DFT and IR spectroscopic study on the conversion of 2-[2-imino-5-nitro-3-(2-oxo-2-phenylethyl)-2,3-dihydro-1h-benzimdazol-1-yl]-1-phenylethanone into radical and anionic products

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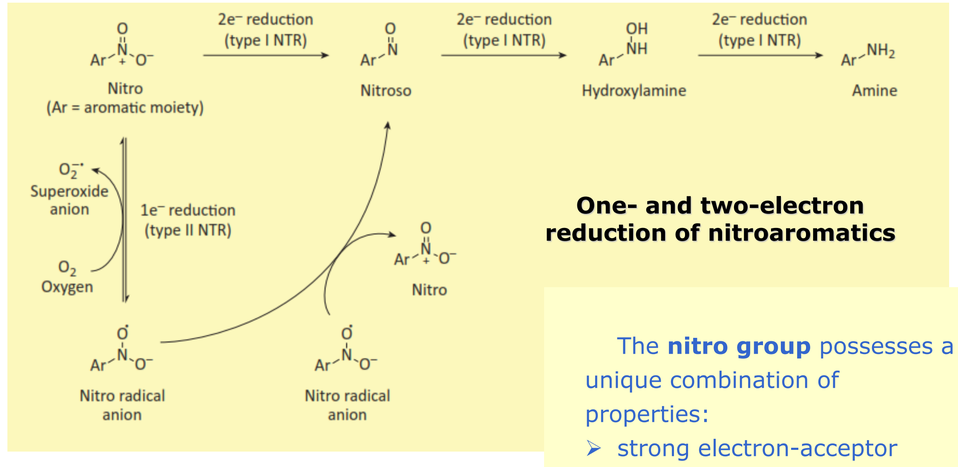
In various studies the presence of a nitro group has demonstrated a beneficial role for anti-cancer action [1,2]. A clinically evaluated 2-nitroimidazole containing lead compound (TH-302) was found to be selectively potent under hypoxia and stable to liver microsomes [3]. Some 5-nitroindazole derivatives have been examined and have shown antineoplastic activity against human colon adenocarcinoma (HT-29), human mammary adenocarcinoma (MCF-7) and human kidney carcinoma (TK-10) cell lines [4].

Studies revealed that nitro-anion radicals were generated by 5-nitroindazole derivatives through a one-electron process at physiological pH and that the mechanism of action was associated with the generation of reduced species of the nitro moiety [4]. The first step in the mechanism of action of nitroheterocyclic drugs as cytotoxic agents for cancer hypoxic cells is the reduction of the nitro group of the drug to the corresponding nitro radical anion [2].

Having in mind the structural similarity between the molecules of 5-nitromidazole, 5-nitroindazole and the 5-nitrobenzimidazole, we have focused our efforts on studying the conversion of nitrobenzimidzole derivatives into anion and radical species.



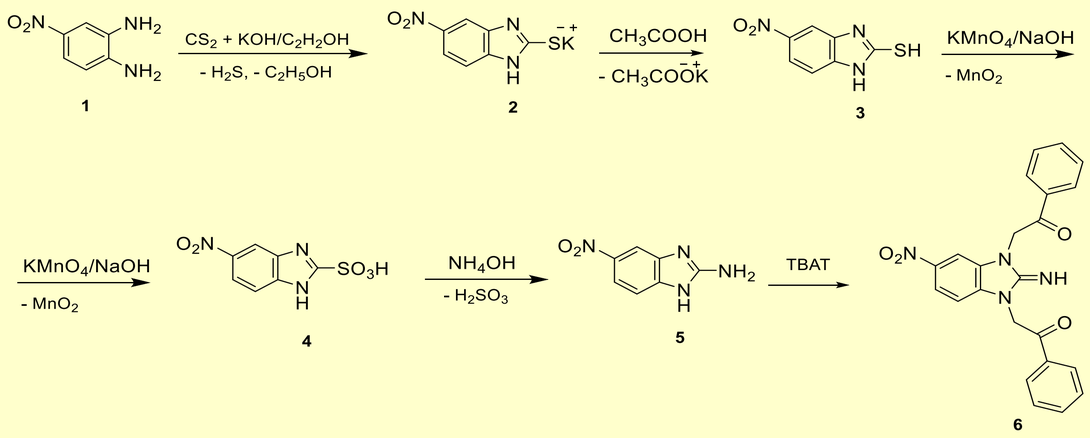
One- and two-electron reduction of nitroaromatics

The **nitro group** possesses a unique combination of properties:

- strong electron-acceptor
- small
- polar
- can form hydrogen bonds
- can be bioactivated by enzymatic reduction to give reactive species i.e. the nitroaromatic compounds act as pro-drugs

