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Indolizine-Squaraines: NIR Fluorescent Materials with Molecular Engineered Stokes Shifts

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Abstract: The development of deep red and near infrared emissive materials with high quantum yields is an important challenge. Several classes of squaraine dyes have demonstrated high quantum yields, but require significantly red-shifted absorptions to access the NIR window. Additionally, squaraine dyes have typically shown narrow Stokes shifts, which limits their use in living biological imaging applications due to dye emission interference with the light source. Through the incorporation of indolizine heterocycles we have synthesized novel indolizine squaraine dyes with increased Stokes shifts (up to >0.119 eV, >50 nm increase) and absorptions substantially further into the NIR region than an indoline squaraine benchmark (726 nm versus 659 nm absorption maxima). These materials have shown significantly enhanced water solubility, which is unique for squaraine dyes without water solubilizing substituents. Absorption, electrochemical, computational and fluorescence studies were undertaken and exceptional fluorescence quantum yields of up 12% were observed with emission curves extending beyond 850 nm.

Introduction

Near-infrared (NIR) absorbing and emitting materials have recently found a number of applications including secure night vision goggles, tissue displays. welding. telecommunications, biological imaging and more.¹ Among NIR materials, several donor-acceptor based designs have emerged including those based on cyanine, BODIPY, and squaraine.² Squaraines have been closely examined in many applications, such as organic solar cell applications, and are of interest for living fluorescence biological imaging applications.^{1a, 3} Ideally, for biological imaging, the NIR material would: (1) be water soluble, (2) absorb and emit photons within the therapeutic window, 700 nm - 1400 nm, and (3) absorb and emit photons which are significantly different in energies.

Popular indoline donor-based squaraines (1) fall short of all of these goals (Figure 1). Their absorption often occurs outside the therapeutic window due to wider than desired optical band gaps and their absorption and emission peak intensities are separated by very small energies (<0.01 eV). Hydrogen bonding approaches have shifted the absorption ranges for benzene based squaraines;⁴ however, these dyes often fall short of the NIR window and have narrow absorption-emission peak separations. Water solubility is especially difficult for many squaraines despite having a charged donor and squaraine core, due to the anionic oxygen often being either shielded with hydrophobic groups or heavily hydrogen bonded with the

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appended donor group. Frequently, the addition of pendant water solubilizing groups are necessary to achieve significant water solubility. Despite these shortcomings, squaraine based dyes remain popular candidates for imaging research purposes as they represent low molecular weight NIR materials with relatively concise syntheses and exceptional molar absorptivities which are commonly greater than 100,000 M⁻¹cm^{-1,5} We sought to address the deficiencies by substituting the indoline donor group with a more electron rich, fully conjugated indolizine donor group. Introduction of a sterically hindered donor with increased electron density may lead to significant separation of absorption and emission spectra, further extension of the absorption range into the therapeutic window, and increased water solubility through allowing hydrogen-bonding access to the charged squaraine core (Figure 1).



Figure 1. Example classes of popular D-A-D squaraines with the donor motifs highlighted in red.

Indolizines have been shown to yield improved performances in dye-sensitized solar cells owing to their exceptional donor strength when compared with common triphenylamine and alkyl amine-based donor groups within the donor-π bridge-acceptor framework.⁶ The increase in relative donor strength is in part due to a nitrogen atom with three separate carbon bonds being incorporated into a building block with a fully planar geometry, a fully conjugated m-system involving the nitrogen loan-pair, reduced resonance stabilization energy of the aromatic indolizine, and the proaromatic nature of the indolizine building block in charge transfer systems. These properties are all attractive when selecting a donor for NIR imaging materials, which rely on strong intramolecular charge transfer events. To investigate the effects of different substituents on the dye's energetic properties and NIR emission, we have targeted twelve donor-acceptor-donor (D-A-D) dyes with the indolizine-squaraine-indolizine (In2SQ) D-A-D core held constant (Figure 2). The substituents were varied from groups without π -electron contributions for steric analysis to a series of aryl substituents of various size baring electron donating, neutral and electron withdrawing groups for analysis of the influence of pendant aryl groups on the core structure. These substituents provide insight into the effects of substituent size, conjugation and electronic contributions on the optical properties (molar absorptivity, excited-state lifetime and fluorescence quantum yield) of this new class of material.

