

Commentary

Biological Monitoring for Isocyanates

JOHN COCKER*

Health and Safety Laboratory, Harpur Hill Buxton S10 3PT, UK

Received 9 August 2010; in final form 7 October 2010

Isocyanates are reactive chemicals and thousands of workers may be exposed to them during their manufacture and use in a wide range of products. They are classed as sensitizers and are a major cause of occupational asthma in the UK. Workplace exposure limits are low and control of exposure often depends on personal respiratory protection. Biological monitoring is increasingly used to assess exposure and the efficacy of control measures, including the behavioural aspects of controls. Biological monitoring methods are available for the most common isocyanates hexamethylene diisocyanate, toluene diisocyanate, isophorone diisocyanate, and methylenediphenyl diisocyanate. They are based on the analysis of hexamethylene diamine, toluene diamine, isophorone diamine, and methylenediamine released after hydrolysis of isocyanate–protein adducts in urine or blood. Volunteer and occupational studies show good correlations between inhalation exposure to isocyanate monomers and isocyanate-derived diamines in urine or blood. However, occupational exposure to isocyanates is often to a mixture of monomers and oligomers so there is some uncertainty comparing biological monitoring results with airborne exposure to ‘total NCO’. Nevertheless, there is a substantial body of work demonstrating the utility of biological monitoring as a tool to assess exposure and the efficacy of controls, including how they are used in practice. Non-health-based biological monitoring guidance values are available to help target when and where further action is required. Occupational hygienists will need to use their knowledge and experience to determine the relative contributions of different routes of exposure and how controls can be improved to reduced the risk of ill health.

Keywords: asthma; biological monitoring; dermal exposure; exposure assessment; isocyanates; urine analysis

Isocyanates are reactive chemicals with a $-N=C=O$ group. They react with hydroxyl groups to form a urethane linkage and with substances containing two or more hydroxyl groups; like polyols, they form polyurethane polymers that have a wide range of properties and applications. Products include rigid and flexible polyurethane foams, insulation materials, thermoplastic elastomers, two-pack spray paints, surface coatings, floor mastics, seals, fillers, and adhesives. Isocyanates also react with amine groups to form ureas and polyureas. The chemical reactivity that makes them so useful also poses problems for biological systems containing hydroxyl and amino

groups. With such a wide range of isocyanate-based products, occupational exposure is widespread and carries a risk of ill health (McDonald *et al.*, 2005). Isocyanates are classified as sensitizers and are the most commonly cited agent for occupational asthma. Vehicle paint sprayers using isocyanate-based paints were reported to be 80 times more likely to get occupational asthma than other workers (HSE, 2009). This may seem surprising given the widespread use of air-fed respiratory protective equipment (RPE) in this industry and raises questions about the actual effectiveness of control measures in practice.

The UK workplace exposure limit (WEL) (HSE, 2005a) for all isocyanates is $20 \mu\text{g NCO m}^{-3}$ for an 8-h time-weighted average (TWA) and $70 \mu\text{g m}^{-3}$ for a 15-min short-term exposure limit (STEL) but these are not health-based limits. The American

*Author to whom correspondence should be addressed.
Tel: +44-0-1298-218429; fax: +44-0-1298-218471;
e-mail: john.cocker@hsl.gov.uk

Conference of Governmental Industrial Hygienists (ACGIH) have recently proposed lowering the threshold limit value (8-h TWA) for toluene diisocyanate (TDI) to $7 \mu\text{g m}^{-3}$ ($3.4 \mu\text{g m}^{-3}$ as NCO), a STEL of $21 \mu\text{g m}^{-3}$ ($10.2 \mu\text{g m}^{-3}$ as NCO), and added a 'skin' notation to signify that dermal absorption of TDI may contribute significantly to the systemic dose. Reducing exposure reduces the risk but even at this level, a risk of respiratory sensitization may remain (ACGIH, 2010). The variety of isocyanates, the mixtures of monomers, oligomers, and their chemical reactivity pose major challenges for sampling and analytical techniques to assess occupational exposure.

