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Effective neutralization of chemical warfare agents (HD, VX) by Me-DABCOF: a small molecule with dual action†

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The ability of mono *N*-methyl-1,4-diazabicyclo[2.2.2]octane fluoride (Me-DABCOF, 1) to act as a bifunctional reagent that effectively and universally neutralizes both the persistent and extremely toxic blister agent HD and the nerve agent VX in nearly neutral aqueous solution, alumina powder or a hydrogel formulation, is described.

Chemical warfare agents (CWAs) pose a significant threat to human life and the environment as evident from their recent usage.[1,2] The most relevant and challenging CWAs in these respects are the extremely toxic and highly environmentally persistent nerve agent VX and the blister agent HD (sulfur mustard). Current decontamination protocols of areas contaminated with these CWAs are based on solutions/formulations holding corrosive reagents such as sodium hydroxide, alkoxide/amine bases, hydrogen peroxide or bleach.[2,3] In addition, these formulations lead to some toxic products, tend to damage the surfaces they are applied on and are considered to be unfriendly to the environment. Development of milder decontamination methodologies is accordingly a current effort in the field. Indeed, active nano-powders,[4] polyoxometalates,[5] metal organic frameworks,[6] enzymes,[7] supramolecular systems,[8] fluoride salts[9] and more,[3] have all been suggested in this regard. Optimally, a decontamination protocol should rely on a facile-made, water-based, non-corrosive and environmentally benign solution, which leads to effective universal decontamination of both types of CWAs. This task is not trivial as for example, even neutral water or mild basic aqueous solutions provide toxic side-products once applied on both types of CWAs *i.e.* VX[10] and HD[11] as shown in Fig. 1a and b.

Previously, we have shown that fluoride salts present in various formulations and matrices provide effective degradation of VX.[9,12,13] However, the decontamination of HD by these fluoride-containing systems was much less effective.[14] As evidently a fluoride nucleophile is not appropriate for HD neutralization, a different nucleophile should be used. We hypothesized that a free amine would react much more effectively with this alkylating agent. However, free amines mostly form corrosive aqueous solutions. DABCO, a rigid scaffold presenting two tertiary nitrogen atoms is a well-known nucleophile as was recently shown in its high activity in alkylations,[15] arylations[16] and epoxide openings.[17] It is also known that after mono *N*-alkylation of DABCO the Lewis basicity of the second free amine function is dramatically decreased,[18] yet, its nucleophilicity is retained. Therefore, aqueous solutions of mono *N*-alkyl-DABCO have almost neutral pH. Aiming to develop a mild universal, non-corrosive water-soluble decontamination agent, we envisioned that Me-DABCOF **1**, which combines two different nucleophilic centres, *i.e.* a non-basic free amine and an ammonium fluoride moiety on one small molecule, would promote effective neutralization of both types of CWAs, *i.e.* HD and VX, respectively

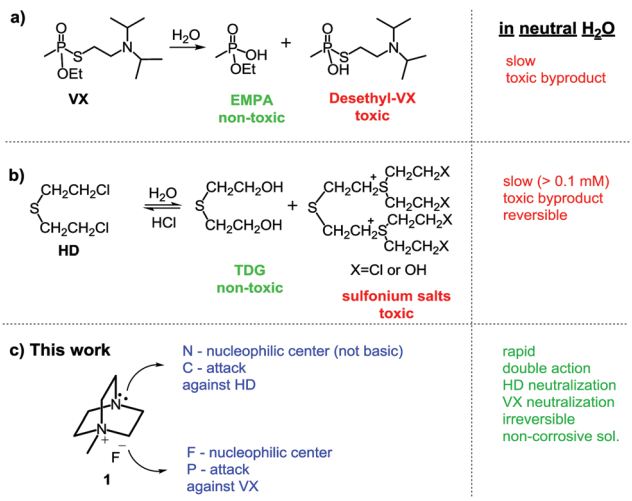

Fig. 1 (a) VX hydrolysis in water. (b) HD hydrolysis in water. (c) Me-DABCOF **1** structure and its dual action in the neutralization of both CWAs in water.

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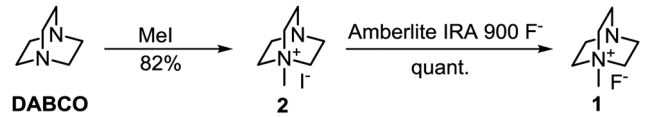

Fig. 2 Preparation of **1** from DABCO.

(Fig. 1c). The excellent performance of Me-DABCOF **1** as a universal decontaminating agent is herein described.

