Article

Differential effects of Oleuropein and Hydroxytyrosol on Aggregation and Stability of CFTR NBD1-ΔF508 Domain

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Supplementary Materials

Table S1. Ligand-mNBD1-ΔF508 docking scores of BIA and Oleuropein full fitness and ΔG values for most stable docked molecules shown in Figure 3.

Name	Full Fitness kcal/mol	ΔG kcal/mol
BIA	-1630.1	-6.37
Oleuropein	-1585.0	-8.36
Hydroxytyrosol	-1626.3	-6.25

Table S2. ANOVA analysis of DSF experiments.

ANOVA Analysis			
mNBD1-ΔF508 *vs Name*	*F*	*P-value*	*F crit*
mNBD1-ΔF508 + 1mM ATP	43.40	0.00275	7.709
mNBD1-ΔF508 + 5mM ATP	302.88	0.00006	7.709
mNBD1-ΔF508 + 1mM BIA	10.86	0.03006	7.709
mNBD1-ΔF508 + 1.5mM ATP + 1mM BIA	194.59	0.00015	7.709
mNBD1-ΔF508 + 100μM Oleuropein	0.29	0.61671	7.709
mNBD1-ΔF508 + 100μM Hydroxy-Tyrosol	0.07	0.79986	7.709
mNBD1-ΔF508 + 500μM Hydroxy-Tyrosol	0.34	0.59343	7.709
mNBD1-ΔF508 + 1mM Hydroxy-Tyrosol	0.85	0.40977	7.709
mNBD1-ΔF508 + 1mM Hydroxy-Tyrosol + 1mM BIA	13.72	0.02076	7.709

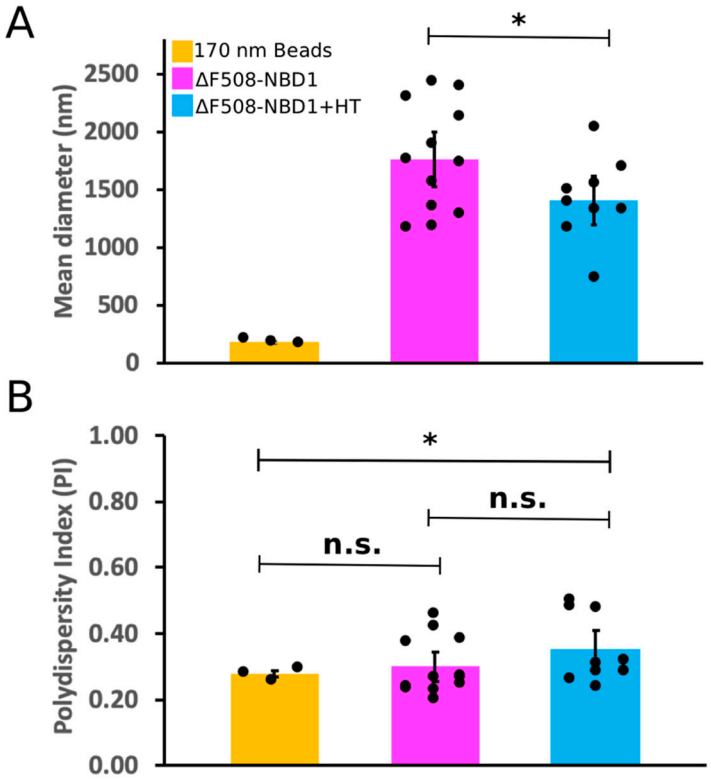

Figure S1. NBD1–ΔF508 aggregates incubated with Hydroxytyrosol are smaller in diameter and mostly monodispersed. A) Mean diameter of NBD1–ΔF508 aggregates measured by dynamic light scattering in the absence (n=4, 12 replicates) or presence of 100 µM Hydroxytyrosol (n=3, 9 replicates), **p*-value = 0.032. Aggregate diameter in absence and presence of Hydroxytyrosol was significantly larger compared to 170 nm beads (3 replicates), *p*-value = 7.17E-08 and *p*-value = 3.21E-06, respectively. **B)** mean polydispersity measurement for control (n=4, 12 replicates) and Hydroxytyrosol incubated (n=3, 9 replicates) NBD1–ΔF508 aggregates compared to 170 nm beads, **p*-value = 0.0387 and n.s. = not significant (*p*-value > 0.05). Data plotted as mean ± SEM. Significant differences were analyzed by unpaired Student's t-test.

