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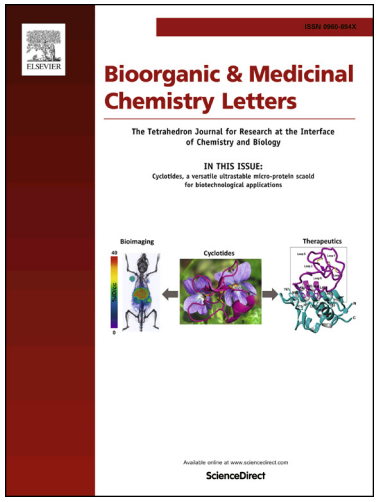

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Synthesis and Investigations into the Anticancer and Antibacterial Activity Studies of β-Carboline Chalcones and their Bromide Salts

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ABSTRACT

A series of sixteen β-carbolines, bearing chalcone moiety at C-1 position, were prepared from easily accessible 1-acetyl-β-carboline and various aldehydes under basic conditions followed by N^2-alkylation using different alkyl bromides. The prepared compounds were evaluated for *in vitro* cytotoxicity against a panel of human tumor cell lines. N^2-Alkylated-β-carboline chalcones **13a-i** represented the interesting anticancer activities compared to N^2-unsubstituted β-carboline chalcones **12a-g**. Off the prepared β-carbolines, **13g** exhibited broad spectrum of activity with IC_{50} values lower than 22.5 μM against all the tested cancer cell lines. Further, the N^2-alkylated-β-carboline chalcone **13g** markedly induced apoptosis in MDA-MB-231 cells by AO/EB staining assay. The most cytotoxic compound **13g** possessed a relatively high drug score of 0.48. Additionally, the prepared β-carboline chalcones displayed moderate antibacterial activity against tested bacterial strains.

β-Carbolines are an important class of nitrogen containing heterocycles due to their widespread biological and pharmacological applications.[1] Particularly, C-1 substituted β-carbolines possess remarkable antimalarial, anti-HIV, antimicrobial, antileishmania, and antitumoral properties.[2-6] Recently, interest in β-carboline alkaloids was stimulated by their potential antitumor and antibacterial activities.[7-9] β-Carbolines exhibit anticancer properties through DNA intercalation, tubulin polymerization, topoisomerase and kinase inhibition.[10,11] For instance, natural β-carboline derivative Harmane **1,** isolated from cured tobacco and its smoke;[12] exhibited moderate anticancer activity through DNA intercalation in addition to anti-bacterial activities.[13,14] Kobayashi et al. reported isolation and cytotoxicity of novel manzamine alkaloid Xestomanzamine A (**2**) from Okinawan marine sponges of *Xestospongia sp.* and exhibited weak cytotoxicity against KB cell lines.[15] Cardellina group isolated Eudistomin T (**3**) from *Eudistoma olivaceum* endowed with antimicrobial and weak phototoxicity.[16-18] In 2008, Cao research group prepared 1-benzylidine-N^2-benzylated-β-carbolinium bromides **4** and **5** with interesting cytotoxic activities (Fig.1 and Fig.2).[19-21] From a series of novel β-carboline-based chalcones Chauhan et al. identified compound **6** with high cytotoxicity (IC_{50} =2.25 μM; MCF-7) against breast cancer cell lines.[22]

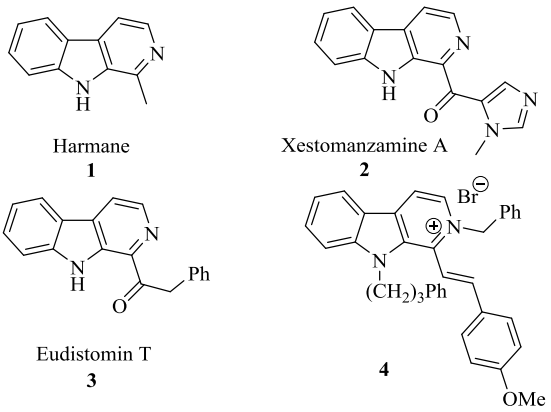

Fig. 1. Pharmacologically interesting C-1 substituted β-carbolines

On the other hand, compounds with enone system continue to be of great interest because of their simple chemistry, easy synthesis and biological importance and usefulness as building blocks in synthetic chemistry.[23] Particularly, wide variety of biological activities associated with chalcones have encouraged organic and medicinal chemists to undertake their structural modifications and chemistry.[24] Chalcones exhibit anti-cancer activities through various mechanisms such as inhibition of multi-drug resistance

