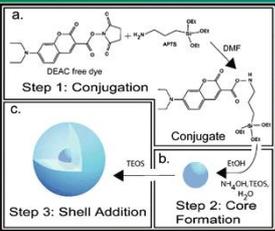


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Fluorescent Triphenyl Substituted Maleimide Derivatives: Synthesis, Spectroscopy and Quantum Chemical Calculations

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Abstract In this paper we describe a semi-empirical quantum method for predicting the wavelength of maximum fluorescence excitation and emission for several known and new maleimide derivatives. All new maleimides, containing a *N*-Benzyl attachment, were successfully synthesised *via* a tandem Suzuki reaction with aryl boronic acids containing either an electron donating, electron withdrawing functional groups. Absorption and emission spectra calculated using the semi-empirical AM1 method with excited state ZINDO calculations proved more reliable than either Hartree-Fock Configuration interaction or time dependent density functional methods. Calculated absorption and emission wavelengths were compared with 26 experimental spectra from known or newly synthesised maleimides and found to have provide reasonable predictions, with an average deviation of less the 6% for absorption maxima and less than 4% for emission peaks. The described method provides a strong benchmark for the accuracy that can be expected from theoretical predictions of fluorescence spectra.

Keywords Maleimide · Quantum chemistry · Semi-empirical calculation · Fluorescence · Synthesis

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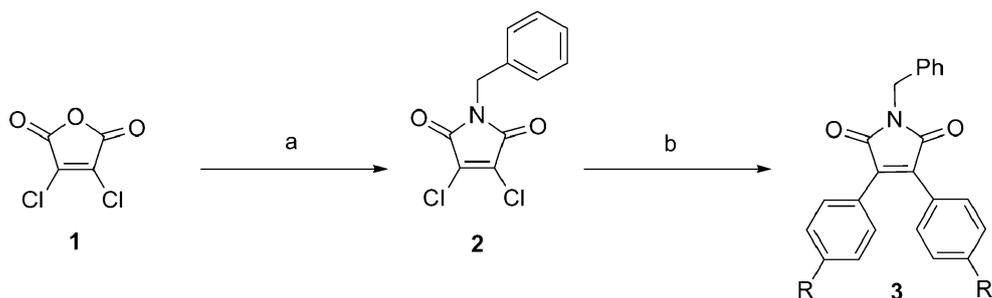
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Introduction

The design and synthesis of new fluorescent molecules is of continuing interest for both understanding the fundamental electronic structure of molecules as well as for many applications in research and industry. Fluorescent dyes have found an enormous number of applications in chemical and biological research, where they can be used for fluorometric detection in high performance liquid chromatography [1, 2], in microscopy and spectrometry to isolate the location of organelles or proteins [3, 4], detect electrical or chemical changes in the environment [5] or to isolate protein interactions or conformational changes [6]. To this end, fluorophores linked to unsubstituted maleimides are often used as thiol reactive probes [7, 8]. Substituted maleimides which have extended conjugation compounds, sometimes naturally occurring, which recently have been shown to inhibit protein kinase C [9].

The ability to design new fluorescent molecules *in silico* before chemical synthesis can expedite the process of constructing dyes with desired properties. Although the effect of the chemical structure on the fluorescent characteristics of the molecules has been studied numerous times [10–25] there is no simple strategy available for accurately predicting these characteristics directly from the molecular structure. From an experimental perspective, reported trends suggest adding electron donating groups to aromatic compounds increases the absorption maximum is as expected. More specifically, reports indicate the addition of a dimethyl amino group $-N(CH_3)_2$ to an aromatic ring increases the wavelength of the maximum in fluorescence intensity, while groups such as methoxy and hydroxyl have more moderate influences. For fluorescence emission spectra it is regularly reported that the addition of an electron donating group increases the wavelength of

Scheme 1 Reagents and conditions: **a** BnNH₂, AcOH, 50 °C, 16 h, 81%; **b** PdCl₂(dppf) (10 mol%), NaOH boronic acid, rt, or Pd₂(dba)₃.CHCl₃, dppf, NaOH, MeNCy₂ and boronic acid, rt, 6–45%



emission. Other studies report that adding electron withdrawing groups, and thus normally conjugation, has a stronger influence on the fluorescence quantum yield, however differences between the type of electron withdrawing functional groups and the excitation and emission maximum varies remarkably [13–15].

As excitation and emission spectra are determined by the electronic energy levels of the molecules, the prediction of accurate fluorescent properties requires accurate electronic structures to be determined. To avoid prohibitive computational demands, such calculations are often done with Hartree–Fock theory (HF), time dependent density functional theory (TDDFT) or semiempirical methods [17]. It is still not clear, however, how accurately each of these approaches can reproduce observed spectra. Semiempirical approaches are the least time consuming and have recently given promising results for maleimide derivatives [18, 19] in addition to many other molecules including porphyrins [20] and benzofurazans. However, recent improvements in computational power has meant that other approaches are feasible, and TDDFT [21–24] and ab-initio configuration interaction [25] approaches have had success in some cases. To help understand the limitations of the methods, we calculate spectra for well characterized AlexaFluor dyes, as well as a maleimide derivative with each of these approaches. We also test the accuracy of less computationally demanding semiempirical AM1/ZINDO approach across all six new maleimide derivatives as well as nineteen previously synthesized molecules [26] to gain an appreciation of the uncertainty in such calculations.

