

MOLECULAR PHARMACOLOGY

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Endoxifen, 4-hydroxytamoxifen and an Estrogenic Derivative Modulate Estrogen Receptor Complex Mediated Apoptosis in Breast Cancer.

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Fig. S1

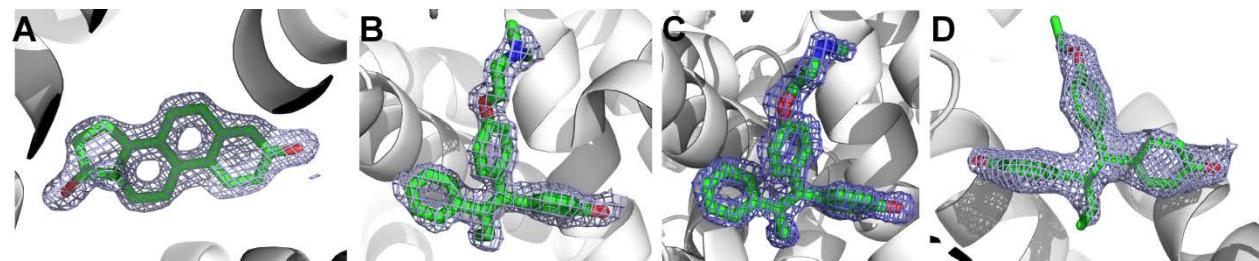


Fig. S2

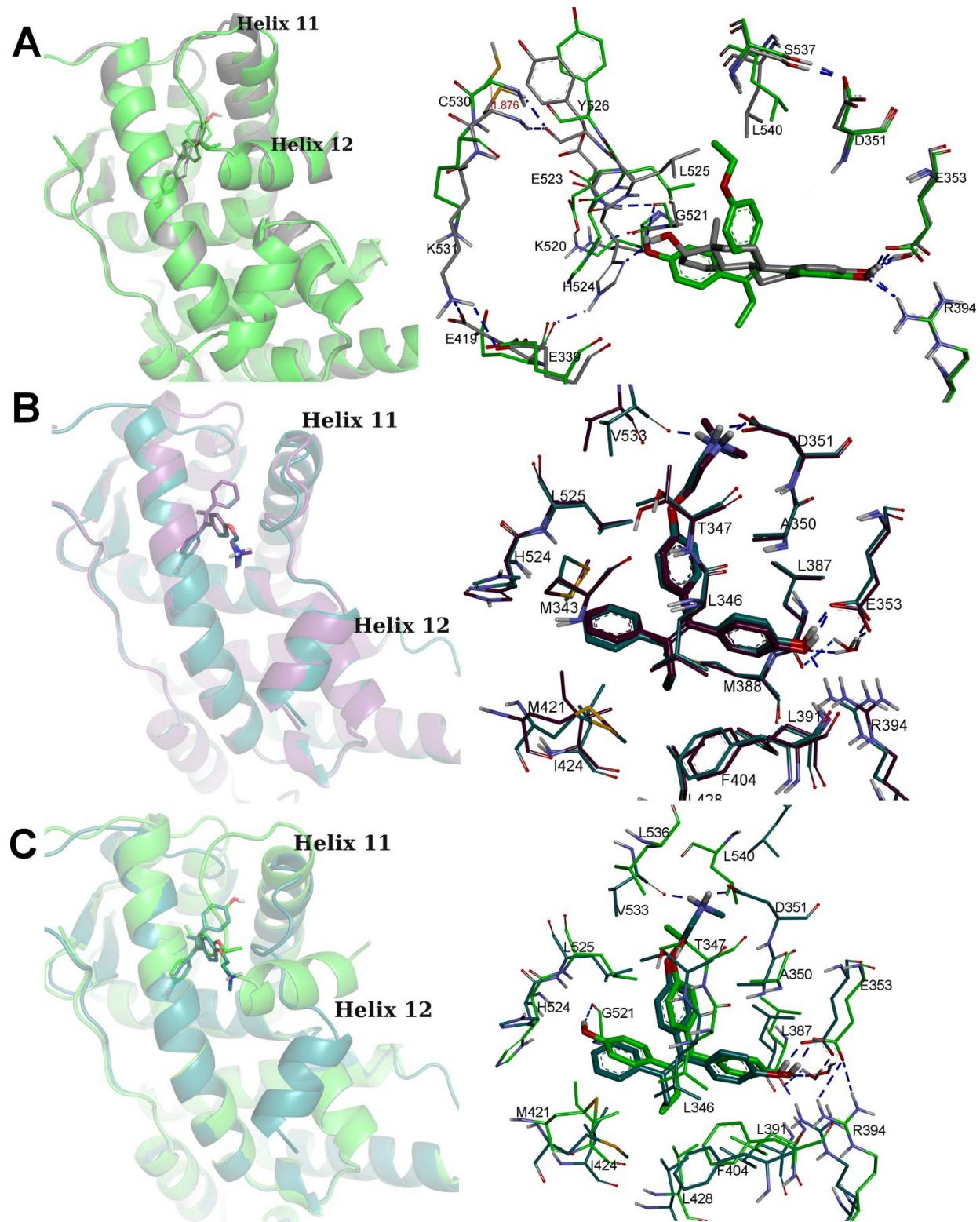


Table S1

	12h	24h	48h
Vehicle	100	100	100
E2	23.47281	12.27021	6.647536
EtOXTPE	49.39486	23.74318	9.740245
4OHT	114.2996	56.27626	61.30403
Endox	78.18556	53.67578	59.9457
ICI	17.67255	10.19532	17.46402

