and analyzed. Additional samples were prepared by wiping glass plates with wipe sampling media moistened with the wetting reagent. The surfaces were not spiked with HDI. The samples were analyzed. No significant interferences were found. (Section 4.6)

3. Analytical Procedure

Adhere to the rules set down in your Chemical Hygiene Plan[14]. Avoid skin contact and inhalation of all chemicals and review all appropriate MSDSs before beginning the analytical procedure.

This analysis closely follows the analytical procedure in OSHA Method 42.[15]

3.1 Apparatus

3.1.1 A high performance liquid chromatograph (HPLC) equipped with fluorescence (FL) and ultraviolet (UV) detectors, liquid chromatograph pump, manual or automatic injector, and chart recorder. A Hewlett-Packard Series 1050 HPLC equipped with a UV detctor and a Kratos Spectroflow 980 FL detector was used in this evaluation.

3.1.2 LC column capable of separating diisocyanate derivatives. A 25-cm × 4.6-mm i.d. Alltech Econosphere CN (5-µm) column was used during this evaluation.

3.1.3 An electronic integrator, or some other suitable method of measuring detector response. A Waters Corporation Millennium[32] (version 3.20) data system was used in this evaluation.

3.1.4 Vials, 2-mL with PTFE-lined caps.

3.1.5 Volumetric flasks, pipets, and syringes.

3.1.6 Micro-analytical balance used to weigh standard preparations.

3.1.7 Centrifuge.

3.2 Reagents

3.2.1 Acetonitrile (ACN), HPLC grade, Lot No. 012683, Fisher Scientific, Fair Lawn, NJ.

3.2.2 Water, HPLC grade. A Millipore Milli-Q system was used to prepare the water for this evaluation.

3.2.3 Ethyl acetate, HPLC grade, Lot No. 924366, Fisher Scientific, Fair Lawn, NJ.

3.2.4 Dimethyl sulfoxide (DMSO), HPLC grade, Lot No. BB965, Baxter, Muskegon, MI.

3.2.5 1,6-Hexamethyl diisocyanate (HDI), Lot No. 01104EU, Aldrich, Milwaukee, WI.

3.2.6 1-(2-pyridyl) piperazine (1-2PP), Lot No. 09914JU, Aldrich, Milwaukee, WI.

3.2.7 Ammonium acetate, Lot No. 897484, Fisher Scientific, Fair Lawn, NJ.

3.2.8 Phosphoric acid, Lot No. 025821, Baker, Phillipsburg, NJ.

---

[14] Occupational Exposure to Chemicals in Laboratories. Code of Federal Regulations, Part 1910.1450, Tittle 29, 1998; http://www.osha-slc.gov/OshStd_data/1910_1450_APP_A.html, (accessed May 2000).

[15] Diisocyanates,1,6-Hexamethylene Diisocyanate (HDI), Toluene-2,6-Diisocyanate, Toluene-2,4-Diisocyanate, Method 42, 1989; http://www.osha-slc.gov/dts/sltc/methods/organic/org042/org042.html, (accessed May 2000).

3.2.9 N,N'-1,6-hexanediylbis[4-(2-pyridinyl)-1-piperazinecarboxyamide], (1-(2-pyridyl)piperazine derivative of HDI), U. S. Department of Labor OSHA Technical Center Salt Lake City, UT. (An external source is: Product number 48146, Supelco, Bellefonte, PA.)

3.3 Standard preparation

3.3.1 All applicable Quality Assurance procedures and accreditation requirements should be observed.

3.3.2 A stock standard solution is prepared by dissolving derivatized HDI in DMSO. The derivatized HDI may be purchased or prepared following Section 3.3.1 of OSHA Method 42[16]. The ratio of the molecular weights of HDI to the HDI derivative is 0.340. Use this factor to express the mass of HDI derivative as the mass of HDI. All dilutions of the stock solutions are made with 90:10 ACN:DMSO to arrive at the working range.

