Theoretical and Experimental Investigations Concerning the Fluorescence of some Azaheterocycles

PhD Thesis Abstract
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Introduction and objectives

Photophysics of five- and six-membered ring aromatic azaheterocycles is dominated by low-lying excited states involving electron promotion either to diffuse $\sigma^*$ N–H antibonding orbitals (in acidic pyrrole and imidazole [1–4]) or from nitrogen lone-pairs orbitals ($n\pi^*$ states in pyridine and all the three diazines, respectively [5–8]). Consequently, most of these molecules lack the efficient fluorescence emission required for practical applications.

Both types of nonradiative dezactivation channels are suppressed in indolizine, a N-bridgehead 5,6-fused ring heterocycle derived formally from pyrrole and pyridine (see Figure 1). Unsubstituted indolizine, as well as the few azaindolizines for which photophysical properties were reported in the literature during the mid 70’s [9–11], exhibit highly efficient fluorescence emission in the near-UV spectral region.

More recent experimental works on functionalized indolizine and pyrrolo[1,2-b]pyridazine show that the emission properties of the heterocycle are preserved by some functional groups, whereas others impact on fluorescence
emission, apparently in an unsystematic fashion [12–17]. In case of the former, potential applications areas suggested by several authors include optoelectronic devices [12] (provided the blue fluorescent emission of pyrrolo[1,2-b]pyridazine derivatives), chemical sensors [18] (by pH- [19, 20] and solvent-dependent spectral shifts) etc.

Although few previous attempts in rationalizing the optical spectral properties of indolizines exist, none has accounted the entire picture and only few addressed different aspects with satisfactory results.

The first theoretical approaches [21, 22] were based on heavily-parametrized SCF methods (either Hückel of Pariser–Par–Pople) and are therefore susceptible to give biased results when extrapolated to larger systems.

None of the more recent theoretical studies [23–25] has gone beyond the vertical approximation in computing the electronic spectra, whereas for these systems it is shown in the present work that modeling the vibronic band shape is crucial for assessing the accuracy of various density functionals.

Solvent effects in the few previous TD-DFT studies on (aza)indolizine derivatives were accounted in the simple but unphysical linear-response formalism (LR-PCM) that predicts solvatochromic shifts in qualitative contrast with experiment. Finally, none of the previous approaches by other authors has attempted to rationalize the effects of substitution based on the nature and the energetics of the corresponding excited state in the unsubstituted heterocycle.

Given the recent interest in exploiting the fluorescence emission of (aza)indolizine for practical applications, original contributions brought by this thesis attempt to rationalize the influence of substitution on their near-UV/Vis spectra. To this end, for the first time *first principles* theoretical investigations at time-dependent density functional theory (TD-DFT) level, including state-specific (SS-PCM) solvation effects [26, 27] and vibronic structure computations [28–30], were used systematically to gain insights into the nature and the energetics of the lowest excited states in unsubstituted (aza)indolizines, whereas specific and nonspecific effects induced by various functional groups were investigated comparatively in several test cases. Experimental studies by steady-state and time-resolved (TCSPC) fluorescence spectroscopy, and also a brief organic synthesis stage followed by X-ray diffraction experiments were used to validate (or infirm) several hypotheses derived theoretically.

In the PhD thesis summarized herein, following the introductory remarks (Chapter I) and a description of the methodology (Chapter II), the original contributions are organized in four main chapters, as following.
Chapter III - Effects of substitution on the lowest excited states of pyrrolo[1,2-b]pyridazine

A first comparative study on two pyrrolo[1,2-b]pyridazine (PP) derivatives, described in chapter III, attempts to elucidate the detrimental effect of the benzoyl group on fluorescence emission, following the experimental observation emphasized by Zbancioc et al. in a recent work [17]. In spite of the apparent similitudes in their absorption characteristics, fluorescence quantum yields of the two derivatives (Figure 2) are 0.80–0.90 in the case of PP2e and about 0.07 in the case of PP2b. TD-DFT computations were performed for the two molecules to unravel the origin intrinsic quenching of fluorescence in PP2b. In addition, steady-state absorption, excitation and fluorescence spectra needed to supplement the experimental picture, were recorded in three aprotic solvents. The lowest excited singlet PP2e analogue, given it’s simpler structure, was subjected to extensive benchmarks using the B3LYP [31] (20% Hartree–Fock exchange energy, HFx), PBE0 [32] (25% HFx), CAM-B3LYP [33] (range-dependent HFx), BH&HLYP (50% HFx) and M06-HF (100% HFx) hybrid exchange-correlation density functionals, as well as the configurational interaction singles (CIS) post Hartree–Fock method.