Table S2

Vehicle	100
E2 1nM	18.37051
EtOXTPE 100 nM	45.63361
E2+4OHT	155.1129
EtOXTPE+4OHT	170.8325
4OHT 1uM	164.2368
ICI 1uM	14.55413

Table S3

	24h	36h	48h
Vehicle	100	100	100
E2 1nM	118.4105	131.7698	143.5935
EtOXTPE 1uM	84.11689	127.8888	147.4219
Endox 1uM	62.318	104.3525	113.713
ICI 1uM	72.89661	99.34927	79.49388
EtOX+E2	51.48969	140.3505	107.118
EtOX+Endox	53.65031	116.031	83.6344
EtOX+ICI	48.65331	105.3464	65.65253
E2+Endox	49.78846	113.2255	89.21531
E2+ICI	59.71132	97.61398	75.9761

Table S4

	ERα LBD Y537S-TPE- GRIP	ERα LBD Y537S-E2- GRIP	ER α LBD C381S, C417S, C530S- 4OHT	ER α LBD C381S, C417S, C530S- Endoxifen
PDB Code	5T1Z	6CBZ	5W9C	5W9D
Data Collection				
Space Group	P2 ₁	P2 ₁	P1	C2
a, b, c (Å)	56.09, 84.21, 58.97	56.13, 84.62, 58.59	58.308, 58.308, 276.947	57.980, 57.980, 277.001
α, β, γ (°)	90, 108.28, 90	90, 108.48, 90	90, 90, 120	90, 90, 120
Resolution Range	50 - 2.1	20 - 1.65	20 – 1.80	20 – 1.65
Number of Reflections				
(all/unique)	28624/6656	61566/10801	56362/51238	56989/15402
I/ σ (highest resolution)	2.18	2.0	1.5	2.0
R _{merge} (highest resolution)	0.08 (0.22)	0.08 (0.48)	0.023 (0.33)	0.06 (0.46)
% Completeness (highest resolution)	98.7 (95.9)	98.6 (95.9)	88.5 (57.9)	90.03 (87.8)
Redundancy (highest resolution)	4.3 (4.3)	5.7 (4.7)	1.6 (1.6)	3.7 (3.7)
Refinement				
Rwork/Rfree (Highest Resolution)	18.61/24.56 (26.90/28.04)	18.45/21.35 (25.18/29.30)	19.16/24.33 (24.77/30.53)	16.75/19.52 (19.66/27.70)
No. Residues/Chain				
ER α LBD	233	214	238	241
GRIP Peptide	13	13		
Protein β -factor	46.31	20.20	24.60	21.23
Water	84	257	220	308
Water β -factor	39.22	32.20	28.50	37.40
Ligand	1	1	1	1
Ligand β -factor	46.89	17.70	19.20	11.45
RMSD				
Bond lengths (Å)	0.02	0.004	0.005	0.018
Bond angles (°)	1.561	0.818	1.040	1.75
Ramachandran plot statistics				
Preferred number (%)	455 (97.64 %)	426 (99.53%)	862 (98.51%)	414 (99.04%)
Additional allowed (%)	11 (2.36%)	2 (0.47%)	13 (1.49%)	4 (0.96%)
Outliers (%)	0	0	0	0

Fig. S1: 2mFo-DFc maps for E2 (A), 4OHT (B), Endoxifen (C), and EtOXTPE (D) in complex with ER α LBD contoured to 1.5 Å.

Fig. S2 Comparisons between the binding modes of EtOXTPE, E₂, Endoxifen and 4OHT at ER α LBD. A – Superposition of ER α with EtOXTPE (green) and E₂ (gray). The aminoacids involved in H-bond contacts with the ligands are shown, together with the residues displaying different orientations between the structures. B – ER α :4OHT complex (purple) superimposed with ER α :Endoxifen complex (cyan). The residues involved in hydrophobic contacts and H-bonds formation with the ligands are shown. C – ER α :Endoxifen (cyan) overlaid with ER α :EtOXTPE complex (green). The H-bonds and the participating aminoacids are shown together with the residues that interact with the ligands.

Table S1. Densitometry data for immunoblot in Fig. 6A.

Table S2. Densitometry data for immunoblot in Fig. 6B.

Table S3. Densitometry data for immunoblot in Fig. 6D.

Table S4. X-ray crystal structure data collection and refinement statistics.