Understanding skin absorption of common aldehyde vapours from exposure during hazardous material incidents

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ABSTRACT

The toxic release of aldehyde vapours during a hazardous materials (HAZMAT) incident primarily results in respiratory concerns for the unprotected public. However, skin absorption may be an important concurrent exposure route that is poorly understood for this scenario. This study provides experimental data on the skin absorption properties of common aldehydes used in industry, including acetaldehyde, acrolein, benzaldehyde and formaldehyde, in gaseous or vapour form using an adapted *in vitro* technique. Two of the four tested aldehydes were found to penetrate the skin in appreciable amounts following 30 minute exposure at HAZMAT relevant atmospheric concentrations; Acetaldehyde (5.29 ± 3.24 $\mu g/cm^2$) and formaldehyde (3.45 ± 2.58 $\mu g/cm^2$). Whereas only low levels of acrolein (0.480 ± 0.417 $\mu g/cm^2$) and benzaldehyde (1.46 ± 0.393 $\mu g/cm^2$) skin penetration was noted. The aldehydes demonstrated differing levels of interaction with fabric. Formaldehyde and acetaldehyde adsorbed strongly to denim whereas benzaldehyde and acrolein displayed no sink properties. However, denim was shown to be an initial protective barrier and reduced penetration outcomes for all aldehydes. This study provides important information to assist first responders and confirms the relevance of using physicochemical properties (e.g. solubility, MW, partition coefficient) to predict skin permeation potential in the absence of empirical data during HAZMAT incidents involving different types of aldehydes.

KEYWORDS

Dermal Exposure, Aldehydes, Vapour, *in vitro,* HAZMAT

INTRODUCTION

Hazardous material (HAZMAT) incidents that lead to human injury most frequently arise from accidental or intentional exposure to toxic gases or vapours [1, 2]. This type of toxic release is usually related to an industrial accident or terrorism-related event. Chemical spill incidents and the deliberate release of toxic materials in terrorism-related events have the potential to affect large numbers of people, especially where these occur in major population centres. In addition to causing widespread fear, they may result in the exposure of the public, emergency first responders, and hospital personnel to chemicals that pose a risk of adverse health effects ranging from mild, through serious to lethal. The respiratory pathway is the most commonly identified hazard route resulting from exposure to gases and vapours; however, skin absorption, arising from dermal exposure, is often an important secondary route that is not well understood for many chemicals. Chemical HAZMAT incidents of this nature are a unique scenario that are typically defined by relatively short exposure timeframes to high concentrations of chemical and usually involve an exposure group with only 'common clothing' offering protection of the skin. Having a greater understanding of the skin absorption properties of toxic gases and vapours under these conditions highlights whether there is potential for this pathway to contribute to the overall toxic burden. Further, it can help inform the decontamination procedures employed by first responders following a HAZMAT incident [3, 4].

Aldehydes are a group of relatively reactive and corrosive organic compounds that are utilised heavily in industry for the manufacture of resins, wood products, paper, plastics, dyes, textiles, carpet and leather goods [5]. The extensive use of volatile aldehydes in these industries along with the associated transportation of large quantities of these chemicals has the

potential to result in the toxic release of aldehyde vapours. For example, a number of HAZMAT-related incidents involving the toxic release of aldehyde vapours in the United States have been documented in government databases [6, 7]. In addition to their industrial use, aldehydes are also a known constituent of combustion smoke and pose an exposure risk to emergency agency personnel responding to bush or structural fires [8, 9]. Previous studies have shown that urinary metabolites of common combustion products (i.e. polycyclic aromatic hydrocarbons (PAHs)) have been detected in firefighters [10-12]. The assumption in these circumstances is that the dose is occurring via skin absorption as in theory respiratory protection by means of self-contained breathing apparatus were always employed. This anecdotal evidence means that the skin permeation profiles of other common combustion products (i.e. aldehyde gases and vapours) are of high importance, and may have practical safety implications, for the firefighting community.

