#### ADVANCED REVIEW







# Cadaver-detection dogs: A review of their capabilities and the volatile organic compound profile of their associated training aids

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#### Abstract

Cadaver-detection dogs (CDDs) are an essential tool for the search and detection of human remains. In order to enhance their search capability, CDDs are regularly trained on natural and synthetic training aids. The odor profile of these training aids comprises a range of volatile organic compounds (VOCs) which is intended to resemble those produced by a decomposing body. It is currently unknown if detector dogs respond to the same stimuli and whether it is a specific VOC or a suite of decomposition-related VOCs as their target odor. This review summarizes the VOCs that have been detected in various CDD training aids such as blood, human remains, decomposition fluid, soil, buried remains, textile, and synthetic formulations. Additionally, it discusses the reported capability of CDDs to respond to each of these training aids. The purpose of this review is to understand the variability of VOCs in CDD training aids and the response of CDDs to this wide range of compounds. Additionally, this review attempts to determine if there is a specific training aid to which CDDs respond preferentially. Such a review will assist to establish better practices for CDD training since no standardized practices exist globally.

This article is categorized under:

Crime Scene Investigation > Special Situations and Investigations Forensic Anthropology > Taphonomic Changes and the Environment Forensic Medicine > Death Scene Investigation

#### KEYWORDS

human remains detection dogs, decomposition, volatile organic, compounds, canine olfaction, search and rescue

# **1** | INTRODUCTION

The use of dogs (*Canis familiaris*) in forensic case work dates to 1888 when bloodhounds were used in the infamous case of Jack the Ripper in Britain (Blum, 2017). Since then, dogs have aided forensic investigation and law enforcement through the detection of live humans, cadavers, other biological specimens (such as blood and semen), explosives, illicit drugs, ignitable liquids, currency, and other contraband (Ensminger & Papet, 2012). The sense of smell in both humans and dogs is achieved via receptor cells and olfactory nerves that facilitate location of target odor. However, the

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difference lies in the fact that dogs have a superior olfactory capability which is reportedly 10,000–100,000 times better than that of humans (Sankaran, Khot, & Panigrahi, 2012; Walker et al., 2003; Walker et al., 2006). Additionally, their short voluminous sniffing and unique nasal airflow pattern allows for greater amounts of odorants to enter dogs' nostrils and generate an enhanced response by triggering the large olfactory bulbs in their brain (Craven, Paterson, & Settles, 2010; Settles, 2005). These odorants comprise of volatile chemical compounds emitted by the odor source.

In recent years, research in forensic volatile organic compound (VOC) analysis has been conducted to identify volatile and semi-volatile compounds that are key in odor detection. Identification of significant compounds could eventually facilitate the construction of a portable field air analysis device that can provide more objective results compared to dogs. However, even with the current advancement in instrumentation, dogs are still considered the most rapid and efficient tool for odor detection in the police community. Their speed, versatility, ruggedness, and discriminating power makes them far superior to portable detection apparatus (Furton, Caraballo, Cerreta, & Holness, 2015; Rust, Nizio, Wand, & Forbes, 2018; Sankaran et al., 2012). The general scent detection capability of dogs can be enhanced to create a desired visual response (barking, sitting down, etc.) in the presence of a target odor such as the "smell of death" in the case of cadaver-detection dogs (CDDs). Therefore, it is essential to appropriately train dogs to illicit a desired response to a target. Several organizations such as The Organisation of Scientific Area Committees for Forensic Science (OSAC) Dogs & Sensors group are focusing on establishing standards and guidelines related to the performance of doghandler teams. Such guidelines have become necessary as a consequence of dog response becoming a questionable evidence in court (Page, 2008; Schoon, 1996, 2005; Schoon, 1998; Schoon & De Bruin, 1994; Taslitz, 1990).

Among the many purposes for which organizations would deploy detector dogs, CDDs are used to assist in the recovery of bodies or body parts. Alternative terminologies including human remains detection (HRD) dogs, victim recovery dogs (VRD), and decomposition dogs are sometimes used interchangeably with CDD. CDDs are typically trained on a vast variety of decomposition odor sources. Depending on availability, these could include human remains, blood, clothing from a decomposed victim, soil, and fluids associated with a decomposing body, gauze placed in contact with decomposing remains, as well as synthetic formulations (referred to as pseudo-scents). The aim of training on these target odors is to reinforce and enhance the dogs' memory to respond to the odorous compounds generated from the training aids (Rebmann & David, 2000).

The process of decomposition results in the evolution of gaseous chemical compounds known as VOCs, which evolve as a result of cellular degradation, microbial activity, breakdown of complex molecules, and insects and larval activity. The decomposition VOC profile is extremely complex with as many as 832 VOCs reported from a pig carcass in a terrestrial ecosystem environment (Dekeirsschieter, Stefanuto, Brasseur, Haubruge, & Focant, 2012). A range of compound classes such as alcohols, ketones, aldehydes, esters, ethers, aromatic hydrocarbons, aliphatic hydrocarbons, halogenated compounds, nitrogen- and sulfur-containing compounds together comprise decomposition odor. Some decomposition-related VOCs are odorants and act as chemical stimuli for CDDs. It is unknown if CDDs respond to a complex combination of VOCs or a core suite of VOCs that are present in the "smell of death" (Stadler et al., 2012). This makes standardization of a CDD training aid difficult. Hence, the most acceptable practice is to expose the dogs to a broad spectrum of decomposition training aids.

This review focuses on summarizing the VOCs reported for the various materials (natural and synthetic) used for CDD training and the dogs' reported capability and performance when training on these range of materials. It aims to bridge the knowledge gap between understanding the VOC profiles produced by a variety of training aids and interpreting the dogs' response to the target compounds. Studies that focus on estimating the dogs' response to, and VOCs released from, the same sample are most effective in understanding decomposition-related compounds potentially detected by the dogs. At this stage, it is difficult to conclude which compounds activate the dogs' olfactory receptors, however, a review of the available literature will summarize the major decomposition-related compounds identified from a range of training aids used for CDDs. This review will also highlight the success rates reported for CDDs in responding to decomposition odor based on the training materials currently used worldwide.

Journal articles that reported VOCs in materials used as CDD training aids (e.g., blood, human remains, soil, decomposition fluid, textile, and chemical formulations) have been included in this review. Animal remains (generally pig remains) are a popular training aid among dog trainers in some countries, mostly due to the ethical and legal restrictions in acquiring and possessing human remains training aids. However, the chemical VOC profile of animal remains is reported to be distinct from human remains (Cablk, Szelagowski, & Sagebiel, 2012; Knobel, Ueland, Nizio, Patel, & Forbes, 2019). For the purpose of this review, only those animal remains studies, which compared and differentiated the VOC profile with human remains have been included. This review has been divided into five sections based on the training aid category: (1) blood; (2) human remains; (3) soil, buried remains, and decomposition fluid; (4) textiles

containing decomposition odor; and (5) synthetic (i.e., chemical) formulations. Each section is further subdivided into two sections discussing the VOCs reported in the literature and detector dogs' reported performances on the specific category of training aid.

# 2 | BLOOD AS A TRAINING AID

Human blood is regularly used as a training aid for blood-detection dogs (BDDs) and CDDs. BDDs are specialized dogs that are trained to respond only to the odor of blood. Fresh blood is an easily available training source to dog handlers as there are fewer legal or ethical issues related to obtaining blood compared to other human tissue samples. It has been reported that blood contains decomposition by-products and therefore, it serves as a suitable training aid for CDDs (Hoffman, Curran, Dulgerian, Stockham, & Eckenrode, 2009).

# 2.1 | VOCs reported in human blood

VOCs in blood result mainly from degradation of proteins, which produce amino acids such as tryptophan. Microbial break down of tryptophan forms volatiles such as indole and skatole (Dent, Forbes, & Stuart, 2004). While indole is a commonly reported VOC, studies have failed to report skatole in blood (Hoffman et al., 2009). Blood at crime scenes can be found at varying stages of degradation and deposited on multiple surface types. This has led to recent studies focusing on understanding changes in the VOC profile as blood ages on various surface types. Some studies have attempted to mimic the blood storage conditions used by law enforcement officials when training BDDs and CDDs. Such studies have contributed to our understanding of the blood VOC spectrum.