Methods

Synthesis

The synthesis of symmetric bis-aryl maleimides has been explored by several synthetic groups including ours [26, 27]. Our synthetic plan was devised on previous work within this group and others involving the cross coupling of halogenated phthalimides [28–30]. Treatment of the commercially available 2,3-dichloromaleic anhydride (**1**) with benzylamine furnished the protected maleimide **2** in

excellent yield (81%). Subjecting this compound to a tandem Suzuki cross coupling reaction afforded the bis-aryl compound in a range of yields, depending on the *para*-substituted R group. The two catalytic conditions, deemed most reliable in the tandem reaction, were PdCl₂(dppf), NaOH, phenylboronic acid (2.5 equiv) and a method applying an in situ formation of Pd(dppf) through Pd₂(dba)₃. Although it was assumed that yields could have been improved by changing the substrate **2** to the diiodo precursor it was not considered necessary for this physical characteristic study [29–31]. The resulting arylated compounds were purified under standard chromatography conditions (Scheme 1).

Calculations

Unless stated elsewhere, absorption and emission spectra were calculated using semi-empirical quantum chemical methods with HyperChem7.5. To calculate absorption spectra, the molecules were first geometrically optimised using the semi-empirical AM1 method [32] (RMS gradient was set to 0.01). Once this was done, a single point excited state calculation was made using the ground state geometry using the ZINDO/S method [33, 34]. To calculate the emission spectrum the molecules geometry optimisation was made in the excited state using the ZINDO/I method [35] (RMS gradient was set to 0.1), with the aim of simulating internal conversion without any emission. After the optimization was completed, the semi-empirical method was set back to ZINDO/S for single-point energy calculation. An emission spectrum was generated when the single-point energy calculation was completed.

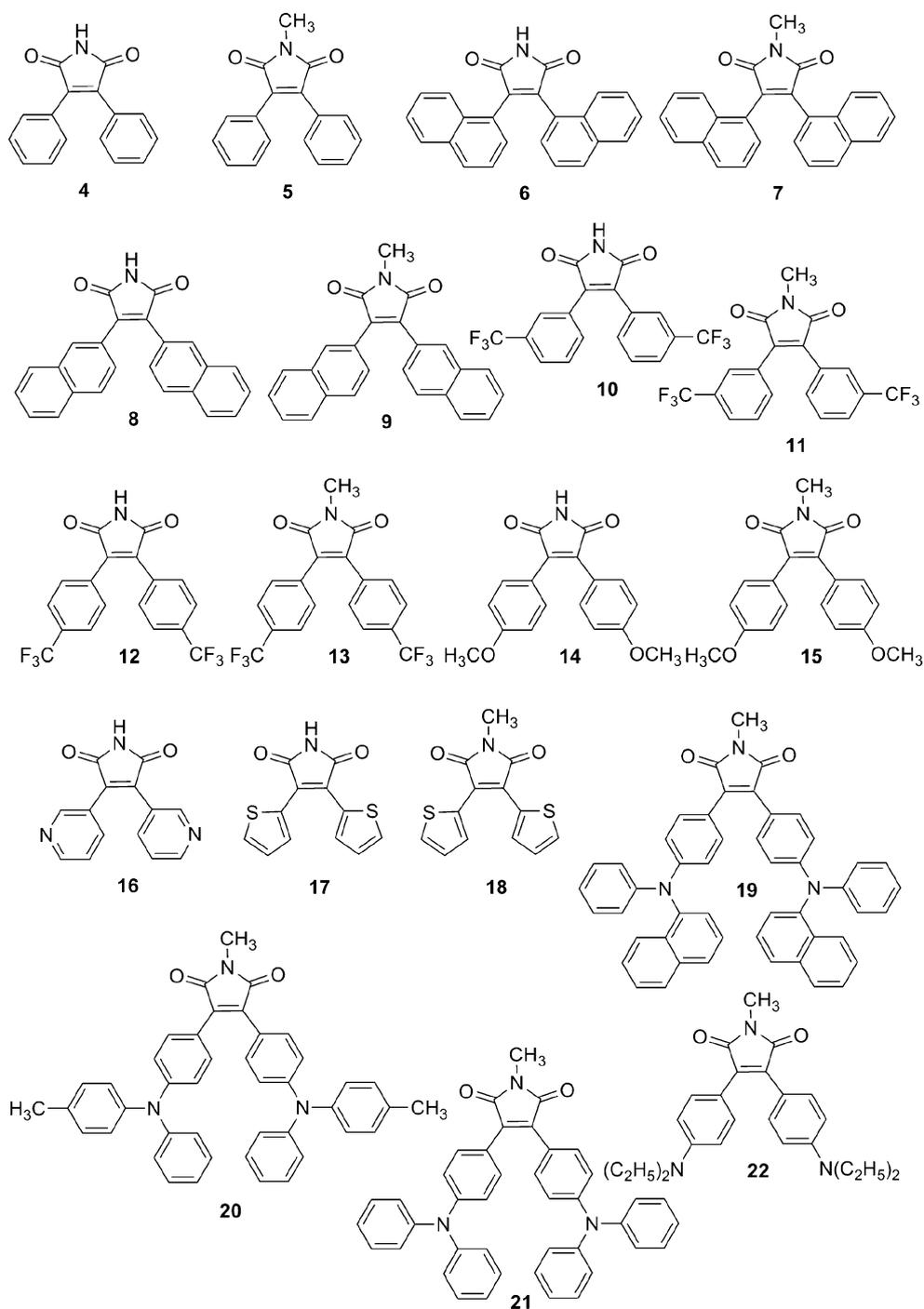
Table 1 Comparison of calculation methods. Wavelength of maximum absorbance calculated by three different methods for three compounds are shown along with the associated experimental value

