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The influence of tetrahydroquinoline rings in dicyanomethylenedihydrofuran (DCDHF) single-molecule fluorophores

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Abstract—We have synthesized several series of DCDHF fluorophores with the amine donor either acyclic or constrained in one or two tetrahydroquinoline rings. The absorption and the fluorescence emission wavelengths and quantum yields have been determined and correlated with the specific donor structures. Generally, inclusion of the donor in a ring annulated to the benzene or naphthalene aromatic (Ar) π-core results in a bathochromic shift of absorption and emission accompanied by an increase in the quantum yield. Thus, the tetrahydroquinoline donor provides an efficient way to tailor the properties of fluorophores with substituted amines as electron-donating groups.

1. Introduction

Fluorescence is a physical property of many materials, which has been exploited in a wide variety of applications. As just one example, organic dyes are extensively employed in biological systems as labels and tags to study the metabolism and other fundamental processes. As a result, there is much current interest in optimizing and tuning the performance of fluorescent dyes for such purposes. In cells, fluorescence detection sensitivity depends on reducing background signals, which arise from autofluorescence or from unbound or nonspecifically bound fluorophores. Because cells autofluorescence with short wavelength excitation, one way to minimize background is to optimize fluorophores for longer wavelength absorption and emission.[1] At the same time this longer wavelength can reduce the light scattering by dense media such as tissues and improve penetration of the excitation light. Finally, optimizing the Stokes shift can result in a greater differentiation between the absorption spectrum and the emission spectrum, which makes it easier to use filters to block excitation light while still collecting most of the emitted fluorescence. It is well-known that polar fluorescent dyes containing an electron-donating group (an amino group in most cases[2–4]) and an electron withdrawing group often exhibit large Stokes shifts because of intramolecular charge-transfer (ICT) states. In the case of an amine donor, replacing the typical dialkylamino groups by a cyclic tetrahydroquinoline group would be one way to shift the absorption and emission to longer wavelengths.

Recently, we have focused on the design and synthesis of a new structure class of donor–acceptor fluorophores, the dicyanomethylenedihydrofuran (DCDHF) dyes.[5–8] The DCDHF fluorophores can be imaged at the single-molecule level with additional beneficial properties such as a viscosity-dependent fluorescence, strong solvatochromism, significant ground-state dipole moment, and moderate hyperpolarizability. One of the most important characteristics of fluorophores for imaging applications is the fluorescence quantum yield (Φ_F), a quantity, which should be as large as possible. In our previous mechanistic studies, intermolecular twisting around the dicyanomethylene bond was shown to strongly affect Φ_F by introducing nonradiative deexcitation pathways.[9–11] Twisting about the amine–phenyl bond might also have an effect on the brightness of the emission. To test this idea, we synthesized several new DCDHF fluorophores with a tetrahydroquinoline ring in an attempt to improve the quantum yield and simultaneously shift the wavelength further to the red.

2. Synthesis

The DCDHF dyes **7–9** that contain a single benzene ring and a single alkene conjugated unit were examined first (Scheme 1). These dyes follow the same synthetic protocol[12] as was used for electro-optical chromophores containing this

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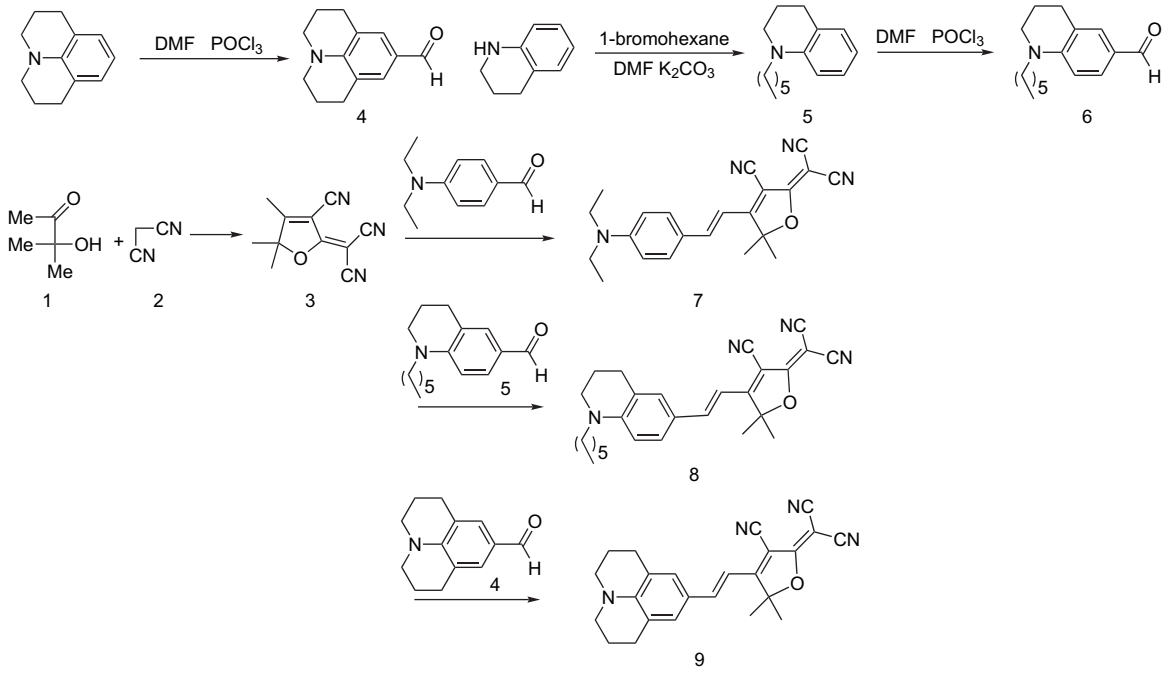
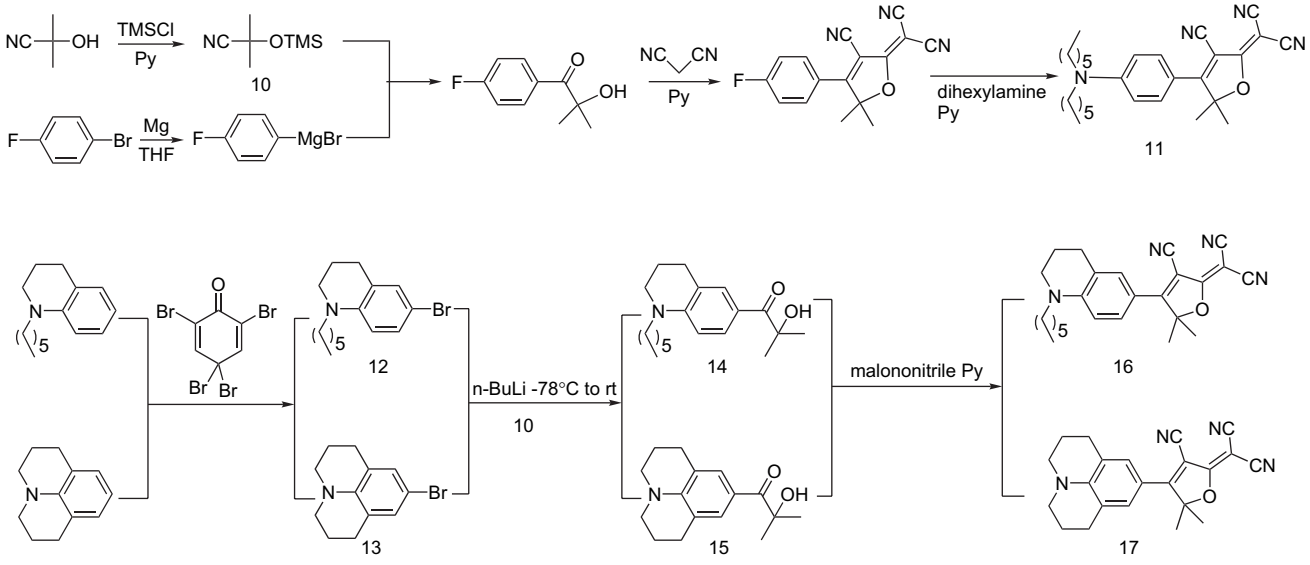


Scheme 1.

