Novel Homo Disubstituted Triphenylethylenes with Potential Proteasomal Inhibition and Anti-Cancer Activity

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Introduction

Protein degradation in eukaryotic cells is divided into two pathways: the lysosomal pathway and the ubiquitin-proteasome pathway. The ubiquitin-proteasome system begins protein degradation by labelling specific proteins with polyubiquitin chains in response to intracellular or extracellular signals.

The 26S proteasome regulates a variety of biological activities, including cell cycle progression, cell growth, proliferation, differentiation, apoptosis, gene transcription and signal transduction, by degrading ubiquitinated proteins. The proteasome's abnormal degradation of essential regulatory proteins disrupts these processes, resulting in uncontrolled cell cycle progression and reduced cell death, both are hallmarks of cancer. [1]

Anticancer medicines based on proteasome inhibitors have been proposed. The 26S proteasome is a huge protein complex with a mass of w2.5 MDa that is divided into two sub complexes: the 19S regulatory complex and the 20S catalytic core. A barrel-shaped protein, the 20S catalytic core is made up of seven α subunits (α 1-7) and seven β subunits (β 1-7). [2]

Proteasome inhibitors' apoptotic effect in tumor cells may be due to inhibition of NFκB activity, altered degradation of cell cycle related proteins, altered pro-apoptotic and anti-apoptotic protein balance, endoplasmic reticulum stress, and inhibition of angiogenesis and DNA repair. [3] RID-F was the most effective of the ridaifen compounds studied, inhibiting all three proteasome functions which are chymotrypsin-like (CT-L), trypsin-like (T-L), and caspase-peptidylglutamyl peptide hydrolase (PGPH). [2]

Herein, we report the synthesis of novel ridaifen analogues that target the three protease activities of the proteasome. Three compounds out of the eleven novel synthesized compounds displayed sub micromolar IC_{50} on CT-L and PGPH activity. Triphenyl based SERMs like TAM, TOR and Clomiphene were repurposed to treat EBOLA virus and inhibit its viral entry or replication.[4] Thus five of our compounds were selected by NIAID for testing on EBOLA virus.

Compound **VI** showed anti-EBOLA virus activity, SI_{50} of 33 and EC_{50} of 0.11 µM. Moreover, compound **VI** showed proteasome inhibition of 2.04 and 2.91 µM on CT-L and PGPH activity respectively.

Results

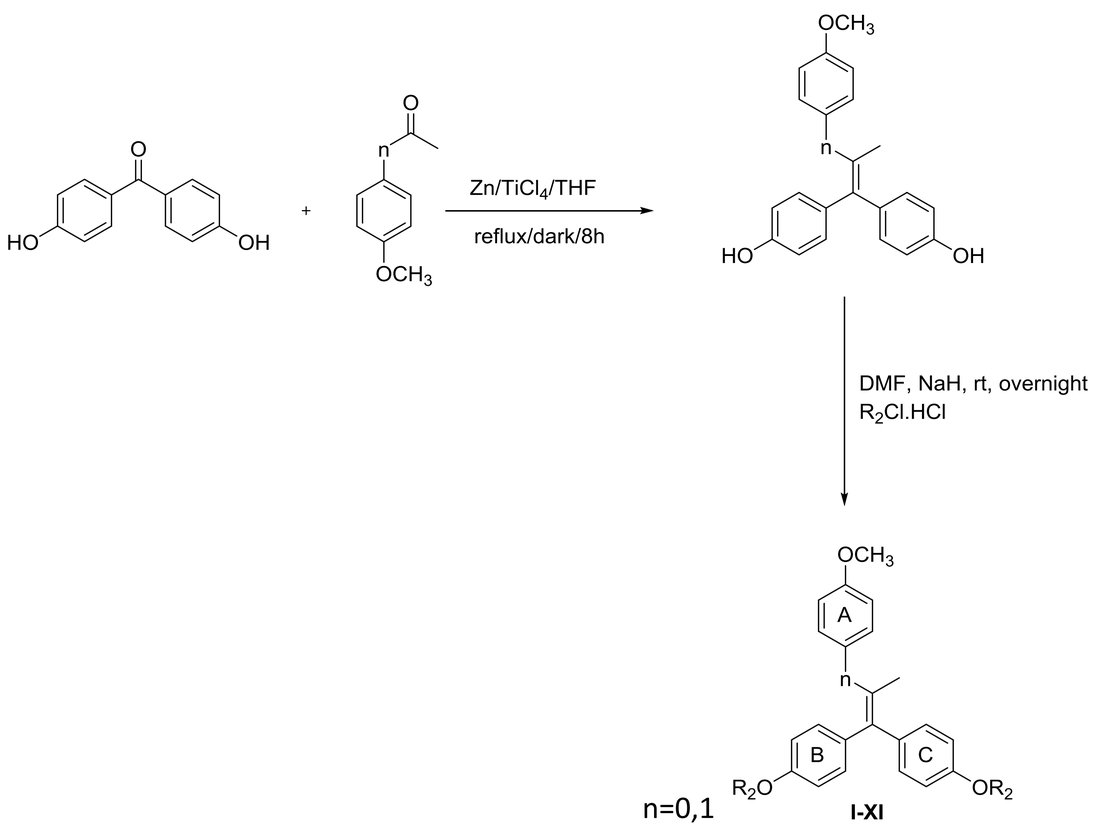
Table 1: Mean GI_{50} (µM) on NCI-60 panel and inhibition of the proteasome activity

