Electronic Supplementary Material (ESI) for Photochemical & Photobiological Sciences

Environment-Sensitive Amphiphilic Fluorophore For Selective Sensing of Protein

Bimlesh Ojha, Gopal Das*

Department of Chemistry, Indian Institute of Technology Guwahati, Assam – 781 039, India

E-mail: gdas@iitg.ernet.in; Tel: +91 361 2582313; Fax: +91 361 2582349

Supplementary Information

Characterization. NMR spectra were recorded on a Varian FT-400 MHz instrument. The chemical shifts were recorded in parts per million (ppm) scale using tetramethylsilane (TMS) as a reference at 298 K.

Compound 1 [8-(butoxy)quinoline]: ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.009 (t, 3H), 1.559 (m, 2 H), 2.019 (m, 2H), 4.238 (t, 2H), 7.056 (m, ArH), 7.409 (m, ArH), 8.114 (m, ArH), 8.941 (m, ArH). ^{13}C NMR (100 MHz, $CDCl_3$): δ 13.939, 19.331, 30.981, 68.612, 108.506, 119.289, 212.463, 126.823, 129.440, 135.823, 140.376, 149.843 and 154.843. Light brown color solid, Melting point 48 ^{o}C.[1]

Compound 2 [8-(octyloxy)quinoline]: ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.854 (t, 3H), 1.328 (m, 10H), 1.478 (t, 2H), 2.007 (t, 2H), 4.205 (m, ArH), 7.032 (m, ArH), 7.384 (m, ArH), 8.094 (m, ArH), 8.921 (m, ArH). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.137, 22.671, 26.072, 28.978, 29.245, 29.435, 31.830, 68.940, 108.513, 119.305, 121.493, 126.702, 129.455, 135.900 149.208 and 154.813. Deep brown color semi solid at RT.[2]

Compound 3 [8-(doecyloxy)quinoline]: ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.837 (t, 3H), 1.178 (m, 18 H), 1.501 (m, 2H), 4.188 (t, 2H), 7.013 (m, ArH), 7.350 (m, ArH), 8.071 (m, ArH), 8.903 (m, ArH). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.244, 22.816, 26.202, 26.339, 26.393, 29.123, 29.703, 29.764, 29.794, 32.051, 69.161, 108.750, 119.457, 121.623, 126.817, 129.638, 135.999, 149.383 and 155.042. Deep brown color semi solid at RT.[2]