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channels such as ABCG2, BCRP, p-glycoprotein and inhibition of protein deacetylation.[25] Also, many research groups either isolated or synthesized natural chalcones with antibacterial as well as anticancer activities. For example, Licochalcone A (**7**) isolated from *Glycyrrhiza inflate* is known to exhibit potent antibacterial activity especially towards *Bacillus subtilis*, *Staphylococcus aureus* and *Micrococcus luteus*.[26] In recent past, Kumar et al. reported synthesis and antitumor activity of indole-based chalcones **8** with IC_{50} values ranging 0.03-0.09 µM against human pancreatic (PaCa-2) carcinoma cells.[27]

In continuation of our efforts to develop potent anticancer agents, recently we have identified indole analogues possessing significant cytotoxicity.[28-33] Inspired by the attractive anticancer properties of β-carboline and chalcone units, herein, we have designed a diverse series of β-carboline chalcones **12a-g** and their bromide salts **13a-i** by incorporating amazing features of β-carboline and chalcone in a single molecule (Fig. 2).

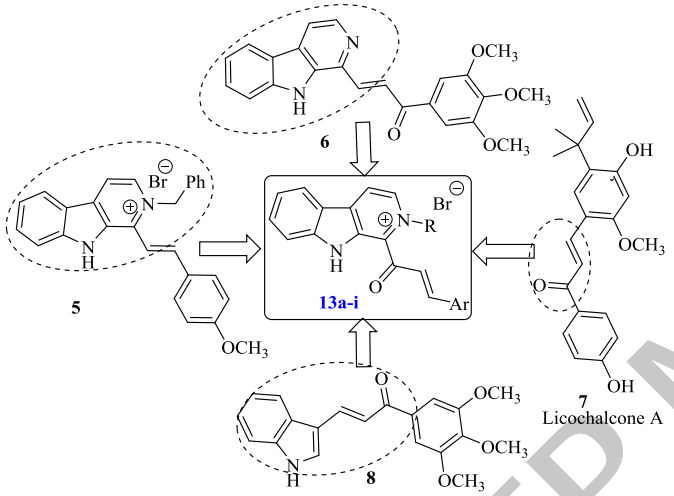
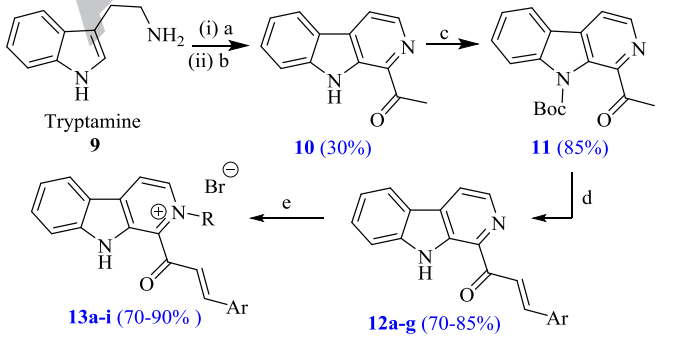


Fig. 2. Rational design of β-carboline chalcones and their bromide salts

Synthesis of novel β-carboline chalcone analogues **12a-g** and **13a-i** is depicted in Scheme 1.[22,34-36] Initially the 1-acetyl-β-carboline **10** was prepared by the Pictet–Spengler reaction of tryptamine with pyruvaldehyde under acidic conditions followed by *in situ* aromatization with 10% palladium on carbon. Later the *N*-H protection of 1-acetyl-β-carboline with di-tert-butyl dicarbonate in the presence of DMAP produced *N*-boc-1-acetyl-β-carboline **11**. Further, reaction of **11** with respective aldehydes in the presence of NaOH solution (20%) in aqueous ethanol for 16 h afforded β-carboline chalcone analogues **12a-g** in good to excellent yields. Finally, the N^2-alkylation of three β-carboline chalcones **12c-e** with various alkylbromides led to the corresponding bromide salts **13a-i** with more than 80% yields.



Scheme 1. Synthesis of β-carboline chalcones **12a-g** and their bromide salts **13a-i**. Reagents and conditions: a) Pyruvaldehyde, TFA, DCM, 30 °C, 48 h, N_2; b) 10% Pd/C, Xylene, reflux, 24 h; c) Di-tert-butyldicarbonate, DMAP, THF, 24 h; d) ArCHO, 20% NaOH, ethanol:water (1:1), 0 °C - 25 °C, 16 h; e) RBr, DMF, 50 °C, 12h.

All the prepared chalcones **12a-g** and their bromide salts **13a-i** were fully characterized using infra-red (IR), NMR (^{1}H and ^{13}C) and mass spectral data. In IR spectra, two characteristic bands at ~3370 cm^{-1} (*N*-H) and ~1650 cm^{-1}(C=O) were observed for all compounds. The ^{1}H NMR spectra for chalcones **12a-g** showed broad singlet at δ ~10.5–12.0 ppm (β-carboline *N*-H proton) and two doublets at δ ~8.4 ppm and ~7.9 ppm (enone HC=CH protons). ^{13}C NMR spectra of **12a-g** and **13a-i** exhibited signals for the carbonyl carbon at δ ~ 190 ppm. HPLC analysis (Column: Waters-C18; 250×4.6 mm; condition: 0.01% TFA in acetonitrile; flow rate = 1 mL/min; and UV detector) showed the purity of the β-carbolines **12a-g** and **13a-i** greater than 97%.