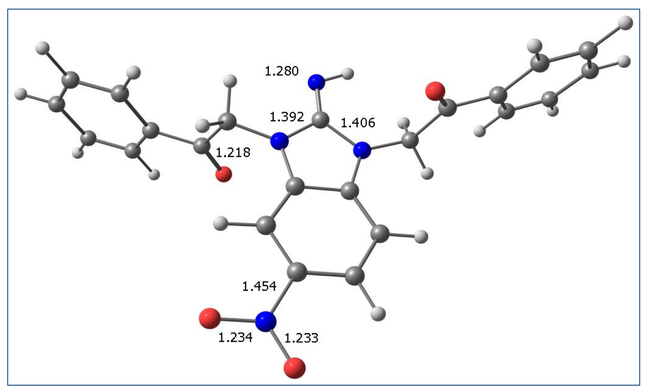
Aim of the study: computational and spectroscopic IR investigation on the electrochemical reduction of 2-[2-imino-5-nitro-3-(2-oxo-2-phenylethyl)-2,3-dihydro-1H-benzimdazol-1-yl]-1-phenylethanone and the resulting products

Synthesis of 2-[2-imino-5-nitro-3-(2-oxo-2-phenylethyl)-2,3-dihydro-1h-benzimdazol-1-yl]-1-phenylethanone (I):



IET-PCM B3LYP/6-311++G** (DMSO)

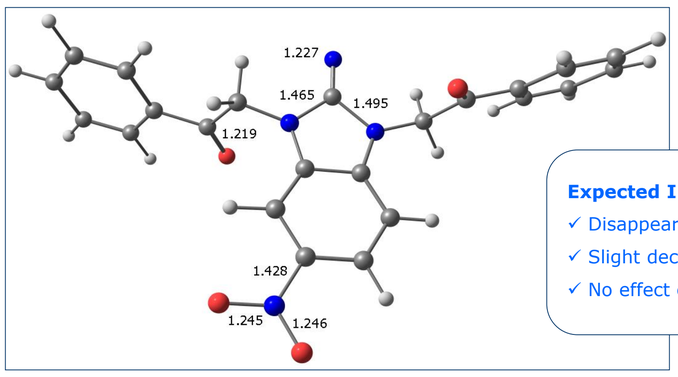
Neutral molecule (I): *selected bond lengths in Å*



According to the calculations, in solvent DMSO the neutral 2-[2-imino-5-nitro-3-(2-oxo-2-phenylethyl)-2,3-dihydro-1h-benzimdazol-1-yl]-1-phenylethanone possesses a planar 2-imino-5-nitrobenzimidazole structure, with the two oxophenyl fragments oriented perpendicularly to it. The carbonyl groups are twisted above and below the plane of the 2-imino-5-nitrobenzimidazole ring.

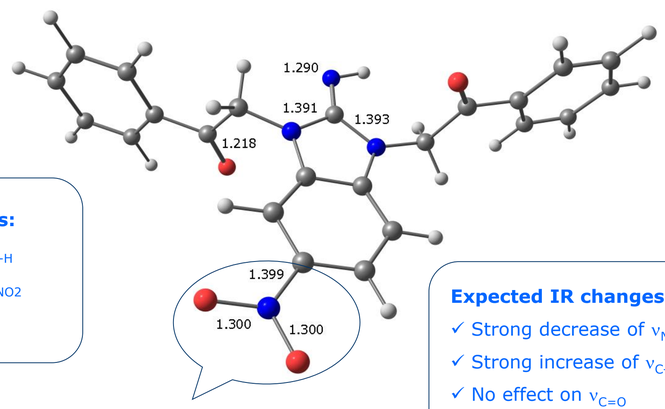
Possible anionic and radical products:

Deprotonation at N3 - anion (II):



Expected IR changes:
- ✓ Disappearance of ν_{N-H}
- ✓ Slight decrease of ν_{NO2}
- ✓ No effect on $\nu_{C=O}$

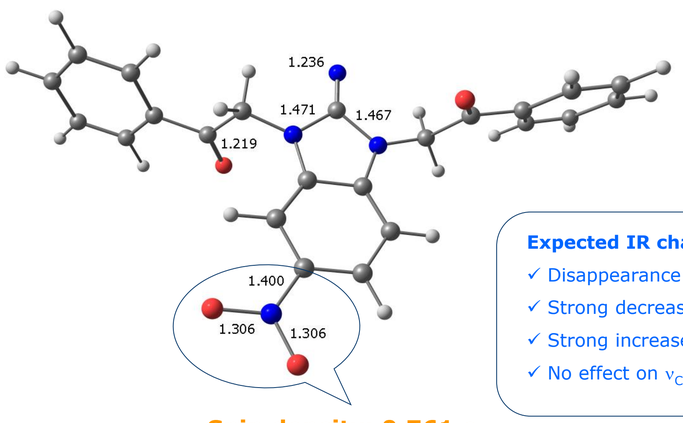
One-electron reduction - radical anion (III):



Spin density: 0.754 e-

Expected IR changes:
- ✓ Strong decrease of ν_{NO2}
- ✓ Strong increase of ν_{C-NO2}
- ✓ No effect on $\nu_{C=O}$

One-electron reduction accompanied by deprotonation at N3 - radical dianion (IV):



