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PhD Thesis Abstract

Synthesis and optical properties study of some new azaheterocyclic dyes

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Contents

Keywords: phenothiazinic dyes, phenothiazinic hydrazones, flavin, microwave assisted synthesis, UV-Vis study, fluorescence, cyclic voltammetry, DFT calculation.

Introduction

This paper presents the results obtains in synthesis and properties study of some heterocyclic aromatic compounds. It is structured in three chapters: first two chapters contain phenothiazine derivatives and the third chapter include flavin derivatives.

Phenothiazines are heterocyclic compounds with many properties which allowed its use in different fields. Due to the pharmaceutical properties, recognized long time ago, phenothiazines are used for clinical purposes, like sedative[1,2], antihelmintics[3,4], anti inflammatory, antimalarials, antibacterial[5,6], anticonvulsants[7], etc. Phenothiazinic derivatives are used as pesticides[8], in analitical chemistry, like redox indicators and reagents in spectrophotometry[9,10], as well as antioxidants for dyes and lubricants[11].

Dyes are organic and inorganic substances used since ancient times for coloring various materials. Over time, dyes have a lot of applications, from the most common, like coloring textiles and vessels to their use in food industry, as catalysts or photosensitizers in medicine. Nowadays, synthetic dyes are used in most fields, while the natural dyes are mainly used in food industry[12].

Synthetic phenothiazinic dyes present a big coloring capacity, due to the push-pull electronic effect of auxochrome groups which induce the expand of chromophore system, as well interesting electrochemical and photophisical properties.

Hydrazones are an important class of compounds in organic chemistry. Due to the ability to react with both electrophiles and nucleophiles reagents, hydrazones are widely used in organic synthesis, especially for the preparation of heterocyclic compounds[13]. Hydrazones derivatives can act as multidentate ligands, as their complexes with transition metals have been used in the treatment of tuberculosis[14,15,16].

Flavins are pteridine compounds which appear in the structure of vitamin B2, of some coenzymes. Synthetic derivatives are used in photocatalysis[17], as photosensitizers[18] or for destruction of patogens by irradiation[19].

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CHAPTER I. PHENOTHIAZINIC DYES

I.1. Overview

Phenothiazinic dyes can be classified into cationic or neutral dyes. Compounds from the first category (ex.: methylene blue, toluidine blue) have applications in textile industry, in optoelectronic devices[1,2]. Particularly, these dyes play an important role in medicine (for marking cells) and biotechnology[3,4,5].

The second class of phenothiazinic dyes with neutral structure posesing vinyl groups exhibits interesting photophysical and photochemical properties. By expanding the π system of the chromophores[6,7,8,9,10] are obtained dyes with significantly red shifted absorbtion maxima.

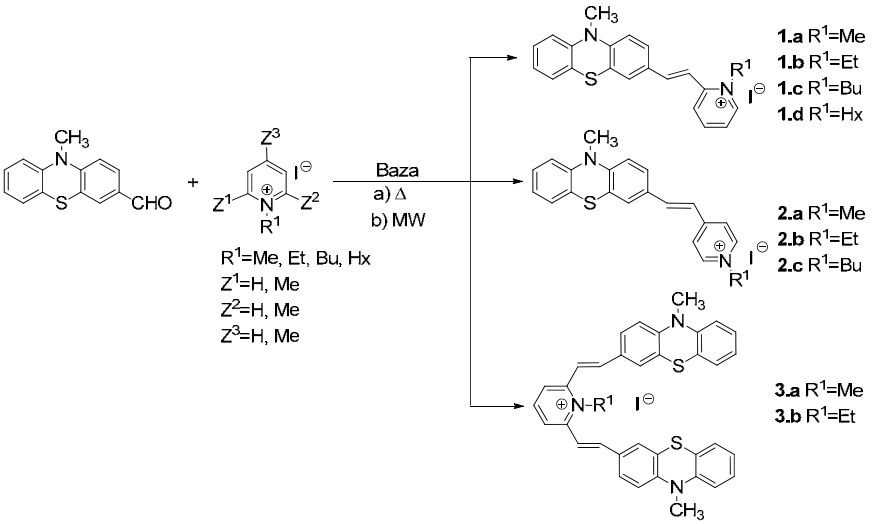
Dyes contain chromophore groups in their structures can be used for solar panels. For this purpose were synthesized phenothiazinic dyes with a cyanoacrylate radical in position 3, acting as electron acceptor, and with a (4-hexyloxy)phenyl group in position 7, as an electron donor. The nitrogen atom from position 10 was substituted with different alkyl groups[11].

I.2. Original results

Below are the results of experimental and theoretical studies on the synthesis as well photophysical and electrochemical properties of phenothiazine derivatives 3- and 3,7-substituted containing extended conjugated π electron systems[40].

Syntheses were carried out both by the classical method and microwave assisted. Microwave radiation is a source of unconventional energy used as a recent method for enabling reactions in organic synthesis[44].

These compounds were synthesized by a Knoevenagel condensation method of 10-methyl-phenothiazine-3-carbaldehyde with alkyl pyridinium salts, in presence of base (Scheme 1.6).



Scheme 1.6. Synthesis of phenothiazinic dyes: reaction conditions: a) piperidine, *i*-Pr-OH, 82^{0}C; b) microwave irradiation in dry media: solid support basic Al_2O_3, 1h, 100^{0}C.

În Table 1.1, we can see that, although yields were, with some exceptions (3.a), comparable to microwave assisted reactions. The reaction time was much shorter than for classical synthesis conditions which proves once again the advantage of using microwave reactors.

Table 1.1. Reaction conditions used in the synthesis of phenothiazinic dyes

Comp.	Temperature [^{o}C]		Time [h]		Yield [%]	
	Δ	MW[a]	Δ	MW[a]	Δ	MW[a]
1.a	82	100	25	1	70	98
1.b	82	100	25	1	35	40
1.c	82	100	30	1	42	50
1.d	82	100	35	1	29	30
2.a	82	100	25	1	69	70
2.b	82	100	25	1	65	65
2.c	82	100	30	1	48	50
3.a	82	100	35	1	45	50
3.b	82	100	35	1	42	45

[a] solid support (basic Al_2O_3)

Structures of merochinoidic dyes with phenothiazine units were established on the basis of NMR spectra. In ^{1}H-NMR spectra of compounds **1.c**, the corresponding signal of protons from

vinyl group are split as a doublet with vicinal coupling constant $^3J_{trans}$= 15.6 Hz, confirming the presence of *trans* geometric isomer (Figure 1.3).

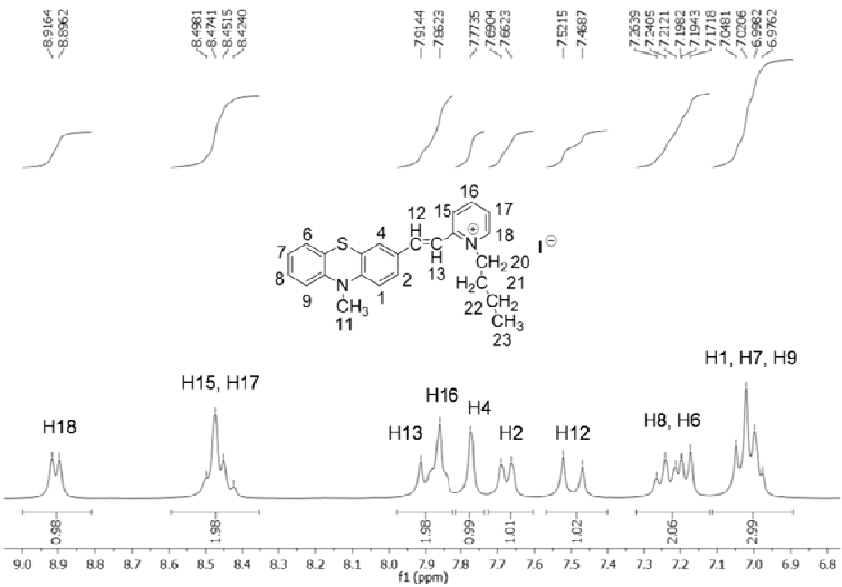
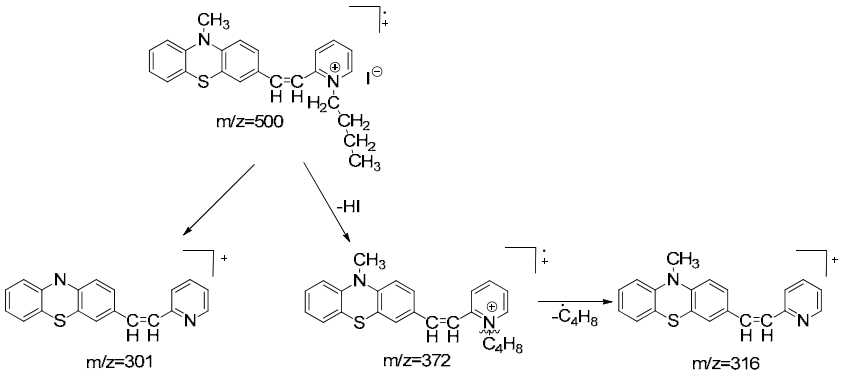