The degree of planarity of the conjugated π -system plays a critical role on inducing significant Stokes-shifts. If the groundand excited-state differ in planarity substantially, a geometry

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reorganization can occur after photoexcitation potentially leading to significant energy differences in the absorption and emission energies. The indolizine donor offers an nucleophilic position for EAS reactions at the 3-position with substituents at the 2position and a fused ring in close proximity to the squaraine core allowing for the introduction of sterically demanding substituents to modulate the degree of planarity of the In_2SQ core and planarity at the 2-position substituent location, which should offer Stokes-shift tunability (Figure 3).



Figure 2. Position numbering convention for the indolizine building block and angles where dihedral steric interactions induce a significant deviation from a planar conjugated system. See Table 2 for calculated twist angles.



Figure 3. Indolizine-squaraine-indolizine (In₂SQ) based target NIR emissive materials.

Meln₂SQ and Phln₂SQ were selected as targets to compare a methyl versus an aryl substituent on the indolizine donors (Figure 2). The methyl substituent offers the smallest group, and a more planarized In₂SQ core. The phenyl group increases steric bulk near the squaraine core and may additionally expand the dye conjugated π -system through incorporating π -electrons from the phenyl ring. Large aryl substituted squaraine dyes (NaphIn₂SQ, Pyln₂SQ, *bis*CF₃Phln₂SQ, *bis*^tBuPhIn₂SQ, MesIn₂SQ) and electronically variable substituted squaraine dyes (MeOPhIn₂SQ, CNPhIn₂SQ, NO₂PhIn₂SQ, HOPhIn₂SQ, CF₃PhIn₂SQ) were targeted to probe steric interactions and evaluate the effect of increased or decreased electron density on the aryl group in order to further probe the photophysical properties of these molecules.

The In₂SQ dye series was synthesized in a straight forward 2-step fashion from ethyl pyridine and alphabromo ketone derivatives (Scheme 1). Upon heating the bromide and pyridine starting materials, N-alkylated salts were awarded which were insoluble in cold acetone. These pyridinium salt intermediates were filtered, dissolved in an aqueous base solution, and heated to reflux. The cyclization-condensation reaction proceeded to give neutral organic indolizine products, which could be isolated through simple filtration upon cooling the solution. Finally, the indolizine heterocycles were heated in the presence of squaric acid as a 2:1 ratio of indolizine:squaraine. The resulting dark green reaction mixture gave an NMR and alumina TLC of analytically pure In2SQ products upon addition of diethyl ether in many cases. In this manner, >1 gram scale was demonstrated with no adverse effects on yield and no need for column chromatography for the PhIn₂SQ derivative.

Results and Discussion

Two possible conformational isomers could result from the double condensation reaction with squaric acid. Given the significant distance the two indolizine building blocks are separated by, NMR may not show a substantial difference in the two possible isomers if both were present as a mixture. To confirm the isomeric purity of the samples, a combined computational IR spectrum prediction and experimental IR spectroscopy approach was employed. The B3LYP density functional was employed with the 6-311g (d,p) basis set in order to calculate the modes and IR intensities of both the cis and trans rotamer of PhIn₂SQ. The calculated modes were scaled by 0.975 in order to correct for the harmonic approximation. Modes were determined experimentally by ATR-IR and compared to calculations of both rotamers (Figure 4). A mixture of the two rotamers would show little difference according to the predicted spectrum since both rotamers are similar except in the region between 1600 and 1800 cm⁻¹. In this region there is noticeably difference between the cis and trans rotamer computationally with better agreement between the experimental IR spectrum and the calculated modes for the cis rotamer, suggesting the cis rotamer is a product of the reaction (scheme 1).

Scheme 1. Synthetic route to In₂SQ dyes.