First, Me-DABCOF **1** was synthesized in two simple steps from the commercially available reagent DABCO, as previously reported by us (Fig. 2, see ESI†).[19]

Aqueous solutions holding various concentrations of **1**, pH of 8.5 (attributed to the ammonium fluoride function), were next applied on both HD and VX and the reactions were followed by ^{13}C and ^{31}P NMR, respectively. In order to facilitate the study of the neutralization of sulfur mustard by **1**, we used ^{13}C-labelled HD (HD*).[14] As hypothesized, we have found that **1** rapidly reacts with HD* to give two final products, which were identified as the ammonium salts **4** (major) and **5** as shown in Fig. 3. For example, in the presence of 20 equivalents of **1**, HD* was converted mostly to the tetra ammonium salt **4** *via* the intermediate **3** (Table 1, run 3, Fig. S5, ESI†). In order to validate these results and fully characterize the product, the reaction that was typically performed using 4.7 mg of HD* was scaled up to 50 mg of non-labelled HD. Slow addition of HD to an aqueous solution of **1** (8 equiv.) led solely to product **4**, which was characterized by ^{1}H-, ^{13}C-, HMQC and HMBC NMR analyses (see Fig. S8–S12, ESI†), as well as HRMS, albeit having a rather unique pattern of multiple quaternary ammonium groups. The intermediate product **3** and the minor by-product **5** were identified and characterized by their ^{13}C-NMR as they hold one arm identical to **4** and one arm identical to HD or TDG, respectively. Remarkably, in spite of the fact that the reactions were performed in nearly neutral aqueous solutions (pH 8.5), in all cases, no toxic sulfonium by-products were observed. As was evident from the analyses, the HD degradation involves two *N*-alkylation steps; the monosubstituted derivative **3** was first quickly obtained and then transformed to the disubstituted product **4** or hydrolysed to **5** in a slower second step (Fig. 3 and Fig. S3–S5, ESI†). Importantly, unlike simple hydrolysis in neutral aqueous solutions where the released HCl

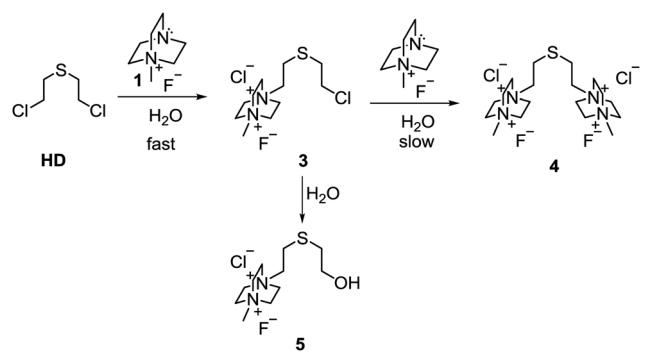

Fig. 3 Reaction of **1** with HD in aqueous solution.

Table 1 HD neutralization in aqueous solutions

Run	Reagent	Equiv.	$t_{\text{total HD}}$[a] (h)	$t_{\text{total 3}}$[b] (h)
1	**1**	3.6	19.1	>27[c]
2	**1**	8	0.9	11.3
3	**1**	20	1.5	12.3
4	DABCO	8	0.9[d]	6.6[e]
5	**6**	3.6	3.6[f]	—[g]

[a] Total conversion of HD to product **3**. [b] Total conversion of **3** to a mixture of products **4** and **5**. [c] At the indicated time the products distribution was: 55% **3**, 25% **4** and 20% TDG. [d] Total conversion to a product analogous to **3**. [e] Total conversion to a product analogous to **4**. [f] A mixture of TDG, sulfonium ions and chlorohydrine was obtained. [g] Intermediate **3** is not relevant in this process.

may react reversibly with TDG to reform HD (Fig. 1b), the formation of the stable ammonium salts **4** and **5** is irreversible.

In respect to the reaction time, the results show that the usage of a large excess of **1** is advantageous over a small excess (compare runs 1 and 3, Table 1). Once a large excess of **1** is used (8 equiv. or more), the rates of the two aforementioned steps are almost the same (compare runs 2 and 3). However, the product distribution is strongly affected by the amount of **1**, as the substitution reaction by **1** competes with hydrolysis. For example, in the case of 8 equiv. of **1**, 56% of product **4** and 44% of the mono hydrolysed product **5** were formed as compared to 85% of **4** and 15% of **5**, which were obtained once 20 equiv. of **1** were applied (Fig. S4 and S5, ESI†). In the presence of 3.6 equiv. of **1**, a mixture of the substitution products **3**, **4** and **5** together with the full hydrolysis product TDG, was obtained (run 1, Fig. S3, ESI†). Expectedly, DABCO itself effectively reacted with HD*, like **1**, *via* *N*-alkylation (run 4, Fig. S6, ESI†). However, DABCO led to much stronger basic conditions than **1** (*i.e.* pH 11.4 *vs.* 8.5, respectively, for the same concentrations), suggesting it is much more corrosive. Next, to further highlight the advantage of **1** over its closed analogues, the activity of *N*-methylquinuclidinium fluoride **6**,[19] which is identical to **1**, but lacks the nucleophilic nitrogen, was also examined. Interestingly, this compound managed to hydrolyse HD* relatively fast; however, toxic sulfonium ions and chlorohydrine were formed, and expectedly, not products like **4** or **5** (run 5, Fig. S7, ESI†).