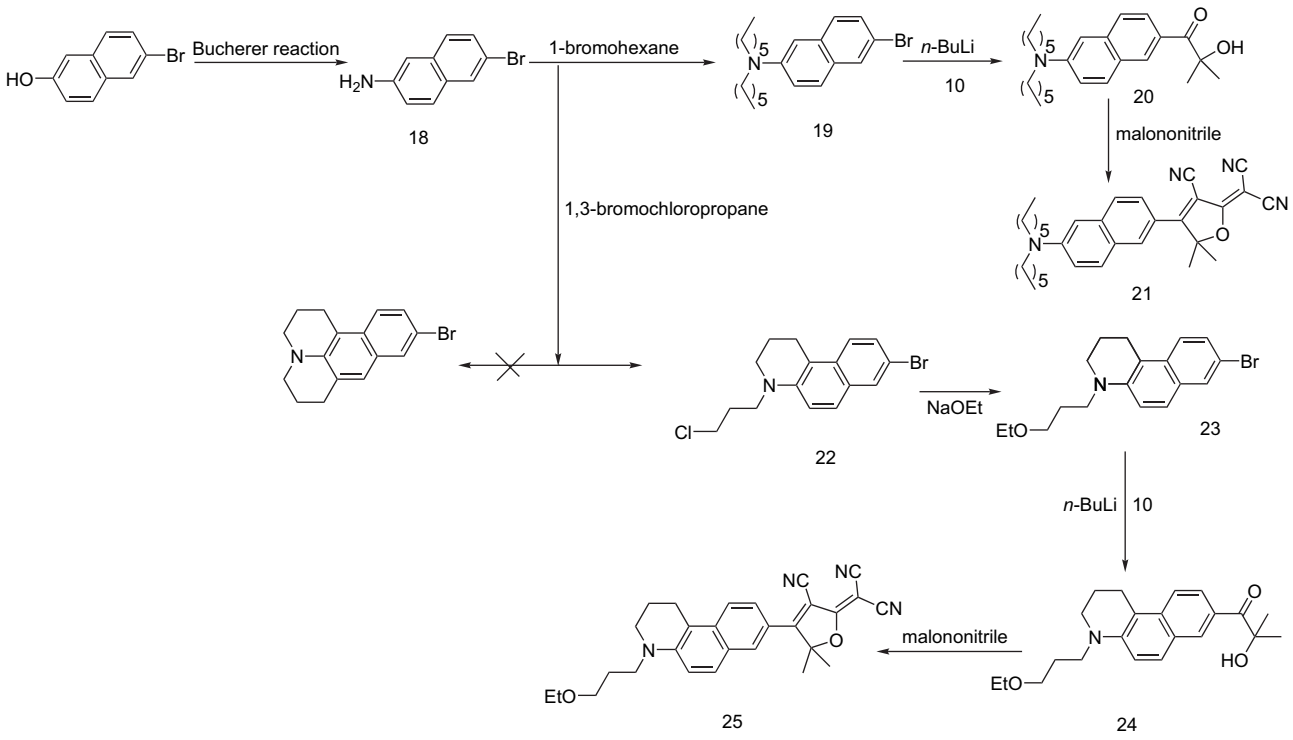
DCDHF acceptor group.[13–17] The precursor **3** was made by condensation of α-hydroxyketone **1** with malononitrile and then reacted with 4-diethylaminobenzaldehyde to give fluorophore **7**; reacted with 1-hexyl-1,2,3,4-tetrahydro-quinoline-6-carbaldehyde **5**, which is prepared by alkylation of tetrahydroquinoline[18] followed by Vilsmeier reaction, to give fluorophore **8**; reacted with 2,3,6,7-tetrahydro-1*H*,5*H*-pyrido[3,2,1-*ij*]quinoline-9-carbaldehyde **4**, which was prepared by Vilsmeier reaction of julolidine,[19] to give fluorophore **9**.

The set of DCDHF dyes without a vinyl link was synthesized originally for photorefractive applications[13,16,20] (Scheme 2). Synthesis of **11** begins with 4-fluorobromobenzene to make the fluoro intermediate, which undergoes efficient aromatic nucleophilic substitution with dihexylamine in pyridine.[13] Compounds **16** and **17** were prepared by bromination of 1-hexyl-1,2,3,4-tetrahydro-quinoline and julolidine[21] followed by lithiation and trapped with **10** to make the respective 4-amino substituted α-ketols. Condensation of the resulting ketols with malononitrile gives the desired DCDHF dyes directly.

When these compounds were evaluated for fluorescence it was immediately apparent that the removal of the alkene resulted in enhanced quantum yields. In the search for more red-shifted dyes that retain high quantum yield, the corresponding naphthalene dyes might be appropriate.



Scheme 2.

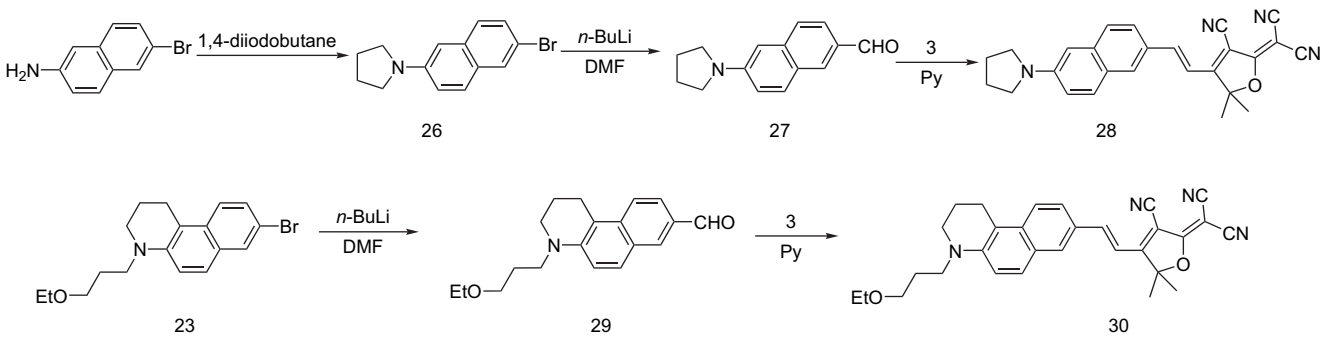


Scheme 3.