	AM1-ZINDO	HF-CIS	TDDFT	Experimental data
4	374.1 (0.479)	255.1(0.429)	371.0(0.141)	350
23	434.9 (0.530)	265.8(0.451)	404.5(0.059)	448
AF488	444.4(0.651)	319	666	493

For comparison, some additional calculations were made using Hartree–Fock Configuration interaction (singles) (HF-CIS) [36] and time dependent density functional theory (TD-DFT) using the B3LYP functional as described in the results. Similar results were obtained using a range of basis sets, however, all results shown employ 6–31G*. All ab initio and DFT calculations were made using Gaussian03 [37].

Experimentally, the fluorescence spectra were all determined in hexane, except for compound **23** where dichloromethane was used for solubility purposes. A consistent choice of solvent is paramount considering reports that fluorescence is highly dependent on solvent polarity. A low solvent with low dielectric constant was chosen for the best comparison with gas phase theoretical calculations.

Fig. 1 Structures of reported maleimide derivatives [25]



Results

Assessing calculation accuracy

To determine the most appropriate method to use for calculating the fluorescent properties of the compounds, we calculated the absorbance properties of two of the maleimide derivatives and one well known organic dye using semi-empirical AM1, HF-CIS and TDDFT methods as shown in Table 1. In all cases, the wavelength of maximum absorption determined using the HF-CIS method was found to be well below the expected value. Altering the basis set (up to 6–311++G**) had little influence on the results suggesting a systematic failure of the approach. Results from TDDFT were more varied, but proved extremely poor for the Alexafluor dye. Again, altering the basis set had little effect. Surprisingly, results from TD-HF theory were much better for AF488 (345 nm) than the equivalent B3LYP result. Semi-empirical calculations appeared more reliable and were utilised for the remainder of the calculations described.

To gain an appreciation of the reliability of the semi-empirical calculations, absorption and emission wavelengths were determined for 19 maleimide derivatives (Fig. 1) that have been previously characterised as shown in Table 2 arranged according to increasing wavelength of absorption (Fig. 2).

In general, the increase in the wavelength of the excitation and emission maxima with the addition of electron donating groups such as -OMe and -NR₂ is shown, as highlighted in the differences in compound 4 in comparison to derivatives 14 and 22. Similarly, the addition of heteroatoms (compounds 16 and 17) and additional conjugation (compounds 6 and 7) has the same effect in the system. In the last instance electron withdrawing groups which do not extend the conjugated system (i.e. compound 13) have the opposite effect to their electron donating counterparts and decrease both excitation and emission wavelengths. In each of these cases the theoretical calculations have been able to predict these changes accurately [38, 39].

The results, presented graphically in 2, highlight the broad level of consistency obtained between the calculated and experimentally determined values. An obvious trend is apparent in the predicted excitation peaks, with the wavelengths being overestimated for compounds absorbing at short wavelengths and underestimated for those absorbing at long wavelengths. While we are not clear as to the origins of this trend, it may be related to the parameterisation of the semiempirical calculations performing less well for molecules with non-local valence electrons. A similar level of agreement is seen in the emission wavelengths as for excitation, but no systematic differences with the experimental data is seen.