In this study we investigate the dermal absorption properties of four common aldehydes, namely, acetaldehyde, acrolein, benzaldehyde and formaldehyde. Acetaldehyde is an aliphatic aldehyde that exists as a colourless, flammable and volatile liquid. Owing to its low boiling point (21 °C) and relatively high vapour pressure (902 mm Hg at 25 °C) it exists as a colourless, pungent gas at room temperature. Whilst low-level sources of acetaldehyde exist in the natural environment (e.g. fruits, beverages, tobacco smoke) it is also used extensively in the manufacture of acetic acid, dyes, explosives, lacquers, plastics and synthetic rubber [13]. Acrolein is also an aliphatic aldehyde that exists as a flammable liquid and has a strong odour. Large quantities of acrolein are synthesised for industrial use and it is used as an intermediate in the manufacture of synthetic glycol, polyurethane, pharmaceuticals and herbicides [14]. Benzaldehyde is an aromatic aldehyde that exists as a clear colourless to slightly yellow liquid and has relatively low volatility (0.127 mm Hg at 25 °C). It is a common ingredient in cosmetics

as a denaturant, flavouring agent and fragrance and is also a key ingredient in food flavouring [15]. Finally, formaldehyde is the smallest aliphatic aldehyde and exists as a colourless strongly-odoured gas at room temperature. It is sold commercially as a 37 %, 44 % or 50 % methanol-stabilised solution called formalin and is commonly used in wood processing, the production of resins, fertilisers, dyes, disinfectants, germicides and as a biological fixative in medical laboratories [16].

There is limited literature surrounding dermal exposure to the nominated aldehydes and whether dermal uptake is a potential contributor to toxicity as a concurrent exposure route to respiratory inhalation. There are no known studies on the potential for dermal uptake of formaldehyde, acetaldehyde or acrolein as a gas or vapour exposure. The limited published studies commonly relate to liquid phase aldehyde exposures, and for exposure times and conditions not relevant to HAZMAT incident scenarios. For example, acrolein is reported to be a dermal, respiratory and ocular irritant with the majority of experimental work relating to inhalation and dermal exposure performed on animals [17]. Lacroix *et al* performed patch testing and observed minimal skin irritation in some cases at low concentration doses (1.0% acrolein) and more severe irritation in all cases at higher concentration doses (10% acrolein) [18]. Faroon *et al* published an extensive review of acrolein health effects which reported that dermal absorption leads to severe local irritation or corrosion at the site of contact and cited dermal LD_{50}'s for rabbits but not humans [19]. No known studies have reported on the dermal uptake potential arising from acrolein liquid or vapour exposure in humans. Similarly, formaldehyde in solution is known to be absorbed into the human skin matrix and is used widely in stabilised solution with methanol (commonly known as formalin) as a biological fixative for tissue samples. Two previous *in vitro* studies have been reported investigating the dermal uptake from formalin solution through human skin with both finding that

formaldehyde was able to penetrate the skin [20, 21]. However, the tendency for formaldehyde to penetrate human skin may be overstated in these studies due to the presence of methanol in the formulation applied to the skin; which is a known penetration enhancer [20]. There are no known studies on the potential for dermal uptake arising from formaldehyde gas exposure.

A number of studies have applied patch testing methodology to evaluate skin allergy and irritation responses to acetaldehyde and reported mixed results with skin sensitisation observed in only some human subjects [22-25]. No studies have reported on the dermal uptake potential of acetaldehyde arising from liquid or vapour exposure in humans. Finally, benzaldehyde is reported to be absorbed through human skin and by the lungs [15]. Barry *et al* performed *in vitro* studies into the liquid and vapour (in a static atmosphere) penetration of benzaldehyde using human abdominal cadaver skin which provided evidence of dermal uptake and reported a permeation flux for both liquid and vapour exposures [26].

In this paper we report the successful application of an adapted *in vitro* dermal exposure technique with dynamic atmosphere gas/vapour generation for the exposure of acetaldehyde, acrolein, benzaldehyde and formaldehyde vapours/gas to human epidermal skin over short-term exposure times relevant to HAZMAT incidents. The empirical data generated addresses an existing knowledge gap surrounding the dermal absorption and penetration properties of each of the aldehydes investigated and can be used to inform decision-making on decontamination procedures following toxic release of these chemicals. This is the first reported study utilising dynamic atmospheres to investigate the dermal absorption properties of aldehyde vapours/gases with human epidermal skin for short-term exposure scenarios consistent with HAZMAT related incidents.