The proposed reduction in the TLV may lead to increased use of RPE and together with the skin notation points to a need for biological monitoring of isocyanate exposure to help assess the efficacy of control measures. Because biological monitoring reflects systemic, rather than external, exposure, it can be used to assess dermal as well as inhalation exposure, control measures like RPE, and also the human behavioural aspects of control.

Biological monitoring for isocyanates has been available for many years (Rosenberg and Saviolainen 1986a,b) and is based on the analysis of isocyanate-derived diamines released by hydrolysis of protein adducts in urine or plasma. The dominant elimination half-lives of the isocyanates-derived diamines in urine are relatively short (2–5 h), which means that urine samples should be collected at the end of exposure and results mostly reflect exposure on the day of collection. The longer half-lives of plasma adducts (20–25 days) offers the potential for looking at integrated exposures over a longer period of time or investigating exposure incidents a short time after exposure has ceased (Sennbro *et al.*, 2004). Recent work shows that methylenediphenyl diisocyanate (MDI) forms adducts with lysine in plasma albumin and may offer potential for assessing risk (Sabbioni *et al.*, 2010). However, the convenience and non-invasive nature of urine sampling make it the most widely used approach for assessing exposure.

Analytical methods are available for the isocyanate-derived diamines from hexamethylene diisocyanate (HDI) (Rosenberg and Saviolainen, 1986a; Brorson *et al.*, 1990; Dalene *et al.*, 1994a; Skarping *et al.*, 1994; Tinnerberg *et al.*, 1995; Maitre *et al.*, 1996; Williams *et al.*, 1999), isophorone diisocyanate (IPDI; Dalene *et al.*, 1994b; Skarping *et al.*, 1994; Tinnerberg *et al.*, 1995), TDI (Sandstrom *et al.*, 1989; Brorson *et al.*, 1991; Skarping *et al.*, 1991; Maitre *et al.*, 1993; Tinnerberg *et al.*, 1995; Carbonnelle *et al.*, 1996; Lind *et al.*, 1996; Kaaria *et al.*, 2001a; Sakai *et al.*, 2002; Sennbro *et al.*, 2003), Naphthalene diiso-

cyanate (NDI; Sennbro *et al.*, 2003), and MDI (Tiljander *et al.*, 1989; Schutze *et al.*, 1995; Sennbro *et al.*, 2003 & 2006; Sepai *et al.*, 1995; Skarping and Dalene, 1995). The methods have a range of hydrolysis procedures, internal standards, extraction methods, derivatives, and detection. However, all the isocyanate-derived diamines in urine can be detected in a single gas chromatography-mass spectrometry method with detection limits around 1 nmol l^{-1} ($\sim 0.1 \mu\text{mol}$ diamine per mol creatinine), and coefficients of variation for within and between day analyses of 5 and 12%, respectively (Creely *et al.*, 2006).

The isocyanate-protein adducts are sufficiently stable for the samples to be sent by post to the laboratory for analysis. The analytical methods are capable of detecting isocyanate exposures well below current occupational exposure limits and a quality assurance scheme for methylenediamine (MDA) is available from the German Society of Occupational Medicine. Because the analysis is based on hydrolysis of conjugates, there is little risk of sample contamination during collection. However, any diamines present during exposure would also be detected and could lead to an overestimate of isocyanates exposure (Pauluhn and Lewalter, 2002). Urinary concentrations of toluene diamine (TDA), NDA, and MDA in people not occupationally exposed are $<0.1 \mu\text{mol mol}^{-1}$ (Rosenberg *et al.*, 2002; Sennbro *et al.*, 2005).

