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ANTICANCER AND ANTI-HIV ACTIVITIES OF SOME PYRIDO/PYRAZINO-BENZIMIDAZOLE DERIVATIVES

BAZI PİRİDO/PİRAZİNO-BENZİMİDAZOL TÜREVLERİNİN ANTİKANSER VE ANTİ-HIV ETKİLERİ

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In this study the anticancer and anti-HIV activities of some 1-hydroxy-3-arylpyrido[1,2-a] benzimidazole and 1-substituted 3-arylpyrazino [1,2-a] benzimidazole derivatives derived from 1-(2-aryl-2-oxoethyl)-2-acetyl/benzoyl-benzimidazole were investigated and appreciable activities were obtained.

Bu çalışmada 1-(2-aril-2-oksoetil)-2-asetil veya benzoil-benzimidazol türevleri ve bu bileşiklerden hareketle elde edilmiş olan 1-hidroksi-3- arilpirido [1,2-a]benzimidazol ve 1-sübstitüe 3-arilpirazino [1,2-a] benzimidazol türevlerinin antikanser ve anti-HIV etkileri değerlendirilmiş ve kayda değer aktivite değerleri bulunmuştur.

Keywords : *Pyrido and pyrazino-benzimidazole; anticancer and anti-HIV activities*

Anahtar kelimeler: *Pirido ve pirazino-benzimidazol; antikanser ve anti-HIV aktivite*

Introduction

The mitomycins (**I**) are an important class of heterocyclic antitumor antibiotics[1-3]. However, the clinical use of mitomycins is limited due to their toxicity. Hence, during the development of less toxic agents, the pyrrolo [1,2-a] benzimidazoles (PBI) (**II**) were designed as a new class of antitumor agents exhibiting activity against a variety of cancer cell lines[4-8]. These results encouraged us to study pyrido or pyrazino-benzimidazole derivatives bearing structural and isosteric relationships to PBI.

Materials and Methods

Chemistry

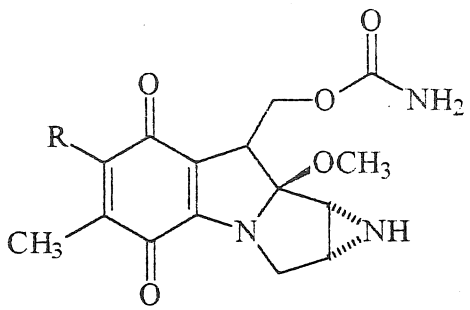
The compounds under investigations were prepared according to the methods reported in an earlier report[9] by us Scheme 1 shows the steps leading to the compounds.

Anticancer Activity

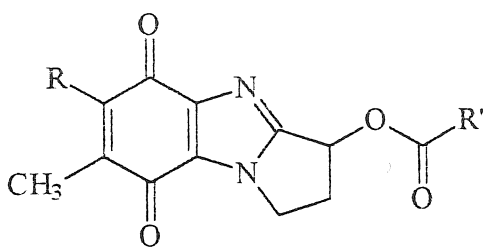
The cytotoxic and/or growth inhibitory effects of the compounds were evaluated *in vitro* against approximately sixty human tumor cell lines drived from nine neoplastic diseases namely; Leucemia (L), Non-small Cell Lung Cancer (NSCLC, Colon Cancer (CC), Melanoma (M), Ovarian Cancer (OC), Renal Cancer (RC), Prostate Cancer (PC), Breast Cancer (BC). The evaluation of anticancer activity was performed at the National Cancer Institute of Bethesda, following the in vitro screening program [10,11] which is based upon the use of multiple panels of 60 human tumor cell lines against which our compounds were tested at 10-fold dilutions of five concentrations ranging from 10^{-4} to 10^{-8} M. The percentage growth was evaluated spcetrophotometrically versus controls not treated with test agents. A 48 h continuous drug exposure protocol was followed and a sulforodamine (SRB) protein assay was used to estimate cell viability of growth.