3.3.3 Bracket sample concentrations with standard concentrations. If, upon analysis, sample concentrations fall outside the range of prepared standards, prepare and analyze additional standards to confirm instrument response, or dilute high samples with the 90:10 ACN:DMSO dilution solution.

3.4 Sample preparation

3.4.1 The surface samples are received in 20-mL vials containing the wipe media and 5.0 mL of derivatizing reagent solution. The vials are placed in a centrifuge that has been set at 2500 rpm for 4 min.

3.4.2 Remove an aliquot of approximately 1 mL and place into an appropriate autosampler vial and seal with a PTFE-lined cap.

3.5 Analysis

3.5.1 HPLC conditions

column:	25-cm × 4.6-mm i.d. Alltech Econosphere CN (5 µm).
mobile phase:	40:60 ACN:water, 0.02 M ammonium acetate adjusted to pH 5.9 with phosphoric acid.
flow rate:	1.0 mL/min for 15 min
FL detector:	240 nm excitation, 370 nm emission
UV detector:	254 nm
injection size:	10 µL
retention time:	9 min
chromatogram:	Figure 3.5.1

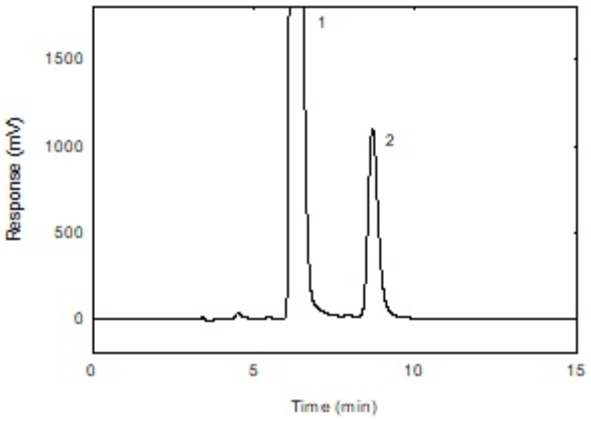

Figure 3.5.1. Chromatogram obtained from samples spiked at the target concentration and diluted 1/50. (1 - excess 1-2PP (derivatizing reagent); 2 - HDI.

[16] Diisocyanates,1,6-Hexamethylene Diisocyanate (HDI), Toluene-2,6-Diisocyanate, Toluene-2,4-Diisocyanate, Method 42, 1989; http://www.osha-slc.gov/dts/sltc/methods/organic/org042/org042.html, (accessed May 2000).

3.5.2 An external standard (ESTD) calibration procedure is used to prepare a calibration curve using at least 2 stock standards from which dilutions are made. The calibration curve is prepared daily. The samples are bracketed with analytical standards.

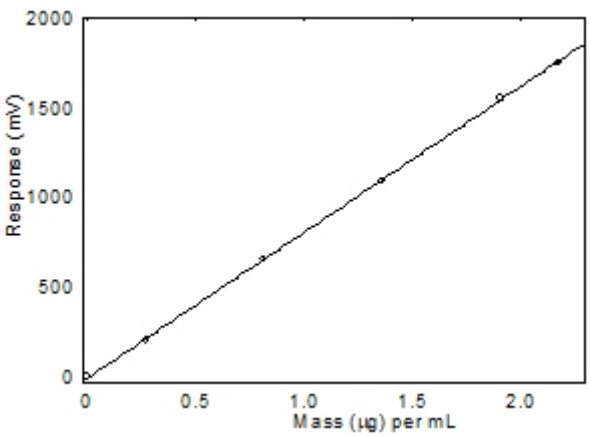

Figure 3.5.2. Calibration curve of HDI.
(Y = 813X - 6.30)