In the vertical approximation, the first excitation energy is overestimated by all the methods employed, B3LYP being apparently the best density functional to reproduce the position of the first absorption maximum. At the same level, fluorescence emission energy is underestimated. Upon modeling the vibronic structure of fluorescence spectrum and the first two ($\pi \rightarrow \pi^*$) absorption bands (see Figure 3), the further step toward a meaningful (physical) description of the absorption phenomenon, the best overall performances are achieved using the PBE0 functional instead of the widely-used B3LYP, the apparent performances of the latter in vertical excitation energy resulting most likely from errors cancellation. Between the two, the former (PBE0, parameter–free) was hence preferred.
Figure 3 – Computed vs. experimental absorption and fluorescence spectra of PP2e to the latter (B3LYP, adjustable parameters, fitted) thoroughly in the subsequent works.

The same (PBE0) global hybrid density functional and also the CAM-B3LYP range-separated hybrid foresee for PP2b in cyclohexan a nπ* excited singlet below the lowest ππ* singlet. The result may relate to the proximity of carbonyl (O) and diazine nitrogen (N1) lone pairs in the ground state (GS) of the most stable conformation at C7 and could explain the low fluorescence emission reported experimentally. Given the small ππ*-nπ* energy difference in Franck-Condon (FC) region, as reproduced consistently by the two functionals, fluorescence emission may occur from the lowest vibrational levels of the allowed singlet (ππ*) whereas excitations to upper vibronic states should trigger ππ* → nπ* internal conversion (IC). Experimental excitation spectra show maxima in the long-wavelength shoulder of the first (mixed, n → π* + π → π*) absorption band. One may expect hence an increase in the emission intensity when passing from nonpolar (cyclohexane) to polar (i.e., chloroform or acetonitrile), subsequent to a destabilized nπ* state in the latter media. Indeed, TD-DFT predicts consistently the interchange of the lowest singlets (nπ* > ππ*) in polar solvents, and the long-wavelength shoulder in the recorded spectra is blue-shifted and vanishes in acetonitrile. However, excitation spectra show negligible variation with solvent
polarity, while fluorescence emission decreases substantially in polar solvents. In the same assumption on the proximity of carbonyl and diazine nitrogen lone-pairs – in a syn orientation of the former (facing the heterocycle) – that stabilizes the lowest \( n\pi^* \) singlet, one may anticipate a conformational-dependent picture of the lowest excited states with respect to the benzoyl group at \( C_7 \). TD-DFT foresee, by most of the functionals used herein (except M06-HF), a reversed order of the two states in the conformer having the aromatic carbonyl from \( C_7 \) oriented in opposite direction (anti), see Figure 4.

The energy of vertical excitation to the lowest (\( \pi\pi^* \)) singlet state of the minor (anti) conformer falls bellow the lowest (\( n\pi^* \)) state of the major (syn) conformation of PP2b and the corresponding \( S_0 \rightarrow S_1 \) (\( \pi \rightarrow \pi^* \)) transition may be tentatively assigned to the longer-wavelength shoulder of the first broad absorption band. Comparison with the excitation spectrum shown in Figure 5 (left) suggests that absorption of radiation leading to fluorescence emission occurs in the minor (anti) conformation, whereas the major conformer is trapped into it’s dark (nonradiative) \( n\pi^* \) state either by direct absorption (unlikely, due to the forbidden character) or via an efficient IC mechanism from the populated \( \pi\pi^* \) upper state, close in energy.

Theoretical results indicate that different orientations of the ester group from \( C_5 \) do not alter significantly the energetics or the character of the lowest excited states. The same conclusion holds for both functional groups of the highly-fluorescent PP2e.