Figure 1.3. ^{1}H-NMR spectra for compound **1.c**, 300 MHz, in DMSO-d_6

To determine more precisely the structure, phenothiazine dyes were investigated also by mass spectrometry. In all cases, we can observe a peak with a low intensity obtained by removing the HI, followed by elimination of the alkyl group from the pyridinic nitrogen. For compound **1.c** in the mass spectra has been identified a peak at m/z=372 generated by the elimination of HI from the molecule of compound. The following peak at m/z=316 corresponds to remove the methyl radical from the phenothiazinic nitrogen atom, which is also the base peak (Schema 1.7).



Scheme 1.7. Fragmentation scheme of compound **1.c**

The same structure with *E* configuration of the double bond connecting the two heterocycles is present also in the solid state as it has been found by X-ray diffraction for compounds **1.c** and **2.c**. (for crystals which were obtained by recrystallization from ethanol and acetonitrile), the bond length C13-C14 being 133.8 (2) pm, respectively 134.3 (3) pm (Figurile 1.5, 1.6).

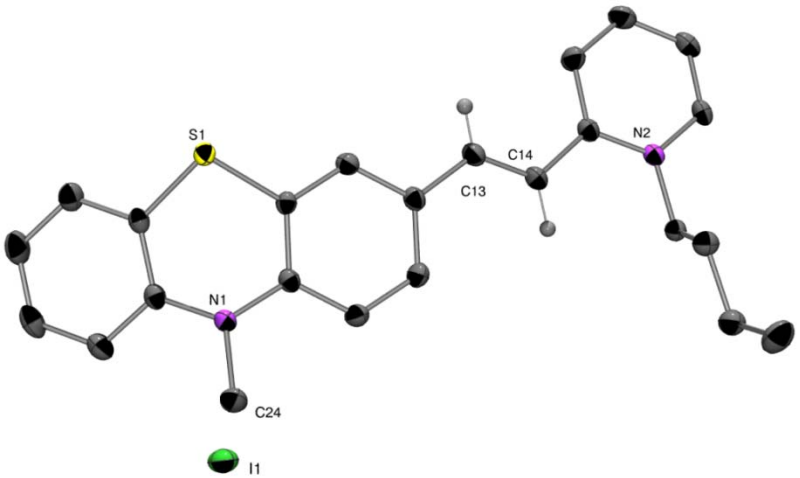


Figure 1.5. ORTEP plot of the molecular structure of compound **1.c**

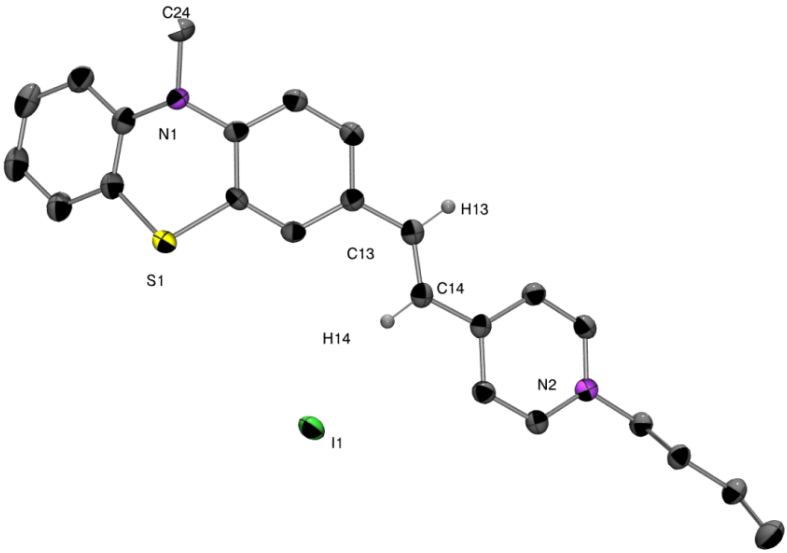


Figure 1.6. ORTEP plot of the molecular structure of compound **2.c**

Compound **1.c** crystallizes in monoclinic system P2(1)/n with four molecules in the unit cell, while compound **2.c** crystallizes in monoclinic system C2/c with eight molecules in the unit cell.

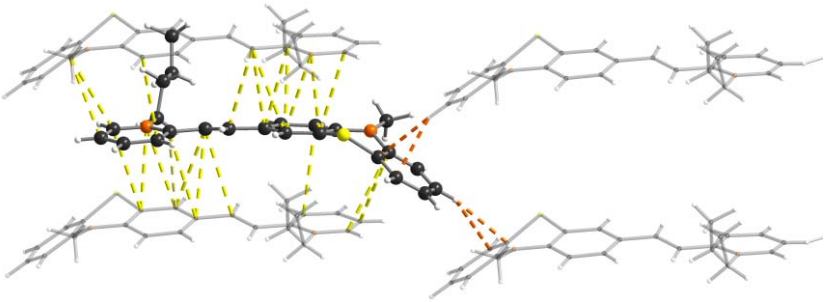


Figure 1.7. Intermolecular π-stacking for compound **1.c**

The electronic absorption spectrum of each phenothiazinic dyes (figure 1.8) contains absorption bands situated in the UV region (200-250, 322–335 nm) as well as in the visible region (444–470 nm). Bathochromic shifts are observable for the visible absorption bands of compounds

2.a-2.c. For compounds **3.a**, **3.b** was observed a hyperchromic effect correlated with conjugation of pyridine units with two chromophore vinyl-phenothiazinic units.

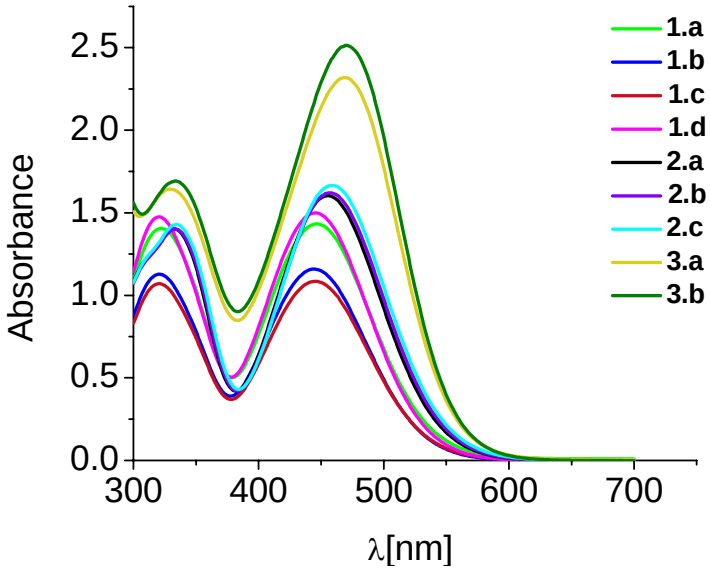


Figure 1.8. UV-Vis spectra in DMSO, 10^{-4}M, for phenothiazinic dyes

The highest value of absorbance were recorded for compound **1.c**. Therefore, were recorded UV-Vis spectra (Figure 1.9) in different solvents to study their influence on spectral properties. It was observed a low solvatochromism effect due to the red shifted of absorbtion maxima with decreasing solvent polarity (λmax = 445 nm in DMSO, and DCM λmax = 470 nm).