Table 2 Calculated (in vacuo) and experimental data [25] (in hexane) wavelengths of maximum absorption and emission of known maleimide derivatives. Calculated oscillator strengths are shown in parentheses

Compound	$\lambda_{\max}^{\text{abs}}$ (nm) calculated	$\lambda_{\max}^{\text{abs}}$ (nm) experimental	Dev. (%)	$\lambda_{\max}^{\text{em}}$ (nm) calculated	$\lambda_{\max}^{\text{em}}$ (nm) experimental	Dev. (%)
4	374.1 (0.479)	350	6.89	473.1 (0.457)	471	0.45
5	377.5 (0.413)	361	4.57	482.2 (0.314)	490	-1.59
6	361.9 (0.290)	373	-2.98	465.4 (0.251)	497	-6.36
7	360.4 (0.288)	377	-4.40	460.5 (0.268)	507	-9.17
8	400.1 (0.573)	391	2.33	525.4 (0.612)	488	7.66
9	402.2 (0.550)	395	1.82	509.7 (0.562)	499	2.14
10	361.2 (0.515)	339	6.55	447.4 (0.480)	460	-2.74
11	364.9 (0.388)	352	3.66	455.1 (0.312)	477	-4.59
12	361.4 (0.519)	339	6.61	449.2 (0.484)	459	-2.14
13	366.4 (0.408)	350	4.69	464.4 (0.322)	475	-2.23
14	396.6 (0.515)	387	2.48	507.3 (0.523)	506	0.26
15	397.8 (0.479)	399	-0.30	512.0 (0.421)	518	-1.16
16	375.8 (0.572)	340	10.53	461.1 (0.532)	464	-0.63
17	449.2 (0.896)	414	8.50	526.2 (0.939)	514	2.37
18	448.7 (0.798)	423	6.08	548.1 (0.652)	530	3.42
19	428.2 (0.614)	474	-9.66	596.0 (0.670)	572	4.20
20	441.0 (0.560)	476	-7.35	581.7 (0.620)	575	1.17
21	437.7 (0.585)	480	-8.81	580.4 (0.690)	581	-0.10
22	418.6 (0.507)	481	-12.97	553.7 (0.550)	581	-4.70

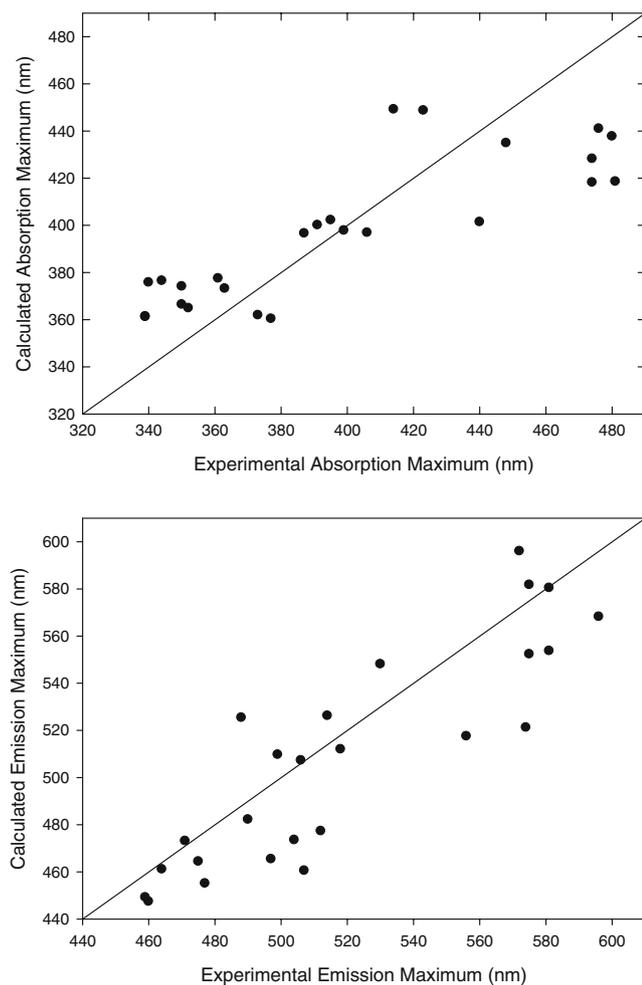


Fig. 2 Tests of calculation accuracy. Calculated wavelengths of maximum excitation (a) and emission (b) are plotted against experimental data for the six new compounds described here as well as the 19 previously reported maleimide derivatives

Excitation and emission spectra of new compounds

In the newly synthesised *N*-Benzyl maleimides (**23–28**) Fig. 3, the inclusion of electron donating substituents such as $-\text{NMe}_2$, $-\text{O}i\text{Pr}$, $-\text{OH}$ (compound **24**, **27** and **28** respectively), greatly increases the wavelength of both the absorbance and emission maximum spectra experimentally when compared to **25** (see Fig. 4 and Table 3), however, the inclusion of the carbonyl moiety has little effect. Such shifts in the wavelengths of the maxima are also observed in work by Hirano on electronic effects in coelenteramide analogues. In most cases these effects are predicted in the semi-empirical quantum calculations with reasonable accuracy, however, the error associated with the absorbance spectra of compound **24** is larger. In the case of the thiophene heterocyclic attachment (compound **23**) and the electron withdrawing acyl derivative (compound **27**) the experimental and calculated values are similar with a slightly larger

deviation in the wavelength of maximum emission (-3.9 and -6.9% respectively). Again a trend is apparent in the predicted excitation peaks, with the wavelengths being overestimated for compounds absorbing at short wavelengths (*c.a.* 340–370 nm λ_{max} i.e. compounds **25** and **27**) and underestimated for those absorbing at long wavelengths (i.e. compound **23** and **24**, **27** and **28**). However, in the fluorescence emission series the calculated values tended to be underestimated in each of the *N*-benzylated cases.

