

Supplementary Information

Binding Mode of CpG Oligodeoxynucleotides to Nanoparticles Regulates Bifurcated Cytokine Induction via Toll-Like Receptor 9

Shanmugavel Chinnathambi¹, Song Chen², Ganesan Singaravelu¹ & Nobutaka Hanagata^{3,4,*}

¹Department of Medical Physics, Anna University, Chennai 600-025, India

²JSPS Research Fellow, 8 Ichibancho, Chiyoda-ku, Tokyo 102-8472, Japan

³Nanotechnology Innovation Station, National Institute for Materials Science, 1-2-1 Sengen, Tsukuba, Ibaraki 305-0047, Japan

⁴Graduate School of Life Science, Hokkaido University, N10W8, Kita-ku, Sapporo 060-0812, Japan

*Corresponding author: HANAGATA.Nobutaka@nims.go.jp

Figure S1: Zeta potential of allylamine-modified Si-NPs. **Figure S2:** EDX spectrum of allylamine-modified Si-NPs. **Figure S3:** Toxicity of allylamine-modified Si-NPs. **Figure S4:** Image of CpG ODN2006x3-PD molecules bound electrostatically onto allylamine-modified Si-NP. **Figure S5:** No release of CpG ODN2006x3-PD molecules from allylamine-modified Si-NPs. **Figure S6:** Image of thiolated CpG ODN2006x3-PD molecules cross-linked to maleimide-modified Si-NP.

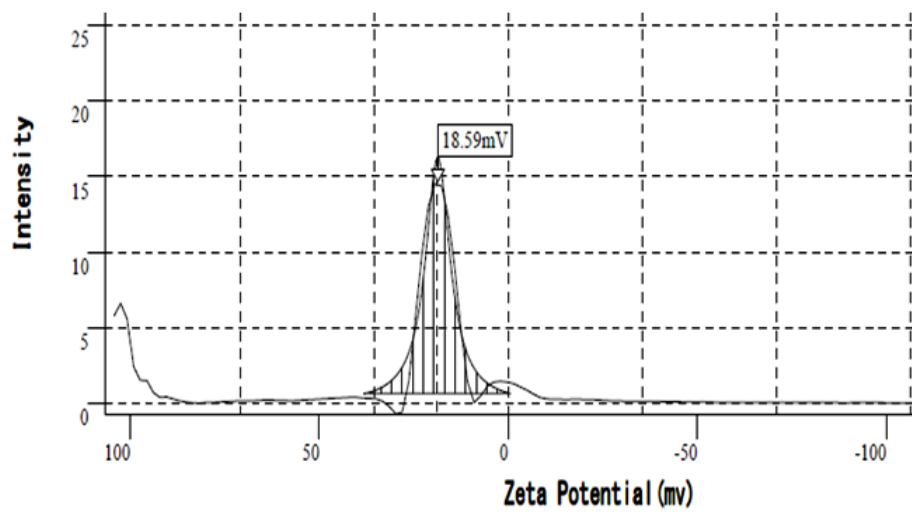


Figure S1. Zeta potential of allylamine-modified Si-NPs.