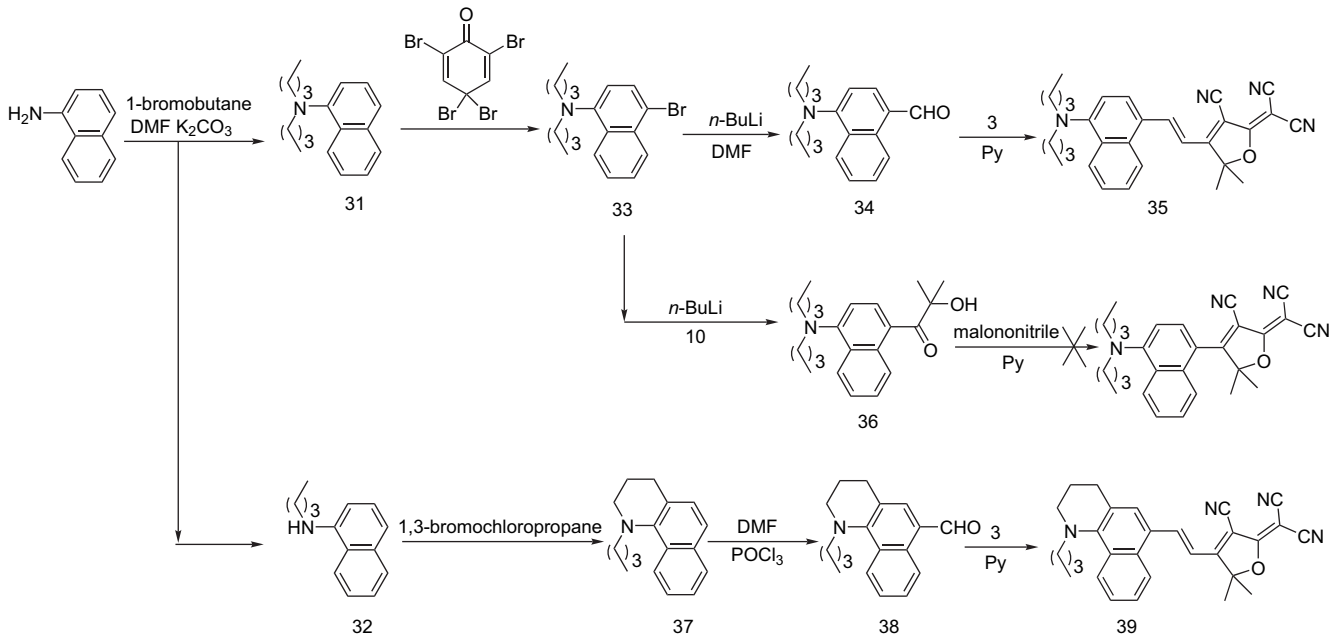
Fluorophore **21** was first synthesized, and found to be excited at 514 nm and was used successfully as single-molecule tracers in the cellular membrane.[8] Although the red shift is not as large as that resulting from addition of an alkene group to the benzene ring, the quantum yield of these dyes is higher than that of their benzene counterparts. With these results in hand, we planned to make 2,6-substituted DCDHF fluorophores with tetrahydroquinoline rings. When we reacted 2-bromo-6-aminonaphthalene (**18**) with 1-bromo-3-chloropropane the product resulting from two-ring closure reactions was expected.[22] However, and to our surprise, instead only the intermediate 8-bromo-4-(3-chloropropyl)-1,2,3,4-tetrahydrobenzo[*f*]quinoline **22** was separated as the major product (63% yield). Proof for this substitution pattern comes from a crystal structure of compound **30** derived from **22** (Fig. 3). All efforts to push the 8-bromo-4-(3-chloropropyl)-1,2,3,4-tetrahydrobenzo[*f*]-quinoline to close a second ring at temperatures up to 200 °C failed. As such, we were unable to prepare 9-bromo-2,3,5,6-tetrahydro-1*H*,4*H*-3a-aza-benzo[*de*]anthracene[23] by this route. However, the chloro functionality in compound **22** is also useful. For example, a nucleophilic attack of ethoxide to displace the chlorine giving **23** was accomplished. A similar 1,2,3,4-tetrahydrobenzo[*f*]quinoline with an alkyl group on the amino group was synthesized in four steps.[24] The ethyl ether product **23** after lithiation leads again to the α-ketol **24** and desired DCDHF dye **25** (Scheme 3).

In addition, both of these dyes with vinyl linkages have also been synthesized (Scheme 4) using the similar method found in Scheme 1. The bromo compounds **26** and **23** were lithiated and trapped with DMF to obtain aldehydes **27** and **29**. These two aldehydes were then condensed with compound **3** to give DCDHF fluorophores **28** and **30**.

We also introduced this tetrahydroquinoline ring onto 1,4-disubstituted naphthalene (Scheme 5). Alkylation of 1-naphthylamine led to both di- and mono-substituted compounds **31** and **32**. Compound **31** was brominated[21] and transformed to α-ketol **36**. However, because of the *peri* steric effect, the



Scheme 4.



Scheme 5.

ring condensation with malononitrile failed with the recovery of reactants. So, we chose to convert this bromo compound **33** to an aldehyde via lithiation and trapping with DMF. Condensation of aldehyde **34** led to the desired fluorophore **35**. On the other hand, mono-substituted compound **32** can undergo ring formation with 1,3-bromochloropropane to give **37** and Vilsmeier reaction to provide aldehyde **38**. Condensation of this aldehyde with heterocycle **3** in pyridine resulted in the desired dye **39**.

3. Results and discussion

The relevant physical properties of the different DCDHF dyes in toluene are summarized in Table 1. As can be seen, the absorption and emission wavelengths systematically increase as the nitrogen donor is constrained by one ring and then by two rings. In the group of benzene substrates with the presence of vinyl group (group A), the absorption wavelength shifts to the red 19 nm with addition of one ring and 32 nm with addition of two rings while the emission wavelength shifts to the red 15 nm with one ring and 25 nm with two rings. In the group of benzene substrates without the presence of vinyl group (group B), the emission shifts are slightly smaller, 10 and 22 nm for the addition of one ring and two rings, respectively. Also, the inclusion of the donor nitrogen in the rings has a substantial influence on the fluorescence quantum yield. It is increased almost five times by comparing **11** with **17** and twice by comparing **7** with **9**.

Table 1. Characterization of DCDHF dyes with and without tetrahydroquinoline rings (all values in toluene)

Group	Entry	λ_{abs}^{max}	λ_{em}^{max} (Stokes shift)	Φ_F Toluene (PMMA)
A	**7**	562	603 (41)	0.02 (0.39)
	8	581	618 (37)	0.028
	9	594	628 (34)	0.053
B	**11**	486	505 (19)	0.044 (0.92)
	16	495	515 (20)	0.10
	17	503	527 (24)	0.21
C	**21**	527	576 (49)	0.85 (0.98)
	25	545	607 (62)	0.64
D	**28**	574	671 (97)	0.0049
	30	600	699 (99)	0.30
E	**35**	538	661 (123)	0.0067
	39	571	677 (106)	0.0096

The same trends were observed in more conjugated systems: groups C, D, and E. In the 2,6-substituted naphthalene systems (groups C and D), the emission wavelength increased upon constraining the amine twist (bathochromic shifts in the emission of 31 and 28 nm, respectively). The quantum yield was also enhanced; this is especially true for group D, which exhibits an increase of almost two orders of magnitude. The 1,4-substituted system (group E) is more like the benzene systems (groups A and B), with 33 and 16 nm bathochromic shifts, in absorption and emission, respectively.

With the spectral data plotted, it is clear that the absorption and emission wavelengths both increase with the increasing constraint of the donor amine twist. A previous theoretical treatment of the electronic properties of the DCDHF dyes[11] showed that amine twist had little effect on the ground- and excited-state energy surface and was not responsible for the large viscosity sensitivity. The experimental results here quantify the degree to which the exact structure of the donor group shifts the spectra (Fig. 1).

An important test for the utility of a fluorophore is its ability to be imaged at the single-molecule level, which requires strong fluorescence, weak coupling with dark states, and photostability. Importantly, single-molecule imaging of derivative **17** demonstrates that we maintained the favorable photophysical properties of the parent molecule as shown

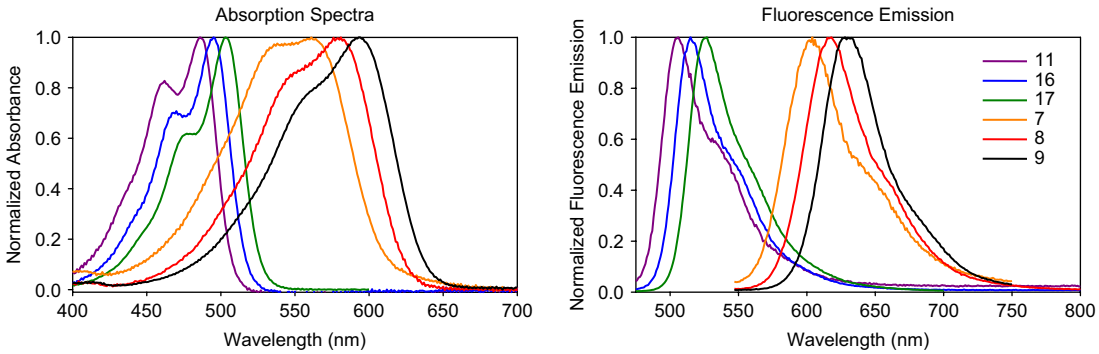


Figure 1. Normalized absorption and fluorescence emission spectra for constrained derivatives and their parent molecules. Increasing the constraint on the amine rotation red shifts the absorbance and emission (for structures see Table 1).

in Figure 2. To characterize the quality of a single-molecule emitter using one simple parameter, we report the total number of photons emitted from a single fluorophore before photobleaching. Measurements on 182 individual molecules yielded an average of $N_{tot,d}=1.5\times10^5$ photons detected per molecule. The photon collection efficiency of our setup is $D=\eta_Q F_{coll} F_{opt} F_{filter}$, which is the product of the camera quantum efficiency, the angular collection factor determined by the objective numerical aperture, the transmission factor through the objective and microscope optics, and the transmission factor through the various filters, respectively.[25] At the emission wavelengths, $\eta_Q=85\%$ for our camera; F_{coll} is assumed to be 50% for our high-NA objective;[26] and we measured F_{opt} and F_{filter} to be 50 and 65%, respectively. After recording single-molecule photobleaching distributions, which are exponential in shape as expected, a fit to the exponential yields the average number of photons. Correcting the $N_{tot,d}$ value for the collection efficiency yields a value for the total number of photons emitted, $N_{tot,e}=1.1\times10^6$ photons per molecule. This value is comparable with the values of DCDHF-6 and R6G (2.4×10^6 and 1.9×10^6 photons emitted per molecule, respectively),[27] both of which are among the best single-molecule fluorophores.