Anti-HIV Activity

The compound was dissolved in dimethylsulfoxide,

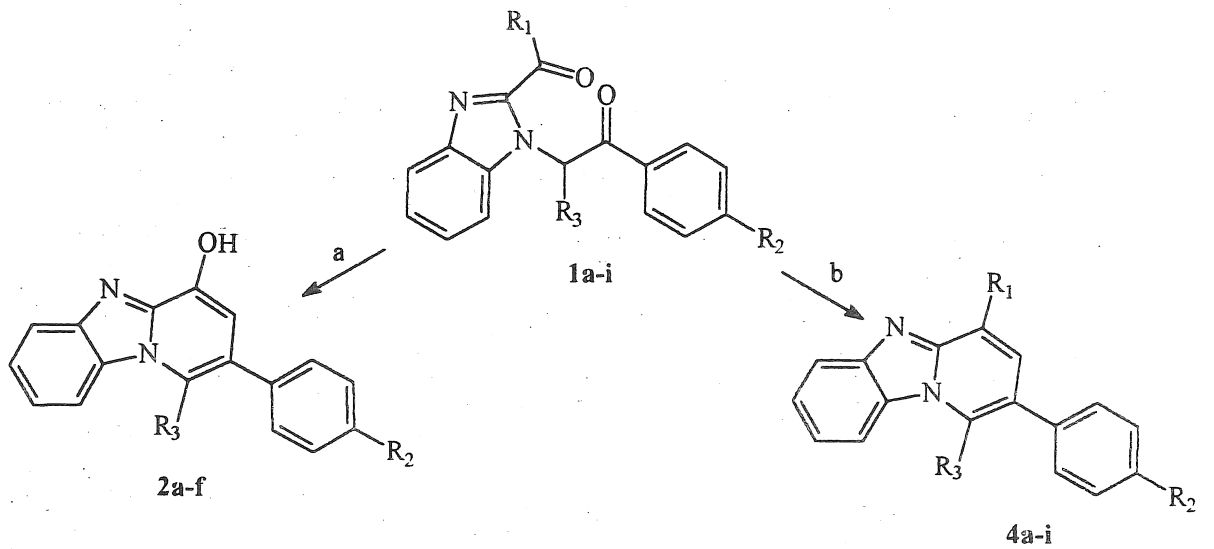


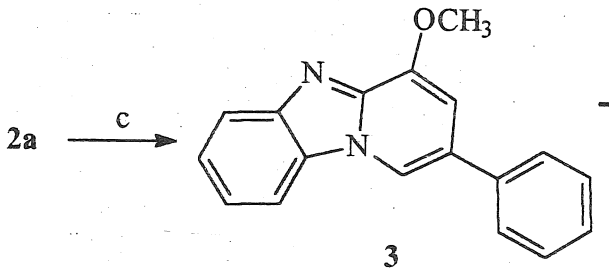
I



II

Formulae I and II





1,2 or 4	R_1	R_2	R_3
a	CH_3	H	H
b	CH_3	CH_3	H
c	CH_3	OCH_3	H
d	CH_3	Cl	H
e	CH_3	NO_2	H
f	CH_3	H	CH_3
g	C_6H_5	H	H
h	C_6H_5	CH_3	H
i	C_6H_5	Cl	H

a : K_2CO_3, 1-C_3H_7OH

b : CH_3COONH_4, CH_3COCH_3

c : CH_3I, K_2CO_3, CH_3COCH_3

Sheme 1

then diluted 1:100 in culture medium before preparing serial half log_{10} dilutions. T4 lymphocytes (CEM cell line) were added and after a brief interval HIV-1 was added resulting in a 1:200 final dilution of the compound. Uninfected cells without the compound serve as basic control, the cultures were incubated at 37°C in 5% carbon dioxide atmosphere for 6 days. The tetrazolium salt XTT was added to all the wells and the cultures are incubated to allow the development of formazan colour by viable cells. Each well is analysed spectrophoto metrically to qualitate formazan production and it is also viewed microscopically for detection of viable cells and confirmation of protective activity. Drug treated virus-infected cells are compared with drug treated non infected cells.

Anticancer and anti-HIV activity results were given in Table 1 and 2.

Table 1. Antiproliferative activity of the compounds $\log_{10} GI_{50}$* Cell type