3.6 Interferences (analytical)

3.6.1 Any compound that produces a fluorescence or UV detector response and has a similar retention time as HDI is a potential interference. If any potential interferences were reported, they should be considered before samples are extracted. Generally, chromatographic conditions can be altered to separate an interference from the analyte. Material Safety Data Sheets (MSDS) identify solvents prevalent in spray painting operations. Three solvents commonly found were separately added to individual media that had been placed in vials, moistened with the wetting reagent and spiked with HDI at the target level. The samples were prepared and analyzed. Benzaldehyde and 2,4-toluene diisocyanate were also tested as potential interferences in the same manner. Neither the solvents or the chemicals caused a discernable interference. (Section 4.6)

3.6.2 When necessary, the identity of an analyte peak may be confirmed with additional analytical data. An absorbance response ratio of UV detector response to fluorescence detector response is determined. The response ratio of samples and standards of similar concentration are compared. (Section 4.8)

3.7 Calculations

The amount of HDI per sampler is obtained from the appropriate calibration curve in terms of micrograms per mL, uncorrected for extraction efficiency. This amount is then adjusted by subtracting the amount (if any) found on the blank and corrected for extraction efficiency. Correct for the 5.0 mL of derivatizing solution that was present in the samples when they were received and any dilutions performed. Perform the calculation using the following formula.

$$M_S = V \frac{M - M_B}{E_E}$$

where M_S is the mass of HDI recovered from the sampled surface (µg)
V is the volume of the derivatizing solution and any dilutions (mL)
M is micrograms per mL from the sample derivatizing solution
M_B is micrograms per mL from the blank derivatizing solution
E_E is the extraction efficiency

The amount may be expressed as micrograms HDI per 100 cm^2 if the surface area that was sampled was provided, by using the following formula.

$$C_S = 100 \frac{M_S}{S}$$

where C_S is the mass (in µg) of HDI per 100 cm^2
M_S is the mass on the sampled surface (µg)
S is the surface area sampled (cm^2)
100 is 100 cm^2

The surface that was sampled may be less ideal (more porous, less smooth) than the surface that was used to evaluate the removal efficiency of the sampling media. In this circumstance, the media will remove the surface contaminant less effectively. There may be significant amounts of contaminant remaining on the surface after sampling. Nevertheless, the amount found in the sample indicates that at least this amount of HDI was present on the surface.

4. Backup Data

General background information about the determination of detection limits and reproducibility of the overall procedure is found in the "Evaluation Guidelines for Surface Sampling Methods".[17]

4.1 Detection limit of the overall procedure (DLOP) and reliable quantitation limit (RQL)

The DLOP is measured as mass per sample. Five samplers were moistened with 0.5 mL wetting solution and spiked with equal descending increments of analyte, such that the highest sampler loading was 3.30 µg/sample. 5.0 mL of derivatizing reagent was added to these spiked samplers, and also a sample blank. The samplers were analyzed with the recommended analytical parameters, and the data obtained used to calculate the required parameters (standard error of estimate and the slope) for the calculation of the DLOP. The data is presented in Table 4.1. Values of 145.5 and 14.0 were obtained for the slope and standard error of estimate, respectively. The DLOP was calculated to be 0.29 µg/sample.

Table 4.1
Detection Limit of the Overall Procedure

mass per sample (µg)	response (mV)
0	35
0.66	102
1.32	193
1.98	304
2.64	400
3.30	506

The RQL is considered the lower limit for precise quantitative measurements. It is determined from the regression line parameters obtained for the calculation of the DLOP, providing 75% to 125% of the analyte is recovered. The RQL is 0.962 µg/sample. Average recovery at this level is 90.5%.