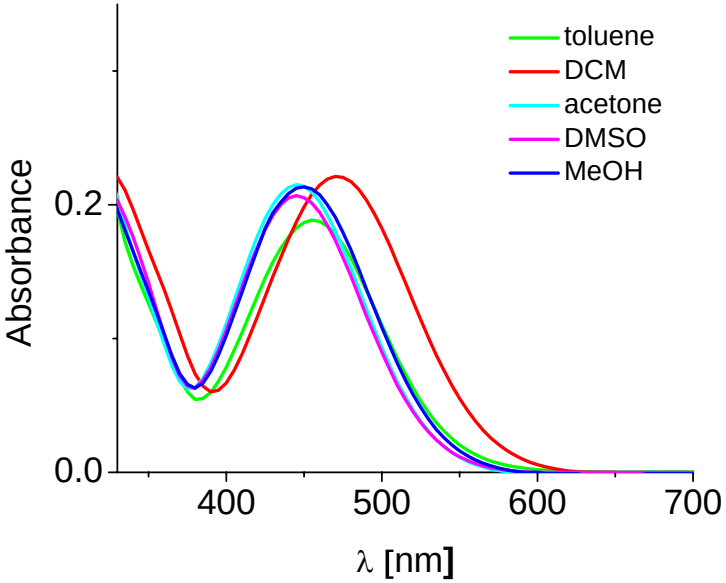


Figure 1.9. UV-Vis spectra in different solvents, for **1.c**

Computational studies:

The optical absorption spectrum was simulated using the time-dependent DFT method (GGA M06-2x(6-31G(d,p)) taking into account the lowest spin-allowed singlet-singlet transitions for the optimized molecular geometries. Frequency analysis has been performed in order to ensure that the optimized geometries are genuine minima.

An inspection of the electron distribution in the molecular orbitals of each compound indicate that the frontier filled orbitals HOMO and HOMO-1 appear located predominantly on the phenothiazine unit, whereas the unoccupied molecular obitals LUMO and LUMO+1 are located predominantly on the pyridine core (in Figure 1.10 plots of the frontier molecular orbitals of **1.a** are depicted). The lowest energy transition originating from HOMO→LUMO is a charge-transfer (CT) transition – hence the extinction coefficient observed experimentally and confirmed by the relatively large computed oscillator strength (f). The higher energy absorptions relying on excitation processes involving HOMO-1→LUMO, HOMO→LUMO+1 and HOMO-2→LUMO transitions respectively, have the same type of CT character. The bands predicted to appear below 300 nm show significant contributions from intra-phenothiazine π→π* transitions; such

contributions being also present in the lower energy bands but distinctly weaker as compared to the CT excitations (7 times less for the 500-nm band, and 2.5 times less for the 300-360 nm bands). The experimental solvatochromism was also corroborated by this theoretical approach. The electron distribution in the LUMO was tentatively assigned to a less polar excited state (with higher electron density on the pyridinium core) and thus better stabilized by the DCM than the more polar ground state represented by electrons distribution in HOMO.

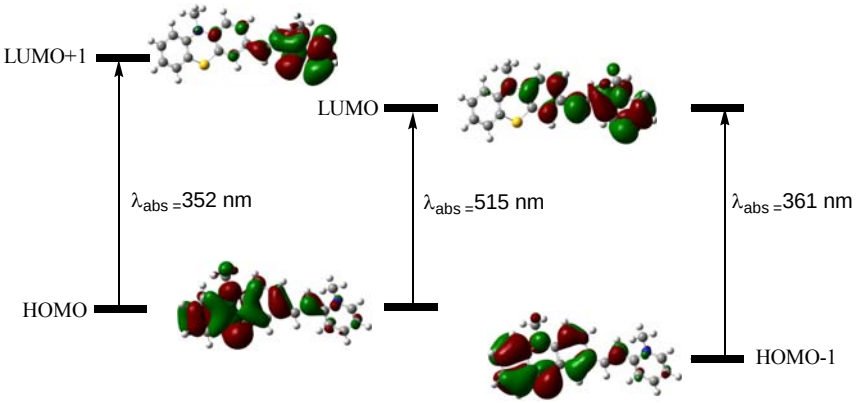


Figure 1.10. Plots of frontier molecular orbitals of **1.a** and maxima for UV-Vis absorption bands originating from the lowest spin-allowed singlet-singlet transitions computed at TDDFT level of theory

Solid state fluorescence spectra of phenothiazinic dyes presents a a single emission band in the range 610-750 nm. In solution, emission band has a low intensity or is missing. Strong non-covalent interactions between molecules may be responsible for the large red shifted emission bands in solid state as compared to the response of these in solution.

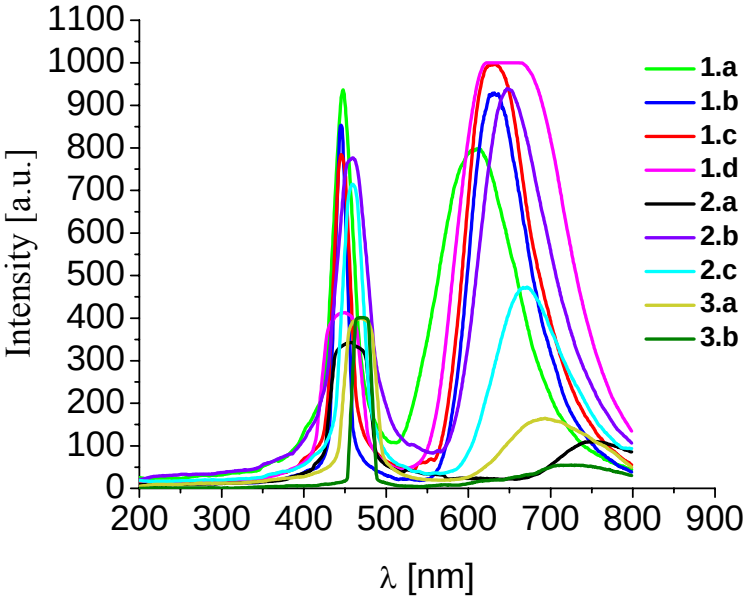


Figure 1.11. UV-Vis excitation and fluorescence emission spectra of phenothiazinic dyes in solid state at ambient temperature

Redox properties of synthesized compounds were collected by means of cyclic voltammetry experiments, in dichloromethane conducted by scanning in the anodic (up to 1.5 V) and cathodic (up to -0.2 V) region. These compounds exhibited up to three oxidation peaks as shown in Figure 1.12. The quasi reversible oxidation peaks (E_{pc}-E_{pa}= 70-120 mV) situated at $E_{1/2}$=806-889 mV are consistent with the typical redox processes of the phenothiazine core and also supported by relevant theoretical predictions such as the shape and energy of the filled frontier orbitals predominantly located on the phenothiazine unit. The enhanced oxidation potential values as compared to 10-methyl-10*H*phenothiazine ($E^{0/+1}$=767 mV) are consistent with an electron donor effect of the phenothiazine moiety.

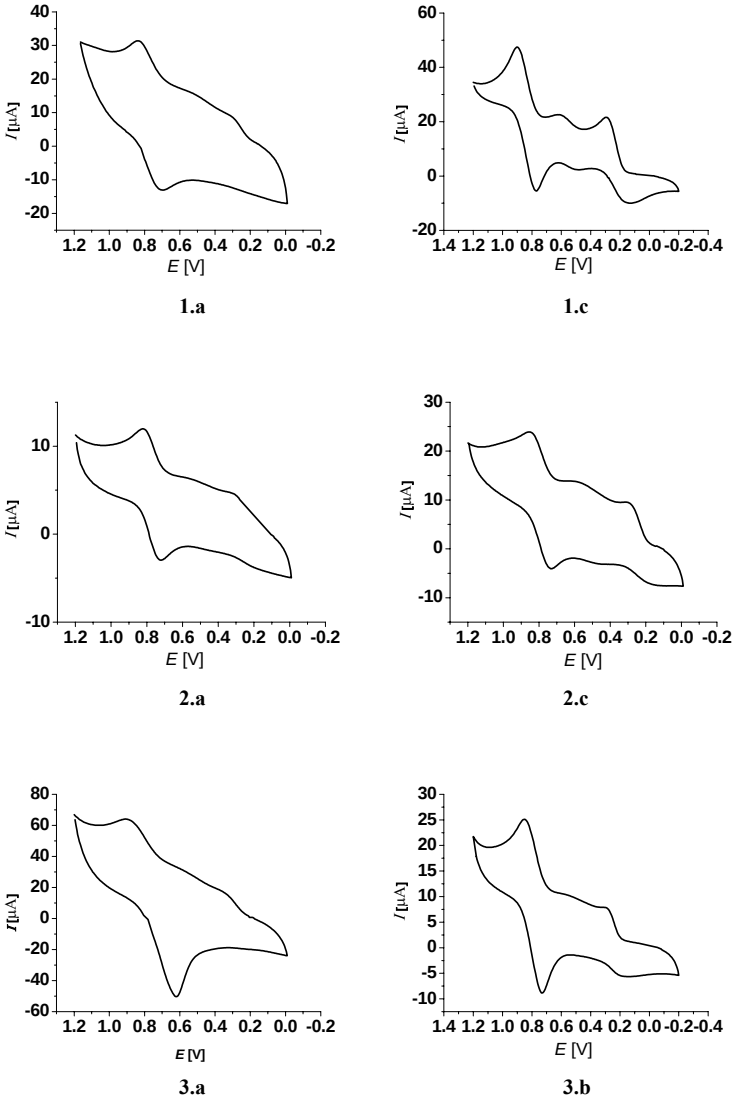


Figure 1.12. Cyclic voltammograms (recorded in DCM, 20 °C, v = 100 mV/s, electrolyte: nBu$_4$N$^+$PF$_6^-$, Pt working electrode, Pt counter electrode, Ag/AgCl reference electrode) using ferrocene/ferrocenium (Fc/Fc$^+$) as internal standard, for phenothiazinic dyes