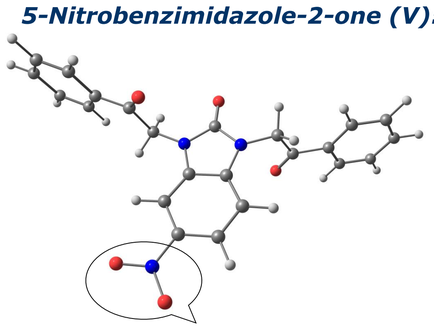
Spin density: 0.761 e-

Expected IR changes:
- ✓ Disappearance of ν_{N-H}
- ✓ Strong decrease of ν_{NO2}
- ✓ Strong increase of ν_{C-NO2}
- ✓ No effect on $\nu_{C=O}$

Comparison to the calculated parameters of other nitroaromatic compounds:

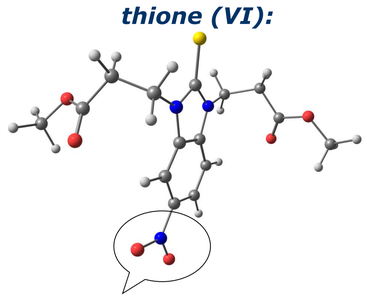
IET-PCM B3LYP/6-311++G** (DMSO)

5-Nitrobenzimidazole-2-one (V):



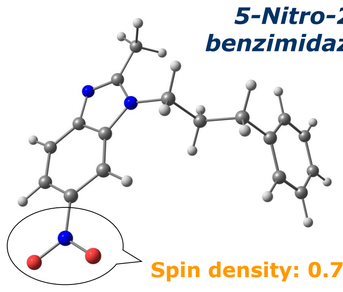
Spin density: 0.751 e-

5-Nitrobenzimidazole-2-thione (VI):



Spin density: 0.741 e-

5-Nitro-2-akhyl-benzimidazole (VII):

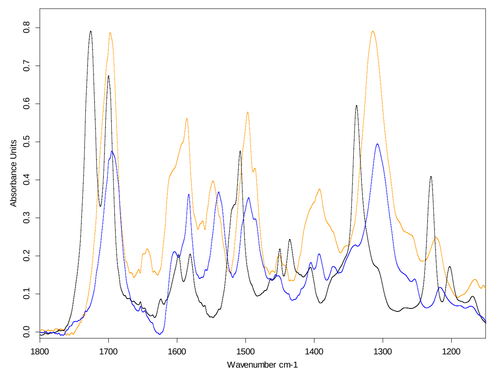


Spin density: 0.746 e-

	EA (kJ.mol^{-1})	$\Delta E_{LUMO-HOMO}$ (eV)
5-nitrobenzimidazole-2-imine (I) → RA (III)	-242.5	3.14
5-nitrobenzimidazole-2-imine (I) → DRA (IV)	-204.9	2.41
5-nitrobenzimidazole-2-one (V) → RA	-247.5	1.56
5-nitrobenzimidazole-2-thione (VI) → RA	-251.6	3.21
5-nitro-2-akhylbenzimidazole (VII) → RA	-244.4	3.85
5-nitrobenzimidazole → RA	-261.1	4.10
nitrobenzene → RA	-257.3	4.66

Electron affinity: $EA = H_{RA} - (H_M + H_{e^-})$

IR frequency changes observed during the electrochemical reduction:



The electrochemical reduction of I was carried out in aprotic solvent DMSO at 4.5 V. During the reduction, the intensity of the band at 1727 cm^{-1} (the IR spectrum of the neutral compound is depicted in black), attributed to the C=N stretching vibration, gradually decreased and completely disappeared (the IR spectrum at t = 60 min - in blue). This indicated deprotonatation of the imine group. In the same time the bands for the nitro stretching vibrations at 1508 and 1338 cm^{-1} did not shift significantly.

The IR spectrum at t = 60 min of the electrochemical reduction was compared with the spectrum of the deprotonated anionic product II, obtained by treatment of I with CD_3ONa (the IR spectrum of the anion - in orange), and showed a great similarity. Therefore, it was concluded that the electrochemical reduction at 4.5 V resulted in the generation of anion II. The generation of a radical anion product will be further attempted by electrochemical reduction at lower current.

Conclusions: The steric and electronic structure of various possible anion and radical anion products of I were characterized by DFT calculations. The calculated adiabatic electron affinities, spin density distributions and HOMO-LUMO energy gaps demonstrated good potential of the iminobenzimidazoles for development of bio-reductive anti-cancer drugs. It was also shown that the propensity to nitro reduction depends strongly on the structural modifications in nitrobenzimidazole ring.

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References: 1. C. P. Guise, A.M. Mowday. A. Ashoorzadeh, Chin. J. Cancer., 2014, 80; 2. M. Gorska, A. Kuban-Jankowska et al., Anticancer Res., 2016, 36(4), 1693; 3. J. X. Duan, H. Jiao, J. Kaizerman, J. Med. Chem., 2008, 2412; 4. V. Aran, C. Ochoa, L. Boiani et al., Bioorg. Med. Chem., 2005, 13, 3197.