In a study by Chilcote, Rust, Nizio, and Forbes (2018), weathering of blood in an outdoor environment collected from a single living donor, was studied for 84 days on a nonporous varnished wooden surface and a porous concrete surface. The weathered blood samples were analyzed for the presence of VOCs and used as part of a standard BDD and CDD training scenario to determine the response of the dogs (Chilcote et al., 2018). Within the first 24 hr, even though the blood was visible on the concrete surface, most blood related VOCs were lost while, on the varnished wooden surface, the number of VOCs varied between each sampling day which was attributed to interaction of the blood with the varnish on the wood, as well as the high temperature and heavy rainfall to which the surface was exposed. A total of 13 compounds for the porous and 10 compounds for the nonporous surface (indicated in Table 1) were deemed statistically significant in differentiating blood from control samples, with one common compound (heptanal) on both surface types. These results highlight the fact that even with the same sample, interaction of the material with the surface can yield variable VOCs.

Variability in VOC due to surface interaction was also reported by Rust et al. (2016) where fresh and degraded blood samples on porous cotton material and nonporous aluminium material were analyzed over 12 months. For both surface types, hydrocarbons and esters were prominent in fresh blood while nitrogen- and sulfur-containing compounds were prominent in degraded blood. The presence of aromatics, hydrocarbons and alcohols dominated the porous samples' profiles while, the nonporous surface profiles were dominated by aromatics and aldehydes. On the porous surface, alcohols were dominant potentially due to bacterial degradation of the cellulose in cotton (Rust et al., 2016). This study indicated that the VOC profile of fresh blood (Day 0–1) was distinctly different from blood degraded longer than one week. Thus, the Chilcote et al. (2018) and Rust et al. (2016) studies established that the blood VOCs vary based on the surface type and over time with the weathering process.

A further study by Rust et al. (2018) investigated the detection limits of BDD and CDD to latent blood evidence on cotton clothing washed up to five times, and compared the dogs' responses to these materials with current presumptive luminol and analytical chromatography techniques. Washing of blood evidence by the offender is a common practice thus, studies such as this one adds value in understanding real scenarios. Results indicated that the luminol test was able to presumptively detect blood washed up to five times while the VOC analysis using advanced chromatography showed difficulty distinguishing between the washed blood VOC profile and the washed negative control VOC profile after the first two washes. The authors proposed that the washing procedure removed blood related VOCs while adding additional VOCs from the laundry powder, becoming trapped within the cotton fibers and resulting in more complex chromatograms.

### **TABLE 1** Summary of Volatile organic compounds detected in dog training aids.

			RI	ood			Human	Remains and	d Decomnosit	ion fluid	Soil and Buried remains		Textile	Synthetic	
			ы				Human Remains and Decomposition fluid				son and bu		. caule	Formulations**	
	(Rendine et al., 2019)	(Chilcote, Rust, Nizio, & Forbes, 2018)	(Rust, Nizio, Wand, & Forbes, 2018)	(Rust et al., 2016)	(Forbes et al., 2014)	(Hoffman, Curran, Dulgerian, Stockham, & Eckenrode, 2009)	(Rosier et al., 2016)	(Rosier et al., 2015)	(Hoffman, Curran, Dulgerian, Stockham, & Eckenrode, 2009)	(Buis et al., 2015)	(Dubois, Stefanuto, Heudt, Focant, & Perrault, 2018)	(Vass, 2012)	(DeGreeff & Furton, 2011)	(Tipple et al., 2014)	(Stadler et al., 2012)
Compounds															
Acids and esters															
Acetic acid													х		
Methyl acetate			~							х					
Ethyl acetate Isobornyl acetate			X X										х		
2-(2-butoxyethoxy)ethyl acetate			x										~		
4-tert-Butylcyclohexyl acetate			х												
Propanoic acid	х							х	х	х			v		
2-Methylpropanoic acid Ethyl propionate							x	x					x		
Propyl propionate							x	X							
2-Methyl-1-(1,1-dimethylethyl)-2-methyl-														x	
1,3-propanediyl propionate Butanoic acid	x					x	-		x	x					
2-Methylbutanoic acid	^					~			^	x					
3-Methylbutanoic acid										X					
Methyl butanoate					x						x		х		
Ethyl 3-methylbutanoate	1			-			x								
Ethyl butanoate							X		x	X					
Propyl butanoate	1						x	x		X					
Butyl butanoate								X X	X X	X X					
Pentanoic acid	1						x	x							
Ethyl pentanoate Hexanoic acid	х						^	^	x	x			x		
2-Ethylhexanoic acid													x		
Ethyl hexanoate	х								Х						
Pentyl hexanoate									х						
Hentyl hexanoate									х	Х					
Octanoic acid									x	x			x		
Undecanoic acid Methyl decanoate			x						^	X					
Methyl 10-methylundecanoate													х		
Methyl dodecanoate			x												
Methyl tridecanoate			X							х					
Methyl tetradecanoate Methyl 14-methylpentadecanoate			x												
Methyl hexadecanoate			~							х		x			
2-Methylpropyl benzoate		х													
Methyl 2-hydroxybenzoate			х										х		
Homomenthyl salicylate			X X												
Hexyl 2-hydroxybenzoate 2-Ethylhexyl tetradecyl oxalate			^										х		
Diethyl phthalate												х			
[2,2,4-Trimethyl-3-(2-															
methylpropanoyloxy)pentyl] 2-			х												
methylpropanoate			x												
Methyl dihydrojasmonate			~												
Aldehydes	1														
Acetaldehyde	х									х		x			
Butanal										X		X			
2-Methyl butanal 3-Methyl butanal	1								-	X X		X X			
Pentanal	1									X		x			
2-Hexenal									x	х					
Hexanal					X				X	X		X		X	
Heptanal	х	x			X				X X	X		x		x	
2-Heptenal 2,4-Heptadienal	1	^		-					X	x					
Octanal	х				x				x	X		x		x	
2-Octenal									Х	Х					
Nonanal	х	х							X	х			х		
2-Nonenal	1								x	v					
2,4-Nonadienal Decanal	1				x				X	X		x	x		
Benzaldehyde	х								x	x	x		x		
Furfuraldehyde													х		
Halogen compounds															
Chloroform Carbon tetrachloride															
Dichlorodifluoromethane	1											x		1	
Dichlorotetrafluoromethane												х			
Trichloromonofluoromethane												х			
Tricholoroethane												~			
Trichloroethylene Tetrachloroethylene	x					x						X X	x		
1,2-Dichloroethene	1					^						x	^		
1,1,2,-Trichloro-1,2,2-trifluoroethane												х			
1,1-Dichloro-1-fluoroethane												х			
1-Chloro-2-methoxy-benzene	1										Х				



### **TABLE 1** (Continued)

			Bl	ood			Human	Remains and	l Decomposit	ion fluid	Soil and Buried remains		Textile	Synthetic Formulations**	
	(Rendine et al., 2019)	(Chilcote, Rust, Nizio, & Forbes, 2018)	(Rust, Nizio, Wand, & Forbes, 2018)	(Rust et al., 2016)	(Forbes et al., 2014)	(Hoffman, Curran, Dulgerian, Stockham, & Eckenrode, 2009)	(Rosier et al., 2016)	(Rosier et al., 2015)	(Hoffman, Curran, Dulgerian, Stockham, & Eckenrode, 2009)	(Buis et al., 2015)	(Dubois, Stefanuto, Heudt, Focant, & Perrault, 2018)	(Vass, 2012)	(DeGreeff & Furton, 2011)	(Tipple et al., 2014)	(Stadler et al., 2012)
Alcohols Ethanol	x							x		x					
2-(2-Methoxyethoxy)ethanol	~												х		
2-(2-Ethoxyethoxy)ethanol			х										х		
2-(Hexyloxy)ethanol				Х											
2-Butoxyethanol													х	х	
1-Propanol	х											X			
2-Methyl-1-propanol			x				x	x		x		х			
1-(1-Methoxypropan-2-yloxy)propan-2-ol 1-Butanol			^				x	x		x				x	
2-Butanol							x	~		~				~	
2-Methyl-1-butanol							х	х							
3-Methyl-1-butanol							х	х		Х					
1-Pentanol					х				х	х				х	
2-Methyl-1-pentanol										х	×				
2-Methyl-3-pentanol 2,2,4-trimethyl-3-penten-1-ol		х									x				
1-Hexanol		^			х	х			x	х					
3-Hexanol				1	~		l			~	1			х	
2-Ethylhexanol					х	x		х	х	x			х	х	
3-Heptanol														х	
3-Heptenol		х													
3-Methyl-3-heptanol											l			X	
3-Ethyl-3-heptanol				-										х	
1-Butoxy-2-propanol 1-Octanol	-		x			v			v	v			~		
1-Octanol 3-Octanol						X			x	X X			x	x	
2-Octen-1-ol					х					^	1			<u>^</u>	
1-Octen-3-ol	х				X	х			x	х	1				
3-Ethyl-1-octanol														х	
2-Hexyl-1-octanol													х		
3,7-dimethyl-3-octanol			х												
2,6-dimethyl-7-octan-2-ol			х										х		
3,7-dimethyl-1,6-octadien-3-ol (Linalool)			x												
Furfuryl alcohol													x		
2-Pentadecyn-1-ol													X		
1-Hexyl-1-decanol 2-Methyl-1-undecanol			x										x		
1-Undecanol			x												
2-Decanol			x												
1-Phenoxy-2-propanol			х												
Benzyl alcohol										Х			х		
Phenol		х								Х			х	х	
4-Isopropenylphenol				х											
4-Ethyl-1,3-benzenediol														X	
2,4-Bis(1,1-dimethylethyl)phenol 2-Phenyl-2-propanol (Cumyl alcohol)												x		x	
5-methyl-2-propan-2-ylcyclohexan-1-ol												^			
(Menthol)			х										х		
Ketones															
Acetone										х				х	
Acetophenone			х							X					
2-Propanone 2-Butanone	x						x x			X X		x		x	
2-Pentanone	^						x			^		^		^	
3-Pentanone							~			х					
3-Methyl-2-pentanone														x	
4-Methyl-2-pentanone														х	
3-Ethyl-2-pentanone														х	
2-dimethyl-4-pentyn-3-one				х							I				
2-Hexanone 3-Hexanone					v					X				x	
3-Hexanone 5-Methyl-2-hexanone		x			x					x	1			^	
3-Methyl-2-hexanone		~								x					
4-Methyl-3-hexanone										x	х				
5-Methyl-3-hexanone										х					
2-Heptanone					х				х					х	
3-Heptanone										х				х	
4-Heptanone				-	х										
3-Methyl-2-heptanone										х	I				
4-Methyl-2-heptanone			x		x						1				
6-Methyl-2-heptanone 6-Methyl-5-hepten-2-one					X						1		x		
3-Octanone					X						1		^		
2,5-Octanedione					X						1				
1-Octen-3-one		х		1							1				
2-Nonanone		x			x					x				x	
2-Decanone														х	
2-Undecanone					х										
2-Tetradecanone			х								I				
1-[4-(1-Hydroxy-1-methylethyl)phenyl]											1		х		
ethanone	-														
1,3,3-trimethylbicyclo[2.2.1]heptan-2-one	1	x									1				
(Fenchone)											I				
6,10-dimethylundeca-5,9-dien-2-one											1		х		
(Geranyl acetone) 3-methyl-6-propan-2-ylcyclohex-2-en-1-one	1			-				-	-	-	1				
3-methyl-6-propan-2-ylcyclohex-2-en-1-one (Piperitone)	1	x									1				
3-methyl-4-(2,6,6-trimethylcyclohex-2-en-1-											1				
yl)but-3-en-2-one (Isomethyl ionone)			х								1		1		
2-Oxolanone (Butyrolactone)				х											х
1-(4-hydroxyphenyl)ethanone				х											
Cyclohexanone	х	х				х			х						
Benzophenone	1										1		1	х	



#### **TABLE 1** (Continued)

	Blood							Human Remains and Decomposition fluid				Soil and Buried remains		Synthetic Formulations**	
	(Rendine et al., 2019)	(Chilcote, Rust, Nizio, & Forbes, 2018)	(Rust, Nizio, Wand, & Forbes, 2018)	(Rust et al., 2016)	(Forbes et al., 2014)	(Hoffman, Curran, Dulgerian, Stockham, & Eckenrode, 2009)	(Rosier et al., 2016)	(Rosier et al., 2015)	(Hoffman, Curran, Dulgerian, Stockham, & Eckenrode, 2009)	(Buis et al., 2015)	(Dubois, Stefanuto, Heudt, Focant, & Perrault, 2018)	(Vass, 2012)	(DeGreeff & Furton, 2011)	(Tipple et al., 2014)	(Stadler et al., 2012)
Cyclic hydrocarbons															
Methylcyclopentane		Х													
(2-Methylpropyl)cyclohexane				x						v		v			
Benzene Toluene	x				x	x			x	X X		X X	x		
Ethylbenzene	^				X	^			^	X	х	X	^		
Propylbenzene			х		~					^	^	~			
1-Methyl-3-propyl benzene	-		x												
1-Methyl-4-propyl benzene			X												
1-Methoxypropyl benzene												х			
(1-Methyldecyl)benzene				х											
1,2-Dimethyl benzene (O-Xylene)												х			
1,4-Dimethyl benzene (P-Xylene)						х			х	х		х			
1-Ethyl-2-methyl benzene												Х			
1-Ethyl-3-methyl benzene			х												
2-Ethyl-1,3-dimethyl benzene			x												
2-Ethyl-1,4-dimethyl benzene	1		X												
4-Ethyl-1,2-dimethyl benzene	1		X X	-					-	-					
(1-Ethylnonyl)-benzene			X	x					-	-					
(1-Pentylheptyl)-benzene 1,2,4-Trimethylbenzene	1		x	*											
1,2,4- I rimethylbenzene 1,2,3-Trimethylbenzene	1		X		x										
1,2,3-i rimetnyibenzene 1,2,3,4-Tetramethylbezene	1		X	-	^	-			-	-			х		-
1,2,3,4-i etrametnylbezene 1,3-Bis(1,1-dimethylethyl)-benzene	1	1	^	-					-				- ^	х	
1,3-Bis(1,1-dimethylethyl)-benzene Naphthalene	1	1		-					-	x		x	х	^	
1-Methyl naphthalene	1									X		~			
Styrene					x					X		х	х		
Cumene			х		X										
1,4-Dioxane														х	
Alpha-pinene			х							х					
Terpinolene											х				
2-Methyl furan										х		х			
2,4-Dimethyl furan														х	
2-Butyl furan										Х					
2-Pentyl furan	х	х			х				х	Х					
Butylcyclohexane			Х												
Propylcyclohexane			Х												
5-Phenylundecane			Х												
Non Cyclic Hydrocarbons															
2-Methylbutane												х			
3-Methylbutane								X				N/			
Pentane			x					x		X		x			
3-Methylpentane												x			
2-Methylpentane 2,4,4-Trimethylpentane		х										^			
4-Methyl-2-pentene	-	~								x					
1,3-Pentadiene										x					
Hexane	х									X		х			
1-Hexene	1									x		~			
3-Ethylhexane	1		х												
Heptane	1				х					X		x		х	
Octane		х			x					X		x		x	
2-Octene					х					х					
3-Octene										Х					
4-Ethyloctane			х												
2,5-Dimethyloctane	1						х								
2,6-Dimethyloctane			X												
2,4,6-Trimethyloctane				X											
Nonane			X							X		X			
2-Methylnonane			X	-			х		-						
Decane	×		x	-	X			X X	-	X		X	x		
Undecane			x	-	x			X	-	X X		x	x		
Tridecane Tetradecane	1	-	X		^					X			x	x	
Pentadecane	1		^	-		-			-	^			X	^	-
Hexadecane	1		x	-		-			-	x			X		-
Hexadecane Heptadecane	1		X	X		-			-	X			X		-
3-Ethyl-2-methyl-heptane	1		^	^						^	х		Â		
2-Methyl-3-heptene	1			x											
4-Methyldecane	1		х												
Dodecane	1	1	X	1	х				1	X					
2,6,10,14-Teramethylheptadecane	1		x							1					
1-Propene	1	1		1					1	1					
2-Methylpropene	1											х			
2-Pentene	1											X			
3,4,4-Trimethyl-2-pentene	1			X											
2,6-Dimethyl-2,6-octadiene	1	х													
3-Decene	1		х				1								
Dodecene	1				х										
Nonadecane	1				х										

