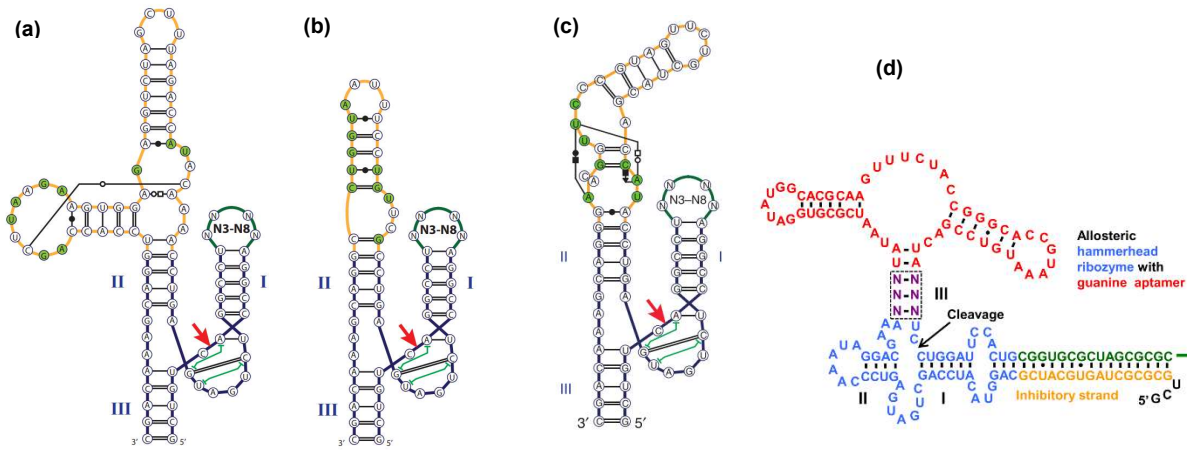
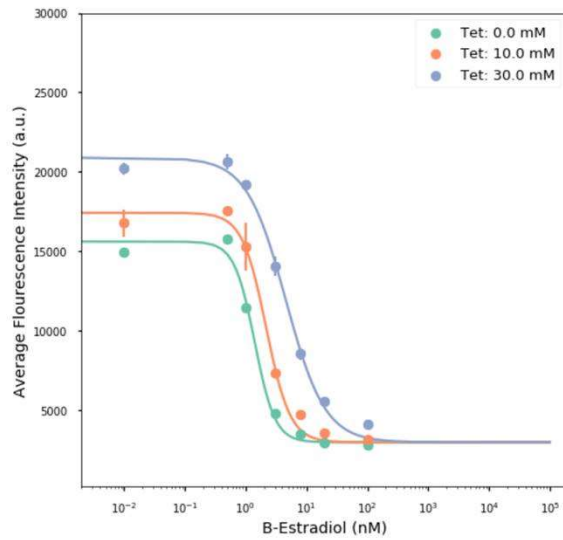