To obtain information about the ground-state configuration of the molecules, an X-ray diffraction structural analysis was performed for fluorophores **16**, **17**, and **30**. Figure 3 shows the numbering scheme for the atoms and the thermal ellipsoids.

The torsion angles of important bonds in all compounds are listed in Table 2. For fluorophores **16** and **17**, the torsion angles involving the amine bonding of the tetrahydroquinoline ring and the benzene ring of these two fluorophores are small and quite comparable (all $<5°$). The angles involving the bonds between the benzene ring and the DCDHF acceptor are likewise very similar (all $<10°$). However, for fluorophore **30**, the tetrahydroquinoline ring becomes more planar with the naphthalene ring (0.73° for the torsion angle involving the amine bonding of tetrahydroquinoline ring and naphthalene ring) and the other chain of the amino group becomes more off plane (−28.95° for the torsion angle involving the bonds between the vinyl group and the DCDHF acceptors).

To investigate the possible charge-transfer between the amine donor and the DCDHF acceptor (the pair of limiting resonance structures is shown in Fig. 4), the bond length between nitrogen–carbon groups present in the six-membered rings would be expected to be shorter than a normal carbon sp^2–nitrogen bond length (1.38 Å).[28] This is the case for **16** (C5–N1, 1.356(4) Å) and **17** (C14–N4, 1.354(6) Å). In addition, the bond length between the carbon of the acceptor and

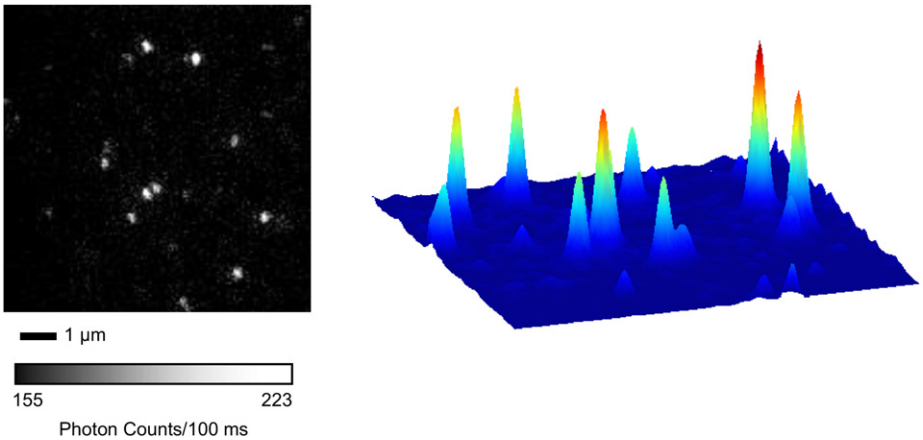


Figure 2. Epifluorescence images of single copies of **17** in a PMMA film imaged at 488-nm excitation. The surface was calculated from two position dimensions and pixel intensity of a Gaussian-smoothed image.

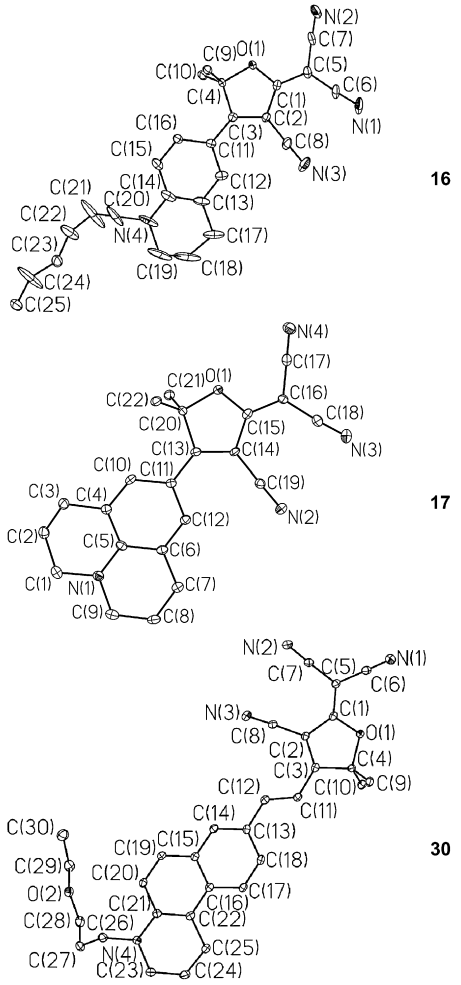


Figure 3. Thermal ellipsoid plots of DCDHF fluorophores **16**, **17**, and **30**. The ellipsoids are drawn at the 30% level.

the carbon of the six-membered ring would also be expected to be shorter than that of a typical carbon sp^2–carbon sp^2 bond (1.48 Å).[28] This is true for fluorophores **16** (C11–C13, 1.430(4) Å) and **17** (C–C 1.429(5) Å).

Our previous calculations on the DCDHF parent molecule showed that furan-dicyano twists on the excited state

Table 2. Torsion angles of fluorophores **16**, **17**, and **30**

Fluorophore	Atoms involved	Torsion angle
16	$C_{(13)}$–$C_{(14)}$–$N_{(4)}$–$C_{(19)}$	−2.57
	$C_{(15)}$–$C_{(14)}$–$N_{(4)}$–$C_{(20)}$	−4.45
	$C_{(16)}$–$C_{(11)}$–$C_{(3)}$–$C_{(4)}$	−8.27
	$C_{(12)}$–$C_{(11)}$–$C_{(3)}$–$C_{(2)}$	−10.65
	$O_{(1)}$–$C_{(1)}$–$C_{(5)}$–$C_{(7)}$	2.58
	$C_{(2)}$–$C_{(1)}$–$C_{(5)}$–$C_{(6)}$	2.85
17	$C_{(6)}$–$C_{(5)}$–$N_{(1)}$–$C_{(9)}$	−2.94
	$C_{(4)}$–$C_{(5)}$–$N_{(1)}$–$C_{(1)}$	−4.10
	$C_{(10)}$–$C_{(11)}$–$C_{(13)}$–$C_{(20)}$	−11.86
	$C_{(12)}$–$C_{(11)}$–$C_{(13)}$–$C_{(14)}$	−10.00
	$O_{(12)}$–$C_{(15)}$–$C_{(16)}$–$C_{(17)}$	4.65
	$C_{(14)}$–$C_{(15)}$–$C_{(16)}$–$C_{(18)}$	5.90
30	$C_{(22)}$–$C_{(21)}$–$N_{(4)}$–$C_{(23)}$	0.73
	$C_{(20)}$–$C_{(21)}$–$N_{(4)}$–$C_{(26)}$	−28.95
	$C_{(12)}$–$C_{(11)}$–$C_{(3)}$-$C_{(4)}$	−172.73
	$C_{(12)}$–$C_{(11)}$–$C_{(3)}$–$C_{(2)}$	2.33

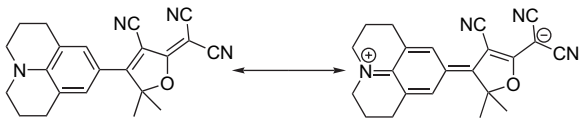


Figure 4. Limiting resonance forms of fluorophore **17**.

manifold predominantly control the changes in emission observed in different hosts.[11] Our current results suggest that twists about the amine–benzene bond also noticeably affect the fluorescence emission. A more detailed solution-phase calculation including excited-state effects needs to be done in the future in order to confirm the exact mechanism.