MATERIALS AND METHODS

Chemicals and chromatography standards

The structural and physicochemical properties of the four aldehydes investigated are shown in Table 1 along with toxicological relevant values. Liquid acetaldehyde (≥99.9%), acrolein (90%) and benzaldehyde (≥99.9%) were all purchased from Sigma Aldrich, Australia and used to produce dynamic aldehyde vapour atmospheres. Solid paraformaldehyde (97%, BDH Chemicals, Australia) was used to produce dynamic formaldehyde atmospheres. Solid 2,4-dinitrophenylhydrazine (≥99, Sigma Aldrich, Australia) was used to create a solution (6 mg/ml in acetonitrile) which was subsequently used to form aldehyde-2,4-dinitrophenylhydrazone derivatives suitable for HPLC-UV analysis. Analytical grade standards of the corresponding aldehyde-2,4-dinitrophenylhydrazones were purchased from Sigma Aldrich, Australia, and used to prepare chromatography standards for HPLC-UV analysis. HPLC solvents used were acetonitrile (>99.5%, Sigma Aldrich, Australia), orthophosphoric acid (HPLC grade, 85% w/w) and Milli-Q water.

NA – not available

Methods

Skin permeation experiments

Skin permeation experiments were performed *in vitro* using static 9 mm Franz diffusion cells with a diffusion-available surface area of 0.64 cm^2 (PermeGear^{TM}, Pennsylvania) and modified for flow-through vapour delivery to the surface of the skin as outlined previously [27, 28]. Human abdominal skin was sourced from cosmetic reduction surgery, with donor consent and ethics approval (SACHR ethics approval #273.10) and the epidermis harvested as described

previously (Gaskin et al, 2014). Human skin donors were female, Caucasian, and ranged in age from 24 to 57 years with no obvious signs of skin damage or scarring/tattooing. Skin electrical impedance testing was utilised to determine barrier integrity *in vitro* pre-exposure [27, 29-31].

Acrolein, acetaldehyde and benzaldehyde vapour were delivered to the skin surface using a previously reported dynamic atmosphere generator [27, 32]. Briefly, liquid aldehydes were injected using a syringe pump in to a mixing chamber and a volatilised fraction subsequently diluted with purified air to achieve the final concentrations. Final test atmosphere concentrations for each aldehyde were: acrolein (153 ppm / 351 mg/m^3), acetaldehyde (996 ppm / 1 794 mg/m^3) and benzaldehyde (167 ppm / 725 mg/m^3). Alternatively, solid paraformaldehyde was used to create formaldehyde atmospheres by placing inside a glass impinger and heating in a water bath (70 °C) to liberate formaldehyde gas. A diluting carrier gas was then passed over the solid surface to produce final formaldehyde test atmosphere concentration (1 000 ppm / 1 227 mg/m^3) for skin exposure. Final exposure concentrations were selected to represent where possible physiologically relevant and within the context of HAZMAT first responder guidance values (i.e. lowest lethal concentration (LC_{LO}) by inhalation) and/or the highest achievable concentration using the dynamic atmosphere generation setup. A constant flow of test atmosphere (500 ml/min) was maintained throughout exposure and all experiments were performed at 22 ± 2 ℃, 30% relative humidity, atmospheric pressure and with maximum skin hydration in order to mimic a real life HAZMAT exposure scenario and avoid any potential variance in exposure rates. Maximum skin hydration was achieved by allowing mounted skin samples to equilibrate for 30 mins prior to exposure according to OECD protocols. Further, the skin under surface maintained constant contact with physiological saline for the extent of each experiment thus maintaining hydration levels throughout.

Skin surfaces were exposed to aldehyde vapour/gas for short-term exposure times (≤ 30 min). The effect of non-protective 'common clothing' on skin permeation outcomes was investigated by placing denim (4 cm^2 cut squares; thickness 0.772 ± 0.011 mm) on top of mounted skin in the Franz diffusion cells. Further, the effects of skin ventilation with fresh air post-exposure was investigated by leaving samples open to atmosphere up to a total time of 60 min (i.e. 30 min exposure plus 30 min ventilation) prior to analysis. This mimics the scenario during a HAZMAT incident where potentially exposed individuals wait in open-air conditions to be decontaminated by emergency responders, with residual contaminant on their skin and clothing. A minimum of three replicates (n ≥ 3; from different donors) for each variable at each exposure time was performed.