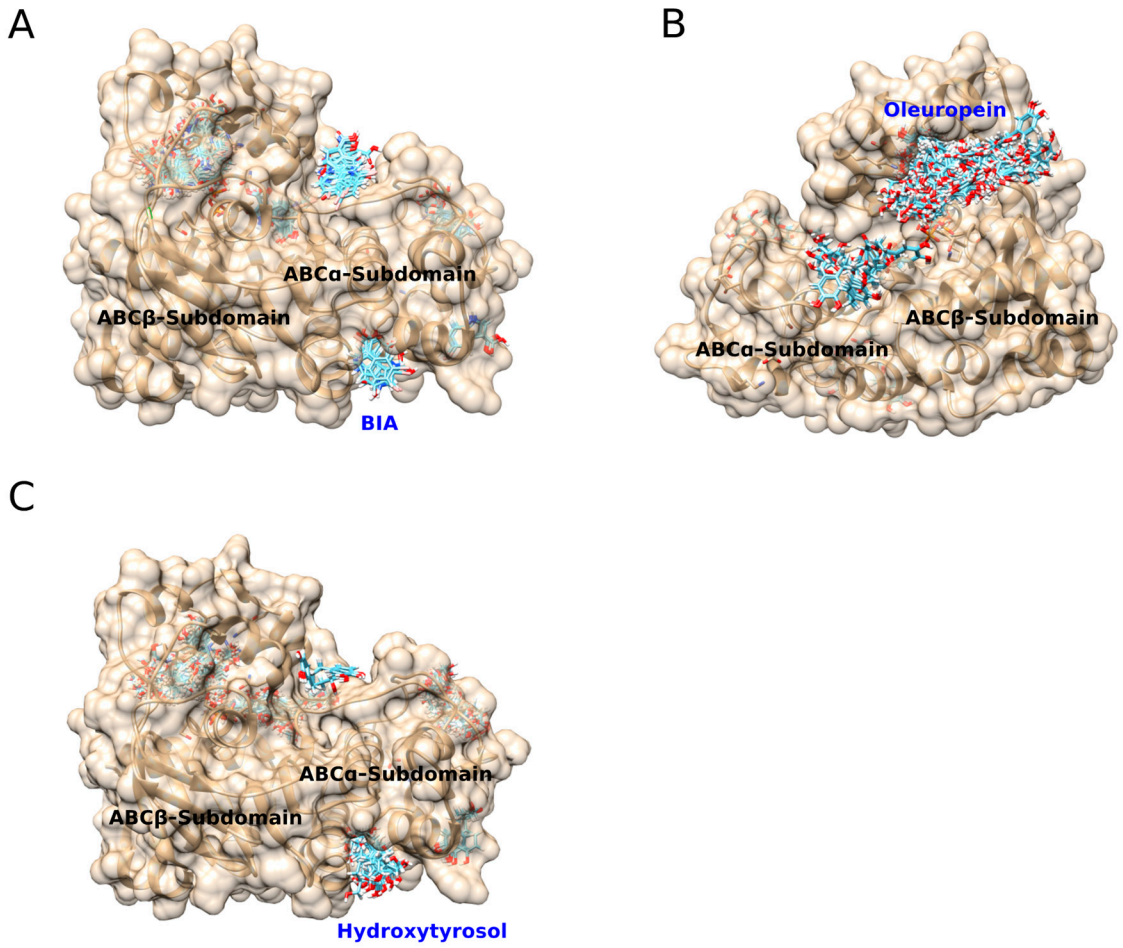

Figure S2. Full SwissDock predicted binding modes for BIA, Oleuropein, and Hydroxytyrosol to mNBD1–ΔF508. Note the majority of predicted binding modes for BIA and Hydroxytyrosol overlap. The majority of Oleuropein binding modes are in the ATP binding cleft. .