Since the majority of the IR spectrum (600-1600 cm^{-1}) is predicted to be similar for the cis and trans rotamers, the obvious peak confirming the cis rotamer presence that is not

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present for the predicted trans rotamer spectrum, cannot be used to exclude the presence of the trans rotamer in the mixture. As such, a variety of crystallization techniques were attempted, and the most successful crystallization attempt led to the formation of small (<0.5 mm) cubic crystals from the vapor diffusion of diethyl ether into chlorobenzene. These crystals, despite the small size, were X-ray quality and proved the presence of the trans rotamer (Figure 5). The crystal structure shows a significant dihedral angle between the phenyl and indolizine planes at 51° and a dihedral angle of 21° between the indolizine and squaraine plane. These results are further compared with computational results below.



Figure 4. IR spectrum of $PhIn_2SQ$ comparing the computational results for the cis rotamer (top, black) and the trans rotamer (bottom, black) to the experimental results (red).

separable via silica gel chromatography with the trans rotamer eluting with a higher Rf value. Separation of these rotamers allowed for the comparison of a purified trans rotamer vibrational spectrum to the cis rotamer vibrational spectrum (Figure S1). For this rotamer, the peak at 1750 cm-1 is present for the cis rotamer similar to that predicted and observed for the PhIn₂SQ cis rotamer. This peak is not present for the trans rotamer experimentally, as predicted. The PyIn₂SQ was uniquely able to be studies as the purified cis rotamer, the purified trans rotamer and as a mixture. It is important to note however, that the rotational barrier to convert from the cis to trans rotamer can be crossed at room temperature on the order of a day timescale. The existence of both rotamers could only be confirmed for PyIn₂SQ since no evidence of the cis isomer was obvious for the other derivatives.

Upon isolation of these novel dyes, the optical properties were probed by UV-Vis-NIR absorption spectroscopy to evaluate the effect the indolizine donor group in relation to the indoline standard (1). Comparison of Meln₂SQ and 1 reveals a near 50 nm red-shift for the indolizine based dye (Figure 6). The 50 nm shift in absorption maxima to lower energy places these dyes in a more ideal range for use with non-invasive in vivo fluorescence imaging applications. Interesting, the absorption curve shape shows a very similar profile for both dyes with a strong, relatively sharp peak at low energy, followed by a weaker vibronic transition at slightly higher energy as a shoulder. Absorption spectra show a characteristic, intense absorption near 720 nm and no substantial absorption between 675 nm and 450 nm (Figures S2 and S3). Meln₂SQ does show an extended absorption peak 1/2 height width of 45 nm (0.108 eV) versus 20 nm (0.076 eV) for benchmark 1 (Figure 6).



Figure 5. X-ray crystal structure representation for $Phln_2SQ$ as the trans rotamer.

The PyIn₂SQ derivative was unique among the synthesized derivatives in that the cis and trans rotamers were



Figure 6. Comparison of UV-Vis-NIR absorption spectra (solid black lines) and emission spectra (dashed red lines) for **1**, Meln₂SQ and Phln₂SQ in toluene (see SI for absorption and emission curves of all derivatives).

Additionally, the indolizine group substituent influence on optical properties was evaluated based on electron donation ability, electron withdrawing ability, steric size and extended π -system size for a variety of aryl groups. Comparison of a methyl versus a phenyl at the 2-position shows a 10 nm red-shift for the aryl substituted PhIn₂SQ dye. Among aryl groups however, a

modest effect on absorption maxima position is observed with a close range varying by only 9 nm (723-732 nm) for the series in toluene. The absorption spectrum in dichloromethane was found to give similar absorption maxima shifted by about 6 nm to higher energy (see SI for DCM results, Table S1). This suggests poor electronic coupling of the aryl group π -system with the squaraine π -system in the ground-state since strongly withdrawing and donating substituents have little effect on the absorption spectrum. A comparison of molar absorptivity of Meln₂SQ and 1 shows a modest reduction in molar absorptivity for the indolizine-based dye (210,000 versus 229,000 M⁻¹cm⁻¹ in toluene). A substantially larger effect on molar absorptivities was observed with indolizine substituent choice ranging from 110,000-262,000 M⁻¹cm⁻¹ in toluene. Replacement of a methyl at the 2-position with a phenyl group reduces the dye's molar absorptivity from 210,000 to 181,000 M⁻cm⁻¹. Expansion of the substituent π -system from phenyl to naphthyl further lowered the molar absorptivity by 31,000 M⁻¹cm⁻¹ (from 181,000 to 150,000 M^{-1} cm⁻¹). Finally, pyrene substitution results in the lowest molar absorptivity of the series at 124,000 M⁻¹cm⁻¹ for the mixture (Figure 7).