The above results clearly show that **1** can effectively neutralize HD by forming the corresponding final ammonium products **4** (mainly) and **5**. HD is a powerful irritant and blistering agent that damages the skin, eyes, respiratory tract and lung membranes. The most prominent toxic effects of this blister agent are on the skin, where it produces severe damage. It is well known that the threat of this persistent and extremely toxic CWA is due to its lipophilicity that leads to fast skin penetration, volatility that results in easy contact with the lung and eyes, and reactivity as an alkylating agent, which causes tissue damage. The ammonium salt products **4** and **5** are polar and highly hydrophilic, not at all volatile and very poor alkylating agents. Therefore, the above-mentioned chemical destruction of HD can be safely considered as a neutralization of sulfur mustard.

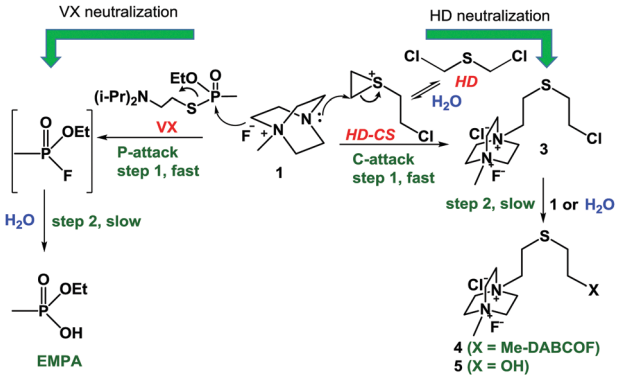

Fig. 4 Mechanisms of VX and HD neutralization by **1** dual action.

The ability of **1** to neutralize VX in an aqueous medium was next examined. Evidently, as seen in the NMR spectra, once a solution of **1** was added to VX (0.5 mL to 2.7–7.6 μL), the "G analogue" intermediate was quickly formed and subsequently hydrolysed to the non-toxic product EMPA while HF was neutralized by the amino thiolate leaving group (Fig. 4 and Fig. S16–S18, ESI†). Importantly, the toxic product desethyl-VX, which is known to be formed once neutral or basic solutions are applied (Fig. 1a), was not detected. Expectedly, as more equivalents of **1** were present, faster reactions were observed (compare runs 1–3, Table 2). However, even a small amount of **1** led to a rather fast VX degradation reaction (run 1). In order to validate the advantage of **1** over DABCO in VX degradation, an aqueous solution of DABCO (pH 11.4) was also applied on this CWA. As can be seen in Table 2, 8 equiv. of this reagent managed only to partially hydrolyse VX to EMPA and the toxic product desethyl-VX after 43 h (run 4, Fig. S19, ESI†). Lacking fluoride ions, this limited activity probably stems from the solution basicity.

Plausible mechanisms for both neutralization processes are depicted in Fig. 4. The mechanism of fluoride-promoted conversion of VX to EMPA *via* the fluoridate intermediate was previously reported by us in detail.[9] In the case of HD, in water, it first converts to the highly reactive cyclic sulfonium (HD-CS) intermediate, which due to its water-solubility and reactivity, is prone to a fast nucleophilic attack by the free tertiary amine of **1**, to afford the water-soluble intermediate **3**. Conversion of **3** to the final product **4** and minutely to **5** by nucleophilic attacks of **1** or water, respectively, occurs at a second stage. This step is slower as the sulfur atom, which now holds the bulky ammonium moiety at the β position, has less propensity for the anchimeric assisted cyclic sulfonium formation.

Table 2 VX neutralization in aqueous solutions

Run	Reagent	Equiv.	$t_{\text{total VX}}$[a] (h)	Desethyl-VX (%)
1	**1**	3.6	2.9	—
2	**1**	8	1.8	—
3	**1**	20	0.9	—
4	DABCO	8	>43[b]	20

[a] Total degradation of VX to give a mixture of the G-analog and EMPA (total conversion to EMPA, with no indication of desethyl VX was observed in runs 1–3 at the readings on the next morning after 20 hours). [b] After 43 hours only partial degradation was obtained: 57% VX, 23% EMPA and 20% desethyl VX.

The high potential aqueous solution of **1** exhibited in degrading CWAs encouraged us to examine the possible incorporation of this bifunctional active molecule in a few other matrices fitted for unique decontamination applications. Specifically, we were interested in the evaluation of an alumina (Al_2O_3) powder and a hydrogel holding **1**, which may be appropriate for the decontamination of delicate instrumentation and vertical surfaces, respectively.