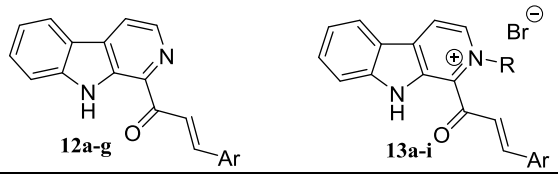
After synthesis and characterization, we studied the *in vitro* anticancer activity of β-carboline chalcones **12a-g** and their bromide salts **13a-i** against six different cancer cell lines by using MTT assay. The tumor cell line panel consisted of pancreatic cancer (BxPC-3), cervical cancer (HeLa), castration-resistant prostate cancer (C4-2), human prostate cancer (PC-3), human embryonic kidney 293 (HEK293T) and breast carcinoma (MDA-MB-231) cells. Doxorubicin was used as the reference drug. Table 1 summaries the cytotoxicity results in terms of IC_{50} values. It is evident that most of the derivatives exhibited moderate to good cytotoxicity against the tested cancer cell lines. The structural changes by varying substituents on β-carboline (R) and aryl (Ar) moieties on enone part produced the sixteen compounds with a wide ranging anticancer activity with IC_{50} values ranging from 15.9 µM to >100 µM. Compound **12a** without any substituents on β-carboline and enone moieties displayed moderate anticancer activity against a panel of cancer cell lines (IC_{50} = 65.5-93.0 µM). Replacement of phenyl group of enone moiety with tolyl, *p*-methoxyphenyl and 3,4-dimethoxyphenyl led to inactive compounds **12b-d** (IC_{50} = >100 µM). Introduction of 3,4,5-trimethoxyphenyl moiety resulted in compound **12e** with improved activity. Cytotoxicity of compounds **12f-g** could not be tested due to solubility problem.

To improve the cytotoxic activity, N^2-alkylated derivatives of β-carbolines **13a-i** were prepared. Interestingly, as shown in Table 1, compared with parent β-carboline chalcones **12c-e** (**12c**: IC_{50} >100 µM, **12d**: IC_{50}>100 µM, **12e**: IC_{50} = 70-100 µM), the N^2-alkylated-β-carbolines **13a-i** showed enhanced cytotoxic activity (IC_{50} = 15-100 µM). Incorporation of benzyl at N^2 of β-carbolines **12c-e** yielded compounds **13a**, **13d** and **13g** shown good to moderate activity (IC_{50} = 15-88 µM). Notably, compound **13g** found to be the most potent analogue of the series with broad cytotoxicity against all the tested cell lines (IC_{50} = 15.9-22.1 µM). Introduction of propargyl moiety in β-carbolines **12c-e** also resulted with similar activity (**13c, 13f** and **13i,** IC_{50} = 19.1->100 µM). However, butylated derivatives **13b**, **13e** and **13h** were inferior in the activity (IC_{50} 25.1->100 µM) when compared to N^2-benzylated analogues **13g**. The activity results suggested that N^2-alkylation of β-carboline moiety is beneficial for the anticancer activity.

We also investigated potential toxicity of compounds **12a-g** and **13a-i** against murine fibroblast NIH3T3 cell line. The results indicate that all the tested compounds **12a-g** and **13a-i** exhibit lower toxicity than the standard drug, Doxorubicin.

Next to determine the preliminary mechanism of cell death, we performed acridine orange/ethidium bromide assay.[37-39] Acridine orange is a vital dye and stains both live and dead cells. Ethidium bromide stains cells that have lost membrane integrity and tinge the nucleus red. Thus, live cells appear uniformly green in acridine orange/ethidium bromide assay. Fig 3 shows that control cells possess normal healthy morphology with intact nuclear architecture and are green in colour.

Table 1. *In vitro* cytotoxicity of β-carboline chalcones **12a-g** and their bromides **13a-i** against a panel of cancer cells and a non-cancerous cell line (IC_{50} in μM)