It has also been tested biocatalytic activity of phenothiazine dyes by oxidation reaction with hydrogen peroxide in the presence of horseradish peroxidase (HRP). For phenothiazine derivatives, such oxidations are known to lead to radical cations and eventually to sulfoxides and sulfones[49].

The ascorbate peroxidase activity and oxygen affinity of hemoglobin (Hb) are enhanced by **3.a**, but no significantly effect was obtained for **1.a** and **2.a**. This difference correlates well with the already noted tendency of **3.a** to aggregate in the HRP reaction, and suggests that binding of **3.a** on the surface of Hb, as due to an increased hydrophobicity and/or tendency to self-aggregate compared to the other compounds, may on one hand accelerate the production of free radicals and on the other hand decrease the affinity of Hb for oxygen.

I.5. References

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CHAPTER II. PHENOTHIAZINIC HYDRAZONES

II.1. Overview

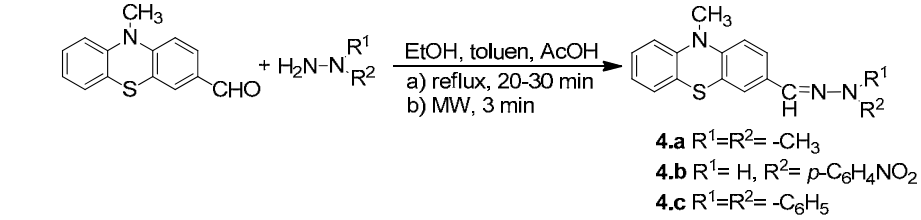
Hydrazones, compounds with formula $R_2C = NNR_2$, and their derivatives represent an important class of compounds in organic chemistry[1,2].

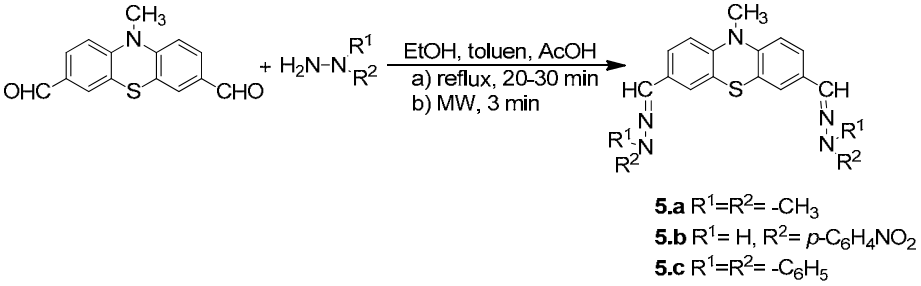
Hydrazones contain two nitrogen atoms bound together, differently hybridized (sp^2, sp^3) and a π C-N bond which is conjugated with the electron pair of the terminal nitrogen atom.

Hydrazones presents interesting biological properties, such as: anti inflammatory, analgesic, anticonvulsant, anti-tumor, anti-HIV, anti-bacterial and anti-tuberculosis activity[1]. This activity (anti-tuberculosis) is attributed to formation of chelate complexes with transition metals that catalyze physiological processes. They also act as herbicides, insecticides, nematicides, rodenticides, plant growth regulators, sterilants for mites. In analytical chemistry, hydrazones are used as multidentate ligands for transition metals in colorimetric and fluorimetric determinations[6].

II.2. Original results

Condensation of 10-methyl-phenothiazine-3-carboxaldehyde with N,N-dimethyl-hydrazine, 4-nitrophenyl-hydrazine and N,N-diphenyl-hydrazine was carried out according to classical condensation method[29], using as solvent a mixture of ethanol: toluene = 1:1 and acetic acid as catalyst. Compounds **4.a-c** (Scheme 2.4) were obtained after the mixture was heated to 80^0C for 30 minutes. A significant increase of reaction rate was observed when the reaction mixture was subjected to microwave irradiation. The reaction of the 10-methyl-phenothiazine-3,7-dicarbaldehide and hydrazine derivatives, used in a molar ratio of 1:2, takes place in a similar conditions to give good yields, obtaining bis-hydrazones **5.a-c** (Scheme 2.4). Therefore, the microwave assisted condensation is a more advantageous method considering that the reaction time is much shorter.





Scheme 2.4

Hidrazones structure **4.a-c** and *bis*- hidrazones **5.a-c** was determined by mass spectrometry (MS), FT-IR, ^{1}H- NMR, ^{13}C-NMR.

În ^{1}H-NMR spectra of compound **4.a** (Figure 2.2) the integrals total correspond to the number of protons from the molecule. Multiplicity of signals is in conformity with structure. As in other cases, the most shielded aromatic protons are H1 and H9, appearing in the spectrum as doublets and the most deshielded protons are H4 and H2 which appearing as doublet and doublet of doublets respectively.

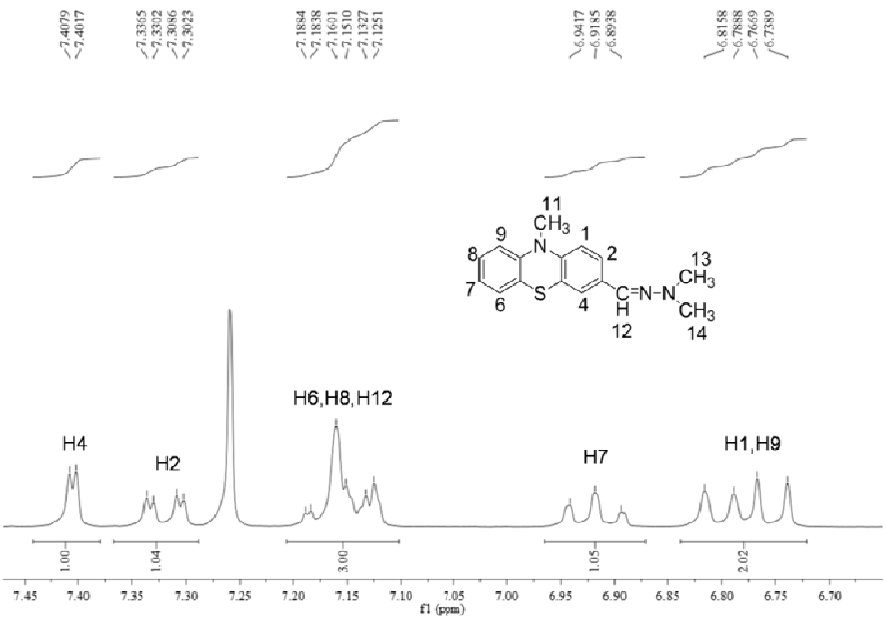
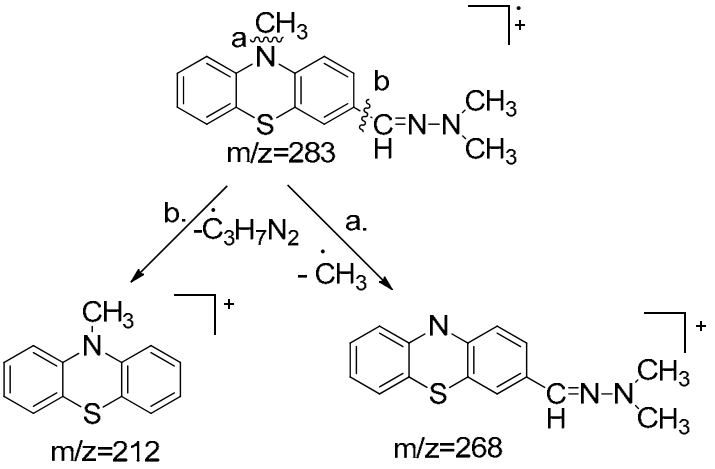


Figure 2.2. ^{1}H-NMR spectra for compound **4.a**, 300 MHz, in $CDCl_3$

Mass spectrometric studies performed on compounds **4.a-c**, **5.a-c** confirmed molecular mass values. In mass spectra of compound **4.a** (Scheme 2.5) are observed three peaks with significant intensity: one correspond to the molecular mass m/z=283, the second correspond to cationic species obtained by removing methyl radical from phenothiazinic nitrogen atom, m/z=268, and the third correspond to N-methyl-phenothiazinyl cation, m/z=212.