There have been a few volunteer studies and numerous occupational studies of exposure to isocyanates involving biological monitoring. All the volunteer studies and the occupational studies with both air and biological monitoring show good correlations between exposure (mostly inhalation) and levels of biomarkers in urine or blood and advocate the utility of biological monitoring. However, so far, only the German Deutsche Forschungsgemeinschaft (DFG, 2009) and the UK (HSE, 2005b) have proposed biological monitoring guidance values (BMGVs) for isocyanates although the ACGIH is working on a biological exposure index for TDI based on the revised TLV (ACGIH, 2010).

The German Commission for the Investigation of Health Hazards of Chemical Compounds in the Work Area have a Biologischer Leit-Wert (BLW) value for MDI of $10 \mu\text{g MDA l}^{-1}$ ($\sim 4 \mu\text{mol MDA}$ per mol creatinine) based on the levels likely to be found in urine after inhalation exposure to $50 \mu\text{g MDI m}^{-3}$ ($\sim 17 \mu\text{g NCO m}^{-3}$) for 8 h (DFG, 2009). The BLW does not exclude the risk of adverse effects on health. An occupational exposure study of MDI in France (Robert *et al.*, 2007) suggested a BMGV of $5 \mu\text{g MDA}$ per g creatinine ($2 \mu\text{mol}$ per mol creatinine).

In 2005, HSE published a BMGV for isocyanates of $1 \mu\text{mol}$ of isocyanates-derived diamine per mol

creatinine in urine samples collected at the end of exposure (HSE, 2005b). The BMGV was based on the 90th percentile of biological monitoring data from workplaces with exposure to HDI, IPDI, TDI, or MDI. It is also not a health-based guidance value but one based on exposure control. Any results exceeding the BMGV should simply trigger an examination of exposure controls and work practice with the intent of reducing exposure.

The comparison of biological data with airborne levels in occupational studies, where exposure may involve polymeric isocyanates, is more complex. An exposure chamber study with five volunteers (Brorson *et al.*, 1990) and a workplace study exposure study of 19 men exposed to HDI monomer (Maitre *et al.*, 1996) found relationships that at the UK, WEL predict levels of hexamethylene diamine (HDA) in urine in the range 10–30 μmol per mol creatinine. This suggests that if workers were exposed to HDI monomer, urine levels at the UK BMGV of 1 μmol per mol creatinine would indicate exposures considerably below the UK WEL. However, the data in HSE's workplace studies of HDI came from paint sprayers using HDI-based paints where the level of HDI monomer in the hardener is $<0.5\%$ and the bulk of the isocyanates is the oligomers. If the HDA measured in sprayers' urine came only from exposure to monomer, then levels of 1 μmol per mol creatinine would represent exposures considerably above the WEL. In view of the widespread use of air-fed RPE by paint sprayers, this seems unlikely. An alternative explanation is that the hydrolysis of urine releases not only HDA from HDI-monomer conjugates but also, at least in part, from the oligomers and/or their conjugates. This seems more likely and is supported by an early study of five paint sprayers who were exposed in a chamber study to HDI-based paints with 0.24% monomer (34% oligomer) at $6\times$ the WEL (as total NCO) for 15 min (Rosenberg and Saviolainen, 1986a). The levels of HDA in urine were roughly $2\text{--}6\times$ the levels predicted at the WEL (as NCO) from the Brorson and Maitre study suggesting that, in the Rosenberg and Saviolainen study, more than just HDI-monomer conjugates were detected. Two volunteer studies with inhalation exposure to mixtures of 2,4- and 2,6-TDI monomers predict urine TDA levels in the range 10–20 μmol per mol creatinine at the UK WEL (Brorson *et al.*, 1991; Skarping *et al.*, 1991). The results found in occupational studies of exposure to TDI during manufacture of polyurethane foams (Maitre *et al.*, 1993; Kaaria *et al.*, 2001a) show a good correlation between airborne TDI and TDA in urine and also suggest that after exposure to TDI at the UK, WEL

urinary TDA will be in the range 10–20 μmol per mol creatinine. So urine TDA below the BMGV of 1 μmol per mol creatinine are again indicative of exposure below the WEL and also below the proposed ACGIH TLV of $7\text{ }\mu\text{g m}^{-3}$, at least for monomeric TDI mixtures. Tinnerberg and Mattsson (2008) recently demonstrated the utility of biological monitoring to demonstrate the effectiveness of control intervention for TDI.

Biological monitoring assesses exposure by inhalation, ingestion, and through the skin and disentangling which is the most significant is the challenge for occupational hygienists. Isocyanates are respiratory and skin sensitizers so both dermal and inhalation exposures need to be controlled. A recurring theme in some published studies is the possibility of dermal absorption contributing to the levels of isocyanate-derived diamines in urine (Maitre *et al.*, 1993; Creely *et al.*, 2006; Austin, 2007; Fent *et al.*, 2008; Flack *et al.*, 2010; Gaines *et al.*, 2010; Liljelind *et al.*, 2010). The good correlations between personal air levels of isocyanates and metabolites in urine, noted in the studies above, suggest that inhalation is the most significant route, at least for the volatile isocyanates or those that are sprayed.

However, there is a clear evidence of dermal deposition and absorption into the skin. Fent *et al.* (2009) measured breathing zone and dermal exposure of spray painters to HDI, uretidone, biuret, and isocyanurate and found dermal concentrations of these isocyanates were higher in painters who did not wear gloves or coveralls. The authors estimated that use of protective equipment was associated with 95% reduction of isocyanurate on skin, but even so isocyanurate (the most significant oligomer) could be detected in 93% of samples from the skin of painters. Bello *et al.* (2006) investigated skin absorption of isocyanates *in vitro* with guinea pig skin and found that polymeric isocyanates can remain on the skin as unreacted species for several hours and that the rate of loss from the skin decreased with increasing molecular weight of the isocyanates. Gaines *et al.* (2010) studied 48 spray painters and found that dermal exposure and inhalation exposure levels, adjusted for the type of respirator worn, were predictors, in their linear mixed models, of urine HDA levels. However, 64% of the dermal samples collected from sprayers were below the limit of detection and the correlation with urine HDA was poor suggesting that the dermal contribution to total exposure in spray painters may be less than inhalation. There is also some evidence that if the air-fed respiratory protection used by paint sprayers is working and being used

correctly, no HDA is found in urine (Rosenberg and Saviolainen, 1986a; Williams *et al.*, 1999).

The significance of the dermal route of exposure may depend on the process as well as the volatility of the isocyanates. Austin (2007) looked at exposure to TDI in two groups of workers manufacturing polyurethane foam. Both groups had similar inhalation exposures but those handling partially cured foam had significantly higher urinary TDA levels suggesting some dermal absorption.

Kaaria *et al.* (2001b) found MDA in 97% of urine samples from workers exposed to MDI making rigid polyurethane foams even though MDI could not be detected in 64% of breathing zone air samples. In those samples where MDI could be detected, the concentrations were <2% of the Finnish occupational exposure limit (OEL). Liljelind *et al.* (2010) measured inhalation and dermal exposure to MDI in iron foundry workers and found airborne levels 50-fold less than the Swedish OEL. Dermal exposure measured using a tape stripping technique on the forefingers, wrists, and forehead found MDI in successive tape strips suggesting that MDI penetrates the skin, but making conservative assumptions for dermal absorption and inhalation, the authors calculated the contribution of dermal exposure would be 20× less than inhalation exposure.

Whether dermal exposure leads to respiratory sensitization is difficult to study in humans. Workplace studies are likely to have both dermal and inhalation exposures and volunteer studies pose ethical problems. Redlich and Herrick (2008) in a review of recent literature on lung and skin connections in occupational lung disease concluded that although further research is needed, it is prudent to reduce both skin and inhalation exposures. In practice, HDI, TDI, IPDI, and MDI are classed as skin sensitizers and therefore, dermal exposure should be well controlled.