Sample analysis

Quantification of skin absorption, penetration and fabric absorption for all aldehydes was achieved using high performance liquid chromatography with ultraviolet/visible detection (HPLC-UV) according to a modified HSE method (MDHS102: Aldehydes in Air). Determination of aldehyde skin penetration was achieved by HPLC analysis of the receptor fluid after exposure and the residual chemical on and in skin (absorption) was measured by placing exposed skin samples in a glass vial with 5 mL of physiological saline and sonicating followed by HPLC analysis of the filtered extract. Prior to HPLC analysis aldehyde samples were reacted for 30 minutes to produce stable 2,4-dinitrophenylhydrazone derivatives with high UV sensitivity. This was achieved via the addition of excess 2,4-dinitrophenylhydrazine derivatising agent to ensure the complete reaction of all aldehydes present in solution. HPLC-UV analysis was performed using a Perkin-Elmer solvent manager and isocratic LC pump connected to a Shimadzu SPD-20A UV-Vis absorbance detector controlled by Perkin Elmer

TotalChrom (v6.2.0.0.1) software. Samples were manually injected (20 µl) and separation was achieved on a Phenomonex Kinetex® C18 column (150 x 4.6 mm 5µm) using optimised chromatographic conditions for each analyte.

Data analysis

Comparisons of skin absorption and penetration outcomes between aldehydes and under different modifiers (e.g. effects of clothing, ventilation) were performed using Analysis of Variance (ANOVA) with Tukey multiple comparisons test, and Independent Samples *T*-tests. Assumption of data normality were checked and met for parametric analysis. Significance for all tests was set at $p \leq 0.05$. Statistical analyses were performed using GraphPad Prism v.7 software.

RESULTS

Overall observations

The results for skin permeation for all four aldehydes for 10, 20 and 30-minute exposures are shown in Fig 1 and Fig 2. After 30 mins exposure the two smallest aliphatic aldehydes, formaldehyde and acetaldehyde, demonstrated the greatest average skin penetration whereas the two larger aldehydes, benzaldehyde and acrolein, demonstrated the least. Skin absorption outcomes revealed that formaldehyde was absorbed to a significantly greater extent ($p = 0.006$) after 30 mins exposure than the other aldehydes, as shown in Figure 2. Post-exposure ventilation up to 60 mins appeared to have minimal effect (no significant difference, $p \geq 0.05$) on both the penetration and absorption outcomes of the aldehydes tested. The presence of denim on top of the skin was generally observed to be protective against skin penetration and absorption, in particular, significantly reducing benzaldehyde

penetration (p = 0.002) and formaldehyde absorption (p = 0.033) compared with unclothed skin. Furthermore, denim fabric was shown to have absorptive capacity particularly for acetaldehyde and formaldehyde.

Acrolein vapour exposure

Exposure of skin to acrolein vapour for up to 30 minutes resulted in a negligible amount penetrating (0.480 ± 0.417 μg/cm^2) and absorbing into (0.887 ± 0.796 μg/cm^2) the skin. Due to the low amounts of acrolein observed to penetrate and absorb into bare skin post-exposure ventilation was not explored in this study. The presence of denim fabric on top of the skin provided a protective barrier and resulted in skin penetration and absorption below the limit of detection (< 0.166 μg/cm^2) (data not shown). Despite denim reducing skin uptake, it retaining only a small amount of the acrolein vapour (0.217 ± 0.157 μg/cm^2).

Acetaldehyde vapour exposure

Skin exposure to acetaldehyde vapour resulted in the penetration of 5.29 ± 3.24 μg/cm^2 after 30 minutes, whereas absorption was shown to be much higher at 22.2 ± 10.4 μg/cm^2 after 30 minutes. The results for post-exposure ventilation and fabric experiments for acetaldehyde are shown in Fig 3A. Post-exposure ventilation up to 60 minutes (i.e. 30 minutes exposure plus 30 minutes ventilation) resulted in no appreciable change to the skin penetration or absorption of acetaldehyde for up to 30 minute exposures. The introduction of denim on to skin resulted in no appreciable change to the amount penetrating the skin across all exposure times; however resulted in reduced amounts absorbed into the skin for longer exposure times (6.28 ± 0.743 μg/cm^2 at 30 mins exposure). Denim also demonstrated absorptive capacity as it retained large quantities of acetaldehyde vapours (155 ± 16.7 μg/cm^2 at 30 mins exposure).

