SYNTHESIS AND BIOLOGICAL ACTIVITY OF NOVEL 2,5-DIAMINO SUBSTITUTED BENZIMIDAZO[1,2-*a*]QUINOLINES

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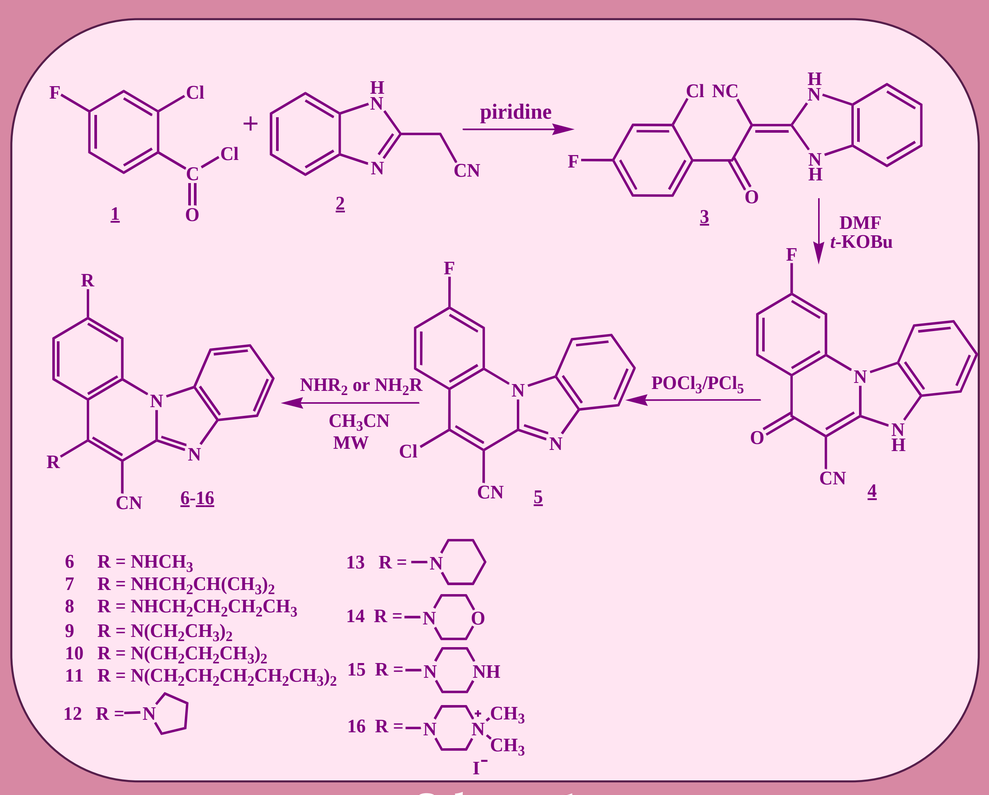
Over the past few years substituted benzimidazoles, as a very important and fundamental building skeletons of various essential synthetic and natural pharmacological compounds, have been one of the most extensively studied classes of heterocyclic compounds due to their well known biological activities. Because of the structural similarity with naturally occurring compounds such as purine, benzimidazole derivatives can easily interact with biomolecules of the living systems. High fluorescence intensity and possibility of interaction with important biomacromolecules of the living systems offer the potential use of azino fused benzimidazoles as fluorescent probes for detection of important molecules as DNA or different proteins in biomedical diagnostics.

Synthesis

As a part of our continuing research in the field of medicinal chemistry, novel 2,5-diamino substituted benzimidazo[1,2-*a*]quinolines were synthesized by uncatalyzed microwave assisted amination. All compounds were characterized by ^{1}H, ^{13}C and NOESY NMR, UV/Vis and fluorimetric spectroscopy and mass spectrometry.



Scheme 1.

Antitumor activity *in vitro*

Antitumor activity *in vitro* of prepared compounds was tested on breast (MCF-7), colon (HCT 116) and lung (H 460) carcinoma cell lines. Considering the influence of length and branching of amino side chains, it can be concluded that the longer side chains strongly and significantly reduce the antiproliferative activity, most probably due to steric hindrance of long and branched side chains. The most active compounds of this series were *i*-butylamino Substituted **7** and piperazinyl substituted derivative **15**.

Compound	GI_{50}[a] (μM) Cell lines		
	HCT 116	MCF-7	H 460
6	7±3	2±1	≥100
7	2±0.2	2±0.6	3±0.2
8	7±1	5±0.8	4±1
9	16±4	22±3	25±2
10	14±5	21±6	29±0.2
11	>100	>100	>100
12	2±0.5	2±0.6	12±1
13	11±3	10±3	15±2
14	11±5	9±1	18±8
15	1.5±0.3	1±0.001	4±0.6
16	>100	>100	>100

Table 1. Antitumor activity *in vitro* ([a]GI_{50}: the concentration that causes 50% growth inhibition)

Topoisomerase I –induced DNA relaxation

To comfort DNA binding results, we used the property of topoisomerase I to solve the constraints generated upon DNA intercalation of circular DNA plasmid. Only compounds **15** and **16** (μM) reveals profiles typical of DNA intercalation. None of the compounds are poisons of topoisomerase as compared with camptothecin (CPT).