4. Conclusion

We have synthesized several examples of DCDHF fluorophores with the amine donor either acyclic or constrained in one or two tetrahydroquinoline rings. Generally, inclusion of the donor in a ring annulated to the benzene or naphthalene aromatic π-core results in a bathochromic shift of absorption and emission accompanied by an increase in the quantum yield. Introduction of the first tetrahydroquinoline ring produces the largest effect, with a smaller effect produced by the second ring. Favorable performance in single-molecule fluorescence imaging is maintained for the newly synthesized derivatives.

5. Experimental

5.1. General

Unless otherwise noted, all chemicals were used as received from commercial suppliers. ^{1}H (400 MHz) and ^{13}C (100 MHz) NMR spectra were obtained as solutions in the indicated solvents, and chemical shifts are reported in parts per million (ppm) downfield from internal standard TMS; ^{1}H and ^{13}C coupling constants are reported in hertz (Hz). Compounds **7**,[5] **11**,[13] and **21**[8] were synthesized according to the methods previously described. The α-(trimethylsiloxy)isobutyronitrile (**10**),[33] 6-bromo-2-naphthylamine (**18**),[29] and 6-bromo-2-(pyrrolidino)naphthalene (**26**)[32] were synthesized following the literature procedures.

Solutions were prepared in toluene and bulk absorption and emission spectra were obtained on Perkin–Elmer Lambda 19 UV–vis spectrometer and a SPEX Fluromax-2 fluorimeter, respectively. Quantum yields were measured against standards with known quantum yields and were corrected for differences in optical density and solvent refractive index.[30]

Single-molecule samples were prepared by spin-casting 1% (by mass) solutions of poly(methyl methacrylate) (PMMA) in distilled toluene, doped with nanomolar fluorophore concentrations, onto plasma-etched glass cover slips. After drying, these samples were studied using a Nikon Diaphot 200 inverted microscope in an epifluorescence configuration.[25] Samples were illuminated using a continuous-wave 488-nm Novalux Protera 488-20 laser; the intensity at the sample was approximately 0.5 kW/cm^2. The emission was collected through a 100×, 1.4 N.A. Nikon microscope

objective, filtered to remove scattered excitation light, and imaged onto a back-illuminated frame-transfer Si CCD camera (Roper Scientific MicroMax) with an integration time of 100 ms.

5.2. Experimental procedure

5.2.1. 1-Hexyl-1,2,3,4-tetrahydroquinoline (5).[18] A mixture of tetrahydroquinoline (4.0 g, 30 mmol), 1-bromohexane (3.8 g, 37 mmol), potassium carbonate (8.31 g, 60 mmol), and a catalytic amount of potassium iodide in DMF (40 ml) was heated to 90 °C overnight. After cooling, the reaction mixture was poured into water and extracted with ether. The organic layer was collected, washed with brine, and dried over magnesium sulfate. The solvent was removed and the residue was purified by flash chromatography (hexane) to give product as a clear liquid (4.8 g, 74%). ^{1}H NMR (400 MHz, $CDCl_3$): 7.06 (m, 1H), 6.95 (d, 1H), 6.57 (m, 2H), 3.30 (t, J=11.2 Hz, 2H), 3.25 (t, J=15.2 Hz, 2H), 2.77 (t, J=12.8 Hz, 2H), 1.97 (m, 2H), 1.59 (m, 2H), 1.35 (m, 6H), 0.93 (t, J=13 Hz, 3H); ^{13}C NMR ($CDCl_3$): 145.5, 129.2, 127.2, 122.2, 115.3, 110.6, 51.7, 49.6, 31.9, 28.4, 27.1, 26.3, 22.9, 22.4, 14.2.

5.2.2. 1-Hexyl-1,2,3,4-tetrahydroquinoline-6-carbaldehyde (6).[18] Phosphorous oxychloride (0.78 g, 5.0 mmol) was added dropwise to anhydrous DMF (1.11 g, 15.2 mmol) under nitrogen with ice-bath cooling. After addition, the mixture was stirred at room temperature for 30 min, transferred to a flask containing 1-hexyl-1,2,3,4-tetrahydroquinoline **5** (1.0 g, 4.6 mmol) and the resulting mixture was heated to 90 °C for 4 h. After cooling, water (100 ml) was added to the reaction mixture and the mixture was neutralized with sodium bicarbonate. The mixture was then extracted with ethyl acetate, washed with brine, and dried over magnesium sulfate. Solvent was removed under vacuum and the residue was purified by flash chromatography (hexane/ethyl acetate, 4:1) to give product as a yellow oil (0.9 g, 80%). ^{1}H NMR (400 MHz, $CDCl_3$): 9.67 (s, 1H), 7.55 (d, J=8.4 Hz, 1H), 7.47 (s, 1H), 6.58 (d, J=8.4 Hz, 1H), 3.41 (t, J=11.2 Hz, 2H), 3.34 (t, J=15.2 Hz, 2H), 2.80 (t, J=12.0 Hz, 2H), 1.97 (m, 2H), 1.62 (m, 2H), 1.36 (m, 6H), 0.88 (t, 3H); ^{13}C NMR ($CDCl_3$): 190.0, 150.3, 131.3, 130.4, 124.5, 121.6, 109.3, 51.5, 49.8, 31.7, 28.0, 26.8, 26.3, 22.6, 21.5, 14.0.

5.2.3. 2-{3-Cyano-4-[(*E*)-2-(1-hexyl-1,2,3,4-tetrahydroquinolin-6-yl)-vinyl]-5,5-dimethyl-5*H*-furan-2-ylidene}-malononitrile (8). A mixture of 1-hexyl-1,2,3,4-tetrahydroquinoline-6-carbaldehyde **6** (0.6 g, 2.45 mmol), 2-dicyanomethylen-3-cyano-4,5,5-trimethyl-2,5-dihydrofuran **3** (0.48 g, 2.40 mmol), pyridine (20 ml), and several drops of acetic acid was stirred at room temperature overnight. Pyridine was distilled out under vacuum. The residue was precipitated from CH_2Cl_2/hexane and recrystallized from CH_2Cl_2/methanol to give the product as blue crystals (0.5 g, 49%). DSC, mp: 208 °C; UV–vis (CH_2Cl_2): λ_{max}=607 nm, ε_{max}=1.74×10^4 l cm^{-1} mol^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$): 7.59 (d, J=15.6 Hz, 1H), 7.38 (dd, J=2.0, 8.8 Hz, 1H), 7.31 (d, 1H), 6.70 (d, J=15.6 Hz, 1H), 6.60 (d, J=8.8 Hz, 1H), 3.48 (t, J=12 Hz, 2H), 3.40 (t, J=15.6 Hz, 2H), 2.80 (t, J=12.4 Hz, 2H), 2.00 (m, 2H), 1.76 (s, 6H), 1.66 (m, 2H), 1.36 (m, 6H), 0.93 (t, J=14.0 Hz, 3H); ^{13}C NMR ($CDCl_3$): 176.4, 174.0, 150.5, 148.4, 132.5, 130.5, 123.2, 121.5, 113.0, 112.2, 111.9, 110.8, 107.8, 96.5, 51.8, 50.1, 31.6, 27.9, 26.8, 26.7, 26.6, 22.6, 21.3, 14.0; IR (cm^{-1}): 2927, 2854, 2218, 1501. Anal. Calcd for $C_{27}H_{30}N_4O$: C, 76.03; H, 7.09; N, 13.13. Found: C, 76.23; H, 7.15; N, 13.47.

5.2.4. 9-Formyl-2,3,6,7-tetrahydro-1*H*,5*H*-benzo[*ij*]-quinolizine (4).[19] Following the procedure described for compound **6**, julolidine (1.0 g, 5.9 mmol) was reacted with phosphorous oxychloride and anhydrous DMF to give the product as a yellow oil (1.1 g, 93%). ^{1}H NMR (400 MHz, $CDCl_3$): 9.61 (s, 1H), 7.31 (s, 2H), 3.31 (t, J=11.6 Hz, 4H), 2.78 (t, J=12.8 Hz, 4H), 1.98 (m, 4H); ^{13}C NMR ($CDCl_3$): 190.1, 147.9, 129.5, 124.0, 120.3, 50.0, 27.7, 21.3.