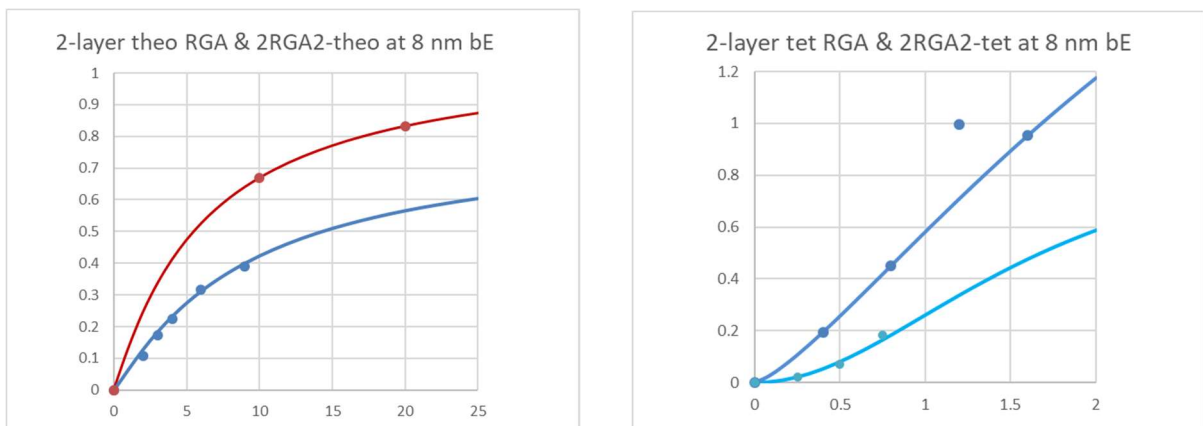
**Supplementary Materials:**



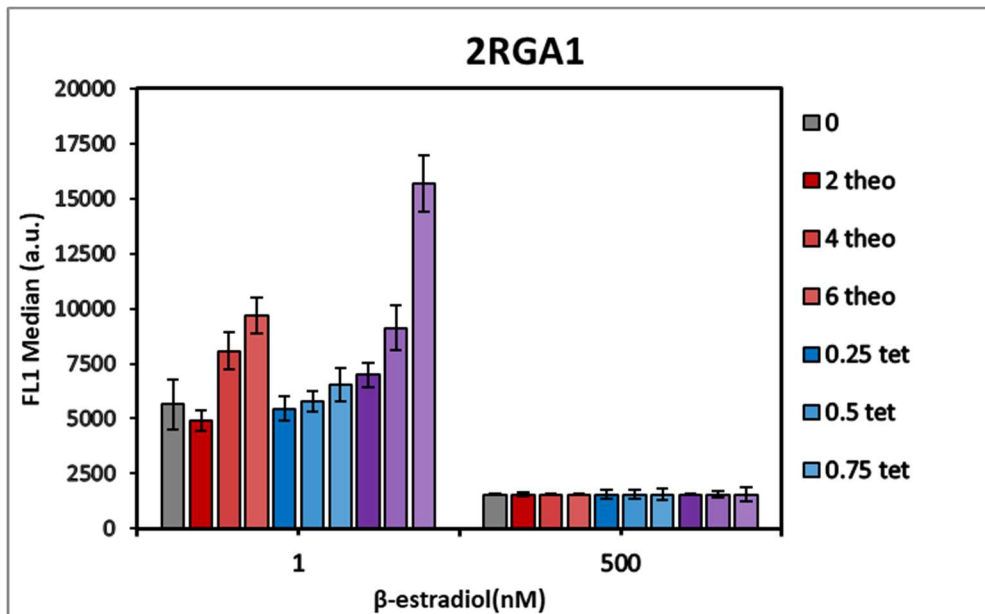
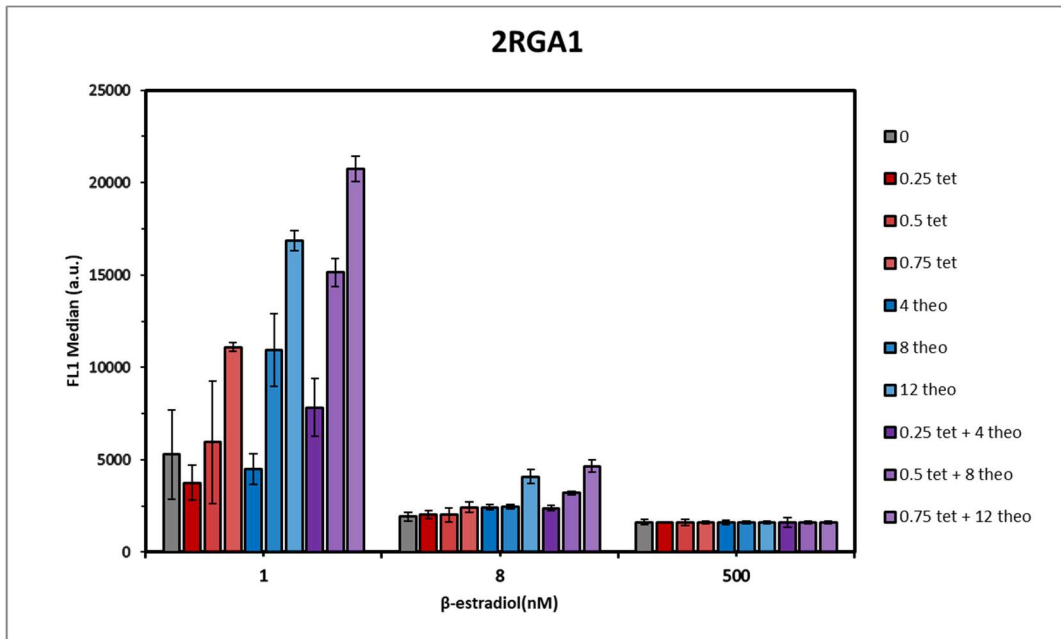
**Figure S1** Secondary structure of (a) theophylline- (b) tetracycline- (c) neomycin- and (d) guanine-responsive tertiary interaction switches.