Benzaldehyde vapour exposure

Minimal penetration was observed at 20 and 30 minutes (1.16 ± 0.375 μg/cm^2 and 1.46 ± 0.393 μg/cm^2 respectively) for skin exposure to benzaldehyde vapour. Further, minimal skin absorption was observed across all exposure timeframes (< 0.878 ± 0.324 μg/cm^2). Results for post-exposure ventilation and fabric experiments are shown in Fig 3B. Post-exposure ventilation had a limited effect (no significant difference) on the amount absorbed into the skin or penetration through the skin across all exposure times. Denim provided a protective effect in regards to skin penetration demonstrated by the significantly (p = 0.002) reduced amount of benzaldehyde detected in the receptor fluid after all exposure times. Denim only retained 0.905 ± 0.428 μg/cm^2 after 30 minutes of vapour exposure.

Formaldehyde gas exposure

Skin exposure to formaldehyde gas for 30 minutes resulted in an appreciable amount of penetration (3.45 ± 2.58 μg/cm^2) whereas, negligible penetration was observed at shorter exposure times. High levels of skin absorption were observed for all exposure times, ranging from 294 ± 146 μg/cm^2 at 10 minutes up to 834 ± 595 μg/cm^2 at 30 minutes, significantly greater (p = 0.006) compared with other aldehydes tested.. Post-exposure ventilation did not appear to have a significant effect on the amount of formaldehyde penetrating the skin. However, the data obtained suggested that ventilation may reduce the skin absorbed amount at 20 minutes (473 ± 319 μg/cm^2) and 30 minutes (458 ± 167 μg/cm^2) exposure. Denim was shown to provide a significant protective effect (p = 0.033) and resulted in a greater than 10-fold decrease in the amount of formaldehyde penetrating the skin after 30 minutes (0.252 ± 0.0421 μg/cm^2) and a greater than 40-fold decrease in the corresponding skin absorbed

amount (18.5 ± 4.92 μg/cm^2). Denim displayed substantial absorptive capacity at both 20 and 30 minute exposures (1657 ± 477 μg/cm^2 and 1949 ± 472 μg/cm^2 respectively).

DISCUSSION

We successfully generated dynamic atmospheres of acetaldehyde, acrolein, benzaldehyde and formaldehyde vapours/gases and applied them to the surface of human epidermis *in vitro* in order to assess their dermal permeation properties for short-term exposures. All aldehyde exposures were performed under constant experimental conditions (flow rate, temperature, humidity and pressure) and with maximum skin hydration. These conditions mimic a controlled real-life exposure scenario and improve the relevance of the data for first responders. This is the first report to assess the skin permeation arising from exposure to dynamic aldehyde vapours and gases under HAZMAT relevant scenarios. The data generated can be used to assist in decision-making for skin decontamination procedures following exposure to these chemicals in HAZMAT incidents.

The overall observation of the ability for formaldehyde and acetaldehyde to penetrate human epidermis in larger quantities than benzaldehyde and acrolein appears to be explained by their relative physicochemical properties, and higher exposure concentrations. Both formaldehyde and acetaldehyde have low molecular weights and partition coefficients (log K_{ow}) allowing them to penetrate the human epidermal barrier more readily. Conversely, benzaldehyde has a much larger molecular weight and partition coefficient and whilst acrolein does have a low partition coefficient its larger molecular weight appears to restrict its ability to penetrate the human epidermal barrier in significant proportions. Further, the overall observation of skin permeation outcomes (acetaldehyde and formaldehyde > acrolein

and benzaldehyde) correlates with the increased water solubility of the smaller aldehydes as shown in Table 1. Hydrophilicity/lipophilicity is an important parameter influencing the affinity of a compound to permeate the lipid-rich stratum corneum. We maintained maximum hydration of the skin during exposure experiments and thus hydrophilic aldehydes may preferentially partition into the hydration layer and be available for uptake. Hydrophilic molecules tend to favour polar pathways for permeation suggesting that the intracellular route of penetration may be important for the aldehydes studied.