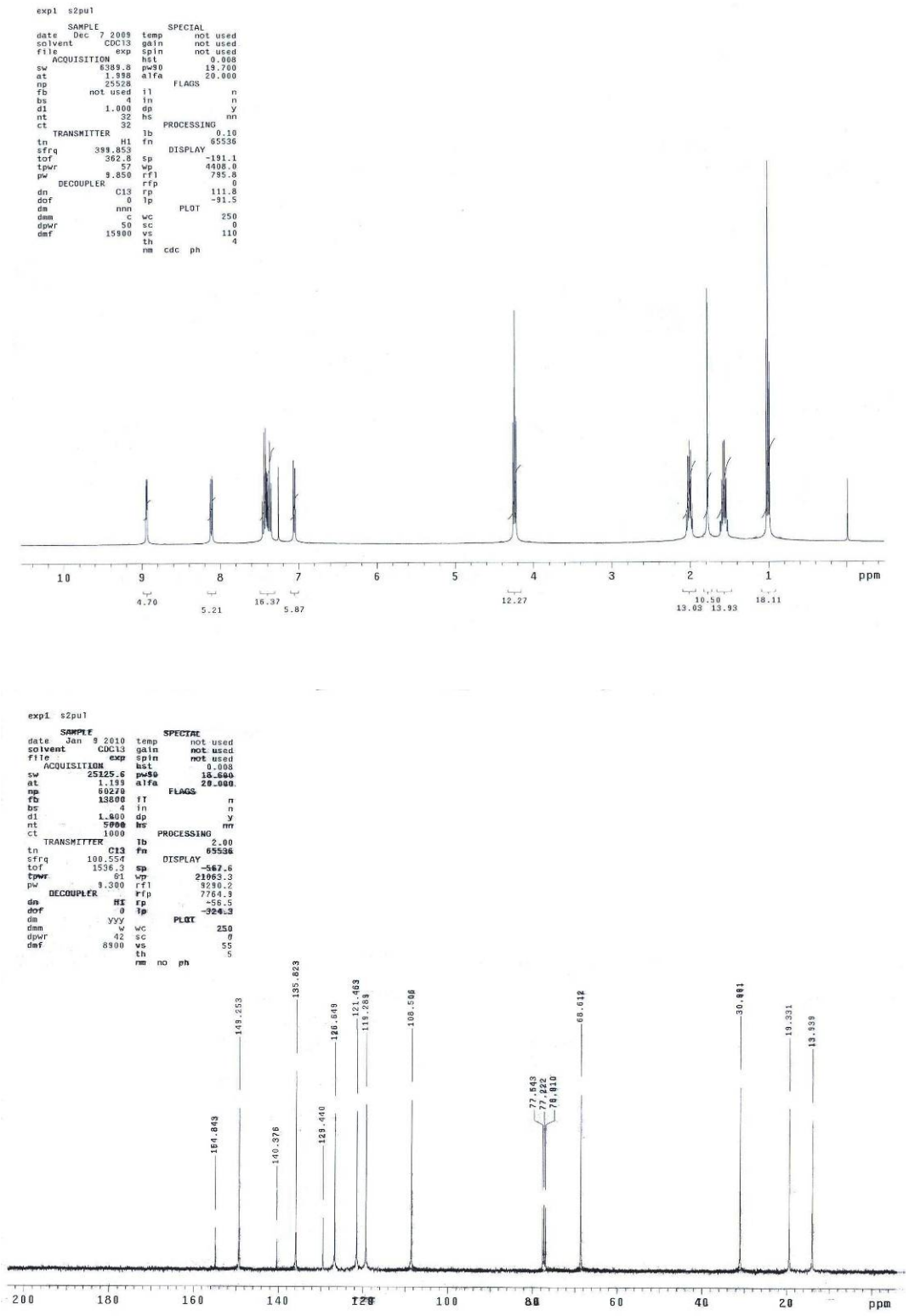

Figure S1. ^{1}H and ^{13}C NMR spectra of Compound **1**.

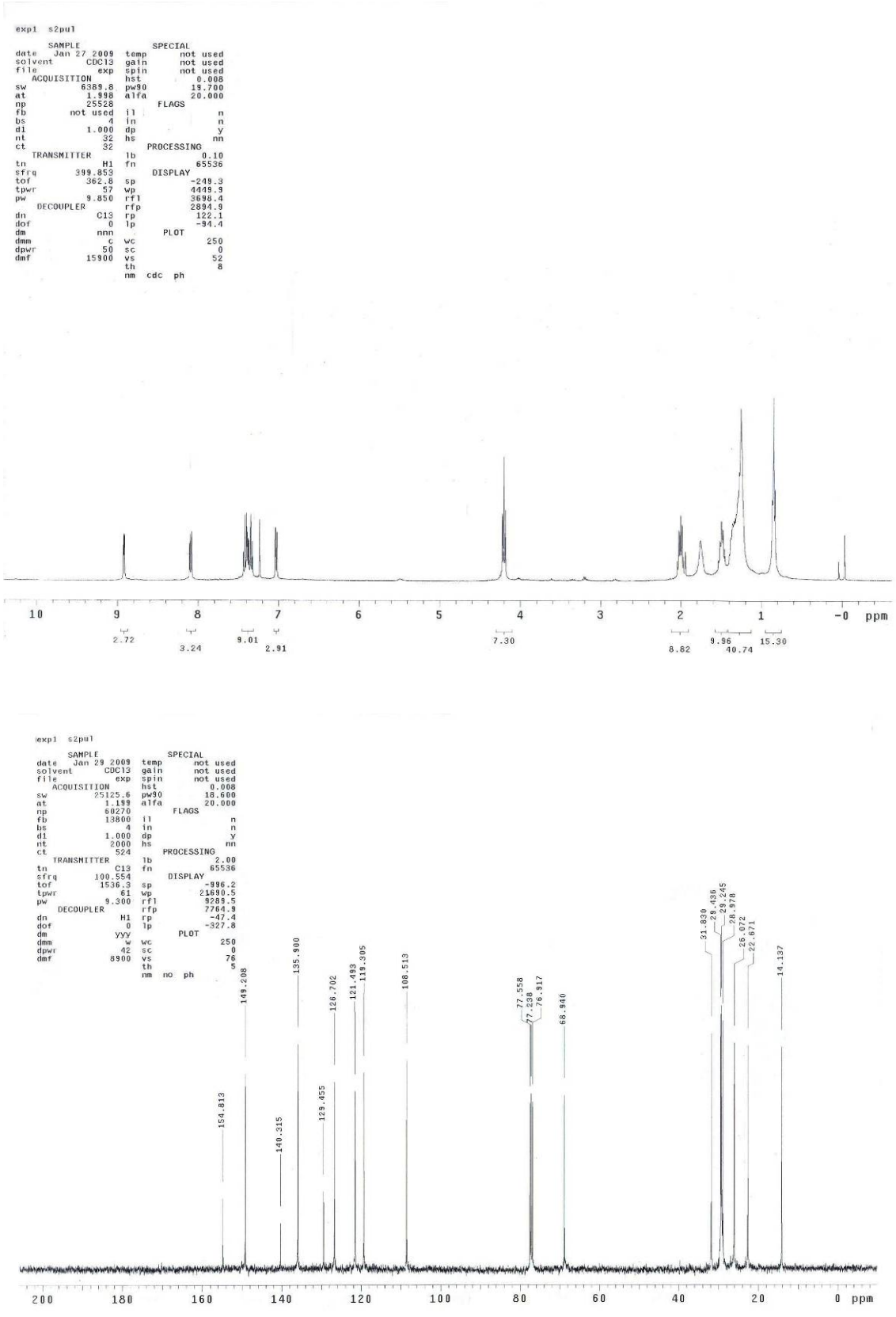

Figure S2. ^{1}H and ^{13}C NMR spectra of Compound **2**.

