

Arene Substitutions in Orchid Bibenzyls: Mechanistic Insights into Glucose Uptake and Lipid Metabolism for Targeting Metabolic Disorders

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Table S1. List of primers used in this study.

Primer	Nucleotide sequence (5' - 3')	Number of nucleotides	ΔG (kcal mol ⁻¹)	T_m (°C)	T_a (°C)
PPAR γ F	GATTCTCCTRTTGACCCAG	19	1.06	51.0	55
PPAR γ R	GARTGSGAGTGGTCTTCCAT	20	-0.95	55.7	55
C/EBP α F	AGTCGGTGGACAAGAACAGC	20	-1.62	57.4	55
C/EBP α R	GTGTCCAGTTCRCGGCTCA	19	0.92	59.0	55
SREBP1 F	YTGCMGACCCTGGTGAGTG	19	-0.32	59.1	55
SREBP1 R	GASCGGTAGCGCTTCTCAAT	20	-1.74	57.4	55
β -Actin F	CCACCATGTACCCWGGCATT	20	0.68	57.6	55
β -Actin R	CGGACTCRTCRTACTCCTGC	20	0.08	57.2	55

Table S2. Three-dimensional coordinates of ligands used for molecular docking analysis.

Ligand ^a	Coordinate		
	X	Y	Z
FAS	10.561091	6.370303	-33.511758
LPL	2.501486	0.863000	102.519514
FABP 4	-4.274389	6.863889	-19.213944

^a The ligands selected for molecular docking included fatty acid synthase (FAS, PDB ID: 4PIV), lipoprotein lipase (LPL, PDB ID:6OB0), and fatty acid binding protein 4 (FABP4, PDB ID: 4NNT). Their three-dimensional structures were obtained from the Protein Data Bank (PDB).

Figure S1. Cytotoxic effects of batatasin III and gigantol in 3T3-L1, PCS-210-010, and L6 cells.

The cytotoxic effects of batatasin III and gigantol were assessed in 3T3-L1, PCS-210-010, and L6 cells during early (A and C) and late (B and D) differentiation stages. Cells were treated for 48 h with varying concentrations of batatasin III (A and B) or gigantol (C and D). The mode of cell death was analyzed through dual nuclear staining with Hoechst 33342 and propidium iodide. Undifferentiated (UC) and differentiated (DC) cells treated with vehicle (0.5% (v/v) dimethyl sulfoxide) served as controls. Representative images were randomly selected from at least three independent experiments (scale bars = 50 μ m).

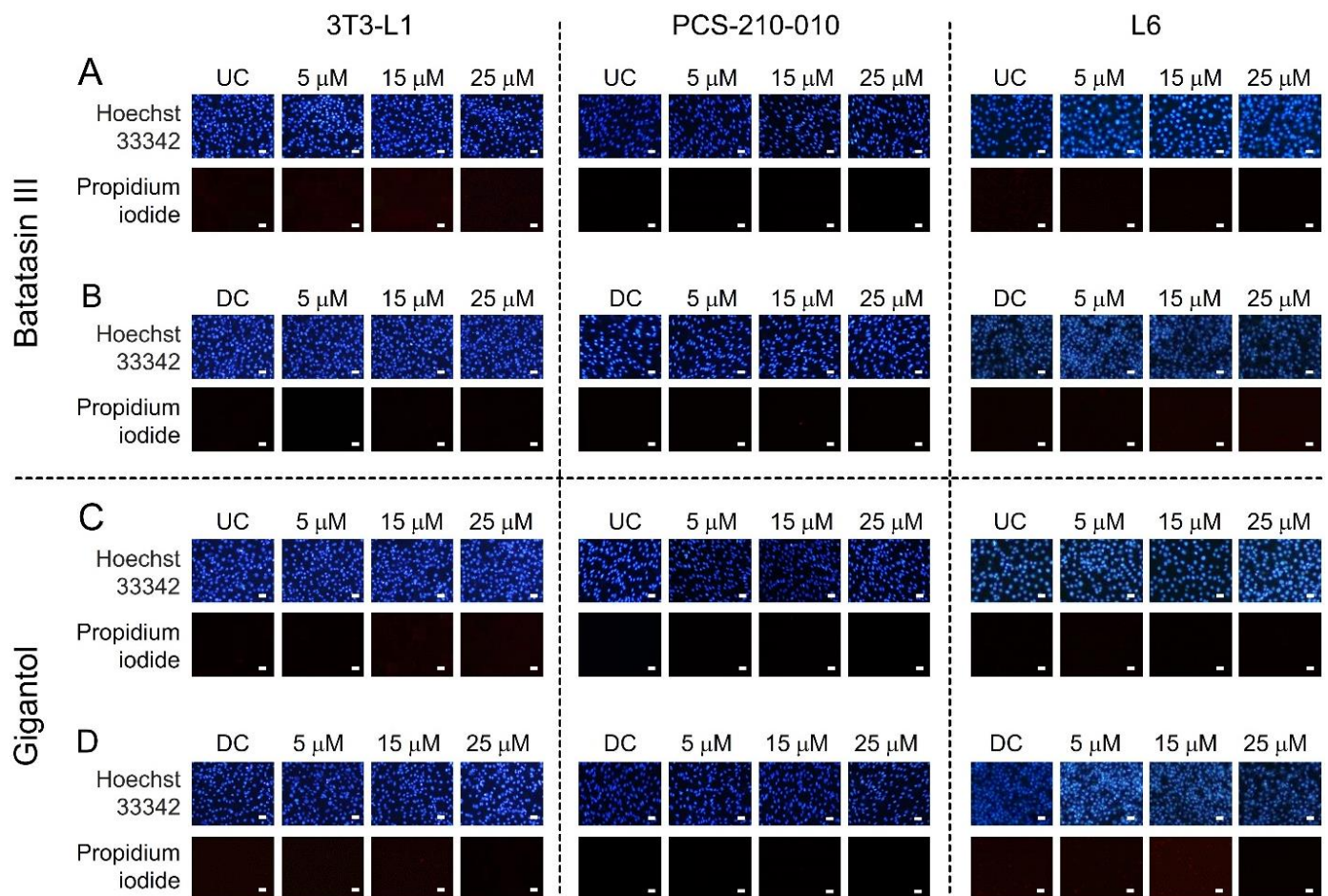


Figure S2. GLUT membrane translocation in 3T3-L1 and L6 cells treated with batatasin III and gigantol. Immunofluorescence imaging was used to assess the localization of GLUT1 (A, E, C, G, I, M, K, and O) and GLUT4 (B, F, D, H, J, N, L, and P) in differentiated 3T3-L1 (A-H) and L6 (I-P) cells treated with non-cytotoxic concentrations of batatasin III and gigantol (up to 25 μ M) for 48 h under both basal and insulin-stimulated conditions. Cells treated with 0.5% (v/v) dimethyl sulfoxide served as vehicle controls. Specific antibodies conjugated to green fluorescence were used to probe GLUT1 and GLUT4, while nuclei were counterstained with Hoechst 33342 (blue fluorescence), illustrating the translocation of GLUT1 and GLUT4 from intracellular compartments to the cell membrane. Images were acquired using a confocal microscope at 20X magnification (scale bar = 20 μ m).