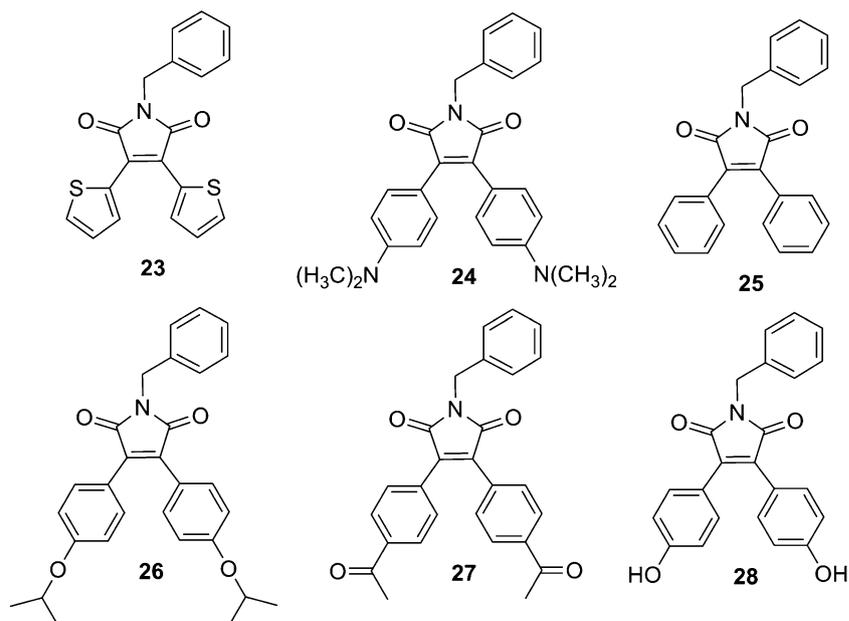
Conclusions

In summary, this paper has compared experimental fluorescence data from known and new maleimide compounds with predicted data. Semi-empirical quantum AM1 method with excited state ZINDO calculations were used for predicting the wavelength of the maximum fluorescence excitation and emission. Calculated absorption and emission wavelengths were within an average deviation of less than 6% for absorption maxima and less than 4% for emission peaks. All new maleimides were successfully synthesised *via* a tandem Suzuki reaction. We expect the described method considering its accuracy can be used for future theoretical predictions of fluorescence spectra.

Experimental

Nuclear Magnetic Resonance (NMR) spectra were recorded on a Bruker ARX 300 (^1H at 300.14 MHz and ^{13}C at 75.47 MHz), Bruker AV 500 (^1H at 500.13 MHz and ^{13}C at 125.8 MHz), Varian 400 (^1H at 399.86 and ^{13}C at 100.54 MHz) or a Bruker AV 600 (^1H at 600.13 MHz and ^{13}C at 150.90 MHz) spectrometer at 25 °C. ^1H and ^{13}C spectra were referenced to the residual (partially) undeuterated solvents. Infrared (IR) spectra were recorded with a PerkinElmer Spectrum One Spectrometer FT-IR spectrometer. Samples were analysed as thin films on NaCl discs. Mass Spectra were collected using electron impact ionisation (EI-MS) on a VG AutoSpec. EI-HRMS was performed with a resolution of approximately 10,000. All air and/or moisture sensitive reactions were performed in flame dried glassware under an argon atmosphere. All solvents were distilled prior to use, and if used in air and/or moisture sensitive reactions, were degassed. Anhydrous solvents were obtained by distillation from the appropriate drying agent. Thin layer chromatography (TLC) was performed on Merck silica gel 60 F₂₅₄, pre-coated aluminium sheets. Visualisation of developed plates was achieved through the use of a 254 nm or 365 nm UV lamp. Column chromatography was performed using silica gel 60 (0.063–0.200 mm) as supplied by Merck with the eluents indicated. Starting materials and

Fig. 3 Structures of new maleimide derivatives (**23–28**)



reagents were generally available from Sigma–Aldrich or Fluka.