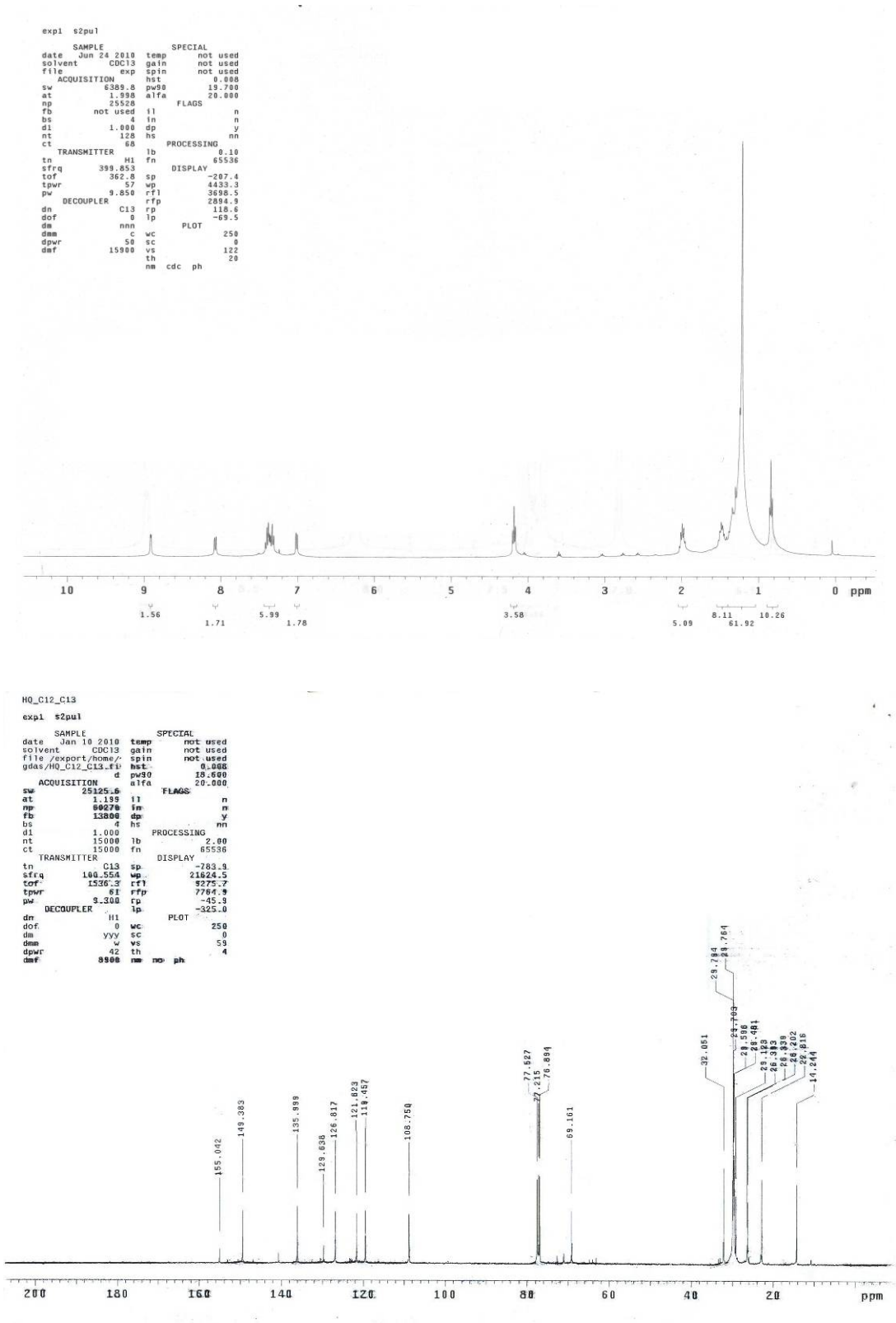

Figure S3. ^{1}H and ^{13}C NMR spectra of Compound **3**.