5.2.5. 2-{3-Cyano-5,5-dimethyl-4-[(*E*)-2-(2,3,6,7-tetrahydro-1*H*,5*H*-pyrido[3,2,1-*ij*]quinolin-9-yl)-vinyl]-5*H*-furan-2-ylidene}-malononitrile (9). Following the procedure described for compound **8**, 9-formyl-2,3,6,7-tetrahydro-1*H*,5*H*-benzo[*ij*]quinolizine (0.5 g, 2.5 mmol) was reacted with 2-dicyanomethylen-3-cyano-4,5,5-trimethyl-2,5-dihydrofuran **3** (0.5 g, 2.4 mmol) in pyridine (20 ml) to give the product as blue crystals (0.4 g, 44%). DSC, mp: 285 °C; UV–vis (CH_2Cl_2): λ_{max}=620 nm, ε_{max}=8.09×10^4 l cm^{-1} mol^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$): 7.53 (d, J=16.0 Hz, 1H), 7.15 (s, 2H), 6.67 (d, J=16.0 Hz, 1H), 3.40 (t, J=11.6 Hz, 4H), 2.78 (t, J=12.4 Hz, 4H), 2.02 (m, 4H), 1.57 (s, 6H); ^{13}C NMR ($CDCl_3$): 173.8, 148.5, 148.4, 130.2, 121.9, 113.2, 112.4, 112.1, 107.3, 96.3, 50.4, 27.5, 26.9, 21.0; IR (cm^{-1}): 2937, 2836, 2204, 1536. Anal. Calcd for $C_{24}H_{22}N_4O$: C, 75.37; H, 5.80; N, 14.65. Found: C, 74.99; H, 5.58; N, 14.97.

5.2.6. 6-Bromo-1-hexyl-1,2,3,4-tetrahydroquinoline (12). A solution of 1-hexyl-1,2,3,4-tetrahydroquinoline **5** (2.0 g, 9.2 mmol) in dichloromethane (20 ml) cooled to −10 °C was treated with small portions of 2,4,4,6-tetrabromo-2,5-cyclohexadien-1-one (3.7 g, 9.2 mmol) such that the temperature of the solution was maintained below 0 °C. The solution was stirred at room temperature for 45 min, washed with aqueous sodium hydroxide (3×50 ml, 2 M) and then washed with brine, and dried over magnesium sulfate. After the removal of the solvent the residue was purified by flash chromatography (hexane) to give the product (2.3 g, 65%). ^{1}H NMR (400 MHz, $CDCl_3$): 7.11 (dd, J=8.8, 2.4 Hz), 7.04 (d, J=2.4 Hz, 1H), 6.43 (d, J=8.8 Hz, 1H), 3.26 (t, 2H), 3.21 (t, 2H), 2.75 (t, 2H), 1.94 (m, 2H), 1.57 (m, 2H), 1.34 (m, 6H), 0.92 (t, 3H); ^{13}C NMR ($CDCl_3$): 144.3, 131.4, 129.6, 127.1, 115.2, 112.0, 51.6, 49.3, 31.8, 28.1, 27.0, 26.0, 22.7, 22.0, 14.1; IR (cm^{-1}): 2927, 2856, 1497. HRMS m/z calcd for $C_{15}H_{22}BrN$ [M+H]: 296.1014. Found: 296.1009.

5.2.7. 1-(1-Hexyl-1,2,3,4-tetrahydroquinolin-6-yl)-2-hydroxy-2-methyl-propan-1-one (14). The 6-bromo-1-hexyl-1,2,3,4-tetrahydroquinoline (1.63 g, 4.2 mmol) was dissolved in anhydrous THF (20 ml) with magnetic stirring. The solution was cooled down to −78 °C and 2.5 M of *n*-BuLi in hexane (2.0 ml, 5.0 mmol) was added. The reaction mixture was stirred at −78 °C for 1 h and 2-trimethylsilyloxy-2-cyano-propane **10** (1.2 g, 7.6 mmol) was added via syringe. The reaction mixture was then allowed to

warm to room temperature and stirred overnight. Next, 10% hydrochloric acid (20 ml) was added to the flask with vigorous stirring for 6 h until TLC shows hydrolysis was complete. Sodium bicarbonate was added carefully to the mixture to neutralize the hydrochloric acid and the resulting mixture was extracted with ether. The organic layer was washed with brine and dried with anhydrous magnesium sulfate. Solvent was removed by rotary evaporator and the residue was purified by flash chromatography (hexane/ethyl acetate, 4:1) to give the product as a light yellow liquid (1.0 g, 78%). ^{1}H NMR (400 MHz, $CDCl_3$): 7.82 (d, J=8.8 Hz, 1H), 7.73 (s, 1H), 6.52 (d, J=8.8 Hz, 1H), 3.40 (t, J=11.2 Hz, 2H), 3.33 (t, J=15.2 Hz, 2H), 2.80 (t, J=12.4 Hz, 2H), 1.99 (m, 2H), 1.65 (s, 6H), 1.62 (m, 2H), 1.36 (m, 6H), 0.93 (t, J=14.0 Hz, 3H); ^{13}C NMR ($CDCl_3$): 201.1, 149.4, 131.7, 131.1, 121.1, 118.7, 108.6, 74.9, 51.4, 49.7, 31.7, 29.3, 29.2, 28.1, 26.8, 26.3, 22.6, 21.6, 14.0; IR (cm^{-1}): 3419, 2928, 1589. HRMS m/z calcd for $C_{19}H_{29}NO_2$ [M+H]: 303.2198. Found: 303.2206.

5.2.8. 3-Cyano-2-dicyanomethylen-4-(1-hexyl-1,2,3,4-tetrahydro-quinolin-6-yl)-5,5-dimethyl-2,5-dihydrofuran (16). A mixture of 1-(1-hexyl-1,2,3,4-tetrahydroquinolin-6-yl)-2-hydroxy-2-methyl-propan-1-one **14** (1.0 g, 3.3 mmol), malononitrile (1.0 g, 15 mmol), and pyridine (50 ml) was stirred overnight at room temperature with several drops of acetic acid. The reaction mixture was poured into ice water (300 ml) with stirring and refrigerated overnight. The precipitate was filtered off and recrystallized from dichloromethane/methanol to give the product as violet crystals (0.5 g, 38%). DSC, mp: 202 °C; UV–vis (CH_2Cl_2): λ_{max}=508 nm, ε_{max}=9.59×10^4 l cm^{-1} mol^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$): 7.87 (dd, J=2.4, 9.2 Hz, 1H), 7.74 (d, J=2.8 Hz, 1H), 6.65 (d, J=9.2 Hz, 1H), 3.50 (t, J=11.6 Hz, 2H), 3.42 (t, J=15.2 Hz, 2H), 2.80 (t, J=12.4 Hz, 2H), 2.01 (m, 2H), 1.84 (s, 6H), 1.65 (m, 2H), 1.35 (m, 6H), 0.93 (t, J=14.0 Hz, 3H); ^{13}C NMR ($CDCl_3$): 177.2, 173.3, 151.0, 131.4, 130.7, 122.7, 113.5, 113.2, 113.1, 112.2, 110.6, 97.0, 51.8, 50.2, 31.6, 28.0, 27.8, 26.7, 26.6, 22.6, 21.1, 14.0; IR (cm^{-1}): 2931, 2858, 2220, 2201, 1563. Anal. Calcd for $C_{25}H_{28}N_4O$: C, 74.97; H, 7.05; N, 13.99. Found: C, 74.58; H, 6.98; N, 13.59.

5.2.9. 9-Bromo-2,3,6,7-tetrahydro-1*H*,5*H*-pyrido[3,2,1-*ij*]quinoline (13).[31] Following the procedure described for compound **12**, julolidine (1.3 g, 7.6 mmol) in dichloromethane (20 ml) was reacted with 2,4,4,6-tetrabromo-2,5-cyclohexadien-1-one (3.05 g, 7.5 mmol) to give the product as clear liquid (1.1 g, 60%). ^{1}H NMR (400 MHz, $CDCl_3$): 6.90 (s, 2H), 3.14 (t, J=11.6 Hz, 4H), 2.75 (t, J=13.2 Hz, 4H), 1.99 (m, 4H); ^{13}C NMR ($CDCl_3$): 143.2, 129.2, 123.5, 107.2, 50.1, 27.7, 21.8.