Cpd	R_2	n	Mean GI_{50} (µM)	IC_{50} (µM) CT-L activity	IC_{50} (µM) PGPH activity	IC_{50} (µM) T-L activity
I	$-C_3H_6N(CH_3)_2$	0	1.62	0.85	0.48	>20
II	$-C_2H_4(C_5H_{10}N)$	0	ND	1.35	1.66	>20
III	$-C_2H_4N(C_2H_5)_2$	0	1.07	1.90	1.60	>20
IV	$-C_2H_4(C_4H_8N)$	0	ND	0.88	0.56	>20
V	$-C_2H_4(C_6H_{12}N)$	0	0.89	3.00	2.43	>20
VI	$-C_2H_4N(CH_3)_2$	0	0.74	2.04	2.91	>20
VII	$-C_3H_6N(CH_3)_2$	1	ND	>20	17.08	>20
VIII	$-C_2H_4(C_5H_{10}N)$	1	ND	8.53	7.95	>20
IX	$-C_2H_4N(C_2H_5)_2$	1	3.63	>20	>20	>20
X	$-C_2H_4(C_4H_8N)$	1	2.45	0.74	0.48	>20
XI	$-C_2H_4N(CH_3)_2$	1	ND	2.16	2.70	>20
RID-F	$-C_2H_4(C_6H_{12}N)$	0	-	0.54	0.61	>10

Table 2: Anti-Ebola activity and selectivity index of the designed analogues

Compound	R_2	n	EC_{50} (µM)	SI_{50}
II	$-C_2H_4(C_5H_{10}N)$	0	2.8	1.6
V	$-C_2H_4(C_6H_{12}N)$	0	2.3	4.4
VI	$-C_2H_4N(CH_3)_2$	0	0.11	33
VII	$-C_3H_6N(CH_3)_2$	1	23	3.9
VIII	$-C_2H_4(C_5H_{10}N)$	1	>10	<2

Chemistry



Conclusion

- ❑ Compounds **I**, **III**, **V**, **VI**, **IX** and **X** showed mean GI_{50}= 1.62, 1.07, 0.89, 0.74, 3.63 and 2.45 µM respectively whereas the mean GI_{50} of TAM = 4.41 µM.
- ❑ Introduction of flexibility with a methoxy group on ring A and dimethylaminopropoxy or dimethylaminoethoxy substituents on rings **B** & **C** lead to significant loss of activity on NCI-60 cancerous cell lines.
- ❑ Comparing compound **IV** with it's congener compound **X**, both compounds bear pyrrolidinylethoxy substituent on rings **B** & **C**. Introduction of flexibility in compound **X** lead to remarkable decrease in the mean growth inhibition over the NCI-60 cancerous cell lines.
- ❑Introduction of piperidinylethoxy substituent on either rigid or flexible scaffolds bearing a methoxy group lead to no inhibition on the NCI-60 cancerous cell lines.
- ❑Three compounds, compounds **I**, **IV** and **X,** having dimethylaminopropoxy and pyrrolidinylethoxy substituent on rings **B** & **C** showed sub micromolar IC_{50} on CT-L activity and PGPH activity.
- ❑ Novel compounds can be optimized for anti-EBOV activity.

Pharmacology

- ❑ Compounds were biologically evaluated for their inhibition of the three protease activities of the proteasome. For each of the screened compounds, IC_{50} was determined, which is the concentration needed to inhibit 50% of the enzymatic action.
- ❑ Compounds were tested for their anti-proliferative activity over NCI-60 panel at 10 µM.
- ❑ Five compounds were subjected to screening for the inhibition of EBOLA virus replication.

References

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