Compd	Ar	R	BxPC-3	HeLa	C4-2	PC-3	HEK293T	MDA-MB-231	NIH3T3
12a	C_6H_5		72.02±2.75	82.36±3.43	80.25±5.46	93.02±3.54	65.58±2.84	71.11±4.12	> 100
12b	4-$CH_3C_6H_4$		> 100	> 100	> 100	> 100	> 100	> 100	> 100
12c	4-$CH_3OC_6H_4$		> 100	> 100	> 100	> 100	> 100	> 100	> 100
12d	3,4-$(CH_3O)_2C_6H_3$		> 100	> 100	> 100	> 100	> 100	> 100	> 100
12e	3,4,5-$(CH_3O)_3C_6H_2$		70.78±4.98	75.2±3.6	71.6±4.87	> 100	82.1±5.09	71.54±4.3	> 100
12f	4-$CF_3C_6H_4$		ND	ND	ND	ND	ND	ND	ND
12g	4-$(CH_3)_2NC_6H_4$		ND	ND	ND	ND	ND	ND	ND
13a	4-$CH_3OC_6H_4$	Benzyl	50.25±4.91	60.14±4.65	65.5±5.58	88.3±7.1	41.4±3.6	52.2±4.95	85.3±5.1
13b	4-$CH_3OC_6H_4$	n-Butyl	50.79±4.91	44.99±4.32	38.79±4.91	64.99±5.5	35.18±4.1	40.9±3.96	74.6±2.50
13c	4-$CH_3OC_6H_4$	Propargyl	> 100	> 100	> 100	> 100	> 100	> 100	> 100
13d	3,4-$(CH_3O)_2C_6H_3$	Benzyl	55.3±3.48	45.25±4.41	42.05±4.35	78.95±6.4	55.14±5.8	48.23±4.69	82.25±3.33
13e	3,4-$(CH_3O)_2C_6H_3$	n-Butyl	> 100	> 100	> 100	> 100	> 100	> 100	> 100
13f	3,4-$(CH_3O)_2C_6H_3$	Propargyl	**30.14±3.41**	**25.32±2.79**	**24.4±2.64**	**29.6±3.1**	**19.14±2.78**	**21.2±3.1**	72.1±4.12
13g	3,4,5-$(CH_3O)_3C_6H_2$	Benzyl	**20±2.1**	**22.1±3.23**	**16.13±4.2**	**22.02±3.25**	**17.18±2.98**	**15.95±3.41**	55.23±5.8
13h	3,4,5-$(CH_3O)_3C_6H_2$	n-Butyl	**25.1±3.65**	**35.56±2.2**	**39.65±2.98**	**40.13±3.5**	**34.4±3.89**	**32.05±4.0**	68.6±6.54
13i	3,4,5-$(CH_3O)_3C_6H_2$	Propargyl	72.02±5.3	70.1±4.9	65.5±5.74	60.98±6.1	55.07±4.4	59.61±4.81	88±3.85
	Doxorubicin		13.56±2.21	5.23±1.92	3.2±1.1	10.2±0.98	3.59±1.1	7.85±1.59	21.32 ±2.56

*The activity data represent mean values ± SD of experiments conducted in triplicates at three independent times

Fluorescence microscopic image of MDA-MB-231 cells treated with **13g** and reference drug-Doxorubicin clearly demonstrate morphological changes characteristic of apoptotic cells formation. This suggest that **13g** induced apoptosis in MDA-MB-231 cancer cell line.

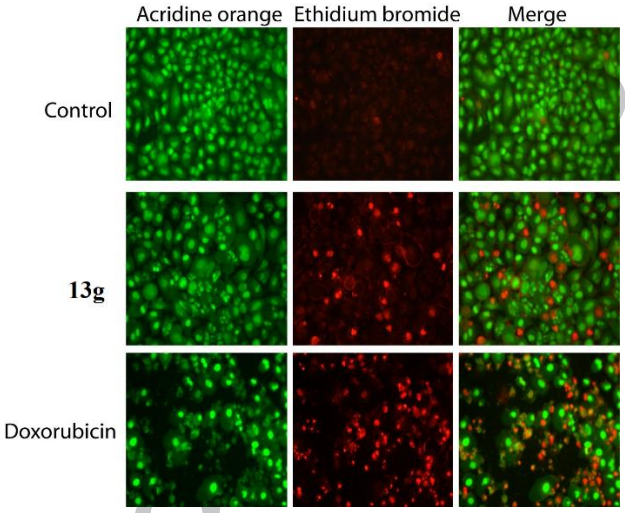


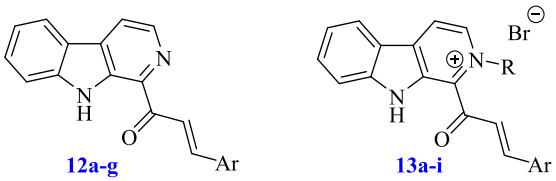
Fig. 3. Morphological assessment of **13g**-treated MDA-MB-231 cells