Scheme 2.5. Fragmentation scheme of compound **4.a**

The molecular structures of **4.a** and **5.a** were examined by X-ray diffraction on suitable crystals obtained by slow evaporation of an ethanol/acetone/chloroform solution, at room temperature during several weeks.

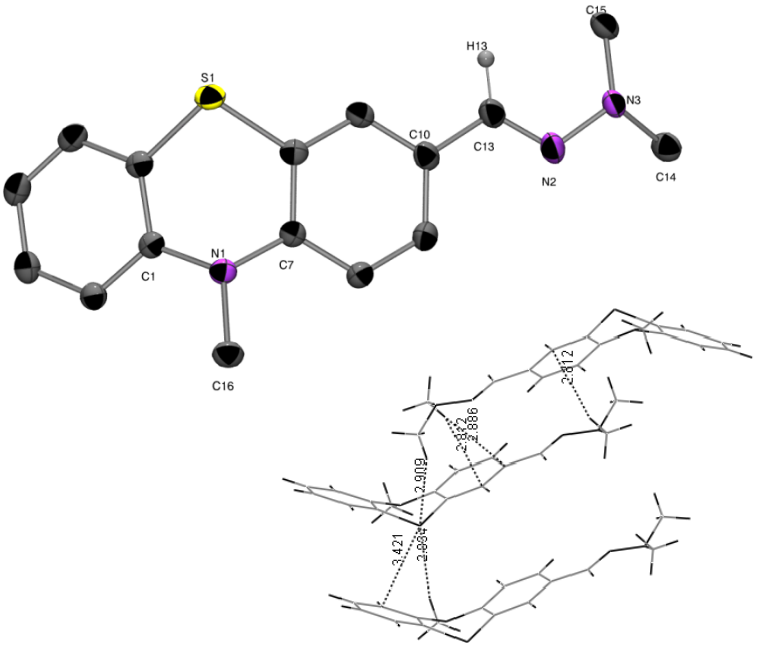


Figure 2.4. Molecular structure of **4.a**. a) Ortep plot, b) Supramolecular associations through intermolecular donor acceptor bonds: S(1)···H–C (2.894–2.909 Å), S···C–Ph (3.421 Å) şi H···C–Ph (2.812–2.886 Å)

Compound **4.a** (Figure 2.4) crystallises in the monoclinic P21/c space group with four molecules in the unit cell. The phenotiazine unit is folded about the S−N axis and the large dihedral angle of 162.2° suggests an electron withdrawing effect of the substituent. The packing diagram of **1a** shows that the molecules arrange in infinite parallel chains connected through intermolecular donor acceptor bonds as shown in Figure 2.4.b.

Bis–hydrazone **5.c** (Figure 2.5) crystallises in the orthorhombic space group P212121 with four molecules in the unit cell. For each compound **1a** and **2c**, *E*-configuration at the C=N bond can be observed.

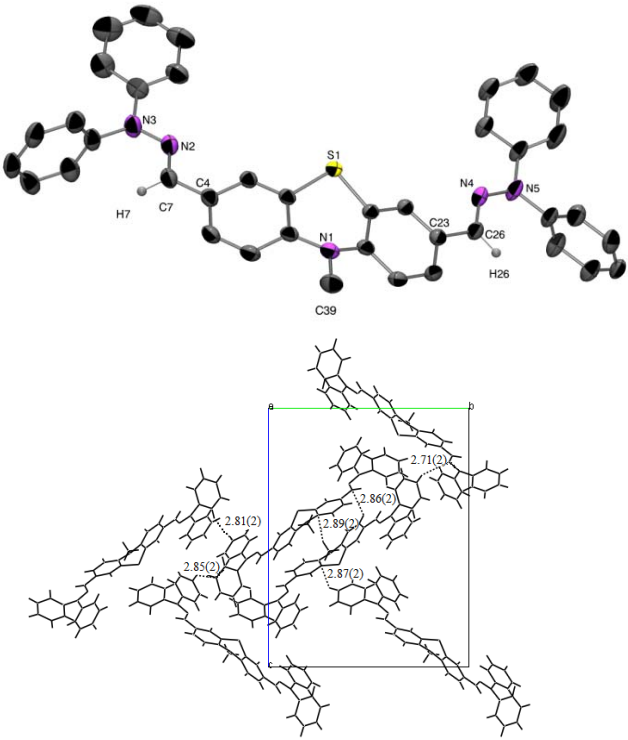
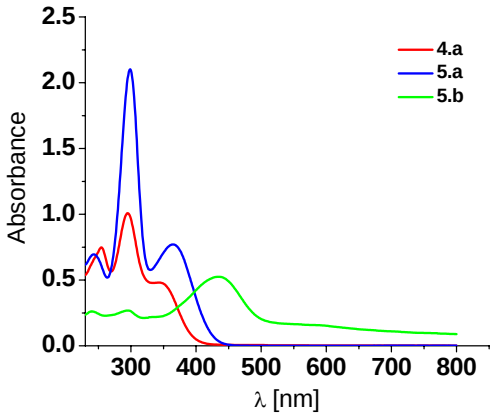


Figure 2.5. Molecular structure of **5.c** (Hydrogen atoms on aromatic rings are omitted for clarity). a) Ortep plot, b) Supramolecular associations by π-stacking were detectable as intermolecular donor

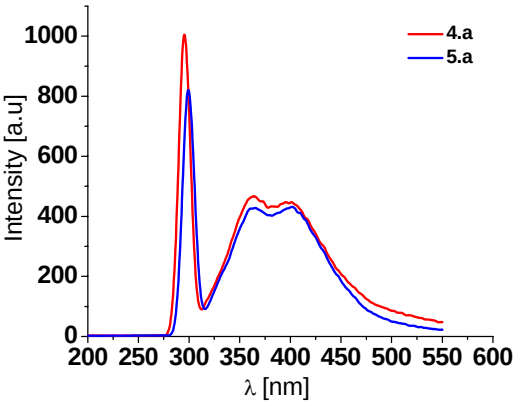
acceptor bonds: C(11)–H(11)···N(4)#2 (2.71(2) Å), C(39)–H(39C)···C(20)#1 (2.89(2) Å), C(30)–H(30)···C(20)#4 (2.87(2) Å) şi C(5)–H(5)···C(26)#1 (2.86(2) Å), C(13)–H(13)···C(17)#3 (2.81(2) Å), C(36)–H(36)···C(12)#5 (2.85(2) Å) (view along a axis)

Electronic properties of the hydrazones were investigated by UV-Vis spectroscopy, fluorescence and cyclic voltammetry. As it may be seen from Figure 2.6.a presenting the UV-Vis spectra of representative hydrazones **4.a**, **5.a** and **5.b**, strong absorption bands appear in the UV region due to the electronic transitions involving the molecular orbitals of the phenothiazine chromophore (**4.a**, **5.a**). A significant bathochromic shift can be observed for the absorption band of **5.b** which contains a *p*-nitrophenyl chromophore.



a

Figure 2.6.a) UV-Vis absorption spectra of hydrazones, 10^{-4} M in acetonitrile



b

Figure 2.6. b) Fluorescence emission spectra of **4.a**, **5.a** in acetonitrile solution upon excitation at 295 nm

Upon irradiation with UV-Vis absorption maxima, hydrazones **4.a** and **5.a** show fluorescence emissions, observed in both diluted solutions and solid states. Figure 2.4.b shows the fluorescence emissions characterized by extremely large Stokes shifts (Table 2.3). Fluorescence was not observed for the other prepared hydrazones and due to the interactions between the phenyl units, which are usually responsible for quenching the emission.