Two ES minima could be located for PP2b, one featuring a syn quasi-planar orientation of the aromatic carbonyl and corresponding to the \( n\pi^* \) singlet (low-
Figure 5 – Experimental absorption spectrum of PP2b and computed vertical excitation spectrum considering only the major conformation (left) and both conformers at C7 (right)

Figure 6 – Energy of the lowest singlet ES of PP2b on the path leading from nπ* (small angles) to ππ* (TICT) minima computed at TD-PBE0 (a) and CIS (b)

severely underestimated emission energies at PBE0 level (up to 1 eV compared to experiment), the conformational dependent qualitative picture of the two lowest excited states is consistent at the PBE0, CAM-B3LYP and CIS levels (see
Figure 6). Search for the conical intersection suggested by twisting curves from Figure 6 at TD-DFT level via the updated branching plane methods locates a crossing of the lowest $n\pi^*$ and $\pi\pi^*$ states in close proximity, with respect to the torsional coordinate at C$_7$, to the GS of the major syn conformer.

Photophysics of PP$_{2b}$ turns to result from an interplay between the near-degeneracy of the lowest excited singlets ($n\pi^*$ and $\pi\pi^*$), strongly depending on the orientation of the carbonyl group at C$_7$, and a strong CT state involving the PP (donor in GS) and benzoyl (acceptor) fragments in a twisted orientation in the relaxed ES geometry [34]. Similar behavior is expected from other PPs bearing pendant aromatic carbonyl groups in position 7 (on the pyrrole ring). Subsequent approaches considered to discriminate these effects included a comparative study on two aliphatic carbonyl derivatives of PP (chapter IV) and a comparative investigation of some newly-synthesized polycyclic diazines featuring two aromatic carbonyl groups constrained in planar orientation with respect to the heterocycle (chapter VI).

Chapter IV - Synthesis, structure and properties of some acetyl–pyrrolodiazines

Due to a smaller spatial extent and hence to a weaker mutual repulsion, the aliphatic carbonyl group of the acetyl-PP derivatives shown in Figure 7 is expected to adopt a planar anti orientation with respect to the pyridazine ring.

PP$_{2k}$ and PP$_{3k}$, as well as their pyrrolo[2,1-$a$]phtalazine analogues, were synthesized in two steps including (i) $N$-alkylation of pyridazine (phtalazine) using $\alpha$–bromoacetone, yielding the corresponding cicloimonium bromide, followed by (ii) a dipolar cycloaddition of the $N$–ylides, generated in situ using triethylamine, to methyl-propilate and DMAD, respectively.

Gas phase DFT (PBE0) and MP2 computations foresee the C$_7$ anti rotamer
Figure 8 – Computed (left) and experimental (right) GS geometries of acetyl-PPs

as most stable in GS for both molecules. Computed internuclear distances are in excellent agreement (r.m.s. 0.007 Å) with experimental counterparts determined from X-ray diffraction (XRD) experiments, whereas the conformational preference of the acetyl group at C7 is reproduced by computations apparently only in **PP3k** (see Figure 8).

In the case of **PP2k**, qualitative discrepancies between theoretical results, predicting systematically a *syn* > *anti* energy order, and experimental evidences suggesting the opposite should reside in strong intermolecular interactions suggested to occur in the more compact crystal packing. Namely, π–π interactions (at about 3.5 Å interplanar spacing, measured) and intermolecular H-bonding may prevail over intramolecular effects in dictating the conformation of **PP2k** in solid state, while the opposite is suggested by the less-compact packing of **PP3k**. Nevertheless, the two different orientations of the acetyl group in the experimental geometry of **PP2k** and **PP3k** represent a strong indication that conformational isomerism occurs at C7.