Two independent studies by Rendine et al. (2019) and Forbes, Rust, Trebilcock, Perrault, and McGrath (2014) analyzed VOC profiles of blood stored in vacutainers which may not ideally replicate a scenario of blood found at a crime scene. However, it is a common practice among dog handlers to expose dogs to blood stored in vials or small containers



#### **TABLE 1** (Continued)

Γ			BI	ood			Human	Remains and	d Decomposit	ion fluid	Soil and Buried remains		Textile	Synthetic Formulations**	
	(Rendine et al., 2019)	(Chilcote, Rust, Nizio, & Forbes, 2018)	(Rust, Nizio, Wand, & Forbes, 2018)	(Rust et al., 2016)	(Forbes et al., 2014)	(Hoffman, Curran, Dulgerian, Stockham, & Eckenrode, 2009)	(Rosier et al., 2016)	(Rosier et al., 2015)	(Hoffman, Curran, Dulgerian, Stockham, & Eckenrode, 2009)	(Buis et al., 2015)	(Dubois, Stefanuto, Heudt, Focant, & Perrault, 2018)	(Vass, 2012)	(DeGreeff & Furton, 2011)	(Tipple et al., 2014)	(Stadler et al., 2012)
Sulphur compounds															
Carbon disulphide	х							Х		Х		Х			
Carbon oxide sulphide												Х			
Sulphur dioxide										Х		Х			
Methanethiol								Х							
3-Methylthio-1-propanol								Х							
Dimethyl sulphide												Х			
Dimethyl disulphide	х					х	х		X	х		х			
Dimethyl trisulphide										х		Х	х		
Diethyl disulphide							х	х							
Methyl(methylthio)ethyl disulphide							x	x							
Methylisobutyl disulphide								X							
Methylhexyl disulphide								X							
Methylethyl disulphide								x		х					
Methylisopropyl disulphide								x		~					
Methyl allyl disulphide								~			x				
1-Methylthioheptane								x			^				
Methylthiopentane								^							
Benzothiazole										x				x	
Senzormazore															
Nitrogen compounds															
Methanamine										х		Х			
Tridecylamine															х
1,4-Diaminobutane (Putrescine)															х
1,5-Diaminopentane (Cadaverine)															х
2,5-Dimethyl-1-pyrrole							х								
Pyridine							x	х		х					
2,6-Dimethylpyridine							X								
2,4,6-Trimethylpyridine														х	
2,5-Dimethylpyrazine								x						~	
2,3,5-Trimethylpyrazine								x							
2,3,5,6-Tetramethylpyrazine								x							
2,5,5,6-retrainethypyrazine 2-Pyrrolidone								^							x
Aniline		х													~
Benzonitrile		^											х		
Methoxy phenyl oxime		х											x	x	
Nitroso methane		^									x		^	^	
Nitroso metnane N.NDibutylformamide		х	×								^				
		x	*												
N-Formyl-N-methylformamide		~													x
				-		-									
4-Aminobutanoic acid (GABA)															х
4-Aminobutanoic acid (GABA) 1,8-Diazabicyclo[5.4.0]undec-7-ene (DBU) Indole	х					X			X	X					

during their training. Rendine et al. (2019) used cadaveric blood and analyzed the VOC profile up to 21 days after collection. Samples from the same days were then used for dog trials with two HRD dogs (referred to as CDD in this review) (Rendine et al., 2019). The authors concluded that the VOC profile of cadaveric fresh blood (aged up to 3 days) was significantly different from degraded blood which is consistent with the previously mentioned results from Rust et al. (2016). Three compounds (1-octanal, hexanoic acid, and ethanol) found in the Rendine et al. study have also been reported in cadaveric blood by another study (Lavigne, 2018).

A study by Forbes et al. (2014) analyzed VOCs in blood stored up to six weeks under three different storage conditions: room temperature, refrigerated, and freezer (Forbes et al., 2014). The study reported that 1-octen-3-ol, 2-heptanone, 4-heptanone, 2-ethyl-1-hexanol, and 2-pentylfuran were responsible for distinguishing degraded from fresh blood samples. Additionally, 1-octen-3-ol remained a dominant compound in all the fresh and degraded blood samples. This study also suggested that blood stored in the freezer produced a distinct VOC profile from that stored at room temperature and in a refrigerator and thus, storing the sample in a freezer was not recommended when used for training purposes.

Based on results from the above-mentioned studies, it appears that alcohols, esters, and aromatic compounds are more prominent in fresh blood samples compared to degraded samples. In contrast, nitrogen- and sulfur-containing compounds were identified only in degraded blood. Hydrocarbons, aldehydes, and ketones may be present throughout the blood degradation process. The VOCs which evolve throughout the process of blood degradation are highly variable and depend upon the surface they are in contact with as well as the surrounding environment. Blood cleaned using a detergent or soap may result in a complex VOC profile by adding external VOCs while simultaneously eliminating some blood-associated VOCs. Thus, it is advisable to use fresh and degraded blood present on various surface types as

training aids for BDDs and CDDs. A summary of VOCs detected in human blood has been listed in Table 1. 2-Pentylfuran is reported in three of the five studies (Chilcote et al., 2018; Forbes et al., 2014; Rendine et al., 2019). Several compounds found in blood such as ethanol, acetaldehyde, cyclohexanone, pentane, hexane, heptane, and octane have also been reported in decomposing human remains (Statheropoulos, Spiliopoulou, & Agapiou, 2005). Future studies should focus on comparing VOCs from cadaveric blood and from living humans to understand any possible variation in VOC profile.

### 2.2 | Dog performance using human blood

Three previously mentioned studies by Chilcote et al. (2018), Rendine et al. (2019), and Rust et al. (2018) that analyzed the VOC profiles of blood, also assessed the performance of BDD and/or CDD on the same blood samples in order to understand the correlation between the VOCs identified using chemical techniques and those that are detected by the dogs.

Chilcote et al. (2018) conducted their study with two BDD and four CDD with blood applied to porous concrete and nonporous varnished wood surfaces. The trials were performed as single-blind experiments, wherein the handler was unaware of the location of the target odors, but the trainer who held the reward knew the location of the target odors. There was no specific trend observed in the dog response accuracy and degradation of blood for any of the surface types. The limit of detection for canines in this scenario was 1 month for blood on concrete and one week for blood on varnished wood samples. From this study it was evident that regular training and exposure to blood on different surface types is required to enhance BDD/CDD capabilities.

In the study by Rust et al. (2018), dogs were exposed to cotton swatches with blood washed up to five times. Even though some dogs were able to detect blood after the fifth wash, overall, the percentage of correct alerts decreased with increasing number of washes. The difficulty in detecting washed blood increased after the second wash for several dogs. Results from the VOC analysis and dog trials during this study suggests that the washing procedure increases the difficulty in blood detection. Thus, when relevant, dogs should be exposed to detergent washed and/or bleach cleaned blood since VOCs from external agents (such as detergent, bleach) can interfere with blood VOC profile, making its detection challenging.