Dye	Abs. Max.	Emis. Max.	Stokes Shift	3	Φ	т
	(nm)	(nm)	(nm, eV)	(M ⁻¹ cm ⁻¹)	(%)	(ns)
MeIn ₂ SQ	712	738	26, 0.055	210,000	8.9	0.42
PhIn₂SQ	723	756	33, 0.075	181,000	3.7	0.45
NaphIn₂SQ	727	766	39, 0.087	150,000	5.3	0.45
PyIn₂SQ	729	757	28, 0.063	124, 000	10.5	1.11
PyIn₂SQ (trans)	730	756	26, 0.058	160,000	5.8	1.03
PyIn₂SQ (cis)	730	754	24, 0.054	110,000	12.1	1.46
MesIn₂SQ	724	735	11, 0.026	262,000	7.3	0.30
<i>Bis-t</i> BuPhIn₂SQ	722	770	48, 0.107	216,000	6.7	0.55
Bis-CF ₃ PhIn ₂ SQ	727	757	30, 0.068	260,000	12.0	0.79
CF₃Phln₂SQ	727	765	38, 0.085	166,000	5.9	0.40
CNPhIn₂SQ	729	756	54, 0.119	213,000	5.8	0.65
NO ₂ PhIn ₂ SQ	732	769	37, 0.082	185,000	11.2	0.65
MeOPhIn ₂ SQ	723	761	38, 0.086	140,000	4.5	0.33
HOPhIn₂SQ*	723	756	33, 0.075	185,000	1.0	0.09
1	633	653	20, 0.050	229,000	16.0	2.40

Table 1. Comparison of Stokes shifts, absorption, emission maxima, molar absorptivity, photoluminescent quantum yield and fluorescent lifetimes in toluene. *Indicates in DMSO





Figure 7. Comparison of molar absorptivities for all arylated dyes.

To probe if the lowering of molar absorptivity is a result of increasing the π -system size or a result of increasing steric bulk MesIn₂SQ, bis-tBuPhIn₂SQ and bis-CF₃PhIn₂SQ were evaluated as neutral, electron rich and electron deficient aryl groups. Increasing steric bulk leads to the highest molar absorptivities observed, exceeding those of the parent indolizine dye and the benchmark **1** by as much as 33,000 M⁻¹cm⁻¹. The MesIn₂SQ and bis-CF₃PhIn₂SQ had similar molar absorptivities of near 260,000 M⁻¹cm⁻¹, while the electron rich *bis-t*BuPhIn₂SQ gave a lower molar absorptivity of 216,000 M⁻¹cm⁻¹. To probe the lower molar absorptivity origin, a variety of electron donating (OMe, OH) and withdrawing (CN, NO₂, CF₃) substituents were evaluated at the para-position on each phenyl of PhIn₂SQ. However, no apparent trend emerged based on electronic considerations (Table 1).



785 nm

Figure 8. Phln₂SQ Arranged in an "M" and covered by white filter paper.

Having established the desired red-shifting of the absorption spectrum in the therapeutic window (~700-1400 nm,