In the view of interesting antibacterial activities of chalcones, the newly prepared β-carboline derivatives **12a-g** and **13a-i** were screened for their antibacterial activity.[23,24,40] *In vitro* antibacterial activity of sixteen β-carboline chalcones **12a-g** and their bromide salts **13a-i** was evaluated against two Gram-positive bacterial strains including *Staphylococcus aureus* (MTCC 96), *Bacillus subtilis* (MTCC 121) and three Gram-negative bacterial strains including *Escherichia coli* (MTCC 1652), *Pseudomonas aeruginosa* (MTCC 424) and *Enterobacter cloacae* (NAIMCC-B-02025) with respect to Chloramphenicol, a standard drug. The Zone of Inhibition (ZOI) and Minimum Inhibitory Concentrations (MICs) for compounds **12a-g** and **13a-i** were determined by the modified broth micro-dilution values method as given in Table 2. The activity results described that β-carboline chalcones exerted moderate antibacterial activity against tested bacterial strains. Among all the compounds, carboline derivative **13a** was found to be most potent analogue against Gram-positive bacterial stain (*S. aureus*) with 15 mm of ZOI and MIC value of 440 μg/mL

For predicting the adsorption, distribution, metabolism and excretion (ADME) properties, computational studies for the compounds **12a-g**, and **13a-i** were performed. Lipinski's rule of five and drug likeness score were used for predicting the physiochemical properties of the molecules.[41-43] The newly prepared β-carboline derivatives **12a-g** and **13a-i** were evaluated for percentage absorption (% ABS) and drug-likeness score (Table 3). β-Carboline chalcones **12a-g** and their bromide salts **13a-i** possess lower logP values suggesting them to be better candidates for bioavailability. This was in accordance with the better cytotoxicity of β-carboline chalcones **12a-g** and **13a-i**. All the β-carbolines, **12a-g** and **13a-i** showed topological polar surface area (TPSA) less than 160 $Å^2$ but greater than 40 $Å^2$, which indicates good intestinal absorption property of these molecules than their Blood-Brain Barrier (BBB) penetration ability. Drug-likeness model score (a combined outcome of physicochemical properties, pharmacokinetics and pharmacodynamics of a compound is represented by a numerical value) was computed by MolSoft software for the synthesized compounds. Computed drug-likeness scores are presented in Table 3. β-carboline chalcones, **12c-e, 13a** and **13d-i** possessed a positive score in the range of 0.06-0.48, which implies them to be good drug candidates. The most cytotoxic compound **13g** [IC_{50} = 15.9 μM against MDA-MB-231 cell line] possessed a relatively high drug score of 0.48.

In conclusion, a library of sixteen simple β-carboline chalcones **12a-g** and bromide salts **13a-i** was prepared in good yields. *In vitro* antitumor activity of newly synthesized β-carbolines **12a-g** and **13a-i** was performed against a panel of cancer cell lines and compound **13g** displayed fairly good anticancer activity against all the tested cancer cells with IC_{50} value ranges 15.9 to 22.1 μM. Activity of N^2-benzylated-β-carboline chalcones were found to be better than that of N^2-unsubstituted-β-carboline chalcones Preliminary mechanism of action studies suggests that **13g** induces apoptosis in breast cancer cells and possess a relatively good drug score of 0.48. Additionally, β-carboline chalcones **12a-g** and **13a-i** demonstrate moderate antibacterial activity against the tested bacterial strains.

Table 2. *In vitro* antibacterial activity of β-carboline chalcones **12a-g** and their bromides **13a-i** against a panel of bacterial strains



Compd	Ar	R	Gram positive				Gram negative			
			B. subtilis		S. aureus		E. cloacae		E. coli	P. aeruginosa
			ZOI (mm)	MIC (μM)	ZOI (mm)	MIC (μM)	ZOI (mm)	MIC (μM)	ZOI (mm)	ZOI (mm)
12a	C_6H_5		9	838	-	-	-	-	-	-
12b	4-$CH_3C_6H_4$		12	720	10	820	10	828	-	-
12c	4-$CH_3OC_6H_4$		8	885	12	733	9	846	-	8
12d	3,4-$(CH_3O)_2C_6H_3$		ND	ND	ND	ND	ND	ND	ND	ND
12e	3,4,5-$(CH_3O)_3C_6H_2$		-	-	7	>900	10	834	-	-
12f	4-$CF_3C_6H_4$		-	-	11	710	11	726	10	-
12g	4-$(CH_3)_2NC_6H_4$		8	870	11	715	8	880	8	-
13a	4-$CH_3OC_6H_4$	Benzyl	8	875	15	440	8	874	8	-
13b	4-$CH_3OC_6H_4$	n-Butyl	-	-	9	-	-	-	-	-
13c	4-$CH_3OC_6H_4$	Propargyl	-	-	9	-	6	-	-	7
13d	3,4-$(CH_3O)_2C_6H_3$	Benzyl	6	-	7	-	8	-	-	-
13e	3,4-$(CH_3O)_2C_6H_3$	n-Butyl	7	-	9	-		-	-	-
13f	3,4-$(CH_3O)_2C_6H_3$	Propargyl	7	-	9	-	6	-	-	7
13g	3,4,5-$(CH_3O)_3C_6H_2$	Benzyl	7	-	9	-	12	726	-	-
13h	3,4,5-$(CH_3O)_3C_6H_2$	n-Butyl	-	-	10	812	10	806	8	12
13i	3,4,5-$(CH_3O)_3C_6H_2$	Propargyl	-	-	9	-	13	590	-	9
	Chloramphenicol		22	32	22	17	20	24	22	21