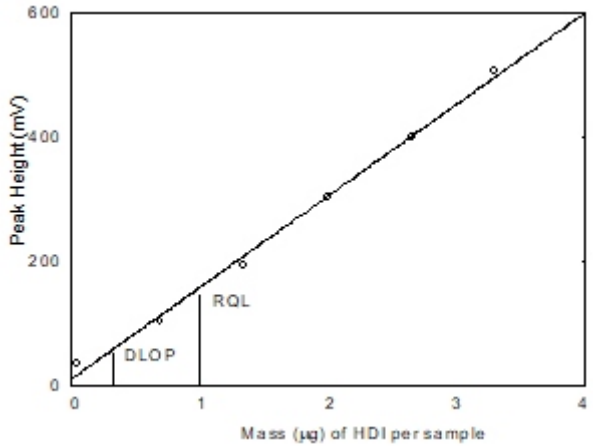

Figure 4.1.1. Plot of data to determine the DLOP/RQL. (Y = 146X + 16.7)

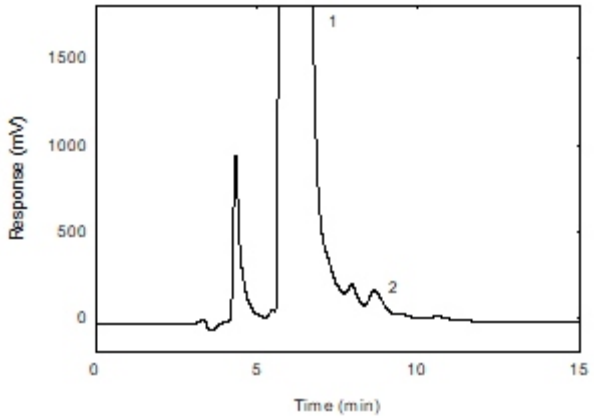

Figure 4.1.2. Chromatogram of the RQL. (1) - derivatizing reagent; (2) - HDI.

4.2 Storage tests

Storage samples were prepared by spiking Ghost Wipes, that had been moistened with the wetting solution, with HDI at the target concentration. Samples were immediately placed in vials containing the derivatizing reagent. Twenty-one storage samples were prepared. Three samples were analyzed

[17] Lawrence, R. *Evaluation Guidelines for Surface Sampling Methods*; OSHA Salt Lake Technical Center, U.S. Department of Labor: Salt Lake City, UT, 2000, unpublished.

on the day prepared. Nine of the samples were stored at reduced temperature (4 °C) and the other nine were stored in a closed drawer at ambient temperature (about 22 °C). At about 5-day intervals, three samples were selected from each of the two storage sets and analyzed. Sample results were not corrected for extraction efficiency.

Table 4.2
Storage Test for HDI

time (days)	ambient storage recovery (%)			refrigerated storage recovery (%)		
0	93.1	96.7	92.4	93.1	96.7	92.4
5	92.3	91.1	92.9	94.3	92.7	93.9
11	85.1	85.9	87.4	87.4	91.5	89.4
15	90.6	90.9	92.1	90.7	92.3	89.4

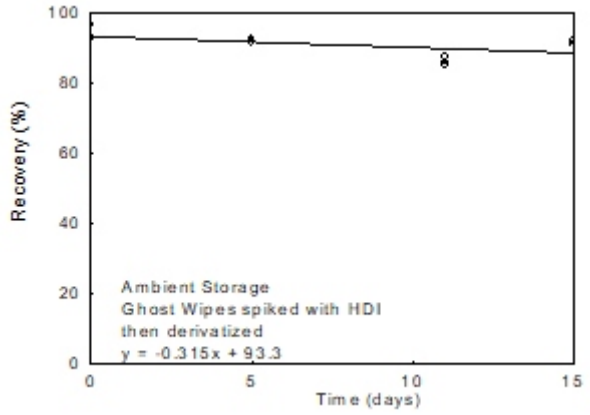

Figure 4.2.1. Ambient storage test for HDI.

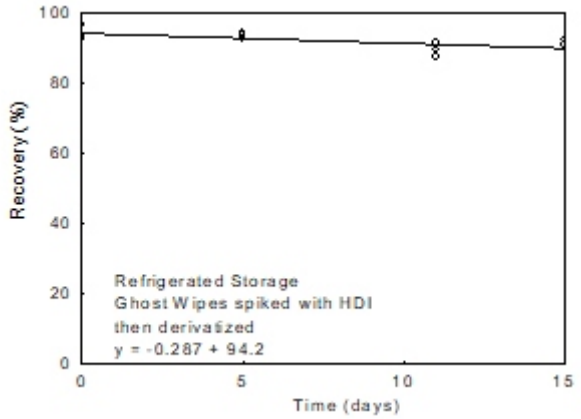

Figure 4.2.2. Refrigerated Storage test for HDI.

