Supporting information

A Selective and Sensitive Chromogenic and Fluorogenic Detection of a Sulfur Mustard Simulant

Vinod Kumar† and Eric V. Anslyn*

Department of Chemistry and Biochemistry, The University of Texas at Austin, Austin, Texas 78712, United States.

Experimental details

General
Fluorescence measurements were carried our using a Photon Technology International Quanta Master spectrofluorimeter with an 814 photomultiplier detection system using a 75W xenon short arc lamp. All chemicals and reagents were bought from Aldrich, Fluka Fisher Scientific and used without further purification.

Caution: CEES is a blistering agent and potential vesicant so it should be handled inside the fuming hood by wearing hand gloves.

Procedure for the naked eye detection of CEES using 1 and SQ solutions
A 1 (0.1 mg, 0.4 mM) was dissolved in 1 mL of methanol containing 3.0 equivalents of K₂CO₃. A solution of SQ dye at 14 µM was dissolved in chloroform using sonicator for 1 hr. 15 µL of 1 solution was able to bleach 1.2 mL of SQ solution instantaneously. A solution of CEES (0.11 mg, 0.89 mM) was allowed to react with a 1 (0.4 mM). 25 µL (200 µM of CEES) of this solution was treated with 1.2 mL of SQ dye solution. These vials were photographed.

Fluorescence titrations of SQ with thiol in chloroform
A stock solution of SQ (2.65 µM) was prepared in CHCl₃. A separate stock solution of 1 (0.4 mM) and 3.0 equivalents of K₂CO₃ was also prepared in methanol. This solution at 0.2 mM (containing SQ was used for titration. A 2 mL aliquot of the SQ solution was transferred to the fluorescence cuvette and the initial fluorescence was measured. The titration was performed by adding successive 50 µL aliquots of the 1 solution to the cuvette and recording the spectrum.

Fluorescence titrations of SQ with the solution of CEES treated with 1 (quantitative analysis)
A stock solution of SQ (2.65 µM) was prepared in CHCl₃. A separate stock solution of 1 (0.4 mM) and 3.0 equivalents of K₂CO₃ was also prepared in methanol. Increasing amount of 20 µM of CEES the 220
µM with respect to 1 was reacted at 80 °C for a minute. Each time, this solution was titrated with 2 mL of stock solution of SQ (2.65 µM) in a fluorescence cuvette and the fluorescence was measured.

**Calibration curve**

The fluorescence response of SQ (2.65 µM) with various concentrations of CEES starting from 20 µM to 220 µM was measured in chloroform. In a similar manner as described above, 1 (0.2 mM) was allowed to react with different concentrations of CEES (from 20 µM to 220 µM) at 80 °C for one minute, followed by the addition of SQ. The magnitude of fluorescence intensity depends on the concentration of CEES. The results indicate that saturation point was achieved with the addition of 164 µM of CEES. Ideally, it should have been 200 µM of CEES. We anticipated that some portion of 1 is getting converted into disulfide under given reaction conditions. Consequently, it is not able to react with CEES, thus leaving it unreacted in solution, which leads to the saturation point at 164 µM. In order to achieve a hypothetical saturation point at 200 µM. We have multiplied all the x-axis values by a factor 1.25 and then plotted the calibration curve. This exercise would scale it to 200 µM of the analyte.

**Chromogenic detection of CEES on surfaces using SQ**

CEES (8 µL) was placed on a surface and absorbed by filter paper. This paper was allowed to react with a solution of 1 (1mg, 4.09 mM) in 1 mL of methanol containing 3.0 equivalent of K2CO3 at 80 °C for 1 min. The solution was cooled to room temperature and then 25 µL of it was mixed with a solution of SQ dye (1.2 mL) at 14 µM in chloroform. Same procedure was followed when there is no mustard on the paper. Both these vials were photographed as shown in figure 1.

![Figure 1. Detection of CEES on paper left (no CEES) and right (with CEES)](image)

**Chromogenic detection of CEES in soil using SQ**

2.0 g of soil was mixed with CEES in diethyl ether (2mL) and was allowed to stand for 20 min. The solvent from soil sample was evaporated by nitrogen blow down. CEES spiked soil sample was reacted
with a solution of 1 (1.0 mg, 4.09 mM) in 1 mL of methanol containing 3.0 equivalent of K$_2$CO$_3$ at 80 °C for 1 min. The solution was cooled to room temperature, centrifuged and then mixed with a solution of SQ dye (1.2 mL) at 14 µM in chloroform. This solution was then photographed.

**Chromogenic detection of CEES in gas phase surfaces using SQ**

1 (1mg, 4.09 mM) was dissolved in 1 mL of methanol containing 3.0 equivalent of K$_2$CO$_3$. 200 µL of it was sprayed over (1” X 2.5”) silica coated TLC and it was dried by blowing of air. The treated TLC plate was cut equally into two parts; one part was used for comparison and another part was kept in vapor generation chamber (Figure 2). The vapor generation chamber was placed on the hot place by providing 80 °C at chamber surface. CEES (10 µL) was placed on the heated surface of chamber. Air was passed through the chamber for 3 min. This pretreated TLC plate was taken out from chamber and drop of SQ (30 µM) was placed on the both the TLC plates (treated with and without CEES). Both the TLC plates were photographed together.

![Gas generation chamber](image)

**Figure 2.** Gas generation chamber