1-Benzyl-3,4-di-thiophen-2-yl-pyrrole-2,5-dione (23) 1,1'-Bis(diphenylphosphino)ferrocene, (134 mg, 0.24 mmol) was added in one portion to a magnetically stirred solution of Pd(OAc)₂ (56 mg, 0.24 mmol) in dry THF (3 mL). The ensuing mixture was stirred for approximately 30 min before being treated with 3,4-dichloromaleimide **2** (281 mg, 1.1 mmol) in one portion and stirred at room temperature for 15 min. 4,4,5,5-Tetramethyl-2-thiophen-2-yl-[1,3,2] dioxaborolane (500 mg, 2.4 mmol) and NaOH (97, 2.4 mmol) were then added in one portion to the reaction mixture. The ensuing solution was then stirred at 46 °C for 16 h. Subjection of the crude material to flash column chromatography (1:19 EtOAc/hexane) afforded the disubstituted maleimide **20** (116 mg, 30%) as an orange solid. M.p 130–132 °C; ¹H NMR (300 MHz, CDCl₃): δ=7.82 (dd, *J*=1.2, 3.9 Hz, 2H, ArH), 7.57 (dd, *J*=1.2, 5.1 Hz, 2H, ArH), 7.46–7.42 (m, 2H, ArH), 7.37–7.28 (m, 3H, ArH), 7.12 (dd, *J*=3.6, 5.1 Hz, 2H, ArH), 4.79 (s, 2H); ¹³C NMR (100.5 MHz, CDCl₃): 170.0 (C=O), 136.3, 131.5, 131.1, 129.8, 128.9, 128.8, 128.1, 127.7, 42.3 (CH₂Ph); IR (neat) ν_{\max} (cm⁻¹): 2,925, 1,765, 1,700 (C=O), 1,432, 1,401, 699; EI-MS *m/z* (relative intensity): 351 (100, [M]⁺), 323 (13), 246 (12), 191 (15), 190 (38); EI-HRMS calcd for C₁₉H₁₃NO₂S₂: 351.0388, found: 351.0386.

1-benzyl-3,4-bis(4-(dimethylamino)phenyl)-1H-pyrrole-2,5-dione (24) 3,4-Dichloromaleimide **2** (400 mg, 1.6 mmol), PdCl₂(dppf).CH₂Cl₂ (127 mg, 0.16 mmol), 4-(*N,N*-dimethylamino)phenyl boronic acid (pinacol ester, 964 mg, 3.9 mmol), NaOH (187 mg, 4.7 mmol, in 0.5 mL water)

in THF (5 mL) was degassed using the freeze pump thaw method. The mixture was heated to 45 °C and left to stir for 16 h. The crude mixture was fused to silica and passed through a plug of coarse silica with 1:1 ethyl acetate/hexane. The filtrate was again fused to silica and subjected to column chromatography eluting with (1:9 → 1:3 ethyl acetate/hexane) to afford the bis-aniline **22** (42 mg, 6%) as a deep red solid, m.p 191–193 °C; *R*_f=0.24 (3:7 ethyl acetate/hexane); ¹H NMR (400 MHz, CDCl₃): δ=7.45–7.38 (m, 4H), 7.38–7.36 (m, 2H), 7.26–7.18 (m, 4H), 6.57 (d, *J*=8.8 Hz, 4H, Ar-H), 4.69 (s, 2H, CH₂), 2.92 (s, 12H, 4 × CH₃N); ¹³C NMR (400 MHz, CDCl₃): δ=171.8 (C=O), 137.2, 131.2, 128.8, 128.7, 127.7, 111.8, 41.7 (CH₂), 40.3 (N(CH₃)₂); IR (film): $\tilde{\nu}$ = 2,895 cm⁻¹, 1,695, 1,604, 1,354, 1,196, 818; MS (EI): *m/z* (%)=425.1 (100, [M]⁺); EI-HRMS calcd for C₂₇H₂₇N₃O₂ 425.2103; found: 425.2107.

1-Benzyl-3,4-diphenyl-1H-pyrrole-2,5-dione (25) 3,4-Dichloromaleimide **2** (400 mg, 1.56 mmol), PdCl₂(dppf).CH₂Cl₂ (127 mg, 0.16 mmol), NaOH (125 mg, 3.1 mmol), phenylboronic acid (415 mg, 3.4 mmol) in THF (5 mL) was degassed using the freeze pump thaw method. The ensuing mixture was stirred at room temperature for 16 h. The crude mixture was fused to silica and subjected to column chromatography, silica gel eluted with (1:19 → 1:4 ethyl acetate/hexane), to afford the bis-aryl compound **23** as a light green solid (243 mg, 46%), *R*_f=0.29 in 1:9 ethyl acetate/hexane, m.p 132–134 °C. ¹H NMR (300 MHz, CDCl₃): δ=7.47–7.44 (m, 6H, Ar-H), 7.36–7.30 (m, 9H, Ar-H), 4.80 (s, 2H, N-CH₂); ¹³C NMR (400 MHz, CDCl₃): δ=170.6 (2C, 2 × C=O), 136.6, 136.3, 130.0, 130.0, 129.0, 128.8, 128.7, 128.7, 128.1, 42.2 (CH₂); IR (film): $\tilde{\nu}$ = 3,060 cm⁻¹, 1,701 (C=O), 1,433,