When compared with the predicted flux values from skin permeation model by Frasch (2002), the extrapolated permeation outcomes for all tested aldehydes were within similar range to predictions (predicted flux values were 0.25 μg/cm^2/hr acetaldehyde, 0.78 μg/cm^2/hr acrolein, 0.85 μg/cm^2/hr benzaldehyde and 1.5 μg/cm^2/hr formaldehyde) [33]. The Frasch mathematical model is used to predict the permeability of chemicals in aqueous vehicles through skin. The experimental data obtained for the aldehydes indicates the robust nature of this model via its ability to predict permeation outcomes for gases and vapours with relative accuracy. Given the short experimental exposure durations investigated in the study, we were unable to calculate a permeability coefficient and steady state rate of transport of aldehydes across the tissue.

It is previously reported that benzaldehyde can be absorbed through human skin [15]. However, our results suggest that benzaldehyde vapour has minimal capacity to either penetrate through or absorb into the epidermal layer of human skin at relatively high atmospheric exposure concentrations (167 ppm) for short-term dynamic exposures (≤ 30 min). Barry *et al* performed *in vitro* studies into benzaldehyde skin absorption using human abdominal cadaver skin (~0.4 mm thick) and reported a permeation flux of 410 (±70) μg/cm^2/h for saturated

vapour resulting from a pure liquid (9 hour exposure time) [26, 34]. The diffusion cells used in their study were a closed-system design containing a donor chamber with liquid reservoir to form a static vapour atmosphere which was reportedly quantified by headspace-GC; however, no actual benzaldehyde vapour exposure concentrations were reported. This is different from the dynamic atmosphere delivery of gas across the skin adopted in the current study, which would mimic typical exposure scenarios in open air environments. A more forensic comparison between the relatively large flux reported by Barry *et al* and the minimal total penetration observed in this study for short-term exposure times is difficult given the unknown exposure concentration in Barry *et al*'s study and the differing experimental setups (dynamic versus static atmosphere) and exposure times. However, the results obtained in the current study have good agreement with predicted model outcomes.

Formaldehyde gas exposure experiments showed a capacity to penetrate the skin, and a high capacity to absorb into the human epidermal layer. The latter is not surprising given formaldehyde is commonly used in solution as a biological fixative and is known to be absorbed into the skin matrix. A high degree of variability was observed in the skin absorption data given the large quantities absorbed and the inherent variability between human epidermal samples. There are no known studies on the potential for dermal uptake of formaldehyde gas exposure. Two previous *in vitro* studies have investigated dermal uptake of formaldehyde from solution. Lodén applied carbon labelled formaldehyde diluted in concentrated formalin solution onto excised human skin mounted in flow through diffusion cells. A resorption rate, equivalent to a permeation flux, was reported as 319 ± 84 μg/cm^2/h, however the presence of methanol (a known penetration-enhancing solvent) in the formalin diluting medium may have enhanced penetration outcomes [20]. Hafeez *et al* applied an aqueous solution of radiolabelled formaldehyde onto human cadaver skin (~0.4 mm thick)

mounted in static diffusion cells to investigate the effects of occlusion on skin absorption and penetration. They stated that absorption and penetration was observed after 1 hr however reported no data under occluded or non-occluded conditions [21]. The results obtained in this study add to the existing body of work by confirming the ability of formaldehyde (in gaseous state) to penetrate human epidermal skin after 30 mins of exposure at high concentrations (1 000 ppm). Gases/vapours are known to have less permeation potential than their liquid counterparts [22], and the outcomes of this study (reduced permeation compared with reported liquid exposures) aligns with that knowledge.

Acetaldehyde vapour was shown to penetrate and absorb into human epidermis after 30 mins at relatively high atmospheric exposure concentrations (996 ppm) demonstrating that it does have a potential dermal uptake pathway. There is a distinct lack of data in the literature surrounding dermal exposure to acetaldehyde vapours. The only previous reports are patch testing studies, primarily investigating alcohol skin sensitivity, to evaluate skin allergy and irritation responses to liquid acetaldehyde [23, 24]. These studies found cross-sensitisation to acetaldehyde in subjects with a demonstrated allergy to alcohols. Two other studies also included patch testing for acetaldehyde as part of a suite of testing and reported mixed results with skin sensitisation observed in only some human subjects [25, 26]. Therefore, this study represents the first empirical data on permeation of acetaldehyde vapour through human epidermis.

