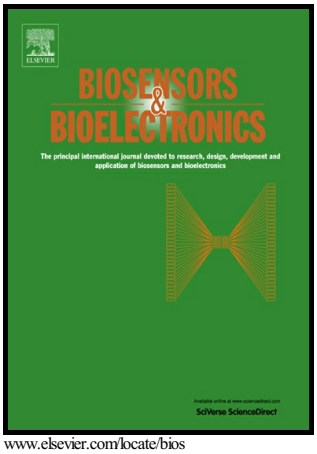

www.elsevier.com/locate/bios

Highly sensitive detection of cysteine over glutathione and homo-cysteine: New insight into the Michael addition of mercapto group to maleimide

Zhaoyang Chen, Qian Sun, Yuhua Yao, Xiaoxiang Fan, Weibing Zhang, Junhong Qian

PII: S0956-5663(17)30013-1
DOI: http://dx.doi.org/10.1016/j.bios.2017.01.013
Reference: BIOS9478

To appear in: *Biosensors and Bioelectronic*

Received date: 15 November 2016
Revised date: 30 December 2016
Accepted date: 6 January 2017

Cite this article as: Zhaoyang Chen, Qian Sun, Yuhua Yao, Xiaoxiang Fan, Weibing Zhang and Junhong Qian, Highly sensitive detection of cysteine ove glutathione and homo-cysteine: New insight into the Michael addition o mercapto group to maleimide, *Biosensors and Bioelectronic* http://dx.doi.org/10.1016/j.bios.2017.01.013

Highly sensitive detection of cysteine over glutathione and homo-cysteine: New insight into the Michael addition of mercapto group to maleimide

Zhaoyang Chen, Qian Sun, Yuhua Yao, Xiaoxiang Fan, Weibing Zhang, Junhong Qian[1*]
Shanghai Key Laboratory of Functional Materials Chemistry, School of Chemistry & Molecular Engineering, East China University of Science & Technology, Shanghai, 200237. China
junhongqian@ecust.edu.cn

Abstract

A fluorescence "off–on" probe CMP for thiols was designed with coumarin as the fluorophore and maleimide as the receptor. The fluorescence of the coumarin was quenched through photoinduced electron transfer (PET) from the fluorophore to maleimide group. The Michael addition of the mercapto group toward maleimide formed a thioether with relatively weak fluorescence. The intramolecular nucleophilic substitution of amino group in cysteine (Cys) to alkylthio produced a much stronger fluorescent amino adduct, which was supported by UPLC-MS and NMR titration. The above sensing mechanism ensured CMP a highly sensitive probe toward Cys over GSH and Hcy. The fluorescence intensity at 495 nm was linear with Cys concentration over the range of 0~10 μM with a detection limit of 14 nM and a rapid response time of 20 min. High selectivity and good competition of the probe toward thiols over other biologically relevant species enabled us to monitor mercapto-containing proteins as well as fluorescence imaging Cys in living cells.

Keywords: Fluorescent probe, Maleimide, Cysteine detection, Sensing mechanism, Bio-imaging application

1. Introduction

Glutathione (GSH), cysteine (Cys) and homo-cysteine (Hcy) are important biological low-weighted thiols in many physiological and pathological processes. They play vital roles in intracellular redox homeostasis through the equilibrium between the free thiols and oxidized disulfides (Kand et al., 2012; Zhang et al., 2004). Abnormal levels of these thiols in biological systems are implicated in many kinds of diseases. GSH serves as the key regulator of the redox state of protein thiols and maintains intracellular redox activities, xenobiotic metabolism, intracellular signal transduction and gene regulation (Dalton et al., 1999). Cys deficiency is reported to be related to slowed growth in children, depigmentation of hair, liver damage, loss of muscle and other health problems (Shahrokhian, 2001). Abnormal levels of intracellular Hcy are involved in cardiovascular and Alzheimer's diseases (Seshadri et al., 2002). Therefore, selective and sensitive detection of thiols is of great significance.

[1] Tel: +86-21-64252331; Fax: +86-21-64233161