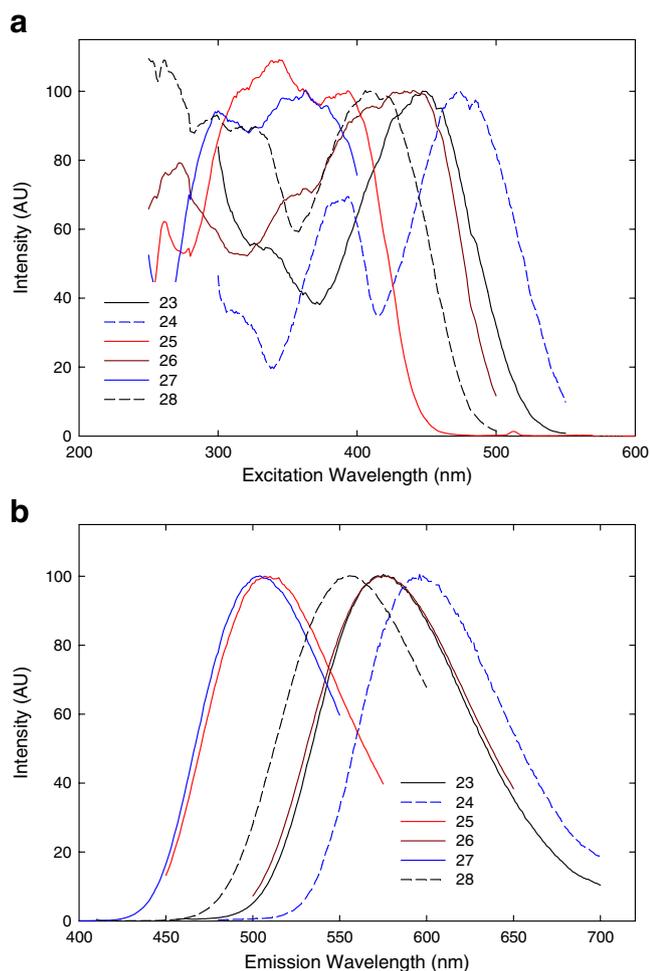


Fig. 4 Measured excitation (a) and emission (b) spectra of new maleimide derivatives

1,401, 1,350, 1,075, 692; MS (EI): m/z (%)=339.1 (100) $[M]^+$, 294.1 (32), 262.1 (21), 178.0 (37); EI-HRMS calcd for $C_{23}H_{17}NO_2$ 339.1259; found: 339.1252.

1-Benzyl-3,4-bis(4-isopropoxyphenyl)-1H-pyrrole-2,5-dione (**26**) 3,4-Dichloromaleimide **2** (400 mg, 1.6 mmol),

$Pd_2(dba)_3 \cdot CHCl_3$ (161 mg, 0.16 mmol), dppf (173 mg, 0.16 mmol), NaOH (62 mg, 1.6 mmol), *N*-methyldicyclohexylamine (334 μ L, 1.6 mmol), 4-isopropoxyphenylboronic acid (617 mg, 3.4 mmol) in THF (5 mL) was degassed using the freeze pump thaw method. The ensuing mixture was stirred at room temperature for 16 h, fused to silica and passed through a plug of coarse silica with 1:1 ethyl acetate/hexane. The concentrated organic washings were subjected to column chromatography, 1:49 ethyl acetate/hexane to afford the bis-ether **24** (69 mg, 9%) as an orange oil. 1H NMR (300 MHz, $CDCl_3$): δ =7.52–7.40 (m, 6H, Ar-H), 7.39–7.24 (m, 3H, Ar-H), 6.86 (m, 4H, Ar-H), 4.81 (s, 2H, NCH_2), 4.60 (septet, J =6.0 Hz, 2H, $2 \times CH$), 1.37 (d, J =6.0 Hz, 12H, $2 \times CH_3$), ^{13}C NMR (300 MHz, $CDCl_3$): δ =171.2, 159.3, 136.8, 134.0, 131.6, 128.9, 128.8, 127.8, 121.1, 115.6, 70.0 ($2 \times CH$), 42.0 (CH_2), 22.1 ($2 \times CH_3$); IR (film): $\tilde{\nu}$ = 3,019 cm^{-1} , 2,980, 1,700 (C=O), 1,251, 1,215, 668; MS (EI): m/z (%)=455.1 (91, $[M]^+$), 371.0 (100), 209.1 (25); EI-HRMS calcd for $C_{29}H_{29}NO_4$ 455.2097; found: 455.2109.

