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Microwave assisted synthesis of tyrosine protected gold nanoparticles for dual (colorimetric and fluorimetric) detection of spermine and spermidine in biological samples

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ABSTRACT

In this work, tyrosine-protected gold nanoparticles (Tyr-Au NPs) were fabricated by one-step reduction of Au^{3+} ion using Tyr as a reducing and capping agent under microwave irradiation. The Tyr-Au NPs were successfully used as a dual probe for colorimetric and fluorescence turn-on assays of spermine and spermidine in biological samples. Upon addition of spermine and spermidine, the characteristic surface plasmon resonance (SPR) band of Tyr-Au NPs was red-shifted to 596 and 616 nm and the emission peak (Tyr) at 410 nm was gradually increased with increasing concentration of both analytes, confirming the aggregation of Tyr-Au NPs induced by spermine and spermidine, which results to restore fluorescence of Tyr on the surfaces of Au NPs. In addition, it shows high selectivity for sensitive detection of prostatic cancer biomarkers spermine and spermidine in real clinical applications with reduced sample preparations.

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1. Introduction

Biogenic polyamines, such as spermine (*N,N′*-bis(3-aminopropyl)butane-1,4-diamine) and spermidine (*N′*-(3-aminopropyl)butane-1,4-diamine), naturally exist in all eukaryotic cells and play important roles in cell growth and proliferation, including the regulation of gene expression, the prevention of endonuclease-mediated DNA fragmentation, and the inhibition of DNA damage, respectively (Seiler et al., 1996). Importantly, spermine and spermidine levels are regulated by multiple pathways, and their levels may be indicative of abnormal biological processes associated with cancer (Clarke et al., 2010). Therefore, elevated levels of spermine and spermidine have been recognized as cancer biomarker for monitoring of the growth of tumors in many types of cancer (van der Graaf et al., 2009). Hence, it is particularly important to develop new health diagnostic tool for selective and sensitive detection of spermine and spermidine in biological samples with reduced samples volumes.

During the past decade, a number of typical analytical techniques has been applied to determine spermine and spermidine in biological samples, including gas chromatography (Niitsu et al., 1993), high performance liquid chromatography (HPLC) (Raza and Al-Shabanah, 2010), capillary electrophoresis (Liu et al., 2003 and Steiner et al., 2009), HPLC coupled with mass spectrometry (Sirocchi et al., 2014) and matrix-assisted laser desorption/ionization mass spectrometry (Sua et al., 2015), respectively. Even though these methods show well ability to detect both spermine and spermidine in complex samples, unfortunately these require tedious sample preparations, and some of them are expensive.

Furthermore, spermine and spermidine are aliphatic in nature and shown neither natural UV absorption nor fluorescence. However, several optical-based assays have been described for the detection of both analytes *via* spectral changes using various derivatization reagents, including small molecule chromophores (Lee et al., 2011), organic polymers (Satrijo and Swager, 2007), biogenic amine (Maynor et al., 2007) and coordination compounds (Chow et al., 2013), respectively. Recently, Fletcher and Bruck used a dicarboxylated ethynylarene as a fluorescent chemosensor for the detection of polyamines in the presence of metal ions (Fletcher and Bruck, 2015). Kostereli and Severin developed amphiphile-based chemosensor for the fluorescence detection of spermine (Köstereli and Severin, 2012). Unfortunately, these methods frequently report results as either non-detectable or higher detection limits. Thus, efficient cleanup and pre-concentration techniques are essentially needed to detect of both analytes with high sensitivity and selectivity.