Rendine et al. (2019) assessed the performance of two HRDs in detecting human cadaveric blood in vials at different stages of decomposition on Days 0, 3, 7, 14, and 21. Interpretation of the results in this study was based on the calculation of the positive predictive value (PPV) and negative predictive value (NPV). PPV is the proportion of the dog's responses to the presence of the target odor that are correct while, NPV is the proportion of the dog's responses to the absence of the target odor that are correct. The PPV in this study ranged between 98.96 and 100% for one dog, and between 99.47 and 100% for the other dog. The NPV ranged between 75 and 100% for the first dog, and between 88.89 and 100% for the second dog. The dog response improved with the more degraded blood samples, and the best response was recorded after 14 and 21 days of degradation of the blood samples. A possible explanation as suggested by the authors was activation of the sulfur-associated neurotransmission pathway in dogs. Sulfur was present in the form of dimethyl disulfide and carbon disulfide which was detected in the VOC profile on Days 14 and 21. Additionally, an increase in the response trend from Day 14 to Day 21 was attributed to a potential quantitative difference in the sulfur compounds which produced an enhanced dog performance. In another study involving two dogs exposed to cadaveric blood in tubes at dilutions up to the ratio of 1:1000,000, statistical results showed a PPV between 89.91 and 100% and NPV between 50 and 100% for various dilutions. This indicated that the dogs were able to detect highly diluted blood samples (Riezzo et al., 2014).

An earlier study by Schoon (2013) reported dogs' accuracy in identifying blood when compared to commonly used presumptive blood tests including luminol, tetrabase, and Kastle–Meyer. Fresh blood collected from a living male was diluted up to 4,000 times with tap water and sprayed on 20 cm<sup>2</sup> vinyl and carpet squares (Schoon, 2013). The dogs had difficulty indicating on blood at dilutions above 2,000 times on smooth nonporous vinyl surfaces compared to porous carpet surfaces. Similar results were observed from the previously mentioned study by Chilcote et al. (2018) who found that the dogs were able to detect blood on porous material longer than that on nonporous material.

Studies to date suggest that a well-trained dog with ongoing exposure to fresh and degraded blood samples will outperform a VOC analysis instrument such as gas chromatography-mass spectrometry (GC-MS) or GCxGC-TOFMS. However, the blood VOC profile is easily altered via surface interactions and procedure such as washing. This could eventually impact the odorous compounds to which the dogs are responding. Thus, it is recommended that, BDDs and CDDs should be trained on blood degraded at different stages and applied on multiple surface types to increase the likelihood of their success in locating latent blood during operational deployment. Exposing dogs to washed and diluted blood could also assist to increase their sensitivity to blood evidence.

# **3** | HUMAN REMAINS AND DECOMPOSITION FLUID AS TRAINING AIDS

In reality, CDDs can be involved in the search of a wide variety of remains including fresh, aged, and dried remains. The nature of human remains could range from whole cadavers, parts of cadaver, cadaveric tissue to even cadaveric fluids. Although, in a field scenario a CDD could be expected to locate intact bodies, it is not practical to train them on whole cadavers. Some scientific evidence suggests that human remains (such as tissue and decomposition fluid) could have an odor profile similar to that of decomposing human bodies (Buis, 2016; Hoffman et al., 2009). Thus, with appropriate ethical considerations, human remains could be considered as an alternative to cadavers for training purposes (Buis et al., 2015; Kapp, 2006). In the current section, cadaver tissues and fluids (excluding blood) have been discussed as potential CDD training aids.

# 3.1 | VOCs reported in human remains and decomposition fluid

The VOC profile of 14 separate tissue samples of human remains that were used for victim recovery (VR) dog training (referred to as CDD in this review) were analyzed in a study by Hoffman et al. (2009). The samples consisted of a blood clot, blood, muscle, testicle, skin, body fat attached to skin, adipocere samples, fat tissue, bone, and teeth (Hoffman et al., 2009). From this study, 17 low-frequency (<33%) VOCs, 15 medium-frequency (33-66%) VOCs, and one highfrequency (67–100%) compound—p-xylene was identified. None of these 33 VOCs are specific to human decomposition (Dekeirsschieter et al., 2009). Even though cyclic hydrocarbons such as p-xylene and toluene are commonly reported in multiple decomposition studies, they are not exclusive to the decomposition process (Inoue et al., 1996; Statheropoulos et al., 2005). For example, these compounds can be detected in the environment as a result of automobile emissions (EPA; PubChem, n.d.). Toluene can be off-gassed from paints, adhesives and nail polish in an indoor setting (EPA, n. d.). Much like cadaver decomposition VOC profiles, the compounds in this study belonged to several chemical classes including acids/acid esters, alcohols, aldehydes, halogens, aromatic hydrocarbons, ketones, and sulfides. Among these, alcohols and aldehydes represented major classes of compounds in terms of frequency of appearance. Since none of the tissue types comprised all 33 VOCs, the authors concluded that a CDD may have to be trained on a variety of human tissues in order to expose them to the entire spectrum of decomposition VOCs. The muscle tissue lacked fatty acids however, the release of free fatty acids during breakdown of muscle tissue has been previously reported (Statheropoulos et al., 2005). This difference could be due to variability in the extent of decomposition and the length of each study since the Hoffmann et al. study analyzed samples for a few weeks while, the Statheropoulos et al. study analyzed samples from a few hours and days to a few months. Twelve of the 33 VOCs were also reported in another human remains study, namely: 1-pentanol, hexanal, butanoic acid, pentanoic acid, heptanal, benzaldehyde, 2-pentryl furan, dimethyl disulfide, hexanoic acid, heptanoic acid, nonanoic acid, and octanoic acid (Lorenzo et al., 2003).

Rosier et al. (2015, 2016, 2017) conducted three studies that aimed to identify VOC markers in human and animal remains (pigs and small mammalian remains) that were decomposed in an aerobic laboratory environment for up to 6 months (in the first study) and up to nine and 12 months (in the second study; Rosier et al., 2015, 2016). During the third study, VOC profiles of larger mammals (lamb and roe remains) were compared to human and pig remains (Rosier et al., 2017). Through the course of the three studies, it was observed that sulfur containing compounds were significant during the first month of decomposition. Based on this, the authors suggested that diethyl disulfide could be used to train CDDs during the first month of decomposition. However, this has not been corroborated by other studies. At the end of the first two studies, it was deduced that seven compounds: propanoic acid, ethyl ester; propanoic acid, propyl ester; butanoic acid, propyl ester; pentanoic acid; ethyl ester; 3-methylthio-1-propanol; methyl(methylthio)ethyl disulfide; and pyridine were present up to 12 months of decomposition and thus, these could be significant in CDD training. However, these seven compounds were not specific to human remains as they were detectable in both human and pig remains. The last of the studies showed that pyridine was of particular significance as it occurred more frequently in human remains than in animals. Pyridine has been reported in animal remains in several studies including Rosier

et al. (2015, 2016) however, none of the previous studies compared the frequency of occurrence of pyridine in animals and humans remains (Dekeirsschieter et al., 2009; Rosier et al., 2015, 2016; Statheropoulos et al., 2011).

Decomposition fluid is the liquified product produced during soft tissue decomposition. A study found that 70% of VOCs detected in decomposed remains were also detected in decomposition fluid (Buis, 2016), suggesting that decomposition fluid could be a CDD training aid substitute. A summary of compounds found in human remains and decomposition fluid tissue has been listed in Table 1. Compounds such as dimethyl disulfide were detected in both human remains and cadaver studies (Hoffman et al., 2009; Statheropoulos et al., 2005). Based on the results of the above mentioned studies, it can be concluded that the VOC profile of human remains is not exclusive to human decomposition as these compounds may be detected in animal remains and in the natural environment. Nevertheless, the similarly in their VOC profile to that of a cadaver favors their use as an alternative CDD training aid once ethical and biohazard issues are addressed. It cannot be concluded from these studies whether a specific human tissue is ideal for training thus, the best practice is to expose dogs to a variety of human tissues, where feasible.