*ZOI (in mm) and MIC (in μg/mL) values. Assay experiments were performed in duplicates at two independent times

Table 3. Calculated drug-like properties of β-carboline chalcones **12a-g** and β-carbolinium chalcone bromides **13a-i**

Compd	Lipinski's parameters				nRB	TPSA[b]	% ABS[c]	No. of violations	Drug-likeness Score[d]
	nHBA (N&O)	nHBD (NH&OH)	LogP[a]	Molecular weight					
12a	3	1	4.49	298.35	3	45.75	93.22	0	-0.28
12b	3	1	4.93	312.37	3	45.75	93.22	0	-0.15
12c	4	1	4.54	328.37	4	54.99	90.03	0	0.06
12d	5	1	4.13	358.40	5	64.22	86.84	0	0.40
12e	6	1	4.12	388.42	6	73.46	83.65	0	0.29
12f	3	1	5.38	366.34	4	45.75	93.22	1	-0.27
12g	4	1	4.59	341.41	4	48.99	92.10	0	-0.32
13a	4	1	1.72	419.50	6	45.98	93.13	0	0.14
13b	4	1	1.56	385.49	7	45.98	93.13	0	-0.05
13c	4	1	0.28	367.43	5	45.98	93.13	0	0.00
13d	5	1	1.31	449.53	7	55.22	89.95	0	0.33
13e	5	1	1.15	415.51	8	55.22	89.95	0	0.16
13f	5	1	-0.13	397.45	6	55.22	89.95	0	0.21
13g	6	1	1.29	479.56	8	64.45	86.76	0	0.48
13h	6	1	1.13	445.54	9	64.45	86.76	0	0.13
13i	6	1	-0.14	427.48	7	64.45	86.76	0	0.16

[a] LogP = Lipophilicity calculated using molinspiration cheminformatics software
[b] TPSA = Topological polar surface area calculated using molinspiration cheminformatics software
[c] % ABS = Percentage absorption calculated using the formula %ABS = 109 - (0.345 x TPSA).
[d] Drug-likeness Score = calculated online using Molsoft.

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Supplementary Material

Supplementary data (general experimental procedures and analytical spectra of final compounds) associated with this article can be found, in the online version, at http://dx.doi.org