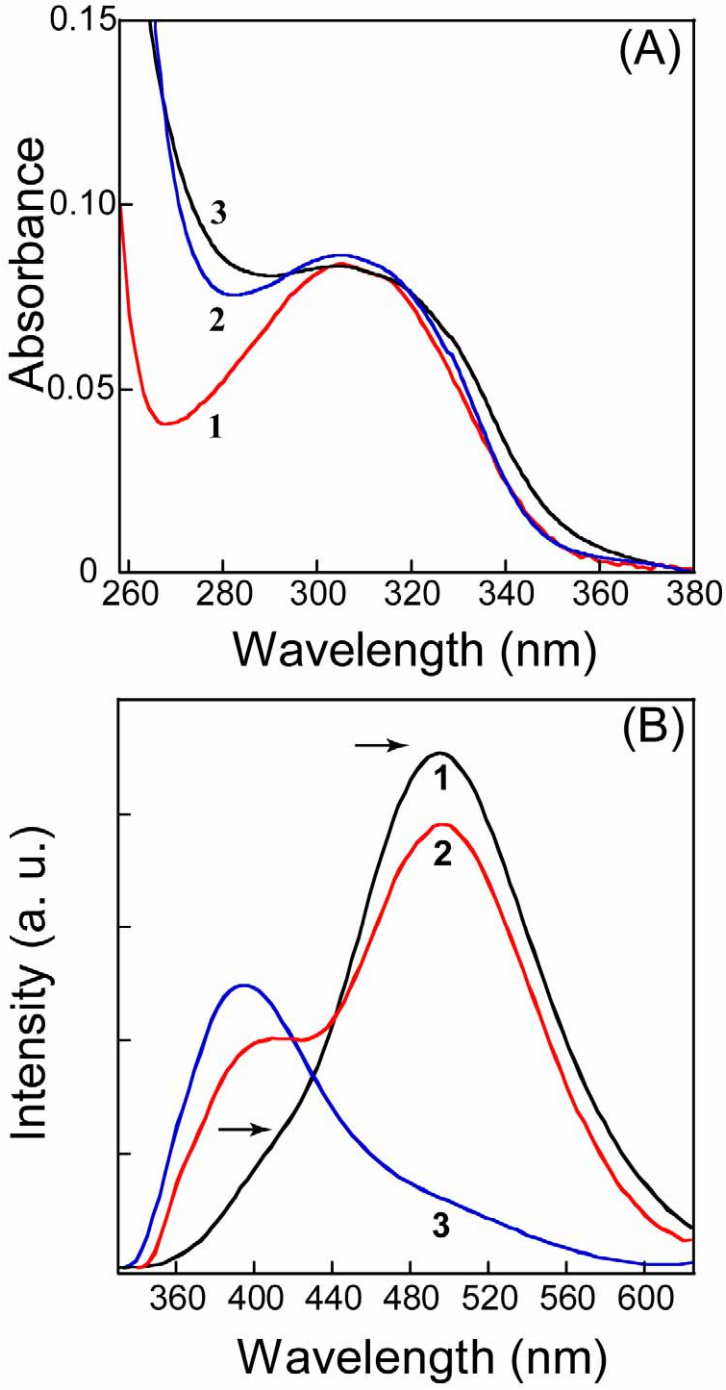

Figure S4. (A) UV-visible absorption and (B) Emission spectra of the compounds (**1**-**3**) in an aqueous buffer solution of pH 7.0.

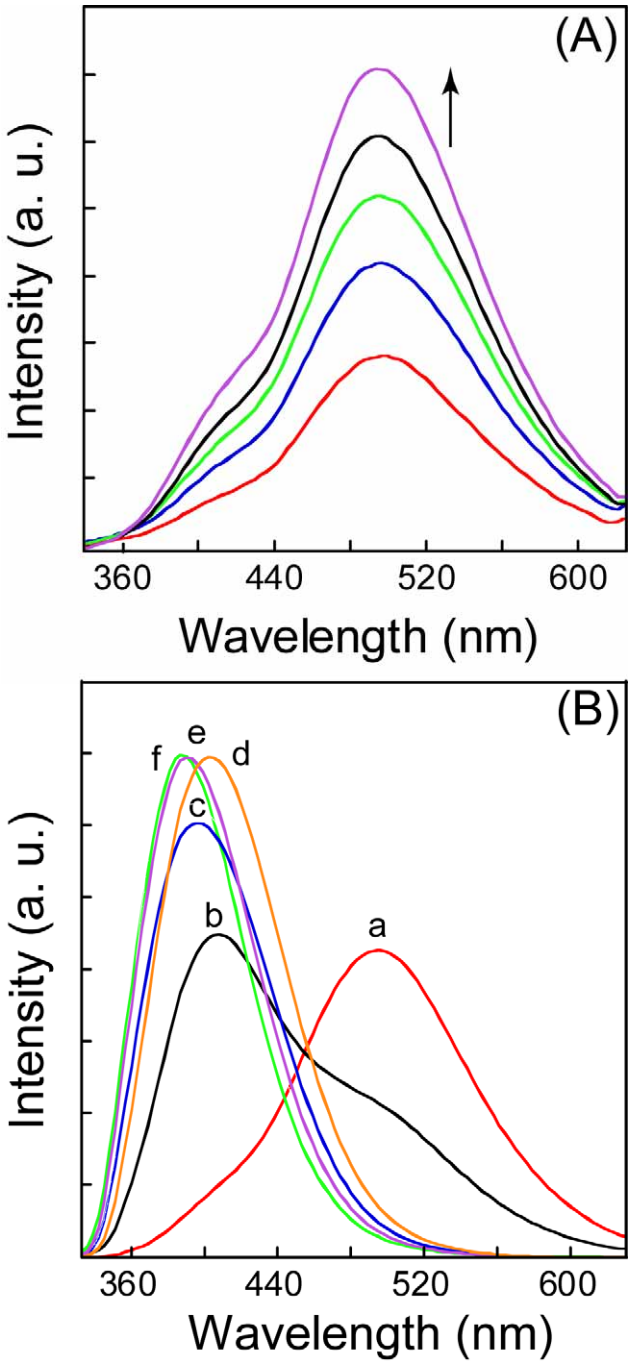

Figure S5. (A) Emission spectra of Compound **1** with increasing concentration from 0 to 50 μM and (B) Effect of different solvent on the emission spectra of the compound **1**; Where Trace a-f: water, methanol, DMSO, acetonitrile, THF and hexane.