In summary, there is still some uncertainty about what exactly is being measured in hydrolysed urine from workers exposed to isocyanates and its relationship to ill health. Nevertheless, there is a substantial body of work demonstrating the utility of biological monitoring as tool to assess exposure to isocyanates and the efficacy of controls, including the human behavioural aspects of control. There are BMGVs but these are not health-based rather than they are an aid to target when and where further investigation is required. Biological monitoring assesses systemic exposure and occupational hygienists will need to use their knowledge and experience to determine the relative contributions of different routes of exposure and how controls can be improved to reduced exposure and the risk of ill health.

REFERENCES

- ACGIH. (2010) TLVs and BEIs Based on the documentation of the threshold limit values for chemical substances and physical agents and biological exposure indices. Cincinnati, OH: American Conference of Governmental Industrial Hygienists ISBN 978-1-607260-19-6.
- Austin S. (2007) Biological monitoring of TDI-derived amines in polyurethane foam production. *Occup Med (Lon)*; 57: 444–8.
- Bello D, Smith TJ, Woski SR *et al.* (2006) An FTIR investigation of isocyanates absorption using in vitro guinea pig skin. *J Environ Monit*; 8: 523–9.
- Brorson T, Skarping G, Nielsen J. (1990) Biological monitoring of isocyanates and related amines II. Test chamber exposure of humans to 1,6 hexamethylenediisocyanate (HDI). *Int Arch Occup Environ Health*; 62: 385–9.
- Brorson T, Skarping G, Sango C. (1991) Biological monitoring of isocyanates and related amines IV. 2,4 and 2,6 toluenediamine in hydrolysed plasma and urine after test chamber exposure of humans to 2,4 and 2,6 toluene diisocyanate. *Int Arch Occup Environ Health*; 63: 253–9.
- Carbonnelle P, Boukourt S, Lison D *et al.* (1996) Determination of toluenediamines in urine of workers occupationally exposed to isocyanates by high-performance liquid chromatography. *Analyst*; 121: 663–9.
- Creely KS, Hughson GW, Cocker J *et al.* (2006) Assessing isocyanate exposure in polyurethane industry sectors using biological and air monitoring methods. *Ann Occup Hyg*; 50: 609–21.
- Dalene M, Skarping G, Tinnerberg H. (1994a) Biological monitoring of hexamethylene diisocyanate by determination of 1,6 hexamethylene diamine as the trifluoroethyl chloroformate derivative using capillary gas chromatography with thermionic and selective-ion detection. *J Chromatogr B Biomed Appl*; 656: 319–28.
- Dalene M, Skarping G, Tinnerberg H. (1994b) Thermospray mass spectrometry of aliphatic diamines derivatised with trifluoroethyl chloroformate, with special reference to the biological monitoring of hexamethylenediisocyanate (HDI) and isophoronediiisocyanate (IPDI). *Chromatographia*; 38: 776–80.
- DFG. (2009) List of MAK and BAT values 2009 Commission for the investigation of health hazards of chemical compounds in the work area. Report No. 45. Weinheim, Germany: Wiley-VCH Verlag GmbH ISBN 978-527-32596-2.
- Fent KW, Gaines LGT, Thomasen JM *et al.* (2009) Quantification and statistical modelling—part II: dermal concentrations of monomeric and polymeric 1,6 hexamethylene diisocyanate. *Ann Occup Hyg*; 53: 691–702.
- Fent KW, Jayaraj K, Ball LM *et al.* (2008) Quantitative monitoring of dermal and inhalation exposure to 1,6-hexamethylene diisocyanate and oligomers. *J Environ Monit*; 10: 500–7.
- Flack SL, Fent KW, Gaines LGT *et al.* (2010) Quantitative plasma biomarker analysis in HDI exposure assessment. *Ann Occ Hyg*; 54: 41–54.
- Gaines LGT, Fent KW, Flack SL *et al.* (2010) Urine 1,6 hexamethylene diamine (HDA) levels among workers exposed to 1,6 hexamethylene diisocyanate (HDI). *Ann Occup Hyg*; 54: 678–91.
- HSE. (2005a) EH40/2005 Workplace exposure limits. Sudbury Suffolk, UK: HSE books.
- HSE. (2005b) WATCH Committee Biological Monitoring for isocyanates. Available at <http://www.hse.gov.uk/aboutus/hsc/iaes/acts/watch/051005/13.pdf>. Accessed 19 December 2010.