In recent years we have been examining the activity of various fluoride impregnated Al_2O_3 supports to effectively degrade CWAs.[12] These active powders were found to be most effective for the neutralization of V and G-type of agents, while much less for HD. Based on our current solution results with **1**, we envisioned that it would exhibit effective bifunctional VX and HD neutralization activity, once impregnated on an Al_2O_3 powder as well. Accordingly, following our previous protocols,[12] a 20 wt% Me-DABCOF/Al_2O_3 powder was prepared and examined for its activity in these types of degradation reactions using solid-state magic angle spinning (SS MAS) NMR monitoring. The results show that similar to solutions of **1**, the dry Me-DABCOF/Al_2O_3 support effectively degraded VX (t_{total} *ca.* 3.5 h, Fig. S20, ESI†). Unlike the solution reactions, however, EMPA was found to be the sole product, as evidently, the "G analogue" intermediate immediately hydrolysed to EMPA on this solid support. Contrary to the trend observed previously with other powders,[20] Me-DABCOF/Al_2O_3 wetted with 10 wt% of water led to ~90 times slower VX degradation than that obtained on the dry powder (Fig. S21, ESI†). The reactions of HD with the dry and wetted Me-DABCOF/Al_2O_3 support were next examined. The dry support led to total degradation of HD within 71 h (Fig. S13, ESI†). Similar to analogous reactions we have explored previously with other R_4NF/Al_2O_3 supports,[21] the elimination products CEVS and DVS were mainly obtained (Fig. 5). On the other hand, the water wetted Me-DABCOF/Al_2O_3 support (10 wt%) led to faster HD degradation with total conversion to the substitution products **4** and **5** after 24 h (Fig. 5 and Fig. S14, ESI†). Therefore, the new solid support Me-DABCOF/Al_2O_3 may be considered as an effective active powder for the universal decontamination of both VX and HD.

Aqueous solutions are optimal for detoxifications of large contaminated areas or for manipulations in a closed container. However, they are not appropriate for the decontamination of small surfaces, specifically, not vertical ones, as they tend to drip from them leaving only a short contact time between the active reagents and the CWA. Active hydrogels, on the other hand, stick to such surfaces and accordingly present an optimal alternative for these types of decontaminations. As compound **1** was not expected to decompose gelators such as polysaccharides, it was envisioned that a hydrogel form of this bifunctional

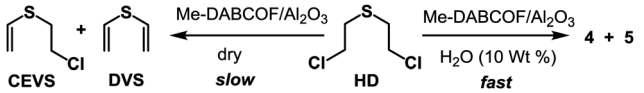

Fig. 5 Substitution *versus* elimination process in Me-DABCOF/Al_2O_3 promoted HD degradation.

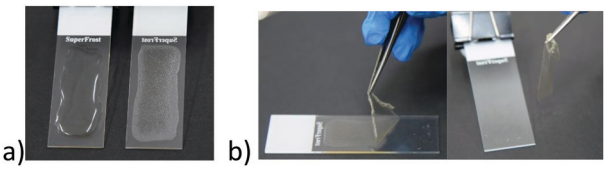

Fig. 6 (a) A gel, directly after application (left) and after 24 hours (right). (b) Peeling the film from a glass surface.

decontaminant may be easily prepared. Indeed, we found that mixing 2.5 wt% of the thickening agent carboxymethylcellulose sodium salt with 6 wt% of compound **1** in water results in the formation of a thixotropic hydrogel. This gel has a pH of 6.8, and thus it is mild and non-corrosive. As can be seen in Fig. 6a, once the hydrogel is applied on a surface it easily sticks to it and does not drip. After 24 h, when *ca.* 90% of the water content has spontaneously evaporated, it turned into a film, which could be simply removed to afford a clean surface (Fig. 6b). The rates and products of VX and HD degradation inside the gel were found to be essentially the same as those obtained with the aqueous solutions. For example, a hydrogel holding 20 equiv. of **1** converted VX solely to the non-toxic final product EMPA within a few hours (<20 h, Fig. S22, ESI†). The degradation rate of HD with the same hydrogel (20 equiv. of **1**) was similarly fast, with conversion to product **3** after 0.6 h and conversion to product **4**, with only a trace amount of **5** after 12 h (Fig. S15, ESI†).

In conclusion, we have shown that Me-DABCOF **1**, featuring two nucleophilic centres, effectively acts as a bifunctional decontaminant for the universal neutralization of CWAs. As it leads to nearly neutral conditions and provides non-toxic products while incorporated in various matrices such as aqueous solution, alumina powder and polysaccharide-based hydrogel, we believe that this simple molecule holds great promise for the future development of CWA decontamination formulations.

Conflicts of interest

The authors have no conflict of interest to declare.

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