Penetration of acrolein vapour through skin and absorption into human epidermis was shown to be minimal after 30 minutes of exposure. Previous studies regarding acrolein dermal exposure have primarily been conducted using animals [17]. The data from the current study suggests that acrolein vapour is unlikely to have a significant dermal pathway at exposure

levels equivalent to atmospheric concentrations that are lethal via inhalation. The only previous human study of dermal acrolein exposure employed patch testing and observed low to severe irritation of the skin resulting from exposure to concentrations ranging from 1 to 10% in solution [18].

The experimental process of post-exposure ventilation of skin resulted in limited influence on skin permeation outcomes for the tested aldehydes. Certainly, no evidence of increased penetration or absorption was noted, and in fact for benzaldehyde decreased penetration was demonstrated for ventilated skin these findings are important for the first responder community in the context of an operational response to HAZMAT incidents. This effect has been noted for other toxic industrial gases (e.g. fumigants) in the context of hazardous material incidents, and has resulted in advice for emergency responders regarding decontamination protocols [1].

Aldehyde gas/vapours demonstrated an interaction with fabric, and a subsequent influence on skin permeation outcomes. Formaldehyde and acetaldehyde strongly adsorbed onto the fabric, whereas less sink properties were noted for benzaldehyde or acrolein. In terms of permeation, denim was largely a protective barrier reducing penetration and absorption outcomes compared with bare skin in most cases (in some instances by up to a factor of 40 (e.g. formaldehyde)). This observation of 'common clothing' fabrics acting as an initial protective barrier to reduce skin permeation outcomes for short term exposures is consistent with previous reports investigating the effect of fabrics in HAZMAT exposure scenarios involving gases and vapours (e.g. fumigants) [3]. The effect of clothing on skin permeation outcomes is linked to the individual chemical affinity with the fabric fibres as well as the physical parameters (thickness, fabric cover and yarn twist) of the fabric. Every-day 'common

clothing' has the potential to act as a barrier to chemical exposure (protecting individuals), as a reservoir (trapping or holding chemicals and facilitating uptake), or as an occlusive barrier to enhance absorption. The impact of clothing on dermal permeation of chemicals (including vapours) has been extensively studied in relation to chemical protective clothing [35-37]. Few studies have been undertaken on the effectiveness of every-day clothing except in relation to spraying of pesticides and, in one case, exposure to sulfur mustard. In a study by Protano *et al* (2009) the impact of different clothing types on reducing skin exposure to a range of pesticides commonly applied using a sprayer was verified. The study showed that cotton clothing had a protection factor of greater than 84% whereas chemical protecting clothing (Tyvek suits) had a performance greater than 97% [38]. Dickson (2008) reviewed two studies on the impact of mustard on the skin undertaken in the mid 1940's (wearing normal military clothing). The chamber studies concluded that normal clothing may give skin protection factors of 1.5 to 2 [39].

The empirical data produced in this study can be translated to assist in decision-making for skin decontamination by HAZMAT first responders. Our results indicate that, under the conditions tested, uptake by intact skin is likely to be an important concurrent exposure route to inhalation for both acetaldehyde and formaldehyde, for high atmospheric exposure concentrations. The data obtained for each aldehyde can be used to estimate the potential dermal uptake arising from total body exposure (18 500 cm^2) for 30 minutes at the tested atmospheric concentrations. The corresponding inhalational uptake arising from exposure to the same concentration can also be estimated by assuming a standard respiration rate of 0.625 m^3 in 30 mins (equivalent to 10 m^3 in 8 hours) and a worst case inhalational absorption rate of 100%. Comparison of these calculated values for each of the aldehydes indicates that dermal exposure would account for between 4% and 8% of the total body uptake in a 30

minute timeframe. In the case of formaldehyde, which exhibited one of the highest skin penetration outcomes, the calculated whole body dermal uptake would be 64 mg compared to an inhalational uptake of 766 mg under the same exposure conditions. Formaldehyde is formed endogenously in the body with a reported concentration in the blood of rats, monkeys and humans of 0.1 mM [40-41]. However, its electrophilic nature makes it reactive towards a variety of endogenous molecules with a half-life of 1 – 1.5 mins and an estimated turnover of 0.61 - 0.91 mg/kg bw per minute [42]. Using our data, the whole body dermal uptake rate of formaldehyde would be roughly 0.5 mg/min, which is well below the background formaldehyde turnover for a 70kg person.