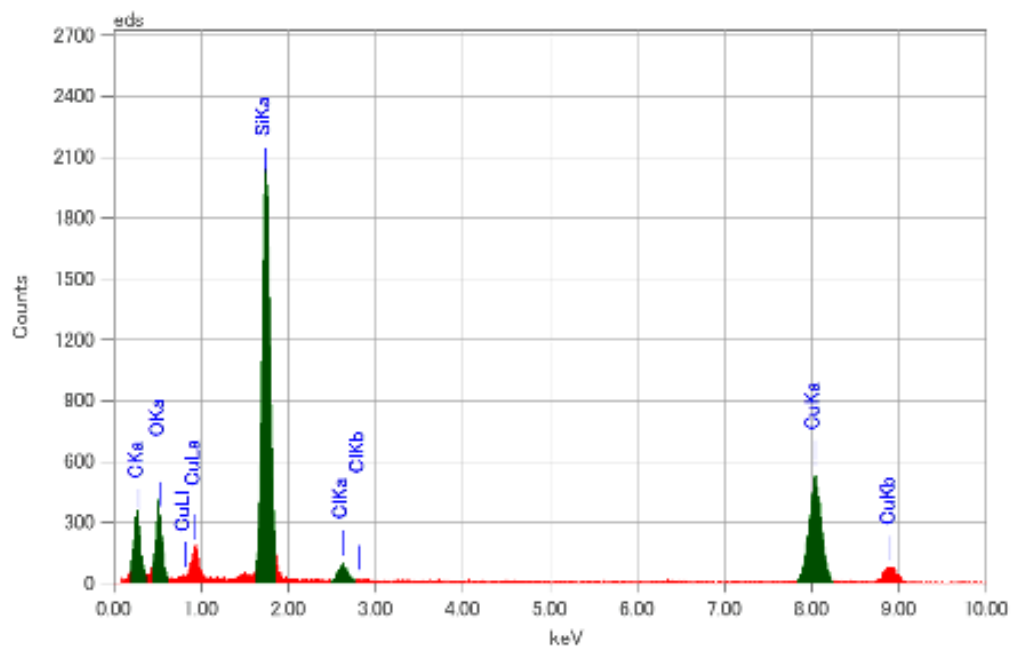


Figure S2. EDX spectrum of allylamine-modified Si-NPs.

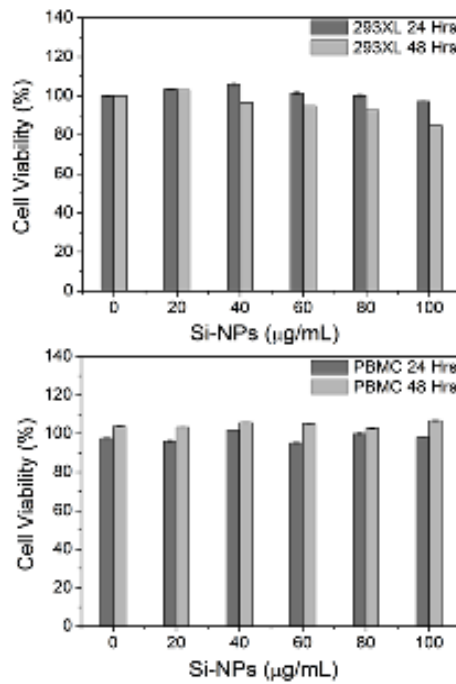


Figure S3. Toxicity of allylamine-modified Si-NPs. (a) 293XL-TLR9 cells were exposure to elevated concentration of allylamine-modified Si-NPs for 24 and 48 h. (b) Peripheral blood mononuclear cells (PBMC) were exposure to elevated concentration of allylamine-modified Si-NPs for 24 and 48 h.

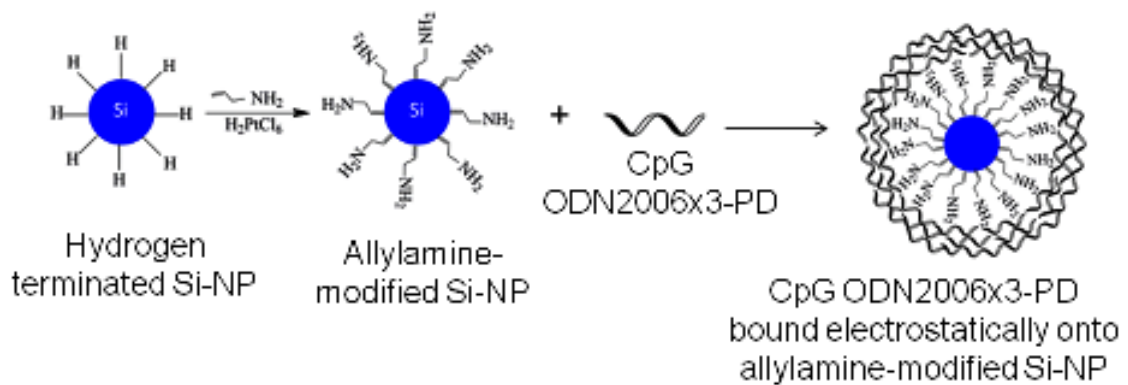


Figure S4. Image of CpG ODN2006x3-PD molecules bound electrostatically onto allylamine-modified Si-NP.