dye absorption onsets ~750 nm) through the use of indolizine donors and retained high molar absorptivities, we next evaluated the emissive properties of the dyes in solution. The fluorescence spectra of the compounds were recorded in toluene and DCM. A large Stokes shift, the difference in energy between the absorption maximum and fluorescence maximum, is desirable for applications requiring significant separation between input and output energy, such as live fluorescence imaging. For this application specifically, molecules should also emit within the therapeutic window (~700-1400 nm). The emission maxima for these dyes were found to range from 756-777 nm with the exception of MesIn₂SQ, which emits at noticeably higher energy (735 nm). This gives Stokes shifts of around 30-50 nm or 0.063-0.119 eV for ln₂SQ series (except Mesln₂SQ 11 nm, 0.026 eV), which is a significantly larger separation of input and output energies than observed for the benchmark 1 (12 nm, 0.034 eV) (Table 1). The absorption and emission of the most water soluble derivative, Meln₂SQ, was also investigated by dissolving the dye in minimal DMSO then diluting with water (Figure S9). A blue shift of the absorption spectrum is noted; however, no evidence of aggregation was observed based on absorption or emission curve shapes since there are no noticeable changes comparing water with organic solvents.

The excited-state lifetimes for the dyes in this series ranged from 0.30 ns to 0.79 ns with the longest excited-state lifetime from *bis*-CF₃PhIn₂SQ (0.79 ns) and the shortest excited-state lifetimes from MesIn₂SQ (0.30 ns) and MeOPhIn₂SQ (0.33 ns). It is noteworthy that the resonance withdrawing substituted dyes CNPhIn₂SQ and NO₂PhIn₂SQ have relatively long-lived excited-states (both at 0.65 ns) and the resonance donating substituted MeOPhIn₂SQ has a relatively short-lived excited-state. While all of the dyes show shorter excited-state lifetimes than the benchmark squaraine **1** (2.40 ns), this is expected as dyes emitting at lower energies typically have reduced excited-state lifetimes due to the Energy Gap Law.

The fluorescence quantum yield is one of the most important metrics for evaluating fluorescent materials, especially in the NIR range where Φ is typically very low, often <1%.⁷ For a number of applications, Φ dictates the amount of dye needed for good image resolution and device performance. Interestingly, PyIn₂SQ was found to have a fluorescence quantum yield of >12%. This is a dramatic enhancement over what is commonly observed for materials emitting photons at energies lower than 800 nm, with notable exceptions⁸. The next highest Φ values were observed for *bis*-CF₃PhIn₂SQ (12%) and NO₂PhIn₂SQ (11%), with the remaining dyes emitting between 3.7%-8.9%. Even the lowest observed Φ for this series is a significant enhancement over that of many materials currently being purposes. evaluated for biological imaging Although substantially larger Stokes-shifts are observed compared to benchmark, 1, with less absorption and emission overlap, we do note that considerable overlap of absorption and emission curves is observed for the indolizine squaraine dyes, which can lead to re-absorption and diminished quantum yields. It is interesting that of the dyes evaluated computationally, PyIn₂SQ had a substantial influence of the HOMO⁻¹ orbital (which is heavily influenced by the π -system of pyrene) on the vertical transition (see below). Since the PyIn₂SQ dye has the highest quantum yield in the series, the modulation of the HOMO⁻¹ to have a significant contribution to vertical transition upon transfer of charge to the LUMO may be a novel strategy for enhancing Φ values for NIR emissive materials. This suggests tuning lower level orbitals could have substantial influences on Φ values.

The solid-state absorption of all the dyes was also measured by diffuse reflectance spectroscopy. All of the dyes showed absorption in the 600-800 nm range in the solid state with several of the dyes' having absorption extending well into the 800-900 nm range (Figure S7). Solid-state emission of dyes in this range is useful for potential applications in secure display technologies (NIR OLEDs). As a demonstration for the use of these materials in as solid emitters, PhIn₂SQ powder was arranged in the shape of an "M", then covered with a white filter. As shown in Figure 8, under visible light irradiation only the filter is visible. However, under NIR irradiation at 785 nm through the white filter, an image of the PhIn₂SQ powder is clearly visible via NIR photon detection at 850 nm. The emission spectra of each dye in the solid state can be found in Figure S8.