3,4-Bis(4-acetylphenyl)-1-benzyl-1H-pyrrole-2,5-dione (**27**) 3,4-Dichloromaleimide **2** (400 mg, 1.6 mmol) $Pd_2(dba)_3 \cdot CHCl_3$ (161 mg, 0.16 mmol), dppf (173 mg, 0.16 mmol), NaOH (62 mg, 1.6 mmol), *N*-methyldicyclohexylamine (334 μ L, 1.6 mmol), 4-acetylphenylboronic acid (562 mg, 3.4 mmol) in THF (5 mL) was degassed using the freeze pump thaw method. The ensuing mixture was left to stir at room temperature for 16 h. The crude mixture was fused to silica and subjected to column chromatography, silica gel eluted with (1:19 \rightarrow 1:4 ethyl acetate/hexane) to afford the bis-aryl compound **25** as a light green solid (295 mg, 45%), R_f =0.30 in (2:3, ethyl acetate/hexane), m.p 149–150 $^\circ C$. 1H NMR (400 MHz, $CDCl_3$): δ =7.92–7.53 (m, 4H, Ar-H), 7.54–7.51 (m, 4H, Ar-H), 7.45–7.41 (m, 2H, Ar-H), 7.32–7.29 (m, 3H, Ar-H), 4.82 (s, 2H, $N-CH_2$), 2.60 (s, 6H, $2 \times CH_3$); ^{13}C NMR (400 MHz, $CDCl_3$): δ =197.4, 169.6, 138.0, 136.52, 136.12, 132.8, 130.2, 129.0, 128.9, 128.58, 128.18, 42.4, 26.8; IR

Table 3 Calculated (in vacuo) and experimental (in hexane) wavelengths of maximum absorption and emission of new compounds. Calculated oscillator strengths are shown in parentheses

Compound	λ_{max}^{abs} (nm) calculated	λ_{max}^{abs} (nm) experimental	Dev. (%)	λ_{max}^{em} (nm) calculated	λ_{max}^{em} (nm) experimental	Dev. (%)
23	434.9 (0.530)	448 ^a	-2.9	552.3 (0.550)	575 ^a	-3.9
24	418.2 (0.496)	474	-11.8	568.2 (0.524)	596	-4.7
25	376.5 (0.450)	344	9.4	477.3 (0.448)	512	-6.8
26	401.4 (0.476)	440	-8.8	521.2 (0.498)	574	-9.2
27	373.2 (0.579)	363	2.8	473.5 (0.543)	504	-6.1
28	396.9 (0.478)	406	-2.2	517.5 (0.487)	556	-6.9

^a in dichloromethane solvent

(film): $\tilde{\nu} = 3,011\text{ cm}^{-1}$, 1,705 (C=O), 1,685 (C=O), 1,401, 1,266, 832, 752; MS (EI): m/z (%)=423.1 (100, $[M]^+$), 408.1 (45), 380.1 (27), 91.0 (34). EI-HRMS calcd for $C_{27}H_{21}NO_4$ 423.1471; found: 423.1465.

1-Benzyl-3,4-bis(4-hydroxyphenyl)-1H-pyrrole-2,5-dione (**28**) 3,4-Dichloromaleimide **2** (400 mg, 1.6 mmol) was $PdCl_2(dppf).CH_2Cl_2$ (191 mg, 0.23 mmol), 4-hydroxyphenyl boronic acid (645 mg, 4.7 mmol), NaOH (187 mg, 4.7 mmol, in 0.5 mL water) in THF (5 mL) was degassed using the freeze pump thaw method. The ensuing mixture was stirred at room temperature for 16 h. To the ensuing mixture was treated with HCl (1.5 mL, 2 M) and the aqueous layer was extracted with ether (4×20 mL). The combined organic layers were dried ($MgSO_4$) and concentrated under reduced pressure. The crude mixture was fused to silica and passed through a plug of coarse silica with 1:1 ethyl acetate/hexane to remove the palladium catalyst. The organic washings were treated with water (50 mL), K_2CO_3 (20 mL, sat aq). Treatment of the separated aqueous layer (HCl, 20 mL, 2 M), extraction with ether (3×50 mL), drying ($MgSO_4$) and concentration of the organic layers afforded diphenol **26** (110 mg, 18%) as an orange oil; 1H NMR (300 MHz, d_6 -acetone): δ =7.46–7.20 (m, 9H, Ar–H), 6.90–6.82 (m, 4H, Ar–H), 4.80 (s, 2H, CH_2); ^{13}C NMR (400 MHz, d_6 -acetone): δ =171.6, 159.6, 138.2, 134.9, 132.4, 129.3, 128.7, 128.2, 121.4, 116.2, 42.1; IR (KBr): $\tilde{\nu} = 3,393\text{ cm}^{-1}$, 1,693, 1,606, 1,351, 1,172, 838; MS (EI): m/z (%)=371.1 (100, $[M]^+$); EI-HRMS calcd for $C_{23}H_{17}NO_4$ 371.1158; found: 371.0146.

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