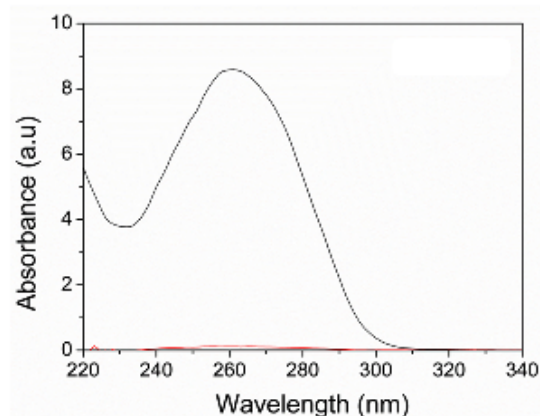


Figure S5. No release of CpG ODN2006x3-PD molecules from allylamine-modified Si-NPs. Allylamine-modified Si-NPs loaded with CpG ODN2006x3-PD were incubated in acetate buffer (pH 5.0) at 37°C. The Si-NPs was removed using Macrospin column G-25 after 24 h. The UV spectrum of acetate buffer after removal of the Si-NPs was measured. Black line, free CpG ODN2006x3-PD molecules in acetate buffer; Red line, acetate buffer after removal of the Si-NPs.

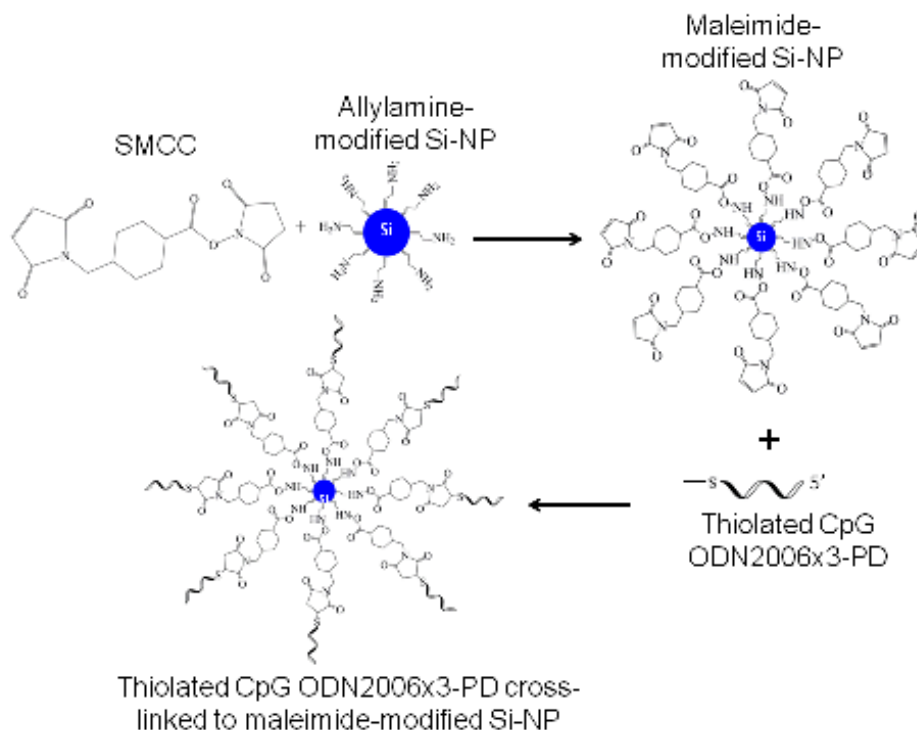


Figure S6. Image of thiolated CpG ODN2006x3-PD molecules cross-linked to maleimide-modified Si-NP. SMCC, 4-(*N*-maleimidomethyl)cyclohexane-1-carboxylic acid *N*-hydroxysuccinimide ester.