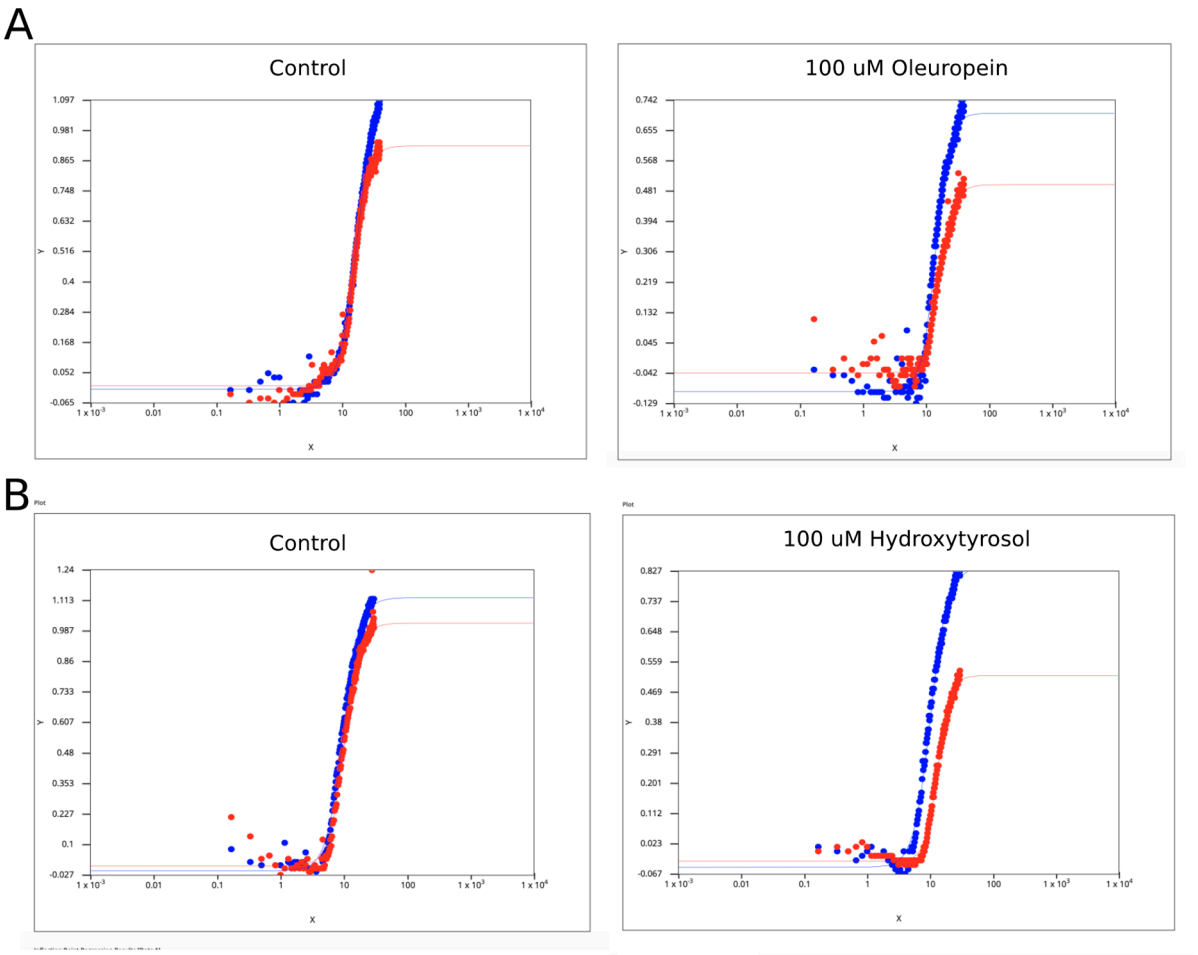

Figure S3. Curve fitting of mNBD1-ΔF508 aggregation data from Figure 1C & D. **A)** Representative results for control (left panel) and 100 µM Oleuropein (right panel) data fitted with logistics equation. **B)** Representative results for control (left panel) and 100 µM Hydroxytyrosol (right panel) data fitted with four parameter logistics equation.