For the *anti* conformer of both **PP2k** and **PP3k**, TD-PBE0 and TD-CAM-B3LYP predict a ππ* character of the lowest singlet excited state (S1), separated by up to 0.30 eV from the lowest 1nπ* (S2) in gas phase and nonpolar solvent (cyclohexane). At the same theory level, energy ordering of the lowest two singlets is interchanged in *syn* conformation (1nπ* < 1ππ*). For **PP2k**, a state-
crossing is located, on the linear interpolated path between \( n\pi^* \) and \( \pi\pi^* \) minima, in a geometry having the acetyl group at 50–60° with respect to the heterocycle (see Figure 9), i.e., between the GS minimum of the \( \text{syn} \) conformer and the transition state for conformational interconversion (90°). As shown in Figure 9, the lowest two excited singlets are strongly influenced by the orientation of the carbonyl group at C\(_7\), the excessive stabilization of the \( n\pi^* \)
state at lower torsional angles (syn) originating from a sudden increase in the energy of a lone–pair orbital contributed from carbonyl oxygen and the nitrogen atom. According to the conformational–dependent picture of the lowest excited singlets, absorption and fluorescence spectra of PP2k in cyclohexane should originate from the \( \pi \rightarrow \pi^* \) transition of the major anti conformation (see Figure 10), whereas the proximity of the second \( n\pi^* \) singlet may impact on the emission intensity. Measured fluorescence quantum yields of PP2k (0.24) and PP3k (0.25) upon excitation at 360 nm, lower than reported for their ester analogues but significantly higher than those of benzoyl–PPs and other carbonyls [35] rationalize with the abundance, at GS equilibrium, of the emitting conformer.

In polar solvents (incl. dichloromethane), both syn and anti conformers feature a \( \pi\pi^* < n\pi^* \) order of the lowest singlet, but different energy separation between the two (0.27 and 0.33, respectively) and hence different emission efficiencies due to the proximity effect. Time–resolved experiments on PP2k in dichloromethane reveal unambiguously a biexponential decay. Photophysi-
cal parameters determined from TCSPC curves ($k_r = 3.3 \times 10^7$ s$^{-1}$ and $\tau_r = 30$ ns) compares well with data previously reported for unsubstituted (aza)indolizines [11].

Chapter V - Comparative investigation of the lowest electronic transition in indolizine and azaindolizines

The energetics and the character of lowest excited states in indolizine (I) and the seven azaindolizines 1–3 and 5–8 shown in Figure 11 were revisited in a first-principles theoretical approach to specifically address the second hypothesis outlined in Chapter III, namely the ICT from PP to aromatic carbonyls, but also to shed light on nonspecific effects from electron–withdrawing/donating groups on the first excitation energy.

Figure 11 – Indolizine (I) and the seven azaindolizines.

TD-DFT (using the B3LYP, PBE0 and CAM-B3LYP density functionals) was employed subsequently (i) in the vertical approach and (ii) modeling the vibronic structure of the first absorption/emission band, both steps being performed in vacuum and in SS-PCM solvent. In an attempt to augment the sparse experimental data with theoretical (ab initio) reference values, vertical excitation energies were also computed in the multiconfigurational complete active space self–consistent field (CASSCF) approach and corrected for dynamic correlation at CAPST2 level. In CASPT2//CASSCF, a full $\pi$–electron active space (10 electrons, 9 orbitals) was considered for indolizine, the lone pair orbital contributed from the additional nitrogen being included in the (12e,10o) active space for each of the azaindolizines.

Among the functionals considered, comparison of TD-DFT vertical excitation energies with CASPT2 reference counterparts apparently recommend the B3LYP (smallest deviations) in modeling the spectra. Compared to experimental (from
wavelengths of absorption maxima), both TD-DFT and CASPT2 approaches not only overrate excitation energies, but are even unable to reproduce most of the observed trends, i.e. the blue-shift of the first band in 3 compared to 1, see Figure 12 (d). Including implicit solvent effects (PCM) does not improve the results for the lowest excitation energy, in agreement with experiments that indicate negligible effects of nonpolar solvents (n-hexane, cyclohexane) on the lowest excitation energy (see Refs. [10] and [9]).

In a further step, the (first) absorption and fluorescence band shapes were modeled in the FC approximation, using vibrational frequencies and normal modes computed at DFT/TD-DFT level for GS/ES. Comparison with available accurate experimental data [10] for 1 in gas-phase allowed first to assess the performances of the B3LYP, PBE0 and CAM-B3LYP density functionals in...
reproducing separately the 0–0 energy for the first singlet transition and the vibrational frequencies in ES (expressed as wavenumbers), respectively.