5.2.10. 2-Hydroxy-2-methyl-1-(2,3,6,7-tetrahydro-1*H*,5*H*-pyrido[3,2,1-*ij*]quinolin-9-yl)-propan-1-one (15). Following the procedure described for compound **10**, 9-bromo-2,3,6,7-tetrahydro-1*H*,5*H*-pyrido[3,2,1-*ij*]quinoline **13** (0.7 g, 2.8 mmol) was reacted with 2.5 M of *n*-BuLi in hexane (1.4 ml, 3.5 mmol) and 2-trimethylsilyloxy-2-cyano-propane (1.2 g, 7.6 mmol) to give the product as a light yellow liquid (0.5 g, 70%). ^{1}H NMR (400 MHz, $CDCl_3$): 7.57 (s, 2H), 3.30 (t, J=11.6 Hz, 4H), 2.77 (t, J=12.8 Hz, 4H), 1.98 (m, 4H), 1.64 (s, 6H); ^{13}C NMR ($CDCl_3$): 201.2, 147.0, 130.0, 119.6, 118.4, 74.8, 49.9, 29.4, 27.8, 21.3; IR (cm^{-1}): 3425, 2915, 2850, 1590. HRMS m/z calcd for $C_{16}H_{21}NO_2$ [M+Na]: 282.1470. Found: 282.1464.

5.2.11. 3-Cyano-2-dicyanomethylene-4-(4-tetrahydroquinoline)-5,5-dimethyl-2,5-dihydrofuran (17). Following the procedure described for compound **16**, 2-hydroxy-2-methyl-1-(2,3,6,7-tetrahydro-1*H*,5*H*-pyrido[3,2,1-*ij*]quinolin-9-yl)-propan-1-one **15** (0.4 g, 1.5 mmol) was reacted with malononitrile (1.0 g, 15 mmol) in pyridine (50 ml) to give the product as violet crystals (0.20 g, 37%). DSC, mp: 247 °C; UV–vis (CH_2Cl_2): λ_{max}=517 nm, ε_{max}=9.04×10^4 l cm^{-1} mol^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$): 7.75 (s, 2H), 3.58 (t, J=12.4 Hz, 4H), 2.94 (t, J=12.4 Hz, 4H), 2.17 (m, 4H), 1.97 (s, 6H); ^{13}C NMR ($CDCl_3$): 177.3, 173.0, 149.2, 129.5, 121.3, 113.6, 113.4, 112.43, 112.39, 96.8, 50.3, 27.7, 27.4, 20.5; IR (cm^{-1}): 2939, 2844, 2218, 2203, 1497. Anal. Calcd for $C_{22}H_{20}N_4O$: C, 74.14; H, 5.66; N, 15.72. Found: C, 74.28; H, 6.08; N, 16.02.

5.2.12. 8-Bromo-4-(3-chloropropyl)-1,2,3,4-tetrahydrobenzo[*f*]quinoline (22). A 200 ml round-bottom flask was equipped with a small glass column filled with molecular sieves (4 Å, 5 g) and a condenser. Next, 6-bromo-2-naphthylamine (3.0 g, 13.56 mmol), sodium carbonate (15.0 g, 54 mmol), and 1,3-bromochloropropane (30.0 g, 191.1 mmol) were added and the reaction mixture was heated with vigorous stirring under an atmosphere of nitrogen while the bath temperature was gradually increased (70 °C/1 h, 100 °C/2 h, 160 °C/12 h). After cooling, the mixture was poured into 1 N potassium hydroxide (150 ml) and extracted with ether. The organic layer was further washed with brine, dried over magnesium sulfate, and purified by flash chromatography (hexane/ethyl acetate: 99:1) to give the product as a yellowish solid (2.9 g, 63%). DSC, mp: 84 °C; ^{1}H NMR (300 MHz, $CDCl_3$): 7.80 (d, J=2.1 Hz, 1H), 7.60 (d, J=9.0 Hz, 1H), 7.49 (d, J=9.0 Hz, 1H), 7.45 (dd, J=11.1 Hz, 1H), 7.10 (d, J=9.0 Hz, 1H), 3.53 (m, 4H), 3.55 (t, J=10.8 Hz, 2H), 3.03 (t, J=12.9 Hz, 2H), 2.11 (m, 4H); ^{13}C NMR ($CDCl_3$): 143.2, 131.9, 130.2, 129.5, 128.0, 126.6, 121.7, 116.1, 115.0, 113.3, 50.6, 49.7, 31.6, 30.4, 23.8; IR (cm^{-1}): 2915, 2850, 1502. HRMS m/z calcd for $C_{16}H_{17}BrClN$ [M+H]: 338.0306. Found: 338.0301.

5.2.13. 8-Bromo-4-(3-ethoxypropyl)-1,2,3,4-tetrahydrobenzo[*f*]quinoline (23). A mixture of 8-bromo-4-(3-chloropropyl)-1,2,3,4-tetrahydrobenzo[*f*]quinoline **22** (1.5 g, 4.4 mmol), sodium ethoxide (0.48 g, 7.5 mmol), and ethanol (20 ml) was refluxed for 24 h. After cooling, the reaction mixture was poured into brine (150 ml) and extracted with ether. The organic layer was collected and dried over magnesium sulfate. The solvent was removed and the residue was purified by flash chromatography (ethyl acetate/hexane, 1:19) to give product as yellow liquid (1.0 g, 65%). ^{1}H NMR (400 MHz, $CDCl_3$): 7.77 (d, J=2.0 Hz, 1H), 7.58 (d, J=9.2 Hz, 1H), 7.45 (d, J=8.8 Hz, 1H), 7.42 (dd, J=11.6 Hz, 1H), 7.11 (d, J=9.2 Hz, 1H), 3.48 (m, 6H), 3.31 (t, J=10.8 Hz, 2H), 3.00 (t, J=13.2 Hz, 2H), 2.07 (m, 2H), 1.86 (m, 2H), 1.24 (t, J=14.0 Hz, 3H); ^{13}C NMR ($CDCl_3$): 143.5, 131.7, 129.9, 129.2, 127.6, 126.2, 123.2, 116.1, 114.4, 112.4, 67.8, 66.3, 49.1, 49.0, 27.6, 23.7, 21.8, 15.3;

IR (cm^{-1}): 2928, 2858, 1588, 1115. HRMS *m/z* calcd for $C_{18}H_{22}BrNO$ [M+Na]: 370.0782. Found: 370.0779.

5.2.14. 1-[4-(3-Ethoxypropyl)-1,2,3,4-tetrahydrobenzo[*f*]-quinolin-8-yl]-2-hydroxy-2-methyl-propan-1-one (24). Following the procedure described for compound **14**, 8-bromo-4-(3-ethoxypropyl)-1,2,3,4-tetrahydrobenzo[*f*]quinoline **23** (0.5 g, 1.44 mmol) was reacted with 2.5 M of *n*-BuLi in hexane (1.5 ml, 3.75 mmol) and 2-trimethylsilyloxy-2-cyano-propane[33] (0.76 g, 4.80 mmol) to give the product as a yellowish liquid (0.35 g, 68%). ^1H NMR (400 MHz, $CDCl_3$): 8.41 (d, J=2.0 Hz, 1H), 7.99 (dd, J=9.2 Hz, 1H), 7.70 (d, J=8.8 Hz, 1H), 7.66 (d, J=9.2 Hz, 1H), 7.16 (d, J=9.2 Hz, 1H), 3.54 (t, J=14.0 Hz, 2H), 3.50 (m, 4H), 3.38 (t, J=10.8 Hz, 2H), 3.03 (t, J=13.2 Hz, 2H), 2.09 (m, 2H), 1.89 (m, 2H), 1.72 (s, 6H), 1.25 (t, J=14.0 Hz, 3H); ^{13}C NMR ($CDCl_3$): 203.4, 145.8, 135.4, 132.7, 129.5, 126.1, 124.9, 124.6, 121.2, 115.5, 111.8, 75.7, 67.6, 66.3, 49.2, 48.7, 29.0, 27.6, 23.5, 21.6, 15.3; IR (cm^{-1}): 3403, 2972, 2929, 2860. HRMS *m/z* calcd for $C_{22}H_{29}NO_3$ [M+Na]: 378.2029. Found: 378.0245.