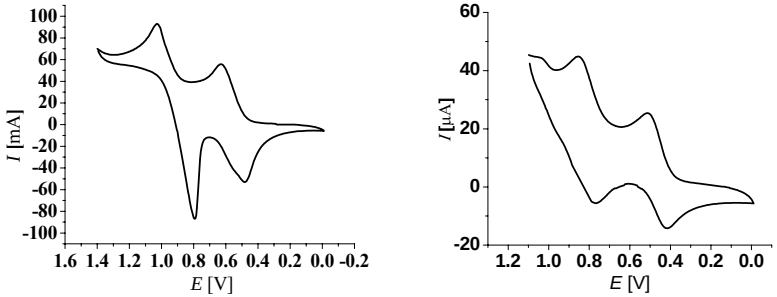
Table 2.3. Electronic properties of hydrazones determined by absorption/emission UV-Vis spectroscopy and cyclic voltammetry

Comp.	**$\lambda_{abs}(\varepsilon_{max})$ [nm][a]**		**λ_{em} [nm][a]**	**Stokes Shift [cm^{-1}]**	**$E_{1/2}^{0/+1}$ [V][b]**	**$E_{1/2}^{+1/+2}$ [V][b]**
4.a	255 (7560)	**295**(10084)	463	12300	0.607	0.986
5.a	**299**(21030)	365 (7010)	490	13000	0.527	0.879
5.b	300 (1310)	435 (5250)	-	-		

[a]solvent: acetonitrile; [b] solvent: dichloromethane

Cyclic voltammetry (CV) experiments were carried out on representative compounds **4.a** and **5.a**, in dichloromethane, using (Fc/Fc+) as internal standard, with scanning in the anodic (up to 1.5 V) and catodic (up to -0.2V) region. Each compound exhibited two oxidation steps as shown in Figure 2.7a,b. In comparison with parent 10-methyl-10*H*-phenothiazine (characterized by

the first oxidation potential $E^{0/+1}$=767 mV), the oxidation of hydrazone **4.a** occurs at lower potential values ($E_{1/2}{}^{0/+1}$=607 mV), while the oxidation of *bis*-hydrazone **5.a** proceeds even easier ($E_{1/2}{}^{0/+1}$=527 mV). The second oxidation peak, which appears at higher oxidation potentials for both compounds (Table 2.3), appears to be a reversible process in the case of *bis*-hydrazone **5.a**.



a b

Figure 2.7. Cyclic voltammograms of **4.a** (a) and **5.a** (b) in CH_2Cl_2, Pt electrode, Ag/AgCl reference, electrolyte $^{n}Bu_4N^{+}PF_6^{-}$, v=100 mV·s^{-1}

II.5. References

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CAPITOLUL III. DERIVAȚI DE FLAVINE

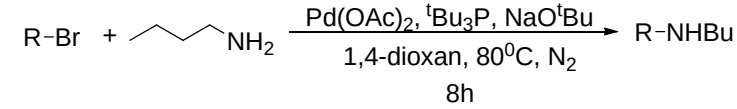
III.1. Overview

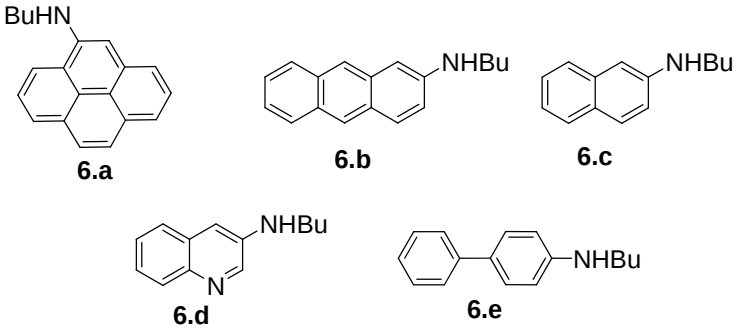
Flavins are isoaloxazinic heterocyclic compounds from pteridine class. Flavin chemistry has attracted much scientific attention, due to the discovery of a large number of flavin-containing enzymes which play a significant role in many biological processes[1]. Flavins are co-factors involved in mediating electron transfer processes[2].

Flavins can exist in three redox states: oxidized, one-electron reduced (semiquinone) and two-electron reduced (flavohydroquinone)[7]. For each oxidation state, flavins may occur in different protonated forms depending on pH[8]. Neutral form can occur in a pH range between 1 and 9, at a pH> 10 the nitrogen atom from position 3 is deprotonated, and when pH <1 is formed a cation. The oxidized flavin form is reduced to semiquinone by accepting an electron. To pH=2, semiquinone is protonated when form a cation and at pH=8 the cation is converted into an anion. Flavin reducing by accepting two electrons leading to a flavohydroquinone which is protonated when pH> 0 at nitrogen atom in position 5 and deprotonated at pH = 6.7 resulting a cationic form, anionic form respectively[4].

III.2. Original results

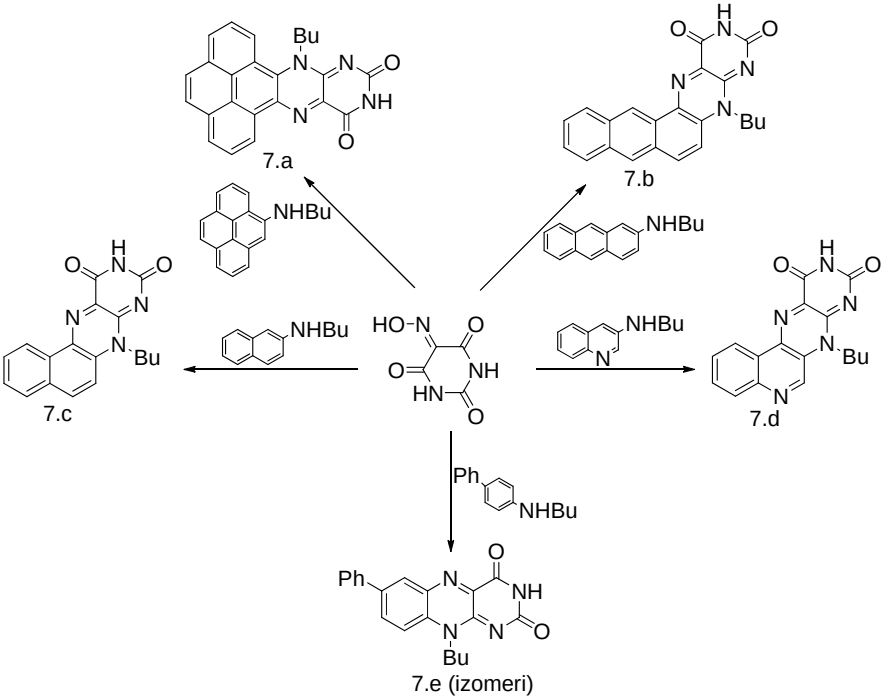
The synthesis of new flavins was accomplished by the condensation[22] of violuric acid with different aromatic amines **6a-e**. The amines intermediates were obtained via Buchwald-Hartwig amination[23] (Scheme 3.5).





Scheme 3.5. Synthesis of the secondary amines **6.a-e**

The condensation of amines **6.a-e** with violuric acid in presence of acetic acid at 118^0C[22], gives the corresponding flavins **7.a-e** in moderate to good yields (Scheme 3.6).



Scheme 3.6. Synthesis of the flavines **7.a-e** (reaction conditions: amines **6.a-e** (1.2 eq.), violuric acid (1 eq), acetic acid, 118^0C, 19 h)

The structures of flavin derivatives **7.a-e** were established by NMR spectroscopy and high resolution mass spectrometry.

In all flavins ^{1}H-NMR spectra appear four signals corresponding to the aliphatic protons of the butyl radical. The proton of the amino group is the most deshielded in all cases. In the case of compound **7.d** (Figure 3.2) the most shielded aromatic protons are H7 and H8 which appear as a multiplet, followed by H6 and H9 which appear as two multiplets and most deshielded protons are H3 and H11 which appear as singlet.