In the harmonic oscillator approximation, the latter (vibrational frequencies) are overrated systematically (see Figure 13, left), that allowing to derive scaling factors for ES vibrational frequencies via a least-squares fit, whereas the "pure" electronic (adiabatic) ES-GS energy difference is substantially underestimated/overestimated by B3LYP/CAM-B3LYP.

Overall, PBE0 gives the best quantitative results among the functionals considered, with errors in 0–0 energies limited to about 0.05 eV and as low as 0.02 eV (for indolizine and 2), values substantially lower than the observed (or predicted) changes induced by azasubstitution, namely a strong blue-shift observed/predicted in 1 and 3 and a red-shift foreseen in 5 and 8 compared to indolizine.

Seeking a rational description of these trends, that would allow further predictions on the effects of substituents, photoinduced reorganization of electron density was analyzed using CT descriptors $q_{CT}$ (amount of transferred charge), $d_{CT}$ (distance to CT) and $\mu_{CT} = q_{CT} \times d_{CT}$ (CT dipole) computed from electron density differences, $\Delta \rho^{ES-GS}$, between the first ES and GS and recently defined [36] as:

$$q_{CT} = \int \rho_+(\mathbf{r}) \, d\mathbf{r} = - \int \rho_- (\mathbf{r}) \, d\mathbf{r}$$  \hspace{1cm} (1)
Figure 14 – ES-GS electron density differences (a) in the first two excited singlets of indolizine computed from unrelaxed one-particle density and (b) for the first excited singlet of indolizine considering the relaxed ES electron density

and

\[ d_{CT} = |R_+ - R_-| = \left| \frac{\int \mathbf{r} \cdot \rho_+(\mathbf{r}) \, d\mathbf{r}}{\int \rho_+(\mathbf{r}) \, d\mathbf{r}} - \frac{\int \mathbf{r} \cdot \rho_-(\mathbf{r}) \, d\mathbf{r}}{\int \rho_-(\mathbf{r}) \, d\mathbf{r}} \right| \]

where

\[ \rho_+(\mathbf{r}) = \begin{cases} \Delta \rho(\mathbf{r}), & \Delta \rho(\mathbf{r}) > 0 \\ 0, & \Delta \rho(\mathbf{r}) < 0 \end{cases}, \quad \rho_-(\mathbf{r}) = \begin{cases} 0, & \Delta \rho(\mathbf{r}) > 0 \\ \Delta \rho(\mathbf{r}), & \Delta \rho(\mathbf{r}) < 0 \end{cases} \]

Density reorganization in the first ES of indolizine (see Figure 14) and azaindolizines reflect an overall CT from the pyrrole to the (di)azine ring. In spite of strong influences induced by azasubstitution simultaneously on the magnitude and orientation of GS and ES dipole moments, the effects on the first excitation energy could not be rationalized in terms of CT dipole, \( \mu_{CT} = |\mu^{ES} - \mu^{GS}| \) (arrow shown in Figure 14), or \( d_{CT}/q_{CT} \). Quantitative descriptors described above do not account for individual (atomic) contribution to the CT, namely the multipoles CT character. As shown in Figure 14, four of the seven centers of indolizine susceptible to (aza)substitution participate to different extents in the CT either as donors (1 and 3) or acceptors (2, 5 and 8), whereas the remaining two (6 and 7) are marginally involved. Essentially the same qualitative conclusion arise when interpreting electron density differences computed from relaxed (Figure 14, b) instead of the approximate (unrelaxed) one-particle ES density (Figure 14, a).