### 3.2 | Dog performance on human remains and decomposition fluid

The success rate of using human remains as training aids has been reported to be as high as 94% during the training session conducted by Royal Canadian Mounted Police handler and dog teams (Dilkie & Veniot, 2017). These results are significant in understanding the impact of obtaining and training CDDs with human remains when possible. One of the earliest studies that estimated reliability of cadaver dogs in the detection of human remains was conducted by Komar (1999) in the form of a 2-month training and research program that used eight dog and handler teams. The training sessions were followed by blind field trial searches in 20 m<sup>2</sup> areas where human bone, animal bone, gauze soaked with human decomposition fluid, and clothing soaked in decay fluids were hidden but not buried. The recovery rate per trial ranged between 55 and 95% and the overall recovery rate in the field trials was 81% (Komar, 1999). The temperature during the search period ranged from -30 to  $10^{\circ}$ C and the snow did not hinder the dogs' performances however, the recovery rates were found to be reduced when the dogs were introduced to dry old human bones. Unlike human tissue, bones have limited organic matter which results in reduced intensity of VOCs (Collins et al., 2002), thus, resulting in less detectable odor compared to human tissue. This makes detection of bones more difficult. Thus, for dogs to be able to locate remains with extended postmortem intervals (PMIs), it is essential to train dogs on both soft and hard tissue.

A study by Cablk and Sagebiel (2011) estimated dogs' capabilities in locating partially buried or hidden individual teeth in an outdoor field setting. Teeth are considered forensically significant as they allow a person's identification through comparison with dental records and individualization through DNA (Bell, 2001). In this study, double blind trials were conducted using three certified HRD teams who were exposed to ten teeth placed 0.9 m apart within 10 m<sup>2</sup> (Cablk & Sagebiel, 2011). Unlike other studies, the trials in this study were conducted with dogs on leash in a guided grid search. During the trial, the recovery by each of the dogs was found to be 78, 61, and 20%. The study concluded that the dogs were able to locate individual human teeth however, each dogs' ability was highly variable. Hoffman et al. (2009) reported that teeth VOCs included 1-pentanol, tetrachloroethylene, toluene, 2-pentylfuran, cyclohexanone, and dimethyl disulfide. These VOCs could represent those responsible for the olfactory response by dogs during the Cablk and Sagebiel (2011) study, however the VOC profile was not studied to confirm this.

When decomposition fluid is released from a body, it is absorbed into the environment around the body such as in soil and textiles, thus, forming an effective source of decomposition odor and a potential training aid for CDDs. Buis et al. (2015) conducted a study that investigated the detection limit of CDDs to decomposition fluid and validated the use of decomposition fluid as a CDD training aid. The fluid was diluted up to one part per trillion (ppt) through serial dilution to generate 13 samples of varying dilutions (Buis et al., 2015). During the first trial, all the dogs responded to the highly concentrated decomposition fluid but at lower concentrations, the positive response rate decreased, and no positive response was observed below  $10^{-5}$  concentration. In the following trials (#2, 3, and 4) the dogs response percentage improved and increased up to 100% (all dogs showed positive alerts) even at the lowest concentration  $(10^{-12})$ . During Trials 5 and 6 which were conducted with a lower volume (0.2 ml) for the same decomposition fluid concentrations, it was observed that the response percentage for the undiluted sample was 100% however, the average correct responses was reduced compared to previous trials when a larger volume (1 ml) of decomposition fluid was used. During Trial 6 for  $10^{-1}$  to  $10^{-12}$  concentrations, 60% of dogs responded correctly. Each of the teams (A, B, and C) showed 80% + correct responses however, Team C showed high error rates during trial 6 because they were absent from Trial

1, 4, and 5 and therefore did not have enough exposure to decomposition fluid. Thus, based on the results it was concluded that with repeated reinforcement of target odor, CDDs could detect 0.2 ml of decomposition fluid even at  $10^{-12}$  concentrations. In a latter study by the authors (Buis et al., 2019), it was found that the correct response rate for decomposition fluid aged over 2 years was 81% (Buis et al., 2019). Results from these studies are in favor of decomposition fluid being used as a CDD training aid. Overall, the dogs have a high success rate in locating human remains however, they may encounter difficulties in cases involving dry remains and/or bones due to a lack of VOCs or reduced odor intensity. A range of tissue, fluids, dry remains, and skeletonized remains should therefore be incorporated as a part of regular maintenance training.

# 4 | SOIL AND BURIED REMAINS AS TRAINING AIDS

When soil is in direct contact with a decomposing body, it is likely that decomposition products will enter into and be contained in the soil. In soil decomposition scenarios, some compounds stay close to the remains while others might leach into the soil plume. This is determined by properties of the soil (porosity) and those of the compound itself (density, volatility, and molecular weight; Osterkamp, 2020). Soil obtained from decomposition sites can be stored by CDD handlers for training purposes. VOC studies on soil and burials have been conducted to understand their suitability and resemblance to the VOC profile of decomposed remains.

# 4.1 | VOCs in soil and buried remains

VOC analysis from soil was conducted by Vass et al. as part of a three series study aiming to build a decomposition odor database (Vass, 2012; Vass et al., 2004, 2008). Four compounds namely toluene, dimethyl disulfide, nonanal, and tetrachloroethylene detected in the Vass et al. (2008)'s study from the surface above human burials were the same as those detected from the CDD training aids (Hoffman et al., 2009). Although some of these compounds are from environmental sources, it is possible that dimethyl disulfide and nonanal could be relevant to the decomposition VOC profile.

In the Vass (2012) study, analysis was conducted on soil samples associated with human decomposition sites obtained from different environments such as desert, deciduous, tropical, marsh/swamp, and grassland biome environments. One hundred and eighty-six soil samples (burial range <1 month-119 years) were collected over 9 years from gravesites (approx. 0.48–2.47 m in depth) which were confirmed using dogs and geophysical instrumentation. Among the 56 significant compounds identified, the author reported several human-specific VOCs including carbon tetrachloride found in the soil plume, pentane, and decane found close to the decomposing remains, and, undecane reported to be from either the plume or the remains. 3-Methyl butanal was reported more frequently than 2-methyl butanal in human decomposition while this trend was reversed for animal remains. Certain compounds such as fluorinated hydrocarbon, benzene, sulfur containing compounds (dimethyl disulfide and dimethyl trisulfide) and some aldehydes (hexanal, heptanal, octantal, and nonanal) were found to be associated with the soil plume. Some of these compounds, particularly the sulfides and aldehydes, could be significant to training CDDs to locate clandestine graves. Halogenated hydrocarbons (fluorinated hydrocarbon and carbon tetrachloride) were detectable in this study due to the presence of contamination from old medical waste deposited on the site. This study also highlighted the fact that the process of decomposition can be highly variable when it occurs near the surface versus in deeper burials as the availability of oxygen varies greatly. Thus, dogs should be trained on materials that have decomposed in both aerobic and anaerobic environment as the volatiles evolved in both the processes can vary.

Dubois et al. (2018) analyzed soil VOCs associated with a decomposing body 7 days after removal of the body (Dubois et al., 2018). Five grams of soil was collected from under the body and at the edge of the cadaver decomposition island (CDI). Several compound classes such as carboxylic acids, esters, aldehydes, alkanes, alkenes, alcohols, cyclic hydrocarbons, halogens, ketones, sulfur-containing compounds, nitrogen-containing compounds, terpenes, and terpenoid derivatives were identified from soil samples. The VOC profile of the soil from the edge of the CDI showed a lower abundance of aldehydes and a higher abundance of alkenes and cyclic hydrocarbons when compared to soil from under the body. The authors suggested that soil samples from beneath the body should be collected for analysis to increase the chance of decomposition-related VOC detection. This could result from decomposition products moving vertically in this soil type as opposed to dispersing horizontally over a large area away from the body.

Ethyl benzene was the only common compound detected in both of the above-mentioned soil studies. It has also been detected in studies associated with decomposition of cadavers in Mediterranean Sea water (Statheropoulos et al., 2005) Differences in VOC profiles from soil and the cadaver could be attributed to the number and concentration of VOCs retained in the soil and variable microbial and entomological activity in the soil. Unlike with scattered or scavenged human remains, soil and burial sites are in contact with the entire body during decomposition and thus, will be exposed to the entire spectrum of VOC's emitted from a human body. However, one drawback to using soil as a CDD training aid is that it may not retain all of the decomposition compounds and may even have additional compounds due to contamination from multiple sources. A summary of compounds found in soil and burial remains has been listed in Table 1.