Substituent selection has relatively minor effects on absorption and emission energies. Electrochemical analysis was used to find if the substituents are effectively isolated from the In_2SQ π -system responsible for the observed absorption and emission energies. The electrochemical properties of the dyes are also critical for applications involving solar cells, display applications, photochemical reactions and for assessing of the stability of the dyes under atmosphere. The dye ground-state oxidation potentials ($E_{(S+/S)}$) ranged from 1.26 to 1.44 V versus ferrocene and the excited-state oxidation potentials $(E_{(S+/S^*)})$, varied from -0.88 V to -1.22 V. The ground-state reduction potentials ($E_{(S/S-)}$) show a similar range of potentials (-1.37 to -1.55 V). Interestingly, this indicates the substituents have tunable control of $E_{(S^+/S)}$, $E_{(S/S^-)}$ and $E_{(S^+/S^*)}$ energy levels without significantly altering the optical band gap energy (E_g^{opt}) which only varies by 0.04 eV between dyes. In general, the dye substituents give the expected changes in energy levels where added π-electron density from aryl groups raise energy levels to more destabilized values and substituents withdrawing melectron density lead to more stabilized energy levels (Table S2)

The observed Stokes shifts are hypothesized to be the result of a geometric reorganization in the excited-state to allow for increased planarization of the π -system. To evaluate this hypothesis, computational analysis was performed on PhIn₂SQ with DFT at the B3LYP/6-311g(*d*,*p*) level. Ground-state and excited-state geometries were first optimized, then TD-DFT at the B3LYP/6-311g(*d*,*p*) level was carried out to predict the vertical excitation (ground-state geometry) and relaxation energies (excited-state geometry), where the difference of these corresponds to a Stokes-shift. Provided reasonable agreement is observed between theory and experiment, the geometry changes from the ground to the excited-state can be analyzed to better understand the origin of the Stokes-shift observed.

In the ground state, the HOMO and LUMO were found to be delocalized throughout the indolizine-squaraine π -system with little orbital contribution from the indolizine substituents (Figure 9). The HOMO⁻¹ orbital was found to be the first orbital with substantial contribution of the indolizine-aryl substituent to the π -system (Figure S10). The indolizine-squaraine backbone is not fully planar and the PhIn₂SQ_{trans} derivative was found to have a twist angle of 10 degrees between the indolizine plane

and the squaraine plane which is in reasonable agreement with the crystallographic data collected (Table 2, Figure 10). Additionally, the aryl substituents have a substantial dihedral angle when compared to the plane of the indolizine (62 degrees for Phln₂SQ_{trans}, in close agreement with crystallographic measurements). The twist angles and calculated energies are



Figure 9. HOMO and LUMO orbitals for PhIn₂SQ as determined by B3LYP/6-311g(d,p) DFT geometry optimization.

similar for PhIn₂SQ_{cis} when compared with PhIn₂SQ_{trans} (Table S3). The PyIn₂SQ_{trans} derivative was also evaluated as it had the largest fluorescence quantum yield (see below). PyIn₂SQ_{trans} has similar dihedral angles to the PhIn₂SQ_{trans} derivative with 8 degrees at the indolizine-squaraine planes and 72 degrees at the pyrene-indolizine planes. Interestingly, PyIn₂SQ_{cis} has a reduced indolizine-squaraine dihedral angle of only 2 degrees and an increased pyrene-indolizine dihedral angle of nearly perpendicular at 87 degrees (see SI for cis rotamer values).

TD-DFT (B3LPY/6-311g(d,p)) was used to assess the orbital contributions to the vertical transition upon photoexcitation of electrons in the ground-state to the excitedstate. The vertical excitation was found to be primarily composed of a HOMO to LUMO contribution (90% for PhIn₂SQ_{trans}, 98% for PhIn₂SQ_{cis}) with some contribution of the HOMO⁻¹ to LUMO transition for the trans rotamers (8% for PhIn₂SQ_{trans}). The contribution of the HOMO⁻¹ to LUMO transition was greatly increased for the large π-system pyrene substituted dye with 58% HOMO to LUMO and 36% HOMO⁻¹ to LUMO for PyIn₂SQ_{trans} and 20% and 72% for the respective orbital transitions for PyIn₂SQ_{cis} (see SI). The substantial involvement of the HOMO⁻¹ orbital highlights the importance of the aryl substituents in controlling the emissive properties of the indolizine-squaraine dye series. The MeIn₂SQ derivative also confirms the importance of the aryl groups as this compound shows a 98% HOMO to LUMO contribution to the lowest energy transition for both the trans and cis confirmers (Table 2).