In the simpler (approximate) approach, all the three CT descriptors derived from electron densities (computed by subtracting GS total density from one-particle ES density) are reproduced by ES-GS differences in Mulliken partial
atomic charges, \( \delta q_i = q_{i,ES} - q_{i,GS} \) in a discrete variant of the formalism given in Eqs. (1) and (2). In indolizine, centers 1 and 3 qualify with similar quantitative extents as donors (\( \delta q_i > 0 \) or \( q_{i,ES} > q_{i,GS} \)), whereas 2, 5 and 8 (\( \delta q_i < 0 \)) involve as acceptors in the photoinduced CT. For centers 1, 2, and 3, the semi-quantitative correlation of the first excitation energy of the corresponding azaindolizine with the amount of charge transferred from (in 1 and 3) or to (in 2) the corresponding center confirms the previous semiempirical systematics proposed by Evleth [22]. Current (non-empirical TD-DFT) results extend the same rationalization to azaindolizines 5 and 8 (see Figure 15, a). The correlation pertains (see Figure 15, b) when the relaxed (instead of unrelaxed, one-particle) ES density is considered, except that in this case differences in ChelpG charges (instead of Mulliken) recover with acceptable accuracy the CT indices derived from electron density.

Except the centers 2, 6 and 7, for which the correlation seems inconsistent between the two approaches, the effect of various electron-withdrawing/donating groups on the energy of the lowest excited state may be rationalized in the same manner.

Deviations of data plotted in Figure 15 from a straight line may relate to the fact that in actual azaindolizines the computed ES-GS absolute differences in partial atomic charges (in both point charge models) are substantially lower than in the parent indolizine. Namely in 1 and 3 / 5 and 8, the additional nitrogen atom in the pyrrole ring involves as donor/acceptor in the CT with a lesser extend than the corresponding carbon atom from the parent indolizine. The blue/red shift of the first absorption band in (aza)indolizines bearing ester/amine groups
on the pyrrole ring in positions adjacent to the bridge confirm the conclusion above.

Chapter VI - Structure and spectral properties of some isoindolo-diazines

Polycyclic diazines shown in Figure 16 (also referred to as isoindolo-diazines) were evaluated in two parallel investigations aiming toward (i) an assessment of their biologic activity and (ii) testing the hypothesis of a photoinduced CT from the pyrrole ring (or the entire PP) to the aromatic carbonyl fragment. In

![Figure 16](image-url)  
*Figure 16 – Structures of polycyclic pyrrolodiazines with aromatic carbonyl groups*

the former approach, the anticancer activity of compounds 1a-c and 2a-c was tested *in vitro* against Leukemia CCRF-CEM, Leukemia MOLT-4, Non-Small Cell Lung Cancer NCI-H460, and Breast Cancer MCF7 cells. All the compounds have significant anticancer activity, expressed by the Percentage Growth Inhibition (PGI) factor, the pentacyclic systems (2a-c) showing the largest values. A plausible hypothesis formulated [37] to explain the observed biological properties designated the compounds as DNA intercalators, relaying on the coplanar molecular geometry of the four(five) condensed rings as proved by XRD experiments [38, 39] regardless the substituent on the remaining position of the pyrrole ring (see Figure 17).

Regarding the influence of the extended π-subsystem on the photophysical properties, structures 1a-c may be ascribed as PPs bearing aromatic carbonyl groups (at the pyrrole ring) that are constrained to adopt coplanar orientation
Figure 17 – Molecular geometry (a) computed for compounds 1a-c and 2a-c and (b) determined by XRD for 1b and 2d

with respect to the PP rings. Consequent to an extended electron delocalization, the first absorption band of all the isoindolo-diazines investigated is red-shifted toward the visible region (410-420 nm).

TD-DFT computations using the PBE0 and CAM-B3LYP functionals foresee in the first ES a photoinduced CT from the PP (PH) to the quinone fragment via the pyrrole ring. Given the small energy separation between the lowest (\(\pi\pi^*\) and \(n\pi^*\)) excited singlets in 1a and 1b, plots of electron density differences reveal a mixed (\(\pi\pi^*+n\pi^*\)) character of the lowest ES (see Figure 18). In contrast, the first ES in 1c and 2c (bearing amine group on the pyrrole ring) has pure \(\pi\pi^*\) character. Assuming a proximity effect, the predictions correlate with measured fluorescence quantum yields, substantially higher in the case of 1c (> 60%) than in 1a,b (< 20%).
Compared to the corresponding unsubstituted four-rings system 1 (quantum yield about 40% [40]) the two opposite trends may be unequivocally assigned to the substituent on the pyrrole ring: the ester group has a detrimental effect whereas the amino group enhances the fluorescence emission. Even in a pre-twisted orientation, the former is expected to stabilize the lowest \( n\pi^* \) singlet \( (S_2) \) by destabilizing the lone pair from either aromatic carbonyl or the pyridazine nitrogen, depending on the conformation adopted in GS. For the lowest excited singlets of 1a, TD-DFT predicts an interchange of the major character \( (\pi\pi^* \leftrightarrow n\pi^*) \) in both nearly-planar orientations.