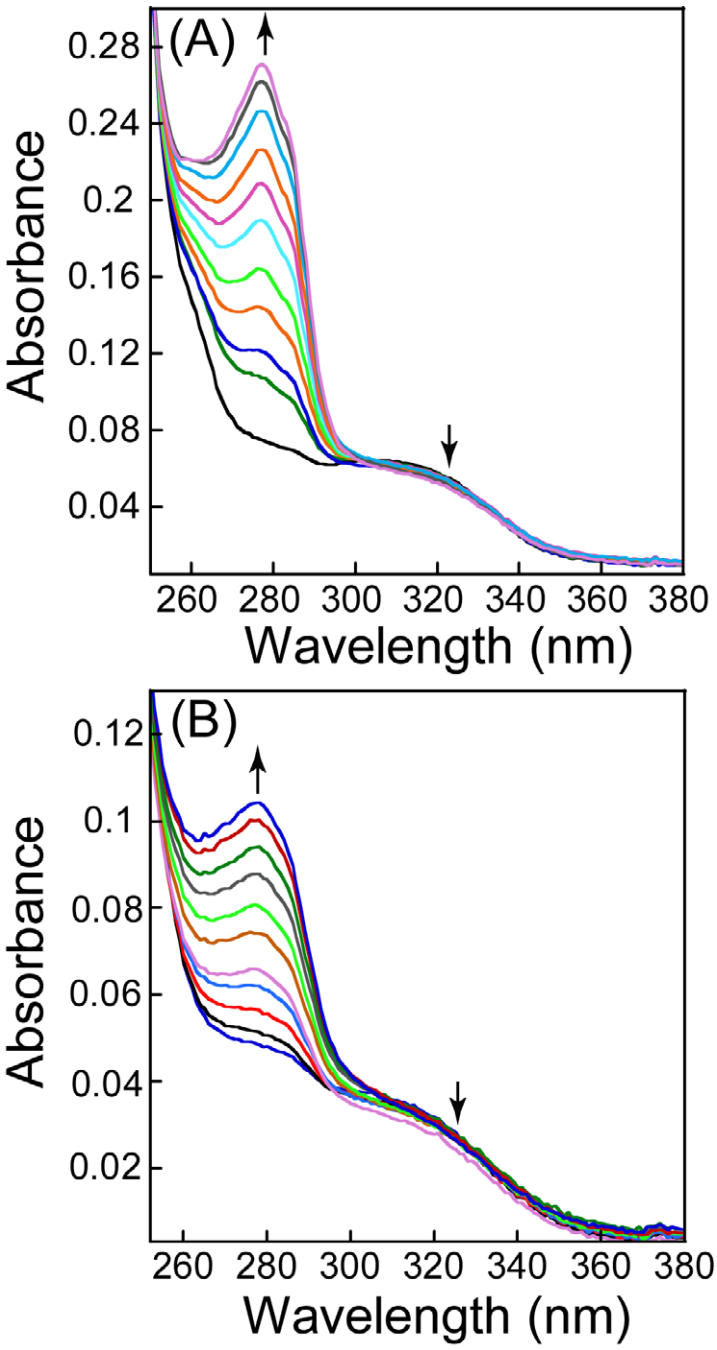

Figure S6. Absorption spectra of (A) Compound **2** and (B) Compound **3** as a function of the BSA concentration ranging from 0 to 100 μg/mL.

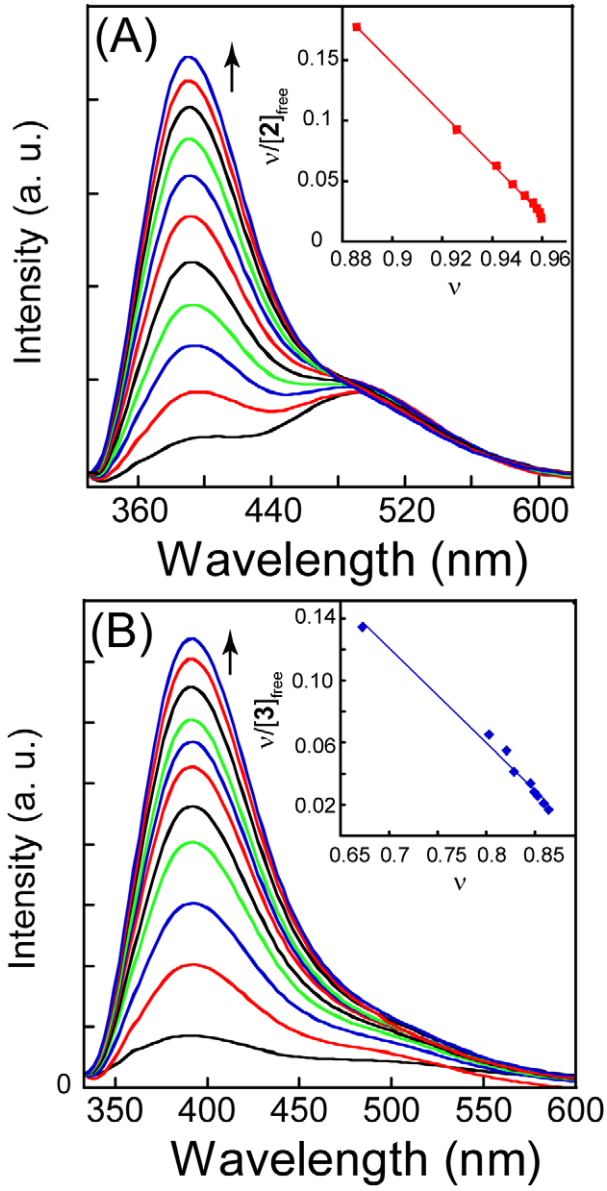

Figure S7. Emission spectra of (A) Compound **2** and (B) Compound **3** with increasing concentration of BSA ranging from 0 to 500 µg/mL in an aqueous buffer of pH 7.0. The compound concentration used was 50 µM and the λ_{exc} was 320 nm and the inset showing the Schatard plot for binding constant.

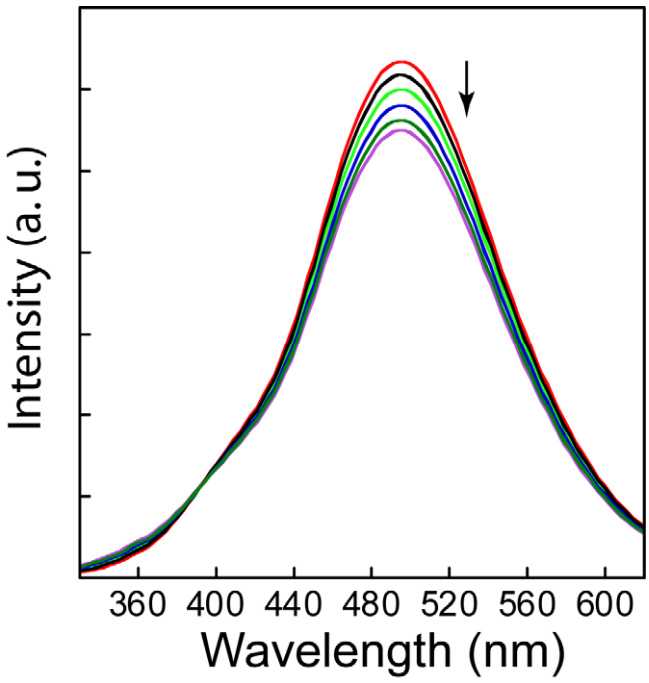

Figure S8. (A) Emission spectra of Compound **1** with increasing concentration of tryptophan amino acid from 0 to 500 µg/mL in an aqueous buffer of pH 7.0.

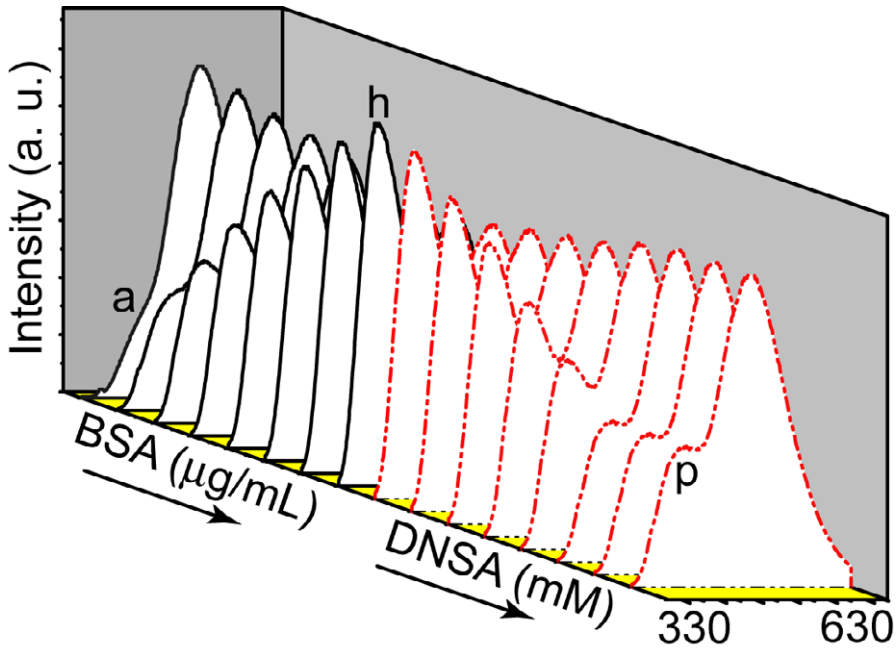

Figure S9. Emission spectra of Compound **1** (50 µM) with increasing concentration of BSA from 0 to 500 µg/mL (solid black, a-h) and with increasing concentration of DNSA dotted (red, i-p) in an aqueous buffer of pH 7.0.

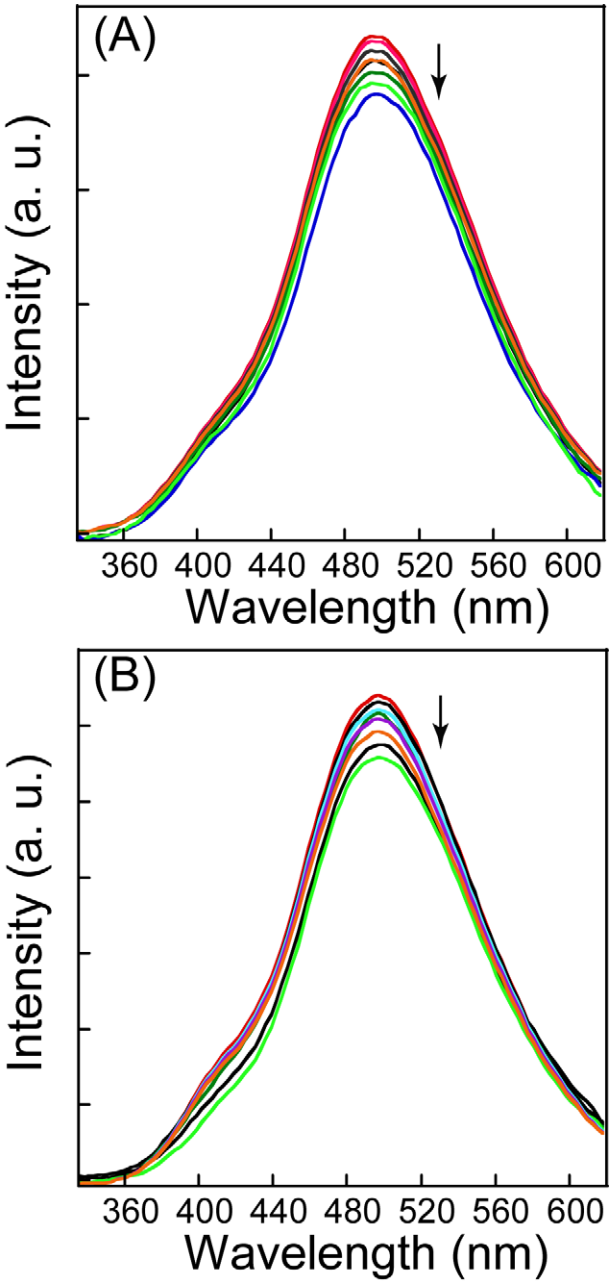

Figure S10. Emission spectra of (A) Compound **1** with increasing concentration of lysozyme (0 to 500 µg/mL) and (B) Compound **1** with increasing concentration of amylase (0 to 500 µg/mL) in an aqueous buffer of pH 7.0.

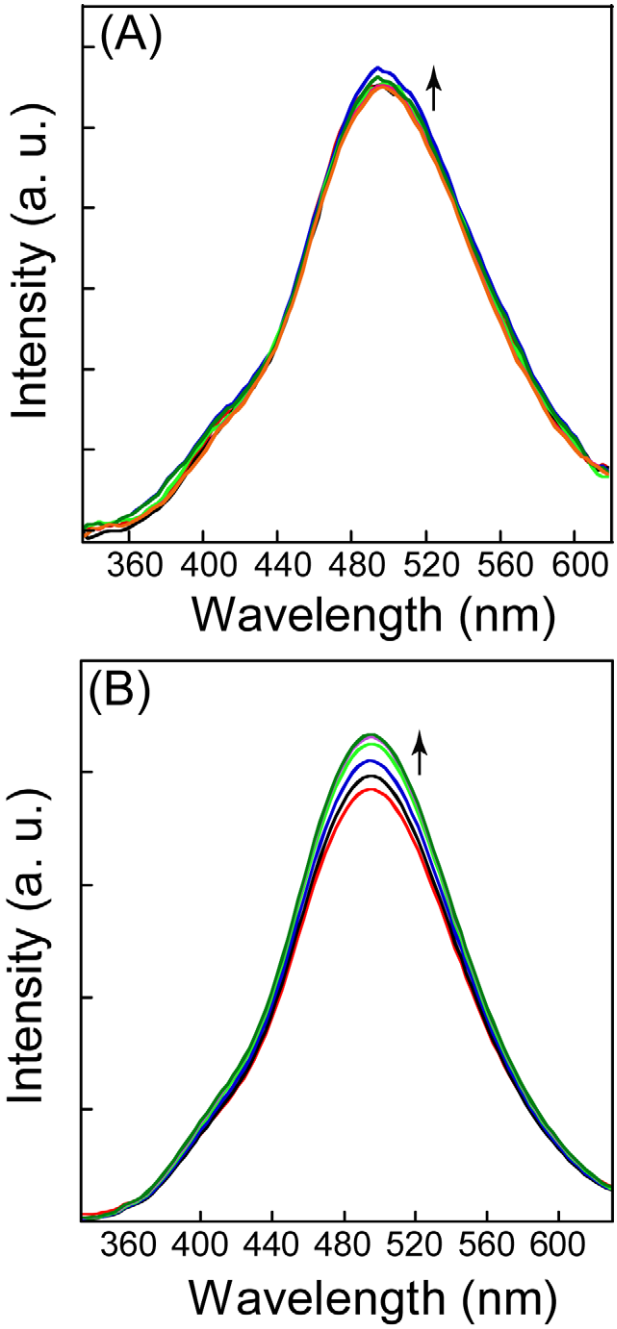

Figure S11. Emission spectra of (A) Compound **1** with increasing concentration of AMG (0 to 500 µg/mL) and (B) Compound **1** with increasing concentration of proteinase K (0 to 500 µg/mL) in an aqueous buffer of pH 7.0. The compound concentration used was 50 µM and the λ_{exc} was 320 nm.