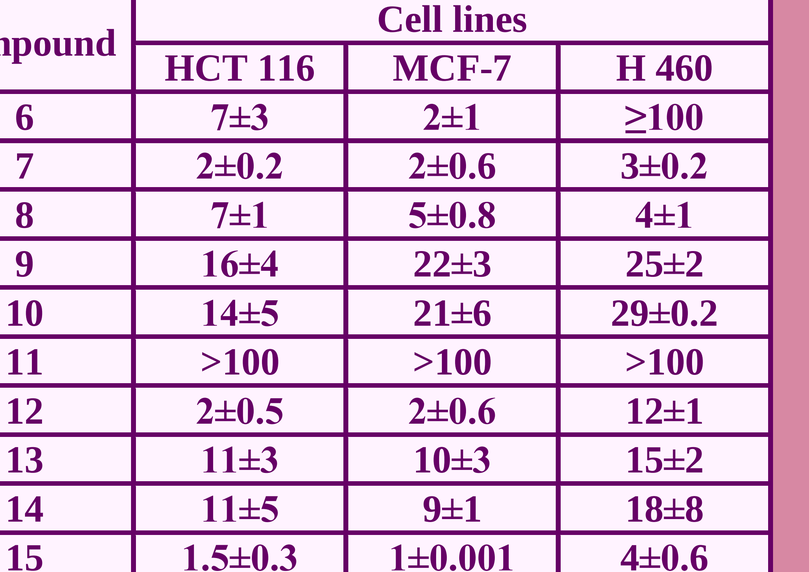


Figure 4. Topoisomerase I –induced DNA relaxation

DNA binding studies

DNA binding studies were studied with ct-DNA on **6-16** by using UV/Vis, fluorescence and CD spectroscopy. Addition of ct-DNA in the buffered solution of compound **15** decreased its absorbance intensity (Figure 1), increased its intrinsic fluorescence intensity (Figure 2) and generated negative induced circular dichroim (ICD), arguing for intercalation of compound **15** between adjacent base pairs of the DNA helix (Figure 3). Similar results were obtained using compound **16**.

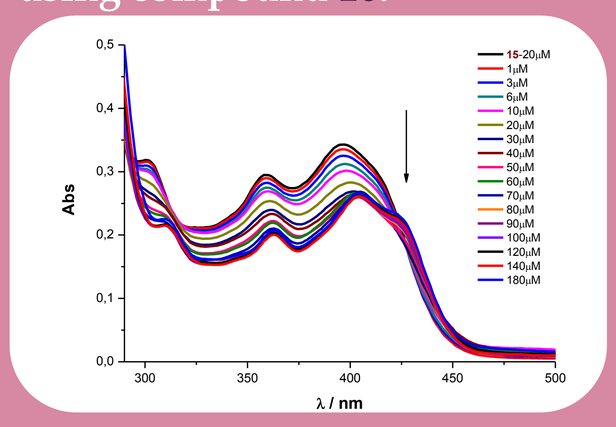


Figure 1. UV/Vis titrations of compound **15** with ct-DNA

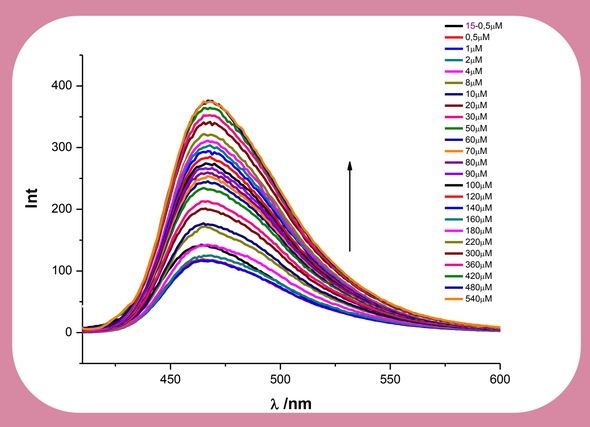


Figure 2. Fluorimetric titrations of **15** with ct-DNA

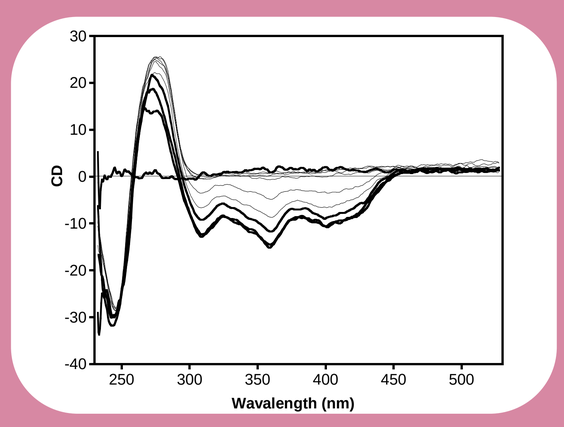


Figure 3. CD titrations of **15** with ct-DNA

To determine the possible mechanisms of biological action of tested compounds, additional biological experiments will be performed (fluorescence microscopy).

References

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3) Perin, N.; Martin-Kleiner, I.; Nhili, R.; Laine, W.; David-Cordonnier, M. H.; Vugrek, O.; Karminski-Zamola, G.; Kralj, M.; Hranjec, M. Med. Chem. Commun. 4 (2013) 1537-1550.

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