In the case of 1c (2c), the pure \( \pi\pi^* \) character of \( S_1 \) and consequently an increased fluorescence intensity may originate from one of the following effects of an intramolecular hydrogen bond suggested by GS equilibrium geometry (see Figure 17): (i) a stabilization of carbonyl lone-pair orbital, and hence an increase in the energy of the lowest \( n\pi^* \) singlet and (ii) a stabilization and enhancement
of the first (ICT) state, the adjacent aromatic carbonyl involved in the hydrogen bond playing as acceptor in the photoinduced CT. Comparison in computed

![Experimental fluorescence spectra of 1a,c and 2a,c recorded in dichloromethane](image)

**Figure 19** – Experimental fluorescence spectra of 1a,c and 2a,c recorded in dichloromethane

excitation and emission energies (TD-DFT) and red-shifted absorption and fluorescence bands of 1a (compared to 1a,b) and 2c (compared to 2a,b), see Figure 19, add theoretical and experimental support for the latter. Namely, intramolecular hydrogen bonds involving aromatic carbonyls enhance and induce a red-shift on fluorescence emission by stabilizing the ICT state. Intermolecular interactions with molecules of biological interest (such as nucleic acids), mostly consisting of H-bonds, are expected to induce similar effects. That opens the route for a reliable spectroscopic approach in investigating non-bonding interactions involved in the biological activity of these systems.

**General conclusions**

The lowest singlet transition in (aza)indolizines is reproduced, at unprecedented accuracy for unsubstituted systems, by TD-DFT using the parameter–free PBE0 density functional using a basis set with moderate complexity. At the same theory level, spectral shifts induced by various groups are reproduced with errors lower than reported in most of the previous works by other authors. Current results recommend this functional/basis set combination in modeling the (lowest) valence singlet transitions in (aza)indolizine derivatives. Particular aspects related to large aromatic groups featuring torsional degrees of freedom and specific solvation effects require additional investigations (i.e. at CAM-B3LYP level).
Excitation to the lowest ES in (aza)indolizines triggers an electron density reorganization (charge transfer) between the two fused rings. Each of the three and four centers of the two rings contribute with different extent either as donor or as acceptor, yielding a multipolar CT character. The first excitation energy in azaindolizines, computed in either the vertical approach or upon modeling the vibronic structure (the latter being strongly required for a reliable comparison with experimental data) correlate qualitatively with ES-GS differences in partial atomic charges computed for the center of indolizine corresponding to azasubstitution. Results presented herein not only confirm a previous systematics proposed for 1-, 2- and 3-azaindolizine (based on heavily-parametrized semiempirical results) but allow the same model to be applied for 5- and 8-azaindolizine as well as for describing/predicting nonspecific effects (of electron-withdrawing/donating groups) on the first excitation energy.

In the case of PP, the functional group on the pyrrole ring (adjacent to the bridgehead nitrogen atom) influences not only the CT ($\pi\pi^*$) state, but also stabilizes the lowest $n\pi^*$ singlet. In effect, a functional group featuring lone-pairs (such as carbonyl) may impact, depending on it’s orientation with respect to the pyridazine ring, on fluorescence emission.

In isoindolo-diazine systems, the relative energy of the lowest two singlets, $\pi\pi^*$ and $n\pi^*$, strongly depends on the substituent on the remaining position on the pyrrole ring. The former (emitting) state features a strong ICT, from the pyrrole ring to the quinone fragment, that may be enhanced by specific intra- or inter-molecular interactions involving the carbonyl group on the acceptor side. In isoindolo-diazines bearing ester groups, the decrease in the fluorescence emission intensity relate to a stabilization of the lowest $n\pi^*$ singlet and hence a proximity effect induced in both nearly-planar orientations of the additional functional group.

Selected references


