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Sensitive turn-on fluorescent detection of tartrazine based on fluorescence resonance energy transfer

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**Materials and Chemicals**

Graphite powders (325 mesh, spectral pure) were obtained from Sinopharm Chemical Reagent Co., Ltd., China. Tartrazine (Tz) was from TCI (Shanghai, China) Development Co., Ltd. The stock solution of Tz \((5.10 \times 10^{-5} \text{ g mL}^{-1})\) was prepared by directly dissolving the commercial product in doubly distilled water and the required concentration of working solution was obtained by diluting the stock solution with doubly distilled water. Fluorescein (Fl) was commercially purchased from Shenyang No. 3 Chemical Reagent Factory (Shenyang, China). A \(4.40 \times 10^{-5} \text{ g mL}^{-1}\) stock solution of Fl was prepared by dissolving solid Fl in ethanol and the working solution was obtained by diluting the stock solution to \(2.20 \times 10^{-7} \text{ g mL}^{-1}\). All other chemicals not mentioned here are of analytical reagent grade and are used as received. Water used throughout was doubly distilled.

**Preparation of Reduced Graphene Oxide**

Graphene oxides (GO) were obtained through natural graphite powder according to the Hummers method,\(^1\) and reduced graphene oxides (rGO) were synthesized by the chemical reduction of GO with hydrazine.\(^2\) The successful synthesis of rGO has been characterized in our previous work.\(^3\) 35.91 \(\mu\)g mL\(^{-1}\) rGO dispersion was used.

**Pretreatment of Sample**

A suitable amount of lemon-flavored sports drink was filtered through a 0.2 \(\mu\)m pore size millipore filter, then 5.5 mL of the filtrate was extracted with ethyl acetate to get rid of some unknown food additives and it was repeated three times. Whereafter, the aqueous solution containing tartrazine was pretreated according to a salting-out assisted liquid–liquid
extraction method. Briefly, 4.5 mL of acetonitrile was added in it for formation of a homogeneous solution. Then 2.500 g of (NH₄)₂SO₄ was added, the mixture solution was shaken until the salt was dissolved, and then it was separated into two clear phases. The target analyte in the sample was extracted into the extraction solvent. Extraction was repeated three times. The extracts were then combined and heated to remove acetonitrile. Then the resulting residue was diluted to 5.0 mL with doubly distilled water for the determination of Tz.

Detection of Tartrazine

150 µL of 0.20 mol L⁻¹ HAc-NaAc buffer solution, pH 5.03, 300 µL of 220.00 ng mL⁻¹ Fl and 40 µL of 35.91 µg mL⁻¹ rGO solution were mixed thoroughly, then 50 µL of working solution containing different concentrations of Tz was added, and the mixture was incubated at room temperature for 1 min. The fluorescence intensities (at 520 nm) of the mixture in the presence of Tz (I) and absence of Tz (I₀) were recorded, respectively. The fluorescence spectra of the mixture were recorded using a Hitachi F-2700 spectrofluorophotometer (Hitachi Ltd., Tokyo, Japan).
**Fig. S1** Fluorescence emission spectra: (A) DCFI (a), DCFI + rGO (b) and DCFI + rGO + Tz (c). Concentrations: DCFI (128.68 ng mL$^{-1}$), Tz (236.11 ng mL$^{-1}$), and rGO (1.09 μg mL$^{-1}$). 0.20 mol L$^{-1}$ HAc–NaAc buffer, pH 5.03; reaction time: 1 min, excitation: 495 nm. (B) EY (a), EY + rGO (b) and EY + rGO + Tz (c). Concentrations: EY (129.16 ng mL$^{-1}$), Tz (236.11 ng mL$^{-1}$), and rGO (0.44 μg mL$^{-1}$). 0.20 mol L$^{-1}$ HAc–NaAc buffer, pH 5.03; reaction time: 1 min, excitation: 500 nm.
**Fig. S2.** (A) The fluorescence spectra of Fl and Tz. The concentrations of Fl and Tz were 122.22 ng mL\(^{-1}\) and 1.59 μg mL\(^{-1}\), respectively. (B) Effect of Tz concentration on the fluorescence of Fl. The concentration of Fl was 122.22 ng mL\(^{-1}\), 0.20 mol L\(^{-1}\) HAc–NaAc buffer, pH 5.03, reaction time: 1 min, excitation: 480 nm, emission: 520 nm.
**Fig. S3.** Effect of rGO concentration on the fluorescence of Fl in the rGO-Fl system. The concentrations: Fl (122.22 ng mL$^{-1}$) and rGO (35.91 µg mL$^{-1}$). 0.20 mol L$^{-1}$ HAc–NaAc buffer, pH 5.03, reaction time: 1 min, excitation: 480 nm, emission: 520 nm.

**Fig. S4.** Effect of the buffer volume on the FEE. Concentrations: Fl (122.22 ng mL$^{-1}$), Tz (212.50 ng mL$^{-1}$), and rGO (2.66 µg mL$^{-1}$), 0.20 mol L$^{-1}$ HAc–NaAc buffer, pH 5.03, reaction time: 1 min, excitation: 480 nm, emission: 520 nm.
**Fig. S5.** Comparison of FEE for the five different reagent addition orders. buffer-Fl-Tz-rGO (a), buffer-Fl-rGO-Tz (b), buffer-Tz-rGO-Fl (c), Tz-Fl-rGO-buffer (d), and buffer-rGO-Fl-Tz (e). Concentrations: Fl (122.22 ng mL\(^{-1}\)), Tz (212.50 ng mL\(^{-1}\)), and rGO (2.66 µg mL\(^{-1}\)), 0.20 mol L\(^{-1}\) HAc–NaAc buffer, pH 5.03, reaction time: 1 min, excitation: 480 nm, emission: 520 nm.

**Fig. S6** Kinetic study for the fluorescence change of the Fl-rGO sensor in the presence of different concentrations of Tz: 0 (1), 18.89 (2), 61.39 (3), 118.06 (4), 188.89 (5) and 212.50 (6) ng mL\(^{-1}\). Fl (122.22 ng mL\(^{-1}\)) and rGO (2.66 µg mL\(^{-1}\)), 0.20 mol L\(^{-1}\) HAc–NaAc buffer, pH 5.03, excitation: 480 nm, emission: 520 nm.
Table S1  Analytical results for Tz in lemon-flavored sports drink sample (n = 5)a.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Found value (ng mL⁻¹)</th>
<th>Standard added (ng mL⁻¹)</th>
<th>Found value (ng mL⁻¹)</th>
<th>Recovery (%)</th>
<th>RSD (%)</th>
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<td>103.89</td>
<td>153.04</td>
<td>102.6</td>
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</tr>
</tbody>
</table>

a 2.00 mL of pretreated lemon-flavored sports drink sample was diluted 5-fold to detect Tz according to the procedure. Concentrations: F1 (122.22 ng mL⁻¹), and rGO (2.66 μg mL⁻¹). 0.20 mol L⁻¹ HAc–NaAc buffer, pH 5.03, excitation: 480 nm, emission: 520 nm.

Supplementary references: