Supplementary Information

Single-stranded DNA probe paired aptasensor with extra dye binding sites to enhance its fluorescence response in the presence of target compound

Seo Won Cho,[1,2], Hyun Jeong Lim[1], Beelee Chua[3,], Ahjeong Son[1,**]*

[1] Department of Environmental Science and Engineering, Ewha Womans University, Seoul 03760, Republic of Korea

[2] Department of Civil and Environmental Engineering, University of Wisconsin-Madison, Madison, WI, 53706, USA

[3]School of Electrical Engineering, Korea University, Seoul 02841, Republic of Korea

*Corresponding Author (B. Chua): 145 Anam-ro, Seongbuk-gu, Korea University, Seoul 02841, Republic of Korea; E-mail. bchua@korea.ac.kr, chuabeelee@gmail.com; Phone. +82 (2) 3290-4639

**Corresponding Author (A. Son): 52 Ewhayeodae-gil, Seodaemun-gu, Ewha Womans University, Seoul 03760, Republic of Korea; E-mail. ason@ewha.ac.kr, ahjeong.son@gmail.com; Phone. +82 (2) 3277-3339; Fax. +82 (2) 3277-3275

Table S1. Environmental concentration (environmentally relevant level) of nonylphenol

Occurrence	Concentrations
Sewage treatment plants	USA Sludge anaerobically stabilized: 754 mg/kg Heat treated sludge: 496 mg/kg Limed sludge: 470 mg/kg Composted sludge: 64 mg/kg
	Sludge anaerobically stabilized: 1100–1800 mg/kg mg/kgmg/kg
	Japan Primary effluent: N.D Secondary effluent: 0.3 μg/l Final effluent: 0.2 μg/l
	Raw influent: 0.1–0.9 μg/l Final effluent: 0.5–1.1 μg/l
River water	0.7 ng/L - 15 μg/L (seasonal variations with higher concentrations in the summer due to an increase in microbial activity at warmer temperatures leading to an enhanced degradation of nonylphenol ethoxylates)
The discharge of STW effluents	~ 790 ng/L in average
In the vicinity of contaminated rivers	0.1–0.8 mg/L
Septic systems	1.2 g/L
Agricultural activities	0.16–0.38 μg/L
Landfill leachate and the discharge of industrial wastewater	< 100–280 ng/L

Sensor selectivity test

Methods: The selectivity of the aptasensor for nonylphenol detection was examined by using 4 nonyl phenol analogs (Table S2), namely, 4-n-octylphenol (OP), 4,4-biphenol, bisphenol A (BPA), and Atenolol. All chemicals were purchased from Sigma-Aldrich. All chemicals (500 mg/L) including nonylphenol as a positive control were subjected to further experiment with the aptasensor. Ultrapure distilled water was used as the negative control. All samples were prepared in triplicate.

Results and Discussion: As shown in Figure S1, all 4 nonylphenol analogs have shown higher normalized fluorescence as compared to that of nonylphenol (NP). The result, depicted by arrow and the square in Figure S1, showed that the fluorescence signal dramatically decreased from negative control to NP. The result also indicates that the developed aptasensor has a higher selectivity for NP over other NP analogs. In Table S2 that listed the structural characteristics of 5 tested chemicals, 4-n-octylphenol (OP) has the most similar structure as the target nonylphenol. Therefore, the fluorescence signal of OP (0.061±0.025) was slightly higher than that of NP (0.034±0.006), in contrast to that of the negative control (0.149±0.039).

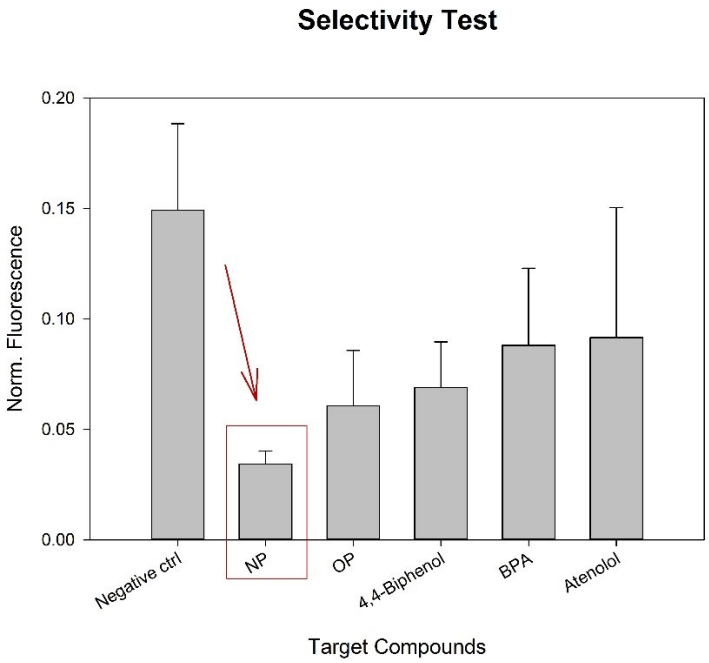
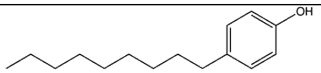
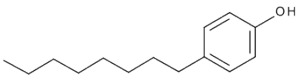
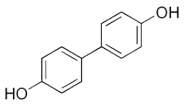
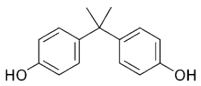


Figure S1. Selectivity verification of aptasensor for nonylphenol (NP) detection by using 4 nonyl phenol analogs (Table S2): 4-n-octylphenol (OP), 4,4-biphenol, bisphenol A (BPA), and Atenolol.

Table S2. Chemicals used for selectivity test: 4-n-octylphenol (OP), 4,4-biphenol, bisphenol A (BPA), and Atenolol

Chemical Name	Chemical Structure
4-n-nonyl phenol (NP)	
4-n-octylphenol (OP)	
4,4-biphenol	
bisphenol A (BPA)	
Atenolol