4.3 Sampler removal efficiency

Six glass surfaces were spiked at the target concentration of HDI, 340 µg /100 cm^2. Samples were collected from each surface using the technique described in Section 2.4 and analyzed. Sample results were corrected for extraction efficiency. The results are shown in Table 4.3.

Table 4.3
Sampler Removal Efficiency
Data for HDI on Ghost Wipes

theoretical (µg/surface)	recovered (µg/sample)	recovery (%)
340	220	64.7
340	246	72.3
340	244	71.7
340	219	64.4
340	228	67.1
340	236	69.3

4.4 Extraction efficiency and stability of extracted samples

The extraction efficiency is dependent on the solvents used in the derivatizing solution.

4.4.1 Extraction efficiency

The extraction efficiencies of HDI were determined by liquid-spiking Ghost Wipes with the HDI at concentrations ranging from the RQL to 10 times the target concentration. The sample media was placed in vials and moistened with the wetting solution. Each sampler was then spiked and the derivatizing reagent solution was added. Four samplers at each concentration were prepared. The samples were placed on a rotator for an hour. These samples were stored overnight at ambient temperature and then analyzed. The mean extraction efficiency over the working range of the RQL to 10 times the target concentration is 96.8%. Note that extraction efficiency also accounts for the additional volume of the wetting reagent.

Table 4.4.1
Extraction Efficiency of HDI from Ghost Wipe

level		sample number				
× target concn	µg per sample	1	2	3	4	mean
RQL	0.99	90.2	88.2	92.0	91.5	90.5
0.1	34.0	100.1	100.9	98.8	94.8	98.7
1.0	340	98.6	98.7	97.9	97.5	98.1
10.0	3400	99.2	100.9	100.1	99.2	99.9

4.4.2 Stability of extracted samples

The stability of extracted samples was investigated by re-analyzing the four target concentration samples 24 h after initial analysis. After the original analysis, two of the auto-sampler vials were recapped with new septa, while the remaining two retained their punctured septa. The samples were re-analyzed with fresh standards. The average percent change was 1.1% for the samples that were resealed and 1.2% for those that were stored with their septa punctured. The septum was punctured 3 times for each injection.

Table 4.4.2
Stability of Extracted Samples for HDI

punctured septa replaced			punctured septa retained		
initial (%)	after one day (%)	difference (%)	initial (%)	after one day (%)	difference (%)
98.6	100.4	1.8	97.9	98.7	0.8
98.7	98.3	0.4	97.5	99.1	1.6

4.5 Reproducibility

4.5.1 Six glass surfaces were spiked at the target level of 340 µg. Two chemists, other than the one developing the method, conducted sampling on the glass surfaces as described in Section 2. The test was repeated with a second chemist performing the sampling. Sample results were corrected for extraction efficiency.

Table 4.5.1.1
Sampling Reproducibility
Data for HDI on Ghost Wipe,
1st Chemist

theoretical (µg/surface)	recovered (µg/sample)	recovery (%)
340	176	51.9
340	212	62.3
340	209	61.4
340	208	61.2
340	222	65.4
340	212	62.2

Table 4.5.1.2
Sampling Reproducibility
Data for HDI on Ghost Wipe,
2nd Chemist

theoretical (µg/surface)	recovered (µg/sample)	recovery (%)
340	200	58.8
340	176	51.7
340	212	62.4
340	177	52.0
340	176	51.7
340	191	56.1

4.5.2 Six samples were prepared by spiking media in the same manner that was used in the preparation of samples for the storage study. The samples were submitted to the OSHA SLTC for analysis. The samples were analyzed after being stored for 22 days at 22°C. Sample results were corrected for extraction efficiency.