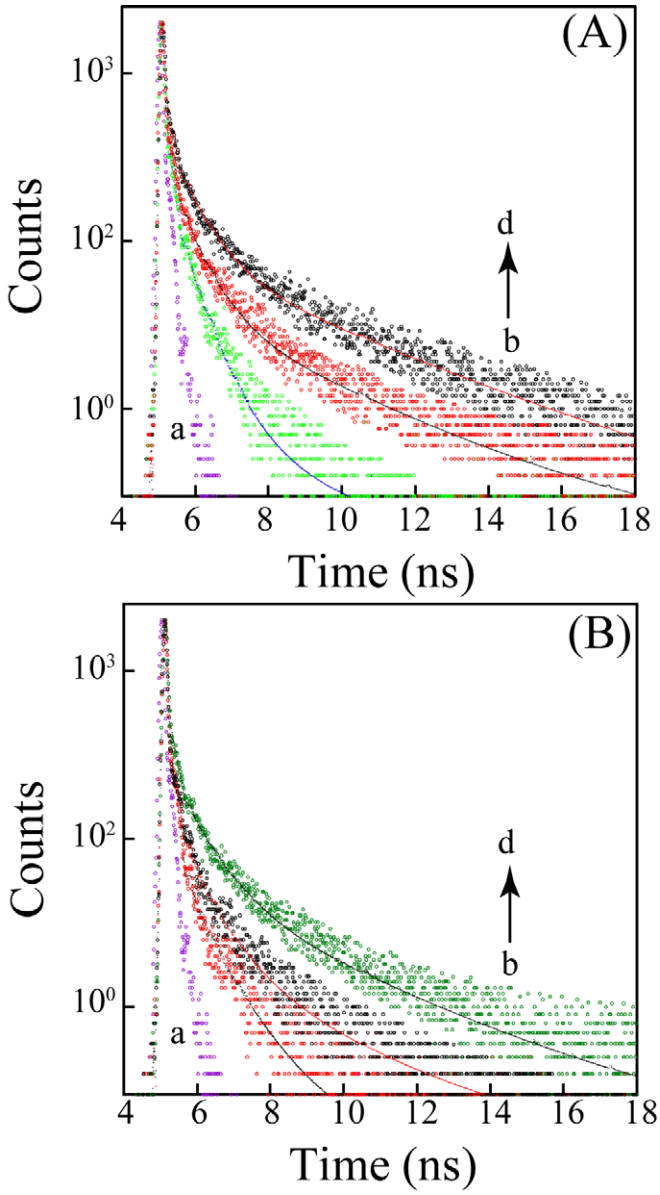

Figure S12. Time resolved fluorescence spectra of (A) Compound **1** and (B) Compound **2** in absence and in presence of BSA; Where Trace a: IRF, Trace b-d: Compound alone, Compound + BSA (250 µg/mL) and Compound + BSA (500 µg/mL) in an aqueous buffer of pH 7.0.

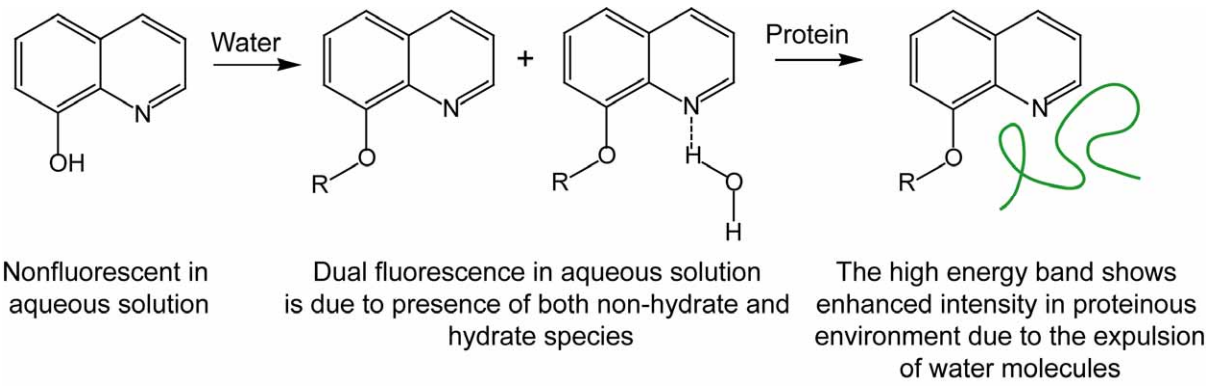

Figure S13. Photophysical responses of compound under different experimental conditions.

Table S1. Physical properties of the different proteins/enzymes.[a]

Protein/Enzyme	Molecular weight (Da)	No. of amino acid residues	No. of aliphatic side chains (Ala, Ile, Leu & Val)	Aliphatic Index[b]
BSA	69323.4	607	Ala-47, Ile-15, Leu-65 & Val-38	77.30
Amylase	55345.1	496	Ala-31, Ile-24, Leu-25 & Val-42	69.33
Proteinase K	28934.9	129	Ala-33, Ile-11, Leu-14 & Val-19	66.52
Lysozyme	14300.1	279	Ala-11, Ile-5, Leu-9 & Val-7	66.59
AMG	12283.3	112	Ala-7, Ile-5, Leu-6 & Val-9	67.86

[a]Walker, John M. *The Proteomics Protocols Handbook*; Humana Press Inc: Totowa, NJ, 2005; [b]Ikai, A. J. *J. Biochem.* **1980**, *88*, 1895.

Table S2. Photophysical Properties of Compounds[a] in Presence of BSA

Sample	τ_1 (ns)	τ_2 (ns)	τ_3 (ns)	α_1	α_2	α_3	τ_m (ns)
1	0.016	0.434	3.554	0.604	0.026	0.004	0.056
2	0.009	0.385	1.134	1.143	0.869	0.002	0.016
3	0.004	0.644	2.760	1.743	0.005	0.004	0.012
1 + BSA[b]	0.053	0.828	5.262	0.194	0.024	0.007	0.297
1 + BSA[c]	0.087	1.087	5.831	0.133	0.026	0.007	0.483
2 + BSA[b]	0.010	0.964	3.292	1.075	0.007	0.001	0.018
2 + BSA[c]	0.013	0.748	4.699	0.836	0.010	0.003	0.038
3 + BSA[b]	0.005	1.697	3.329	1.652	0.003	0.003	0.014
3 + BSA[c]	0.012	0.882	4.121	0.878	0.006	0.002	0.028

[a][Compound]: 50 μM, [b][BSA]: 250 μg/mL and [c][BSA]: 500 μg/mL

References

(1) Shchukina, M. N.; Savitskaya, N. V. S. Ordzhonikidze *All-Union Chem. Pharm. Inst.* Moscow, USSR. Zhurnal Obshchei Khimii, 1952, **22**, 1218.

(2) Andre, L.; Jean, S.; Farchid, V. Z. *Bull. Soc. Chim. Fr.* 1987, **6**, 1027.