Figure 10. Comparison of the geometry of $PhIn_2SQ$ in the ground and excitedstate along the indolizine-squaraine-indolizine edge (a and b), along the indolizine edge (c and d), and of the π -system face (e and f), respectively.

The excited-state geometries were optimized with TD-DFT calculations on the PhIn₂SQ and PyIn₂SQ derivatives. In both cases (trans or cis), the indolizine substituent adopts a more planarized configuration with a 6-8 degree reduction in the dihedral angle. This observation suggests the aryl substituents may have a significant influence on the dye excited-state properties. Interestingly, the indolizine-squaraine dihedral angle substantially *increases* in the excited state. A substantial increase of the indolizine-squaraine dihedral angle is observed for PhIn₂SQ_{trans} (17[°]), PhIn₂SQ_{cis} (12[°]), PyIn₂SQ_{trans} (7[°]) and

Table 2. Dihedral angles, orbital contributions to vertical transitions, vertical transition energies and oscillator strengths computed with DFT and TD-DFT analysis at the B3LYP/6-311g(d,p) level.

	-					
dye	angle 1	angle 2	transition	contrib.	vert. trans.	oscillator
	(°)	(°)		(%)	(nm eV)	strength
MeIn ₂ SQ	9°	-	H→L	98%	569 2.18	1.0118
PhIn₂SQ	10° (21°)ª	62° (<u>51°)</u> ª	H→L	90%	607 2.04	0.7524
			H-1 → L	8%		
PyIn₂SQ	8*	72°	H→L	58%	628 1.97	0.4171
			H-1 → L	36%		
			H-3 → L	5%		
MeIn ₂ SQ*	25	-	H→L	83%	676 1.83	0.5317
			H-1 → L	14%		
Phin ₂ SQ*	27°	56°	H→L	78%	733 1.69	0.3689
			H-1 → L	19%		
PyIn ₂ SQ*	13°	66°	H→L	38%	758 1.64	0.1019
			H-1 → L	56%		
			H-3 → L	4%		

PyIn₂SQ_{cis} (4°). The reduced reorganization of the excited-state geometry for PyIn₂SQ relative to the ground-state geometry suggests the possibility of a higher fluorescence quantum yield for this dye is due to a better geometry overlap between the ground- and excited-states. Importantly, upon computational analysis of the MeIn₂SQ derivative, an increase in dihedral angle is still observed in the excited state (9° to 25°) indicating the geometry reorganization is not the direct result of the aryl group on the indolizine. Additionally, no significant shifts in bond length are observed between the ground and excited-states, which indicates the driving force for the reorganization computationally observed is subtle.

A series of indolizine-squaraine based NIR fluorescent compounds, with emission maxima ranging from 730 to 770 nm,

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were synthesized and the optical properties studied. The dyes were studied with X-ray crystallography as well as IR spectroscopy to assess the presence of structural rotamers. The NIR dyes were analyzed with electrochemical studies, UV-Vis-NIR spectroscopy, and fluorometry in a variety of solutions as well as in the solid-state. Compared to a benchmark indolinesquaraine compound, these materials are significantly redshifted in absorption and emission spectrum values. Solution absorption and emission curves show Stokes shifts up to 50 nm and some of the highest fluorescence quantum yields known for emissive small molecules in this NIR region with significant Stokes-shifts (Ф>12%). Large Stokes shifts are desirable for living biological imaging applications to avoid background signals from the excitation source, and the squaraine-based dyes examined have demonstrated a large shift and high PLQE. DFT computational analysis shows the origin of this Stokes-shift to come from the molecular engineering of large dihedral angles within the π -system, which undergo significant changes when the excited-state geometry reorganizes.

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Keywords: NIR dyes • fluorescence quantum yield • squaraine • indolizine • emissive materials

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Twelve novel squaraine dyes were prepared with an indolizine donor with molecularly engineered Stoke-shifts and strong emissions into the NIR region. Computational analysis is provided to aid in understanding these parameters.



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