Table 4.5.2
Analytical Reproducibility
Data for HDI on Ghost Wipes

theoretical (µg/surface)	recovered (µg/sample)	recovery (%)
340	382	112
340	370	109
340	375	110
340	365	107
340	393	116
340	377	111

4.6 Interferences

4.6.1 Media

Tests were conducted to determine interference due to contamination of the prepared media. Two blank wipe sampling media were placed in vials, moistened with the recommended solvent. The derivatizing reagent solution was added and the samples were processed and analyzed.

Two additional samples were prepared by wiping the same type of surface that was used for the removal efficiency test (glass plate) with media moistened with 50:50 isopropanol:DI water. The surfaces were not spiked with HDI. The samples were placed in vials containing the derivatizing reagent solution, processed and analyzed. The results are shown in Table 4.6.1.

Table 4.6.1
Interference to the Analysis of HDI
from the Media or Surface (µg found)

sample	1	2	mean
blank	0.00	0.29	0.14
from surface	0.00	0.00	0.00

4.6.2 Tests were conducted to determine the effects of potential interference from three solvents that are commonly found in auto-body repair shops (toluene, 2-heptanone and petroleum distillate) and also, two additional chemicals (benzaldehyde and 2,4-toluene diisocyanate). Three samplers were prepared for each compound tested. The sample media was placed in vials and moistened with 0.5 mL of the wetting solution. Each sampler was then spiked with 340 µg HDI and 5.0 mL of the derivatizing reagent. A potential interfering compound was then added. The samples were processed and analyzed. The amounts of interfering compound added and the results are shown in Table 4.6.2. None of the compounds tested caused a significant interference.

Table 4.6.2
Interference to the Analysis of HDI, with an interferant compound added.
(µg found, not corrected for extraction efficiency)

potential interferant	amount of interferant spiked (µg)	amount of HDI spiked (µg)	amount of HDI recovered			
			1	2	3	mean
toluene	173400	340	319	320	315	318
2-heptanone	164000	340	305	307	305	306
petroleum distillate	145300	340	309	307	311	309
benzaldehyde	650	340	297	318	313	309
2,4-TDI	650	340	312	320	318	317

4.7 Analyte confirmation or qualitative analysis

Diisocyanate qualitative analysis confirmation is not performed by GC/MS because the derivatized diisocyanate is not sufficiently volatile for gas chromatography. Diisocyanate may be confirmed by a peak ratio technique. The analysis is conducted with fluorescence and UV detectors in series. A ratio is established between fluorescence detector response and UV detector response of a standard that is the approximate concentration of the sample. A similar ratio is established between fluorescence detector response and UV detector response of the sample. The ratio of the sample is compared to the ratio of the standard for diisocyanate confirmation. Any required laboratory confirmation criteria must be met before the analyte confirmation is reported.

4.8 Derivatizing reagent storage test

Derivatizing reagent solution was prepared as in Section 2.3 and divided into two portions. One portion was placed in a refrigerator. The other portion was placed in a fire cabinet at ambient temperature. After sixty days fresh derivatizing reagent was also prepared. Standards were prepared at 0.1, 1.0, and 10 times the target concentration using fresh derivatizing reagent. Spiked samples were prepared at 0.1, 1.0, and 10 times the target concentration using the ambient and refrigerated derivatizing reagent solutions. Excess derivatizing reagent peak was noted on the chromatograms for all samples and standards. Samples prepared with the derivatizing reagent solution that was stored at ambient temperatures for sixty days gave average results that were 93.4%, 96.4%, 99.2% of the 0.1, 1.0, and 10 times the target concentration, respectively. Samples prepared with the derivatizing reagent solution that was stored at refrigerated temperatures for sixty days gave average results that were 88.4%, 97.4%, 99.6% of the 0.1, 1.0, and 10 times the target concentration, respectively. The results are shown in Table 4.8. The effect of light on the derivatizing solution was not tested. The derivatizing reagent solution may be stored in a dark place with adequate ventilation, at room temperature for at least two months. No refrigeration is required for storage.