### 4.2 | Dog performance on soil and buried remains

One of the earliest studies testing the ability of cadaver dogs to locate buried human remains was conducted in Alabama by Lasseter et al. During this trial, fresh and skeletonized human and animal remains were buried at 1 and 2 ft. depths. Fresh human remains odor was obtained in the form of gauze pads placed inside bodies during autopsy (Lasseter, Jacobi, Farley, & Hensel, 2003). Only one alert was recorded for skeletal human remains at 2 ft depth and another alert to fresh human remains at 1 ft depth throughout the trial. This study could not distinguish between dog responses based on animal and human odor as the dogs responded with either no alert or a "narrow" alert during the trial. Despite having varied years of experience (10 years–1 year 8 months), the percentage of no alert was high (53%), potentially due to a lack of odor from the burials. A more recent study by Glavaš and Pintar (2019) showed that dogs could locate older buried remains as they aided discovery of five new tombs that dated to the iron age in Europe.

Alexander, Hodges, Bytheway, and Aitkenhead-Peterson (2015) studied the sensitivity and accuracy of eight certified HRD dogs (four dual purpose and four single purpose) on grave soil previously associated with human remains. Ten soil samples were collected from graves with residual odor after removal of human remains, and six were from graves with human remains (Alexander et al., 2015). The PMI of bodies ranged from 18 to 915 days and the number of days since the bodies had been removed from the graves (denoted as RCDI) ranged between 18 and 667 days. Trials were conducted on 30 g of soil from graves with and without human remains; 3 g of soil unextracted and extracted at a 1:10 soil/water ratio; and oven dried latex free sterile pads impregnated with water obtained from extracted soil solutions. An overall 92.8% response accuracy was indicated on grave soils. The dogs were able to correctly identify the oldest PMI of 915 days with 100% accuracy and the oldest RCDI of 667 days with 85.7% accuracy. This suggests that detecting residual odor in soil might be more difficult compared to soil with remains. However, there was no specific trend in response accuracy with increasing number of RCDI thus, even though the dogs responded to residual odor in soil, no conclusions could be drawn about the time elapsed before dogs stop responding to residual odor. In contrast to a common belief among dog handlers that single purpose dogs tend to perform better in field, this study observed that overall, accuracy of dual-purpose dogs (92%) was not different from that of single purpose dogs (91%). Thus, soil in contact with a decomposing body could be a useful training material since CDDs can identify residual human remains odor in the soil. A subsequent study conducted by the authors indicated that soil porosity could affect dogs' performance while locating remains (Alexander, Hodges, Wescott, & Aitkenhead-Peterson, 2016). This was indicated through the fact that the mean response time by the dogs in sand was much quicker than clay soil as the dogs showed a faster change in initial behavior. This was likely due to the ease of movement of highly volatile compounds in highly porous sandy soil from the soil toward the surface thus, making them more readily detectable. However, the dogs had difficulty pinpointing the exact location of the odor in the sandy soil which could be attributed to the high porosity of the soil as volatiles might become more easily dispersed over larger areas.

From these studies, it can be understood that even though the soil may not retain all decomposition compounds, it can retain some VOCs long after the remains have been removed, often referred to as residual odor. However, this is highly influenced by the soil porosity. Highly porous soil may allow leaching of compounds but will also disperse and release VOCs much more readily than less porous soil. Thus, the handler needs to consider soil properties when collecting and storing decomposition soil for training purposes. Fewer compounds are detectable in soil long after the remains have been removed. Thus, if the soil is used from a site which once had remains, it is likely that the dogs will only be exposed to a small spectrum of the entire decomposition VOC profile.

# 5 | TEXTILE AS A TRAINING AID

Textiles associated with a decomposing body can be found at the crime scene in the form of clothing or body wrappings (Hoffman et al., 2009; Komar, 1998). Depending on the type of material, decomposition products will be absorbed and retained by some textiles. Thus, it is thought that the textile contains the odor from the body and is suitable as an alternative training aid to human remains such as bones, blood etc. (Komar, 1998). Textiles are used in instances where live scent tracking is required (Kalmus, 1955; Settle, Sommerville, McCormick, & Broom, 1994; Stockham, Slavin, & Kift, 2004). Typically, dogs are first exposed to the missing individual's clothing containing the individual's live scent from sebaceous gland secretions and then the dog will be released in a specific location to locate similar scent in the vicinity (Jones, Dashfield, Downend, & Otto, 2004).

# 5.1 | VOCs in textile associated with decomposing human remains

A study by Nizio, Ueland, Stuart, and Forbes (2017) proposed that the VOCs retained by degrading textile through the various decomposition stages formed a reliable representation of cadaveric odor. These results were based on a study involving buried porcine remains covered with 100% cotton t-shirts. Such a study has not been replicated with decomposing human remains however, textile in the form of gauze pads trapped with decomposing human remains odor was studied by DeGreeff and Furton (2011). A dynamic headspace concentration device called the Scent Transfer Unit (STU-100) was used for odor collection. STU-100 is a modified vacuum pump that was originally designed to collect scent from evidence at crime scenes and living humans. DeGreeff et al applied the STU-100 for cadaveric scent collection for VOC analysis from 21 bodies in the early to mid-stage of decomposition and compared it with the dogs' performance on pads concentrated with cadaveric odor (DeGreeff & Furton, 2011). In this study, samples were collected onto Dukal cotton gauze, Johnson & Johnson cotton-blend gauze, and polyester material at different flow rates. The VOC analysis results indicated that Dukal gauze at the low flow rate setting on the STU-100 collected the highest number of VOCs. Thus, a cotton-based gauze or a blend of cotton with polyester gauze was concluded to be optimal for VOC collection. These results are consistent with the current practices among law enforcement agencies that use cotton-based gauze material for odor collection (Goss, 2019). A summary of the compounds found in textile associated with human remains has been included in Table 1. Similar to STU-100, SEKR, and the human scent collection system have been developed which are used to generate scent pads that could be used by detector dogs for explosives, narcotics, and live humans (Furton et al., 2015). However, to date their application in the collection of cadaveric odor has not been validated.

# 5.2 | Dog performance on textile associated with decomposing human remains

Oesterhelweg et al. (2008) were the first to report on the duration of contact between a cadaver and textile material for the odor to be detected by a dog, as well as the duration that the odor will be contained in a fabric. In their study, two deceased individuals were wrapped in cotton blankets and placed on top of 20 cm<sup>2</sup> carpets for two and 10 min intervals, respectively (Oesterhelweg et al., 2008). The performance of three BDD and CDD was tested in an indoor environment using carpet squares "contaminated" with decomposition odor for 2 and 10 min. Results from the study indicated that the dog could identify the carpet squares with 94% accuracy (2 min) and 98% accuracy (10 min), respectively. The authors concluded that the dogs' accuracy to detect textile contaminated with cadaveric odor does not degrade with time (at least for the period studied) and that trained CDDs would be able to recognize a decomposing tissue after PMIs of just 2 h.

DeGreeff, Weakley-Jones, and Furton (2012) proposed that gauze pads containing cadaveric odor could replace conventional training aids such as blood, human remains, and so on. This further study focused on understanding the efficiency of gauze to trap cadaveric odor using the STU-100 and to be subsequently detected by HRD dogs. Results from this study indicated that cadaver odor collected onto gauze pads using the STU-100 can be used as a training aid for up to 12 h after collecting the odor. Exposure of gauze pads to open air for more than 24 hr reduced the positive detection rate to 50%. Eighty-six percent of the dog alerts were correct. In comparison, the previously mentioned Oesterhelweg et al. (2008) study produced better results with 94% of dogs alerting correctly with 2 min contaminated carpet and 98%

of dogs alerting correctly with 10 min contaminated carpet. This suggests that direct contact of the textile with decomposing remains elicits a better response to decomposition odor by CDDs as opposed to using the STU-100.