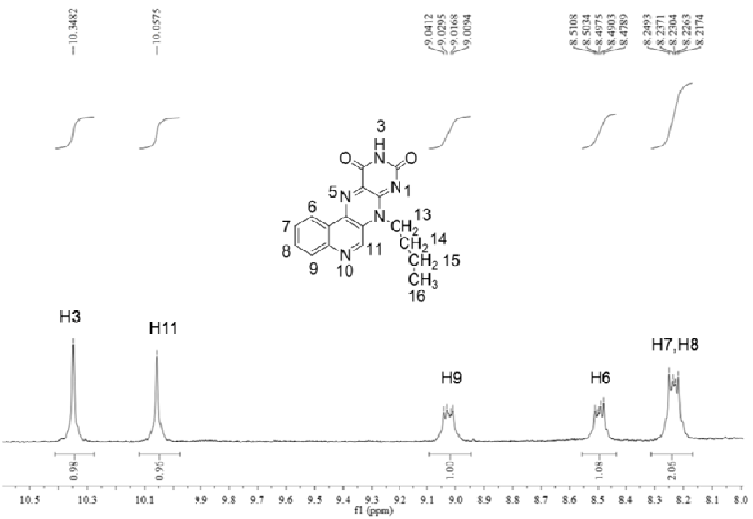


Figure 3.2. ^{1}H-RMN spectra for compound **7.d**, 300 MHz, in F_3C-COOD

The electronic properties of the flavin derivatives **7.a-e** were investigated by absorption and emission spectroscopy (Table 3.1.) and cyclic voltammetry (Table 3.2). Furthermore, the fluorescence quantum yields (Φ) of these chromophores were determined with riboflavin in ethanol as standard.

Table 3.1. Electronic properties of flavins **7.a-e**

Flavins	$\lambda_{max,\ abs}$**[nm]**		$\lambda_{max,em}$ **[nm]**		$\Phi_{fluore.}$**[%]**[a]		Φ^1O_2**[%]**[c]
	CH_3CN	CH_2Cl_2	CH_3CN	CH_2Cl_2	CH_2Cl_2	CH_3CN	EtOH
7.a	240,270, 338, 353, 494, **529**	242,281, 340, 355 503, **543**	608	611	61	38 30[b]	11
7.b	232, 258, 291, 356, **493**, 525	295, 361, 505, **542**	585	585	24	11	27
7.c	260, 304, **454**, 481	254, 262, 306, **462**, 490	508, 540 sh	511, 544 sh	19	6	35

7.d	255, 271, 302, 428, **456**	257, 309, 433, **460**	479, 508 sh	480, 509 sh	35	16	36
7.e	280, 328, **438**	283, 414, **438**	472,492	467 sh, 530	-	-	-
8	268, 341 **441**, 468	270, 347, 422, **447**, 475	505, 529 sh	505, 532 sh	31	38	93

[a] Determined with riboflavine as standard, [b] determined under argon atmosphere, [c] determined with TMPyP as standard in ethanol.

The UV absorption and fluorescence spectral data of flavin in two different organic solvents are summarized in Table 3.1. The present flavin exhibits three intense absorption bands within the range 304 – 530 nm. The energy of the long wavelength absorption band decreases by about 206 eV on going from CH_2Cl_2 to CH_3CN. These results suggest a strongly allowed π–π* transition with a charge transfer characters. Also, a pronounced red shift (4–6 nm) is observed in absorption spectra of flavins **7.a-d**, from acetonitrile to dichoromethane, and a slightly bathochromic shift in the case of fluorescence of flavins **7.a-e**. The extinction coefficient is lower for the expanded flavin **7.a-e** in comparison with butylflavin, **8** (Figure 3.3). All compounds display intense yellow daylight fluorescence, except pyren- and antracen cromophore which display orange daylight fluorescence, with a fluorescence quantum yields in dicholoromethane between 19 and 61 %, respectively between 6 and 38% in acetonitrile (Figure 3.4). In particular, the emission data can be attributed to the presence of a pyrene moiety which seems to be the dominant fluorophore in both solvents.

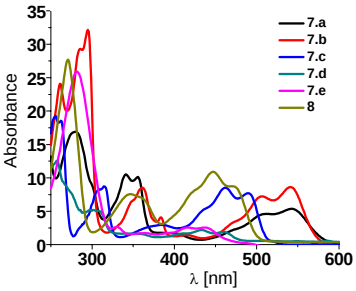


Figure 3.3. UV-Vis spectra of flavins **7.a-e** in CH_2Cl_2

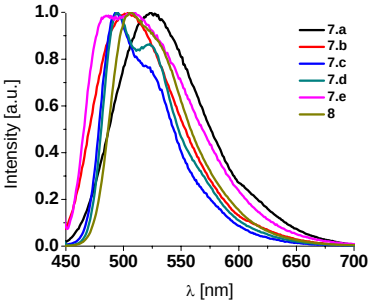


Figure 3.4. Fluorescence spectra of chromophores **7.a-e** in CH_2Cl_2

Furthermore, it was investigated the singlet oxygen generation and the photostability of flavins **7.a-e**. It was observed that flavin derivatives have less singlet oxygen generation with quantum yields between 11- 38 % in comparison with butylflavin, **8** (Table 3.1). The choromophores **7.a, 7.b, 7.c**,**7.e** seems to be more photostable then flavin **8**, except quinolineflavin,**7.d**, which is bleaching after 5s of irradiation (Figure 3.5).

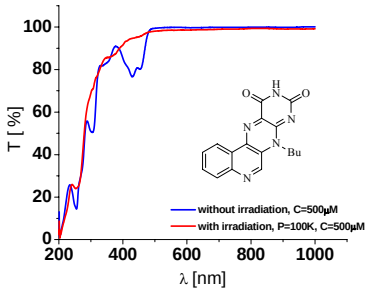


Figure 3.5. Photostability of flavin **7.d** after 5 s of laser irradiation

Fluorescence variation for compound 7.d in the presence of acids and bases

It was investigated the emission of flavin **7.d**, in CH_2Cl_2, in presence of small portions of TFA (trifluoroacetic acid) and TEA (triethylamine). Emission spectra of the titration experiments are shown in Figures 3.6, 3.7, 3.8.

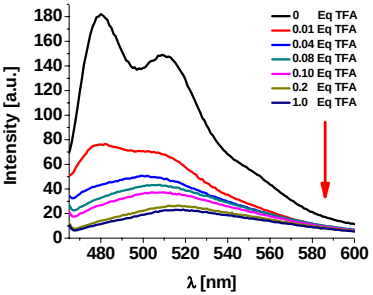


Figure 3.6. Emission spectra of **7.d** in the presence of TFA

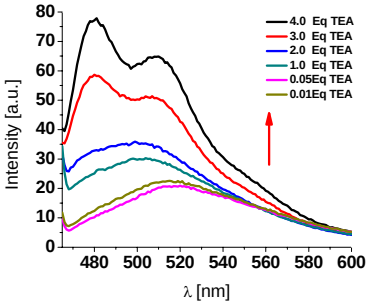


Figure 3.7. Emission spectra of **7.d** in the presence of TEA (0.01- 4.0 eq TEA)

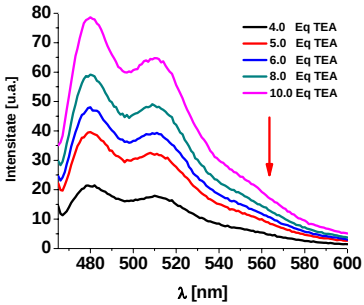


Figure 3.8. Emission spectra of **7.d** in the presence of TEA (4.0- 10.0 eq TEA)

Compound **7.d** display an emission at 408 nm (Figure 3.6). After the addition, in small portions, of 0.1 eq- 2 eq of TFA the emission is quenched. We assume that the pyridine unit are protonated, leading to a species with a very low band gap, as the LUMO is stabilized by protonation. This species are nonfluorescent as there apparently are effective paths available for non-radiative excited-state deactivation, in line with energy gap law[25]. Upon further addition of TFA, the flavine units are likewise protonated. However, this process is reversible, since the neutralization of the acidic sample with a base (triethylamine) led back to the fluorescence signal (Figure 3.7). However, the corresponding emission signal is not identical to the original, because after adding 4 eq of TEA (Figure 3.8) emission is quenched again, assuming that flavin **7.d** was reduced. This shows that the fluorescent properties of quinolinflavin, **7.d**, can be modified depending on its protonation or deprotonation.