In practice, those exposed individuals who are asymptomatic on scene may have been exposed to lower concentrations. Notwithstanding, secondary uptake through the skin via contact with clothing may be an important exposure parameter to consider on-scene for decontamination. Based on this, it would be recommended that for short-term (10-30 min) exposures to high concentration gas/vapour decontamination of the skin in line with agency policy should be considered for both of these chemicals. Furthermore, since denim demonstrated significant absorptive capacity for both acetaldehyde and formaldehyde, a recommendation to remove and bag bulky outer clothing to reduce the potential for secondary inhalational exposure resulting from off-gassing is also warranted. Data for both acrolein and benzaldehyde indicated that dermal uptake is unlikely to be a significant pathway under the conditions tested, however given the inherent toxicity of acrolein skin decontamination may still be appropriate.

We have presented the first empirical data on the skin permeation potential of four common industrial aldehyde gases/vapours using HAZMAT relevant exposure scenarios. Acrolein and

benzaldehyde demonstrated little capacity to penetrate human epidermis whereas acetaldehyde and formaldehyde displayed high potential to penetrate skin at high atmospheric concentrations (1 000 ppm) and for short exposure times (≤ 30 mins). Further research is needed to quantify the potential contribution towards total body burden that results from skin permeation. Despite commonality between the tested chemicals their individual skin permeation properties were different. Permeation results were, however, in alignment with predicted outcomes based on models using physicochemical properties and other parameters. This suggests that grouping 'like' chemicals for common advice to emergency responders regarding potential for skin uptake may not necessarily be appropriate. Rather, prediction of skin permeation potential of chemicals on an individual basis using important physicochemical properties (e.g. MW, K_{ow}, solubility), in the absence of empirical permeation data, may be more accurate and caution should be taken when determining the need for skin decontamination after dermal exposure resulting from a HAZMAT toxic release incident.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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Table 1: Structural and physicochemical properties of the aldehydes investigated in this study (source: chemspider database)

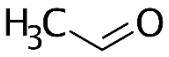
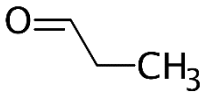
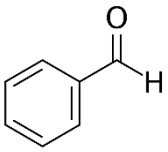
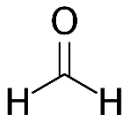
Property	Acetaldehyde	Acrolein	Benzaldehyde	Formaldehyde
Structure				
CAS number	75-07-0	107-02-8	100-52-7	50-00-0
Molecular Weight ($g.mol^{-1}$)	45.05	56.07	106.13	30.03
Boiling point (°C)	21	52.5	179	-19.5
Vapour Pressure (mm Hg at 25 °C)	902	274	0.127	3 890
Partition Coefficient (Log K_{ow})	-0.34	-0.01	1.48	0.35
Solubility (g/L at 25°C)	1 000	212	6.95	400
IDLH (ppm)	2 000	2	NA	20
LC_{LO} (ppm)	NA	153	NA	NA
OSHA PEL (ppm)	200	0.1	NA	0.75

Figure 1: Penetration profiles of acrolein (153 ppm), acetaldehyde (996 ppm), benzaldehyde (167 ppm) and formaldehyde (1 000 ppm) vapour/gas through human abdominal skin. Values are mean ± SD, n ≥ 3. LOD 0.166 μg/cm^2. ND, not determined.

Figure 2: Absorption profiles of acrolein (153 ppm), acetaldehyde (996 ppm), benzaldehyde (167 ppm) and formaldehyde (1 000 ppm) vapour/gas into human abdominal skin. Values are mean ± SD, n ≥ 3. LOD 0.166 μg/cm^2. ND, not determined.

Figure 3: The influence of denim and post-exposure ventilation on skin penetration (left) and absorption (right) of (**a**) acetaldehyde vapour (996 ppm), (**b**) benzaldehyde vapour (167 ppm) and (**c**) formaldehyde gas (1 000 ppm). Values are mean ± SD (n ≥ 3). ND, not determined.