- HSE. (2009) Safety in motor vehicle repair: working with isocyanates paints. INDG 388(rev1). Available at <http://www.hse.gov.uk/pubns/indg388.pdf>. Accessed 15 June 2010.
- Karria K, Hirvonen A, Norppa H *et al.* (2001a) Exposure to 2,4- and 2,6-toluene diisocyanate during production of flexible foam: determination of airborne TDI and urinary 2,4- and 2,6-toluenediamine (TDA). *Analyst*; 126: 1025–31.
- Karria K, Hirvonen A, Norppa H *et al.* (2001b) Exposure to 2,4- and 2,6-toluene diisocyanate (MDI) during moulding polyurethane foam: determination of airborne MDI and urinary 4,4'-methylenedianiline (MDA). *Analyst*; 126: 476–9.
- Liljelind I, Norberg C, Egelrun L *et al.* (2010) Dermal and inhalation exposure to methylene bisphenyl isocyanates (MDI) in iron foundry workers. *Ann Occup Hyg*; 54: 31–40.
- Lind P, Dalene M, Skarping G *et al.* (1996) Toxicokinetics of 2,4 and 2,6 toluene diamine in hydrolysed urine and plasma after occupational exposure to 2,3 and 2,6 toluene diisocyanate. *Occup Environ Med*; 53: 94–9.
- Maitre A, Berode M, Perdix A *et al.* (1993) Biological monitoring of occupational exposure to toluene diisocyanate. *Int Arch Occup Environ Health*; 65: 97–100.
- Maitre A, Berode M, Perdix A *et al.* (1996) Urinary hexane diamine as an indicator of occupational exposure to hexamethylenediisocyanate. *Int Arch Occup Environ Health*; 69: 65–8.
- McDonald JC, Chen Y, Zekveld C *et al.* (2005) Incidence by occupation and industry of acute work related respiratory diseases in the UK, 1992–2001. *Occup Environ Med*; 62: 836–42.
- Pauluhn J, Lewalter J. (2002) Analysis of markers of exposure to polymeric methylene-diphenyl diisocyanate (pMDI) in rats: a comparison of dermal and inhalation exposure. *Exp Toxic Pathol*; 53: 135–46.
- Redlich CA, Herrick CA. (2008) Lung/skin connections in occupational lung disease. *Curr Opin Allergy Clin Immunol*; 8: 115–9.
- Robert A, Ducos P, Francin JM *et al.* (2007) Biological monitoring of workers exposed to 4,4'-methylenediphenyl diisocyanate (MDI) in 19 French polyurethane industries. *Int Arch Occup Environ Health*; 80: 412–22.
- Rosenberg C, Saviolainen H. (1986a) Determination in urine of diisocyanate-derived amines from occupational exposure by gas chromatography-mass spectrometry. *Analyst*; 111: 1069–71.
- Rosenberg C, Saviolainen H. (1986b) Determination of occupational exposure to toluene diisocyanate by biological monitoring. *J Chromatogr*; 367: 385–92.
- Rosenberg C, Nikkila K, Henriks-Eckerman M *et al.* (2002) Biological monitoring of aromatic diisocyanates in workers exposed to thermal degradation products of polyurethanes. *J Environ Monit*; 4: 711–6.
- Sabbioni G, Dongari N, Kumar A. (2010) Determination of a new biomarker in subjects exposed to 4,4'-methylenediphenyl diisocyanate. *Biomarkers*; 15: 508–15.
- Sakai T, Morita Y, Kim Y *et al.* (2002) LC-MS determination of urinary toluenediamine in workers exposed to toluenediisocyanate. *Toxicol Lett*; 134: 259–64.