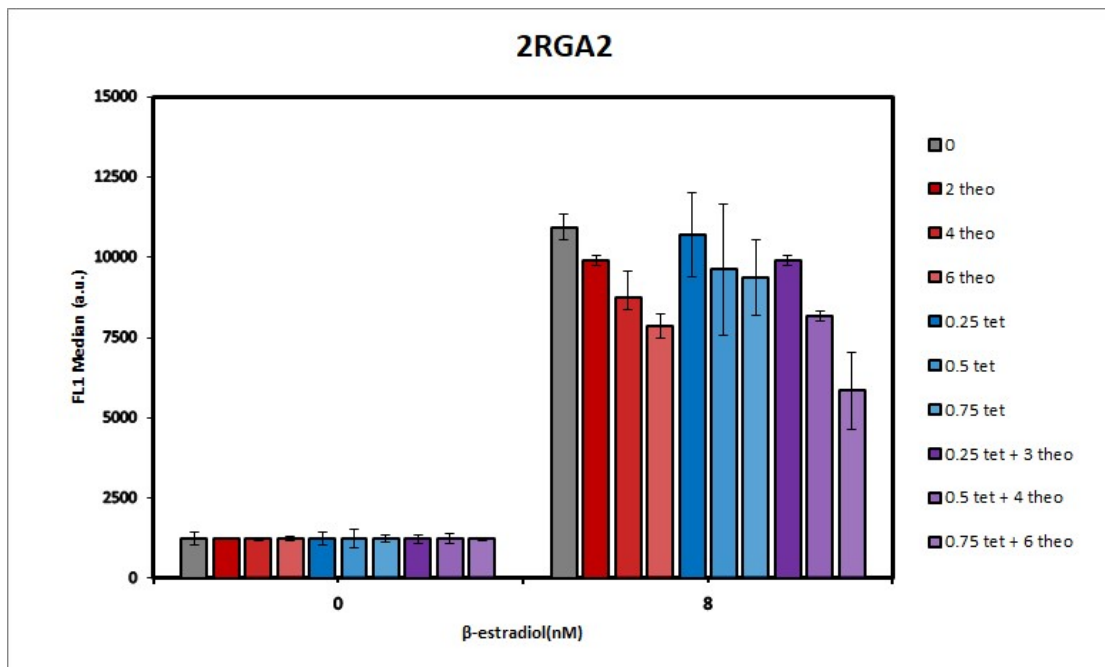
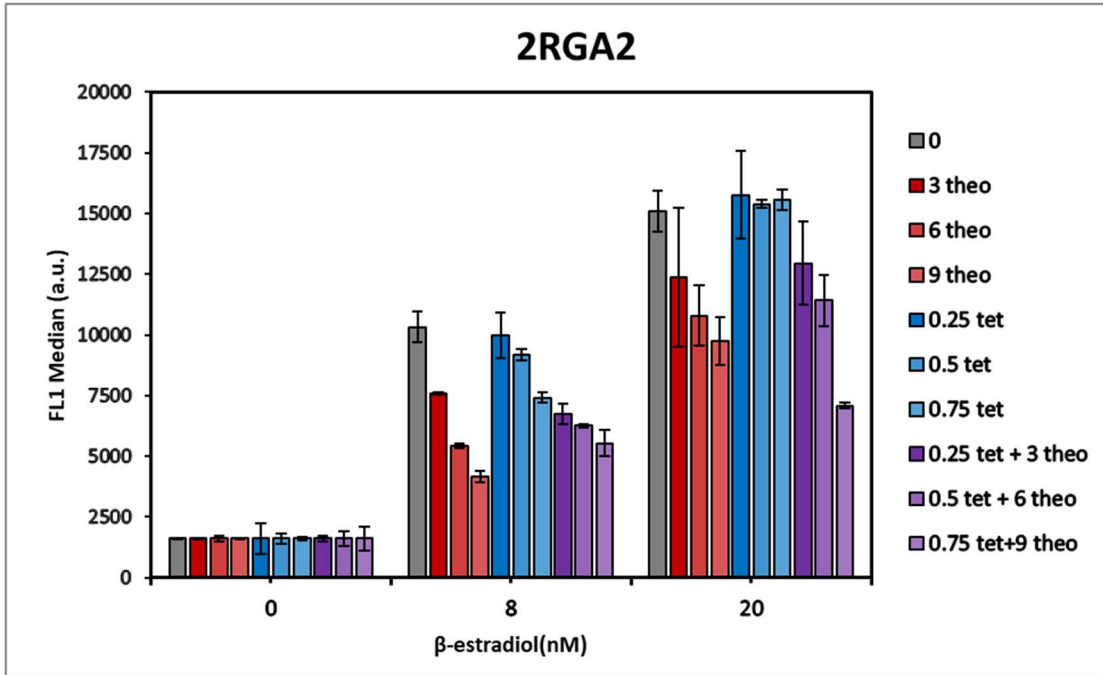
**Figure S2.** The responsive curves of control RGR-w8 within high Theophylline ligands and concentrations.



**Figure S3.** Plots to compare the inhibition abilities of each ligand in two-layer RGAs and 2RGA2 at 8 nm bE induction level (a) Theophylline (b) Tetracycline



**Figure S4.** The fluorescence response plots of 2RGA1 device in mixture buffer with selective Theo and Tet concentration at 1 and 8 nM bE induction levels. Use 500 bE induction condition as a negative control. (a) Theo (0, 4, 8, 12 mM) and Tet (0, 0.25, 0.5, 0.75 mM) (b) Theo (0, 2, 4, 6 mM) and Tet (0, 0.25, 0.5, 0.75 mM)



**Figure S5.** The fluorescence response plots of 2RGA2 device in mixture buffer with selective Theo and Tet concentration at 8 and 20 nM bE induction levels. Use 0 bE induction condition as a positive control. (a) Theo (0, 3, 6, 9 mM) and Tet (0, 0.25, 0.5, 0.75 mM) (b) Theo (0, 2, 4, 6 mM) and Tet (0, 0.25, 0.5, 0.75 mM)