Comp.	L	NSCLCS	CC	CNSC	M	OC	RC	PC	BC	MG-MID
1a	-5.60	-5.18	-5.23	-5.14	-5.34	-5.22	-5.11	-5.24	-5.85	-5.34
c	-5.15	-4.73	-4.94	-4.45	-4.88	-4.99	-4.70	-4.86	-5.2	-4.89
d	-5.51	-5.25	-5.27	-5.21	-5.45	-5.32	-4.90	-5.06	-5.46	-5.29
e	-5.03	-4.84	-4.82	-4.70	-4.65	-4.88	-4.72	-4.74	-4.82	4.82
f	-4.42	-4.26	-4.16	-4.19	-4.31	-4.40	-4.17	-4.16	-4.33	-4.28
g	-4.00	-4.10	-4.30	-4.01	-4.08	-4.00	-4.05	-4.00	-4.50	-4.14
i	-5.69	-4.74	-5.33	-4.88	-5.42	-5.05	-4.93	-5.36	-5.80	-5.25
2a	-5.85	-5.34	-5.63	-5.48	-5.75	-5.39	-5.20	-5.40	-6.01	-5.57
b	>-4.00	>-4.15	>-4.00	>-4.00	>-4.02	>-4.05	>-4.00	>-4.00	>-4.16	-4.05
c	>-4.00	>-4.00	>-4.00	>-4.00	>-4.00	>-4.00	>-4.00	-4.00	-4.01	-4.00
d	-5.61	-4.49	-4.41	-4.37	-4.37	-4.35	-4.08	-4.00	-4.51	-4.41
e	-4.00	-4.07	-4.00	-4.37	-4.00	-4.10	-4.01	-4.00	-4.00	-4.06
f	-4.24	-4.49	-4.57	-4.52	-4.37	-4.28	-4.30	-4.25	-4.62	-4.52
3	-4.66	-4.70	-4.71	-4.65	-4.69	-4.69	-4.75	-4.66	-4.69	-4.69
4a	-4.26	-4.67	-4.62	-4.71	-4.66	-4.85	-4.68	-4.49	-5.04	-4.69
b	-4.50	-4.58	-4.40	-4.54	-4.47	-4.53	-4.61	-4.49	-4.93	-4.58
c	-4.07	-4.28	-4.00	-4.36	-4.16	-4.45	-4.30	-4.34	-4.52	-4.28
d	-4.31	-4.10	-4.22	-4.07	-4.19	-4.35	-4.22	-4.00	-4.47	-4.23
e	-4.00	-4.07	-4.00	-4.21	-4.20	-4.12	-4.07	-4.00	-4.18	-4.11
f	-4.64	-4.55	-4.53	-4.51	-4.54	-4.65	-4.55	-4.52	-4.61	-4.57
g	-4.00	-4.42	-4.06	-4.84	-4.27	-4.78	-4.50	-4.24	-4.43	-4.41
h	4.00	-4.21	-4.02	-4.70	-4.24	-4.31	-4.44	-4.00	-4.32	-4.28
i	-4.05	-4.67	-4.40	-4.86	-4.63	-4.91	-4.70	-4.50	-4.66	-4.61

*Cells were exposed for 48 h to serial dilutions of compounds tested. Growth inhibition was evaluated by SRB assay 3. The optical density was read with a titertek microplate reader at 540 nm. Subsequent data analysis was performed with Statistica Microsoft software. Growth inhibition dose(GI_{50};M) represents the concentrations at which the percentage growth ii+50 as compared with control untreated cells (+100). The compounds, Whose $\log_{10}GI_{50}$ was higher than -4 were considered as not active.

Table 2. Anti-HIV activity of the compounds* at 2.00 10^{-4}

Compound No	Infected Response (%) of Control	Uninfected response (%) of Control
1a-i	inactive	-
2a	inactive	-
2b	15.08	64.22
2c	9.10	85.75
2d	8.56	13.58
2e	30.82	123.74
2f	4.40	4.86
3	inactive	-
4a-c	inactive	-
4d	78.50	17299
4e	79.14	107.72
4f	inactive	-
4g	18.32	36.19
4h	33.46	39.28
4i	10.13	8.72

*Control: 3'-azido-3'-deoxythymidine (AZT)

Conclusion

Considering the compounds, whose $\log_{10}GI_{50}$ was higher than -4 were considered as not active, it could be pronounced that almost all compounds exhibited appreciable anticancer activity.

The most significant increase of activity was observed for the compound **2a** (i.e.MG-MID is -5.57). The increased activity of **2a** may be due to its greater resemblance of PBI. The presence of 1-hydroxy group is the most important function in this resemblance.

The other compounds **2b-h** in the same serie which includes several aryl groups at the third position exhibited lower activity than that of **2a**. The corresponding hydroxy group in PBI derivatives were acylated. However, it is known that the activity was decreased when the hydroxy group was converted to ether function. The similar situation was observed for the compound **2h**. Considering these results, further modification of the pyridobenzimidazole structure may lead to compounds with higher anticancer activity. These modifications will be achieved in our future studies.

Some of the pyrazinobenzimidazole derivatives 4, including chlorine and nitro groups showed considerable anti-HIV activity. The compounds 4d and 4e proved to have highest activity against CEM cell lines as shown in Table 2.

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