- Sandstrom JF, Skarping G, Dalene M. (1989) Chromatographic determination of amines in biological fluids with special reference to the biological monitoring of isocyanates and amines. II determination of 2,4 and 2,6 toluenediamine using glass capillary gas chromatography and selected ion monitoring. *J Chromatogr*; 479: 135–43.
- Schutze D, Sepai O, Lewalter J *et al.* (1995) Biomonitoring of workers exposed to 4,4'-methylenedianiline or 4,4'-methylenediphenyl diisocyanate. *Carcinogenesis*; 16: 573–82.
- Sennbro CJ, Lindh CH, Mattsson C *et al.* (2006) Biological monitoring of exposure to 1,5-naphthalene diisocyanate and 4,4'-methylenediphenyl diisocyanate. *Int Arch Occup Environ Health*; 79: 647–53.
- Sennbro CJ, Lindh CH, Tinnerberg H *et al.* (2003) Development, validation and characterization of an analytical method for the quantification of hydrolysable urinary metabolites and plasma protein adducts of 2,4- and 2,6-toluene diisocyanate, 1,5 naphthalene diisocyanate and 4,4'-methylenediphenyl diisocyanate. *Biomarkers*; 8: 204–17.
- Sennbro CJ, Lindh CH, Tinnerberg H *et al.* (2004) Biological monitoring of exposure to toluene diisocyanate. *Scand J Work Environ Health*; 30: 371–8.
- Sennbro CJ, Littorin M, Tinnerberg H *et al.* (2005) Upper reference limits for biomarkers of exposure to aromatic diisocyanates. *Int Arch Occup Environ Health*; 78: 541–6.
- Sepai O, Schutze D, Heinrich U *et al.* (1995) Hemoglobin adducts and urine metabolites of 4,4'-methylenedianiline after 4,4'-methylenediphenyl diisocyanate exposure of rats. *Chem Biol Interact*; 97: 185–98.
- Skarping G, Brorson T, Sango C. (1991) Biological monitoring of isocyanates and related amines III. Test chamber exposures of humans to toluene diisocyanate. *Int Arch Occup Environ Health*; 63: 83–8.
- Skarping G, Dalene M. (1995) Determination of 4,4'-methylenediphenyldianiline (MDA) and identification of isomers in technical-grade MDA in hydrolysed plasma and urine from workers exposed to methylenediphenyldiisocyanate by gas chromatography-mass spectrometry. *J Chromatogr B Biomed Appl*; 663: 209–16.
- Skarping G, Dalene M, Tinnerberg H. (1994) Biological monitoring of hexamethylene and isophorone-diisocyanate by the determination of hexamethylenediamine and isophoronediamine in hydrolysed urine using liquid chromatography and mass spectrometry. *Analyst*; 119: 2051–5.
- Tiljander A, Skarping G, Dalene M. (1989) Chromatographic determination of amines in biological fluids with special reference to the biological monitoring of isocyanates and amines III determination of 4,4'-methylenedianiline in hydrolysed human urine using derivatization and capillary gas chromatography with selected ion monitoring. *J Chromatogr*; 479: 142–52.
- Tinnerberg H, Mattsson C. (2008) Usage of air monitoring and biomarkers of isocyanates exposure to assess the effect of a control intervention. *Ann Occup Hyg*; 52: 187–94.
- Tinnerberg H, Skarping G, Dalene M *et al.* (1995) Test chamber exposure of humans to 1,6 hexamethylene diisocyanate and isophorone diisocyanate. *Int Arch Occup Environ Health*; 67: 367–74.
- Williams NR, Jones K, Cocker J. (1999) Biological monitoring to assess exposure from isocyanate use in motor vehicle repair. *Occup Environ Med*; 56: 598–601.