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Figures

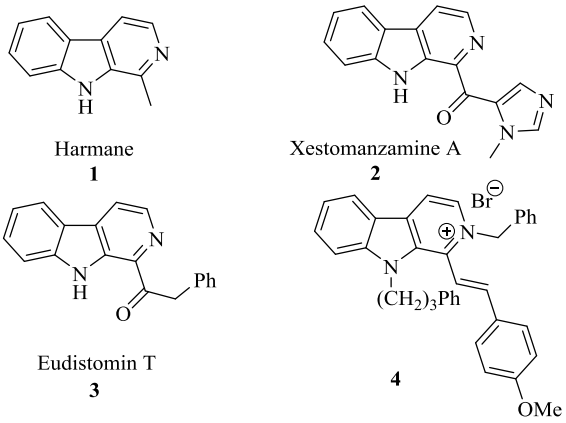


Fig. 1. Pharmacologically interesting C-1 substituted β-carbolines

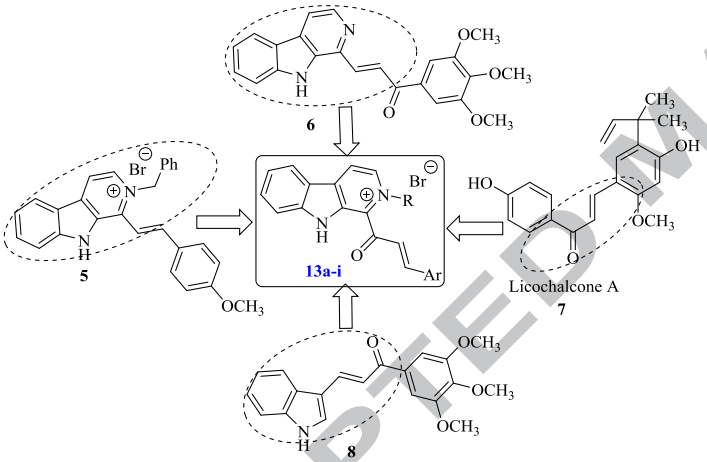


Fig. 2: Rational design of β-carboline chalcones and their bromide salts

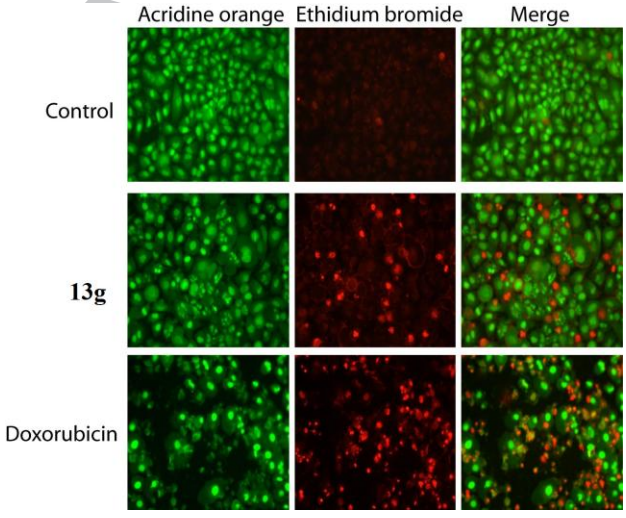
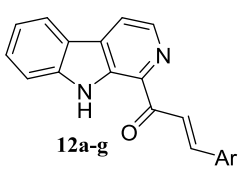
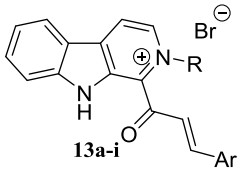


Fig. 3. Morphological assessment of **13g**-treated MDA-MB-231 cells

Tables

Table 1. *In vitro* cytotoxicity of β-carboline chalcones **12a-g** and their bromides **13a-i** against a panel of cancer cells and a non-cancerous cell line (IC_{50} in μM)

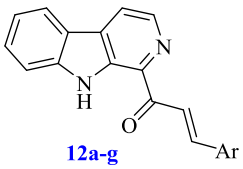


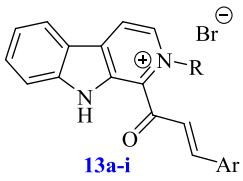


Compd	Ar	R	BxPC-3	HeLa	C4-2	PC-3	HEK293T	MDA-MB-231	NIH3T3
12a	C_6H_5		72.02±2.75	82.36±3.43	80.25±5.46	93.02±3.54	65.58±2.84	71.11±4.12	> 100
12b	4-$CH_3C_6H_4$		> 100	> 100	> 100	> 100	> 100	> 100	> 100
12c	4-$CH_3OC_6H_4$		> 100	> 100	> 100	> 100	> 100	> 100	> 100
12d	3,4-$(CH_3O)_2C_6H_3$		> 100	> 100	> 100	> 100	> 100	> 100	> 100
12e	3,4,5-$(CH_3O)_3C_6H_2$		70.78±4.98	75.2±3.6	71.6±4.87	> 100	82.1±5.09	71.54±4.3	> 100
12f	4-$CF_3C_6H_4$		ND	ND	ND	ND	ND	ND	ND
12g	4-$(CH_3)_2NC_6H_4$		ND	ND	ND	ND	ND	ND	ND
13a	4-$CH_3OC_6H_4$	Benzyl	50.25±4.91	60.14±4.65	65.5±5.58	88.3±7.1	41.4±3.6	52.2±4.95	85.3±5.1
13b	4-$CH_3OC_6H_4$	n-Butyl	50.79±4.91	44.99±4.32	38.79±4.91	64.99±5.5	35.18±4.1	40.9±3.96	74.6±2.50
13c	4-$CH_3OC_6H_4$	Propargyl	> 100	> 100	> 100	> 100	> 100	> 100	> 100
13d	3,4-$(CH_3O)_2C_6H_3$	Benzyl	55.3±3.48	45.25±4.41	42.05±4.35	78.95±6.4	55.14±5.8	48.23±4.69	82.25±3.33
13e	3,4-$(CH_3O)_2C_6H_3$	n-Butyl	> 100	> 100	> 100	> 100	> 100	> 100	> 100
13f	3,4-$(CH_3O)_2C_6H_3$	Propargyl	**30.14±3.41**	**25.32±2.79**	**24.4±2.64**	**29.6±3.1**	**19.14±2.78**	**21.2±3.1**	72.1±4.12
13g	3,4,5-$(CH_3O)_3C_6H_2$	Benzyl	**20±2.1**	**22.1±3.23**	**16.13±4.2**	**22.02±3.25**	**17.18±2.98**	**15.95±3.41**	55.23±5.8
13h	3,4,5-$(CH_3O)_3C_6H_2$	n-Butyl	**25.1±3.65**	**35.56±2.2**	**39.65±2.98**	**40.13±3.5**	**34.4±3.89**	**32.05±4.0**	68.6±6.54
13i	3,4,5-$(CH_3O)_3C_6H_2$	Propargyl	72.02±5.3	70.1±4.9	65.5±5.74	60.98±6.1	55.07±4.4	59.61±4.81	88±3.85
	Doxorubicin		13.56±2.21	5.23±1.92	3.2±1.1	10.2±0.98	3.59±1.1	7.85±1.59	21.32 ±2.56