Table 4.8
Derivatizing Reagent Storage Test (60 days)

× target concn	ambient storage recovery (%)	refrigerated storage recovery (%)
0.1	93.4	88.4
1.0	96.4	97.4
10.0	99.2	99.6

4.9 Reaction time study

Many texts and articles indicate that water and alcohols will react with isocyanates. One published study determined that the reaction rate of water with isocyanates was at least 5 orders of magnitude (100,000) times slower than the reaction rate of the derivatizing reagent, 1,2-pyridyl piperazine with isocyanates.[18] The use of a wetting solution consisting of 50:50 isopropanol:water in this method required a reaction rate test to be performed. The test was to determine whether the water and isopropanol in the wetting reagent reacts with HDI rapidly enough to be a concern, relative to the amount of time it takes to collect a sample and deposit it into a vial containing the derivatizing reagent. Vials containing 0.5 mL 50:50 isopropanol:water (wetting solution) were spiked with HDI at 1.0, 0.5 and 0.1 times the target concentration. Two vial at each concentration were

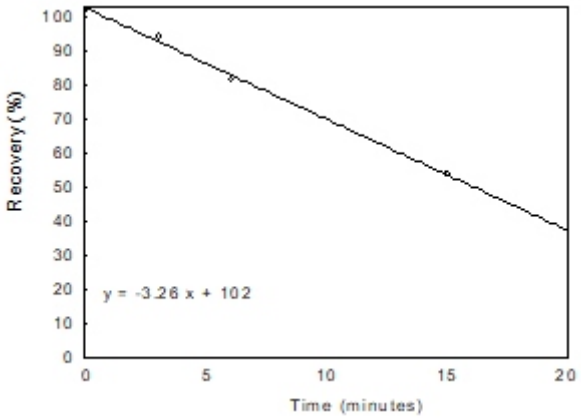

Figure 4.9. Average recovery of HDI at timed reaction intervals with the wetting solution.

[18] Wu, W., et al. Application of Tryptamine as a Derivatizing Agent for Airborne Isocyanate Determination Part 4. Evaluation of Major High-performance Liquid Chromatographic Methods Regarding Airborne Isocyanate Determination With Specific Investigation of the Competitive Rate of Derivatization, *Analyst* **1991**, 116, p 24.

prepared. Five milliliters of derivatizing reagent was added as quickly as possible. The vials were capped and agitated to insure mixing. The procedure was repeated three more times, except intervals of 3, 6 and 15 minutes were allowed to pass before adding the derivatizing reagent. Standards bracketing the concentrations were prepared with the same spiking solution in a similar manner, with no wetting solution, but in 5.5 mL of derivatizing reagent. Analysis was conducted and the result are presented in Table 4.9. The concentration of the spike did not appear to effect the recovery but the amount of time the spike was allowed to remain in the wetting reagent before being derivatized did effect the recovery. The percent recovery for all concentrations was averaged for each reaction time interval and plotted against time in minutes (Figure 4.9). Using the equation of the line, the recovery can be predicted for a given reaction time. The average recovery at 3.5 minutes is 90.6%. Based on this information, the amount of time it takes to collect a sample is important. The time interval, from beginning to collect the sample, until the sample is placed in the vial containing the derivatizing reagent, should not exceed three minutes.

Table 4.9
Percent Recovery of HDI After Timed Reaction Intervals With the Wetting Solution

µg spiked	"Immediate"	3 minutes	6 minutes	15 minutes
34.0	105.4	97.7	89.0	62.5
34.0	105.6	100.3	87.2	59.2
170.0	99.7	92.2	78.3	49.8
170.0	97.9	88.9	77.3	50.4
340.0	101.6	92.5	79.5	50.4
340.0	101.7	92.4	77.9	49.5
Average	101.9	94.0	81.5	53.7