In recent years, Au NPs with well-defined nanostructure have

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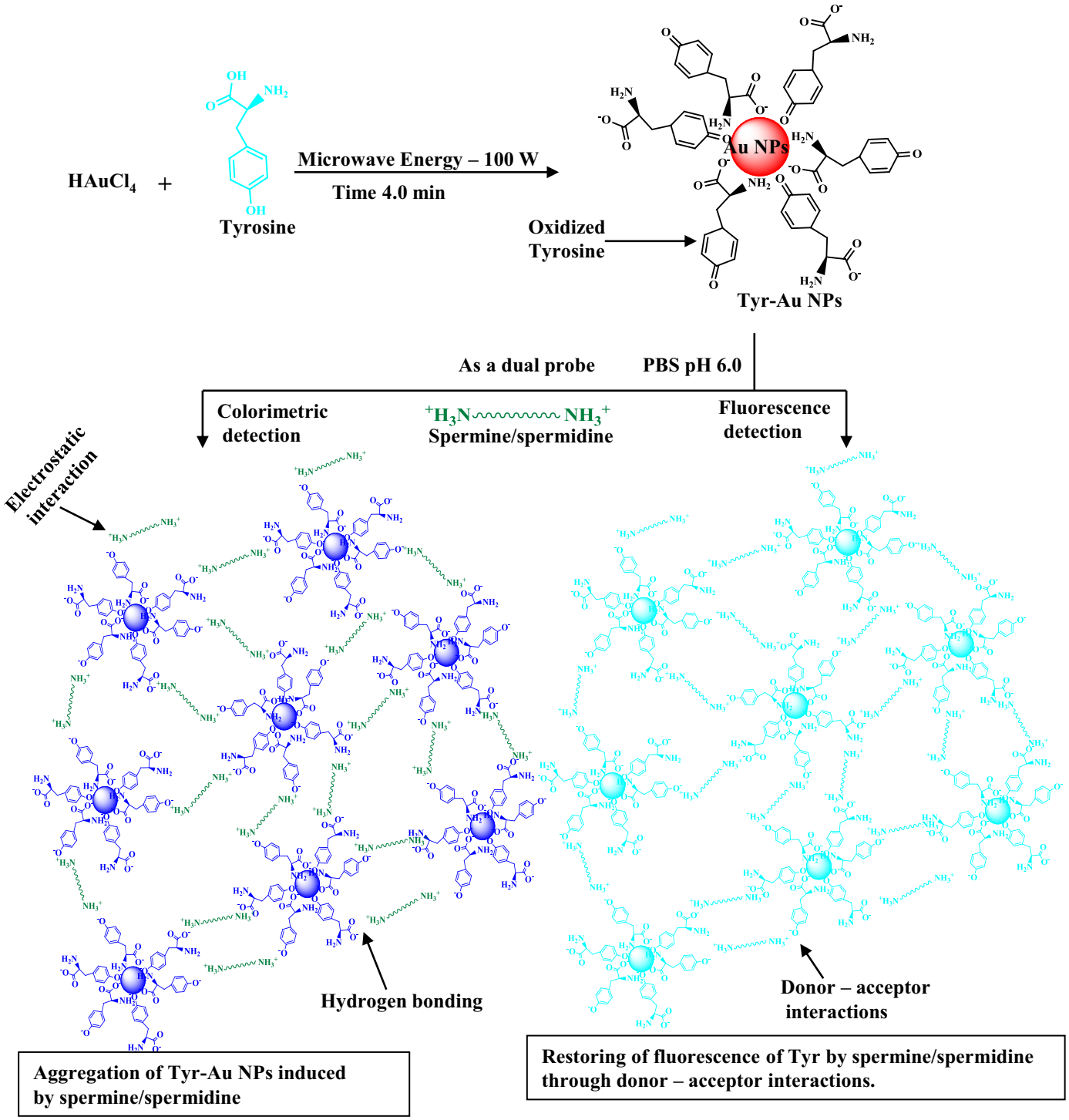
emerged as prompt visual reporters and received great attention to use them as probes for assay of a wide variety of chemical species owing to their efficient integration of the unique optical properties and the excellent surface/interface recognition ability afforded from the rational design of their surface chemistry (Saha et al., 2012; Dykman and Khlebtsov, 2014; Zhou et al., 2015). In general, the simplest way of synthesizing Au NPs is to reduce chloroauric acid by using various organic molecules as reducing agents (Yeh et al., 2012). To alter the applications of Au NPs, the surface functionalization of Au NPs should be performed for improving various factors such as biocompatibility, reduce toxicity, and fabrication of Au NPs with molecular probes for assaying of various biospecies. As a result, various organic molecules such as Tyr (Daima et al., 2014; Selvakannan et al., 2004), cyclodextrins (Jin et al., 2015), gallic acid (Yoosaf et al., 2007), poly(*N*-vinyl-2-pyrrolidone) (Hoppe et al., 2006), pyrogallol acid (Jiang and Yu, 2010), 4-amino-3-(d-gluco-pentitol-1-yl)-4,5-dihydro-1,2,4-triazole-5-thione (Salman et al., 2012), *N*-(2-hydroxyethyl) -*N*-methyl morpholinium tetrafluoroborate (Kim et al., 2006), chitosan (Bhumkar et al., 2007), alkanethiol (Farrell et al., 2013), *N*-acetylglucosamine (Song et al., 2012), histidine (Zhao and Huang, 2014), ampicillin (Hur et al., 2014), 4-amino antipyrine (Rawat



Scheme 1. Dual (colorimetric and fluorimetric) sensing mechanism for assay of spermine and spermidine using Tyr-Au NPs as a probe.

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