*The activity data represent mean values ± SD of experiments conducted in triplicates at three independent times

Table 2. *In vitro* antibacterial activity of β-carboline chalcones **12a-g** and their bromides **13a-i** against a panel of bacterial strains





Compd	Ar	R	Gram positive				Gram negative			
			B. subtilis		S. aureus		E. cloacae		E. coli	P. aeruginosa
			ZOI (mm)	MIC (μM)	ZOI (mm)	MIC (μM)	ZOI (mm)	MIC (μM)	ZOI (mm)	ZOI (mm)
12a	C_6H_5		9	838	-	-	-	-	-	-
12b	4-$CH_3C_6H_4$		12	720	10	820	10	828	-	-
12c	4-$CH_3OC_6H_4$		8	885	12	733	9	846	-	8
12d	3,4-$(CH_3O)_2C_6H_3$		ND	ND	ND	ND	ND	ND	ND	ND
12e	3,4,5-$(CH_3O)_3C_6H_2$		-	-	7	>900	10	834	-	-
12f	4-$CF_3C_6H_4$		-	-	11	710	11	726	10	-
12g	4-$(CH_3)_2NC_6H_4$		8	870	11	715	8	880	8	-
13a	4-$CH_3OC_6H_4$	Benzyl	8	875	15	440	8	874	8	-
13b	4-$CH_3OC_6H_4$	n-Butyl	-	-	9	-	-	-	-	-
13c	4-$CH_3OC_6H_4$	Propargyl	-	-	9	-	6	-	-	7
13d	3,4-$(CH_3O)_2C_6H_3$	Benzyl	6	-	7	-	8	-	-	-
13e	3,4-$(CH_3O)_2C_6H_3$	n-Butyl	7	-	9	-		-	-	-
13f	3,4-$(CH_3O)_2C_6H_3$	Propargyl	7	-	9	-	6	-	-	7
13g	3,4,5-$(CH_3O)_3C_6H_2$	Benzyl	7	-	9	-	12	726	-	-
13h	3,4,5-$(CH_3O)_3C_6H_2$	n-Butyl	-	-	10	812	10	806	8	12
13i	3,4,5-$(CH_3O)_3C_6H_2$	Propargyl	-	-	9	-	13	590	-	9
	Chloramphenicol		22	32	22	17	20	24	22	21

*ZOI (in mm) and MIC (in µg/mL) values. Assay experiments were performed in duplicates at two independent times

Table 3. Calculated drug-like properties of β-carboline chalcones **12a-g** and β-carbolinium chalcone bromides **13a-i**

Compd	Lipinski's parameters				nRB	TPSA[b]	% ABS[c]	No. of violations	Drug-likeness Score[d]
	nHBA (N&O)	nHBD (NH&OH)	LogP[a]	Molecular weight					
12a	3	1	4.49	298.35	3	45.75	93.22	0	-0.28
12b	3	1	4.93	312.37	3	45.75	93.22	0	-0.15
12c	4	1	4.54	328.37	4	54.99	90.03	0	0.06
12d	5	1	4.13	358.40	5	64.22	86.84	0	0.40
12e	6	1	4.12	388.42	6	73.46	83.65	0	0.29
12f	3	1	5.38	366.34	4	45.75	93.22	1	-0.27
12g	4	1	4.59	341.41	4	48.99	92.10	0	-0.32
13a	4	1	1.72	419.50	6	45.98	93.13	0	0.14
13b	4	1	1.56	385.49	7	45.98	93.13	0	-0.05
13c	4	1	0.28	367.43	5	45.98	93.13	0	0.00
13d	5	1	1.31	449.53	7	55.22	89.95	0	0.33
13e	5	1	1.15	415.51	8	55.22	89.95	0	0.16
13f	5	1	-0.13	397.45	6	55.22	89.95	0	0.21
13g	6	1	1.29	479.56	8	64.45	86.76	0	0.48
13h	6	1	1.13	445.54	9	64.45	86.76	0	0.13
13i	6	1	-0.14	427.48	7	64.45	86.76	0	0.16

[a] LogP = Lipophilicity calculated using molinspiration cheminformatics software

[b] TPSA = Topological polar surface area calculated using molinspiration cheminformatics software

[c] % ABS = Percentage absorption calculated using the formula %ABS = 109 - (0.345 x TPSA).

[d] Drug-likeness Score = calculated using Molsoft.