Cyclic voltammetry measurements

Table 3.2. Redox potentials of flavins **7.a- e**

Flavins	$\mathbf{E_0^{0/-1}}$ [mV][a]	$\mathbf{E_0^{-1/-2}}$[mV][a]	$\mathbf{E_0^{-2/-3}}$ [mV][a]	$\mathbf{E_0^{-3/-4}}$[mV][a]
7.a	-614[b]	-1258 (ΔE=60)[b]	-2050 (ΔE=60)[b]	-
7.b	-658	-1178 (ΔE=63)	-1405[c]	-1973 (ΔE=60)
7.c	-705	-1423[c]	-	-
7.d	-550[c]	-781	-1.211[c]	-
7.d TFA	-62[c]	-213[c]	-608[c]	-707
7.e	-340[c]	-606	-1.196	-
8	-716	-1409[c]	-	-

[a] Recorded in AcCN, 20 °C, v = 250 mV/s, electrolyte: $^{n}Bu_4N^{+}$ PF_6^{-}, Glassy carbon working electrode, Pt counter electrode, Ag wire reference electrode, internal standard Fc/Fc^{+}; [b] recorded in DMSO

According to cyclic voltammograms we can observe 2-4 reduction peaks for compounds **7.a-e** (Table 3.2, Figure 3.9). The first reduction potential, $E_0^{0/-1}$, is dependent on the aromatic units structure, its values being in the range -340 and -705 mV (Table 3.2). Compared to butylflavin (reference flavin), **8**, ($E_0^{0/-1}$= -716 mV, $E_0^{-1/-2}$= -1409 mV), the first reduction potential is reversible and shifted to anodic area (**7.a**: $E_0^{0/-1}$= -614 mV, **7.b**: $E_0^{0/-1}$= -658 mV, **7.c**.: $E_0^{0/-1}$= -705 mV, **7.d**: $E_0^{0/-1}$= -550 mV, **7.e**: $E_0^{0/-1}$= -340 mV). The second reduction step occurs at higher potentials (**7.a**: $E_0^{-1/-2}$= -1258 mV, **7.b**: $E_0^{-1/-2}$= -1178 mV, **7.d**: $E_0^{-1/-2}$= -781 mV, **7.e**: $E_0^{-1/-2}$= -606 mV), with the exception of flavin **7.c**.: ($E_0^{-1/-2}$= -1423 mV), which has the second potential almost identical to butylflavin **8**.

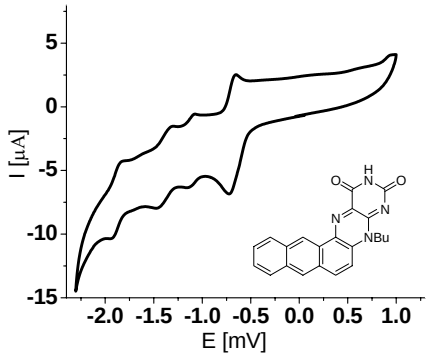
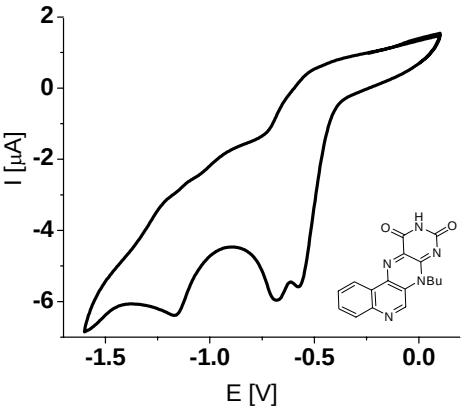
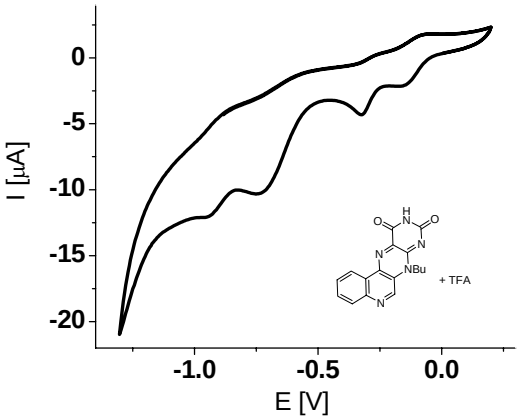


Figure 3.9. Cyclic voltammetry of **7.b** recorded in CH_3CN, 20 °C, electrolyte: $^{n}Bu_4N^+PF_6^-$, Pt working electrode, Pt counter electrode, Ag wire as reference electrode, internal standard Fc/Fc^+, v = 50 mV/s

Using cyclic voltammetry was studied redox behavior of quinolineflavin, **7.d**, in the presence of TFA (Figure 3.10). It should be noted that the presence of the protonated form of quinoline unit is reflected in the reduction potential values. For example, the trianionic species of flavin **7.d**, is reduced to a potential shifted to the anodic area, compared to quinolineflavin free base. Even more, by the addition of TFA, it was observed a fourth reduction potential.



a



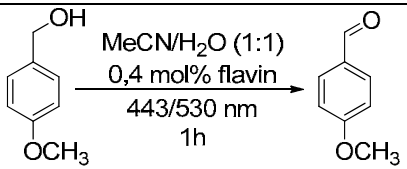
b

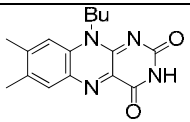
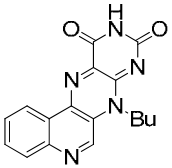
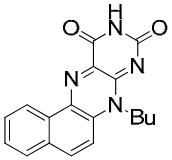
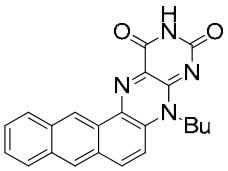
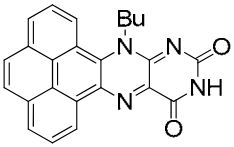
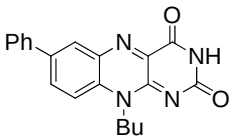
Figure 3.10. Cyclic voltammetry of **7.d** (a) and **7.d+TFA** (b), recorded in CH_3CN, 20 °C, electrolyte: $^{n}Bu_4N^+PF_6^-$, Pt working electrode, Pt counter electrode, Ag wire as reference electrode, internal standard Fc/Fc^+, v = 50 mV/s

Photocatalytic oxidation reactions with flavin derivatives

The flavins which were described before were tested as catalysts in some photocatalytic oxidation reactions. The *p*-methoxy benzyl alcohol was used as a substrate for check the flavins activity (Tabel 3.3). Also, butylflavin (position 1, Table 3.3) was used as reference.

Table 3.3. Photocatalytic oxidation of *p*-metoxy benzyl alcohol



Catalyst	443 nm η %[a]	530 nm η %[a]	Φ^1O_2%
	24	-	0.2
	**40** **57**[b]	-	**1.3**
	6	-	0.04
	-	1	0.03
	-	<1	0.008
	2	-	0.05

a) yields determined from GC-FID method; b) 0.01 eq. TFA; conditions: $C_{alcohol}$=0.2M, GC internal standard=CB (0.2 M)

From Table 3.3. it can be observed that the best yield and quantum yield was obtained for quinolinflavin (**7.d**, second position). Butylflavin (**8**, first position) gave also a moderate yield, while the conversion for the other flavins is low. In the case of quinolinflavin (**7.d**) it was added 0.01 eq. TFA for protonating the nitrogen atom from the pyridine unit. For this reaction it was obtained a higher yield than the first, which means that the protonated form of quinolinflavin is a better oxidant than quinolinflavin (**7.d**).

III.5. References

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General Conclusions

This paper presents the synthesis and investigation structure and properties of 37 compounds, 21 of them being new substances.

The aim of this work was to obtain new organic compounds with potential biological and photochemical potential. Therefor have been made various condensation reactions to obtain hydrazones, phenothiazine dyes (Knoevenagel condensation), and flavin derivatives.

For the synthesis of hydrazones and phenothiazinic dyes were used as starting materials 10-methyl-10*H*-phenothiazine-3-carbaldehyde and 10-methyl-10*H*-phenothiazine-3,7-dicarbaldehyde and for the flavin derivatives was used violuric acid.

In the case of phenothiazinic dyes and hydrazones, reactions were carried out both by the classical method (convective heating) and by microwave irradiation, applying irradiation technique in solvent or "dry media" in the presence of solid support (for the phenothiazinic dyes). In these conditions were obtained similar yields and reaction time was significantly reduced.

Electronic properties were determined by UV-Vis absorption and fluorescence spectroscopy, cyclic voltammetry, highlighting the isoaloxazinic (for flavins) and phenothiazine rings characteristics.

The synthesized compounds were characterized by spectroscopic, spectrometric and electrochemical methods: UV-Vis, fluorescence, IR, NMR, MS, cyclic voltammetry and elemental analysis.

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