### **6** | SYNTHETIC TRAINING AID

Synthetic training aids for CDDs were developed as a consequence of the increasing difficulty in ethical and legal procurement of natural human remains as well as the biohazardous nature of these training aids. Synthetic training aids are reported to comprise the key VOCs of the "scent of death." Sigma–Aldrich<sup>®</sup> and SOKKS<sup>®</sup> provide commercially-available cadaver scent formulations. Sigma–Aldrich<sup>®</sup> formulations include Sigma Pseudo<sup>TM</sup> Corpse Scent Formulation I (PSI) which is reported to resemble early decomposition scent, Sigma Pseudo<sup>TM</sup> Corpse Scent Formulation II (PSII) which is reported to resemble late decomposition scent and Sigma Pseudo<sup>TM</sup> Corpse Scent, drowned victim scent (PSDV) which is reported to resemble the scent of a drowned victim. Unlike PSI and PSII, PSDV is no longer commercially available. SOKKS-MPTS<sup>®</sup> claims "SOKKS-Cadaver is REAL human odor and will enable the dogs to detect humans at ALL stages of decomposition" (SOKKS-MPTS<sup>®</sup>, n.d.). It is designed to cater for both land and water decomposition scenarios however, no published study has validated this claim. To date, there have been only two studies that aimed to characterize the composition of the Sigma Pseudo<sup>TM</sup> Corpse Scents, one of which also studied the dogs' response in the field.

### 6.1 | VOCs in synthetic training aids

The first study to analyze the Sigma-Aldrich® pseudo scents was conducted by Stadler et al. (2012) and qualitatively analyzed PSI and PSII (Stadler et al., 2012). A second study by Tipple et al. (2014) quantitatively analyzed PSDV in addition to the PSI and PSII formulations (Tipple et al., 2014). Both studies demonstrated consistent results. The major compounds reported in PSI were 2-pyrrolidone and 4-aminobutanoic acid while the minor compounds reported were 1,8-Diazabicyclo[5.4.0]undec-7-ene (DBU), butyrolactone and tridecylamine in the Stadler et al. study and, 3-methyl-2-pyrrolidinone and 4-methyl-2-pyrrolidinone in Tipple et al. study. Differences in the minor compounds reported could have occurred because methyl-2-pyrrolidinone is produced in the presence of butyrolactone and amine (Harreus et al., 2011). PSII was reported to be more complex in its composition compared to PSI as it comprised 1,4-diaminobutane (putrescine) and 1,5-diaminopentane (cadaverine) in addition to the two major and minor compounds reported for PSI. Cadaverine and putrescine are highly polar volatile products of amino acid degradation. Only one study so far has recorded them when analyzing decomposition VOC profiles (Vass, 2001). Most studies have failed to record these compounds potentially due to their breakdown by bacterial activity (Rosier et al., 2015). In the Tipple et al. study,  $GC \times GC$  analysis of the samples revealed low concentrations of acetone and isopropanol in PSI and 5-amino-pentanol, ethanol, butyrolactone, acetone, methanol, and 1,4-dioxane in PSII. PSDV was reported to have the same compound composition as PSI. The Material Safety Data Sheet (MSDS) (Version 5.1) published by Sigma-Aldrich for PSDV reports its composition as 11.8% 4-aminobutanoic acid and 88.2% Nalco. A summary of compounds found in synthetic formulations used for CDD training has been included in Table 1.

Evidently, the VOC profile of synthetic training aids is an oversimplified representation of the actual decomposition VOC profile. Several significant factors such as the interaction of decomposing remains with the soil environment, and the microbial community which influences the VOC profile produced, are absent in the preparation of synthetic training aids. Thus, this limits their use among the CDD handler community. Converse to the results from the previously mentioned study, a study in 2018 was unable to detect cadaverine or putrescine in the formulation (Lavigne, 2018). The author suspected this could be due to a potential change in formulation by the company considering the fact that their formulations were simplistic. This study also found two compounds (naphthalene and cyclobutanol) common between the two synthetic formulations and deer liver remains. Such results directly indicate a limitation of using synthetic formulations as CDDs trained using them could produce false positive results in the field.

# 6.2 | Dog performance on synthetic training aids

Initially, dog handlers reported that CDDs trained on cadaverine or putrescine were able to perform successfully even in the field (Rebmann & David, 2000). However, as the complexity of the decomposition VOC profile was better

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understood, the popularity of the simplistic synthetic training aids reduced among the dog handler community. Cadaverine and putrescine are toxic thus, their exposure to dogs and handlers must be controlled and avoided when possible (Lewis, 1998). A dog's performance to synthetic training aids was recorded in the Tipple et al. (2014) study in both indoor and outdoor settings. A VRD (referred to as CDD in this review) that had been previously trained on a wide range of human remains odor was deployed for this study (Tipple et al., 2014). For the outdoor environment study, PSI and PSII samples were applied directly on a rock and three capsules of PSDV were dissolved in lake water 1 m from the shoreline. For the indoor trials, gauze pads impregnated with PSI and PSII samples were placed inside metal cans situated at screening stations. PSDV samples were placed in a bucket at the screening stations by dissolving them in tap water. At the end of the trials, it was observed that the dog showed no significant positive response toward any of the formulations. Thus, it was concluded that synthetic formulations are not an ideal representation of human decomposition odor. Currently, there is not enough empirical evidence to assess the suitability of synthetic training aids and further enhancement of these synthetic training aid formulations is required before they can be appropriately implemented into CDD training.

# 7 | CONCLUSION

CDDs are exposed to a complex array of VOCs through various training material including blood, human remains, soil or textile associated with human remains, decomposition fluid and synthetic formulations. The various classes of compounds that have been identified in the training aids include sulfur and nitrogen-containing compounds, carboxylic acids, ester, alcohols, aldehydes, ketones, cyclic hydrocarbons, aliphatic hydrocarbons, and halogenated compounds. None of the training aids comprise a single compound profile thus, it is not ideal to expose the CDDs to only one compound or one type of training aid for training purposes. In this regard, synthetic formulations which contain a limited number of compounds are not recommended as training aids for CDDs. Human remains tissue and decomposition fluid appear to be the most similar in decomposition VOC spectrum to human cadavers and dogs trained on these aids report high alert rates. Soil beneath decomposing remains can also be useful to train dogs on residual odor however, their major drawback is that they may not retain all decomposition products, thus limiting their effectiveness as a sole training aid. Blood at various ages should be included in the training protocol, especially because not all crime scenes involve human remains. Some scenes can involve blood that has been cleaned and thus, it is recommended that CDDs are exposed to washed/cleaned blood applied on multiple surface types. The VOC profile of textile directly in contact with a decomposing body needs to be studied further to determine its VOC profile. Among the complex mixture of decomposition VOCs, some compounds have been reported by multiple (five or more) studies in different training aids. These include 2-ethyl hexanol, 1-octen-3-ol, benzaldehyde, heptanal, octanal, nonanal, 2-butanone, dimethyl disulfide, 2-ethyl hexane, octane, decane, and toluene. These compounds have been reported in the decomposition VOC profile from human cadavers as well (Martin & Verheggen, 2018). It is difficult to conclude if the compounds identified as "significant" through modern analysis and instrumentation are the same that the dogs respond to but, since they have been repeatedly reported, it is likely that the dogs are constantly exposed to them and may be associating them with decomposition activity. These compounds could potentially be used to create a substitute for natural training aids with further research in the future. Since the decomposition VOC spectrum is highly variable and complex for enhanced dog performance, it is recommended to expose CDDs to as many decomposition-related odorous compounds as ethically available.

Further research is needed to understand how the VOC profile of training aids differ from human cadavers since, the training aids are subjected to various storage conditions (room temperature vs. refrigerated vs. freezer) and environmental conditions (burial vs. surface vs. water decomposition) during CDD training. It is not known if the odor profile varies greatly between cadavers and human tissue training aids, if the odor changes over time, and the length of time it is ideal to use human tissue as a training aid. Once these aspects are better known, the current CDD training practices can be further enhanced and standardized.

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#### **CONFLICT OF INTEREST**

The authors have no conflict of interests to declare.

#### **AUTHOR CONTRIBUTIONS**

**Rushali Dargan:** Conceptualization; data curation; formal analysis; resources; validation; writing-original draft; writing-review and editing. **Shari Forbes:** Conceptualization; formal analysis; funding acquisition; project administration; resources; supervision; validation; writing-review and editing.

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