5.2.15. 8-(3-Dicyanomethylen-2-cyano-5,5-dimethyl-3,5-dihydrofuryl)-4-(3-ethoxypropyl)-1,2,3,4-tetrahydrobenzo[*f*]quinoline (21). Following the procedure described for compound **16**, 8-(2-methyl-2-hydroxy-1-oxopropyl)-4-(3-ethoxypropyl)-1,2,3,4-tetrahydrobenzo[*f*]quinoline (0.3 g, 0.84 mmol) was reacted with malononitrile (0.4 g, 6.0 mmol) in pyridine (50 ml) to give the product as a green solid (0.12 g, 32%). DSC, mp: 216 °C; UV–vis (CH_2Cl_2): λ_{max}=569 nm, $\varepsilon_{max}=4.09\times10^4\,l\,cm^{-1}\,mol^{-1}$; ^1H NMR (400 MHz, $CDCl_3$): 8.30 (d, J=2.4 Hz, 1H), 7.92 (dd, J=9.2 Hz, 1H), 7.75 (d, J=9.6 Hz, 1H), 7.66 (d, J=9.2 Hz, 1H), 7.21 (d, J=9.2 Hz, 1H), 3.59 (t, J=6.8 Hz, 2H), 3.47 (m, 6H), 3.02 (t, J=12.8 Hz, 2H), 2.11 (m, 2H), 1.91 (m, 8H), 1.25 (t, J=14.0 Hz, 3H); ^{13}C NMR ($CDCl_3$): 176.6, 175.4, 147.7, 135.7, 132.8, 130.6, 125.1, 124.5, 122.6, 118.2, 116.1, 112.6, 112.5, 112.4, 67.3, 66.4, 49.4, 48.8, 27.7, 27.5, 23.2, 21.2, 15.3; IR (cm^{-1}): 2941, 2857, 2219, 1518. HRMS *m/z* calcd for $C_{28}H_{28}N_4O_2$ [M+Na]: 475.2110. Found: 475.2111.

5.2.16. 2-Pyrrolidino-6-naphthaldehyde (27). In a dry 50 ml round-bottom flask were placed 6-bromo-2-(pyrrolidino)naphthalene (1.0 g, 3.6 mmol) and 40 ml anhydrous diethyl ether. The mixture was cooled to −78 °C and 2.5 ml (5.0 mmol) of *n*-BuLi in hexane (2.5 M) was added to this stirred mixture over 10 min. The resulting mixture was stirred for an additional 1 h at −78 °C and then DMF (1 ml, 12.9 mmol) was added to the resulting mixture via a syringe over 5 min. After completion of addition the temperature was allowed to increase to room temperature and stirred for 8 h. The reaction was quenched with 30 ml water and the mixture was extracted with dichloromethane and the organic layer was washed with water, brine, and dried with anhydrous $MgSO_4$. Solvent was removed by rotary evaporation and the residue was purified by silica gel chromatography with hexane and a hexane/CH_2Cl_2 (9:1) mixture as eluants to give a yellow solid (0.68 g, 84%). ^1H NMR (400 MHz, $CDCl_3$): 10.0 (s, 1H), 8.15 (s, 1H), 7.83 (dd, J=8.8, 2.0 Hz, 2H), 7.65 (d, J=8.8 Hz, 1H), 7.04 (dd, J=9.2, 2.4 Hz, 1H), 6.75 (d, J=2.0 Hz, 1H), 3.47 (m, 2H), 2.10 (m, 2H); ^{13}C NMR ($CDCl_3$): 191.8, 148.2, 138.9, 135.1, 130.9, 130.1, 126.4, 124.8, 123.5, 116.3, 104.6, 47.7, 25.5; IR (cm^{-1}): 2945, 2862, 1678, 1561. Anal. Calcd for $C_{15}H_{15}NO$: C, 79.97; H, 6.71; N, 6.22. Found: C, 80.30; H, 6.81; N, 6.15.

5.2.17. 1-(3-Cyano-2-dicyanomethylen-5,5-dimethyl-2,5-dihydrofuran-4-yl)-2-[2-(6-pyrrolidinonaphthyl)]ethane (28). 2-Pyrrolidino-6-naphthaldehyde (225 mg, 1.0 mmol) and 3-cyano-2-dicyanomethylen-4,5,5-trimethyl-2,5-dihydrofuran (200 mg, 1.0 mmol) were dissolved in 10 ml of pyridine. Two drops of acetic acid was added to the solution and the mixture was stirred for 24 h at room temperature. Pyridine was removed by an efficient oil pump with a dry ice trap at room temperature. The residue was purified by flash chromatography with hexane/dichloromethane mixture as eluant (1:1 to pure dichloromethane) to give a blue solid (330 mg, 72%). UV–vis (CH_2Cl_2): λ_{max}=605 nm, $\varepsilon_{max}=5.81\times10^4\,l\,cm^{-1}\,mol^{-1}$; ^1H NMR (400 MHz, $CDCl_3$) 7.91 (s, 1H), 7.80 (d, J=16 Hz, 1H), 7.75 (d, J=8.8 Hz, 1H), 7.63 (d, J=1.2 Hz, 1H), 7.04 (dd, J=9.2, 2.4 Hz, 1H), 7.00 (d, J=15.6 Hz, 1H), 6.75 (d, J=2.4 Hz, 1H), 3.51 (m, 4H), 2.13 (m, 4H), 1.82 (s, 6H); ^{13}C NMR ($CDCl_3$): 174.0, 148.6, 148.4, 138.1, 134.2, 131.1, 127.2, 126.8, 125.5, 123.7, 116.7, 112.3, 111.5, 111.4, 111.0, 105.1, 97.1, 47.9, 26.7, 25.5; IR (cm^{-1}): 2950, 2848, 2220, 1556. Anal. Calcd for $C_{26}H_{22}N_4O$: C, 76.83; H, 5.46; N, 13.78. Found: C, 76.77; H, 6.58; N, 13.64.

5.2.18. 1-(3-Ethoxypropyl)-1,2,3,4-tetrahydrobenzo[*g*]-quinoline-7-carbaldehyde (29). 8-Bromo-4-(3-ethoxypropyl)-1,2,3,4-tetrahydrobenzo[*f*]quinoline **23** (0.5 g, 1.44 mmol) was dissolved in anhydrous THF (20 ml) with magnetic stirring. The solution was cooled down to −78 °C and 2.5 M of *n*-BuLi in hexane (0.8 ml, 2 mmol) was added. The reaction mixture was stirred at −78 °C for 1 h. Next, anhydrous DMF (0.8 ml, 11.7 mmol) was added via syringe. The reaction mixture was then allowed to warm gradually to room temperature and stirred overnight. Hydrochloric acid (10%, 20 ml) was added to the flask and the mixture was stirred vigorously for 6 h until TLC showed that the hydrolysis was complete. Sodium bicarbonate was added carefully to the mixture to neutralize hydrochloric acid and the resulting mixture was extracted with ether. The organic layer was washed with brine and dried with anhydrous magnesium sulfate. Solvent was removed by rotary evaporation and the residue was purified by flash chromatography (hexane/ethyl acetate, 19:1) to give the product as a light yellow liquid (0.30 g, 55%). ^1H NMR (400 MHz, $CDCl_3$): 10.01 (s, 1H), 8.10 (d, J=1.6 Hz), 7.85 (dd, J=8.8, 1.6 Hz, 1H), 7.75 (d, J=8.4 Hz, 1H), 7.70 (d, J=8.4 Hz, 1H), 3.56 (t, J=14.4 Hz, 2H), 3.50 (m, 4H), 3.40 (t, J=10.8 Hz, 2H), 3.05 (t, J=13.2 Hz, 2H), 2.10 (m, 2H), 1.91 (m, 2H), 1.27 (t, J=14.0 Hz, 3H); ^{13}C NMR ($CDCl_3$): 191.8, 146.0, 136.4, 135.3, 129.7, 129.3, 125.0, 123.5, 121.9, 115.4, 112.2, 67.6, 66.3, 49.2, 48.8, 27.6, 23.7, 21.6, 15.3; IR (cm^{-1}): 2929, 2859, 1679, 1599. HRMS *m/z* calcd for $C_{19}H_{23}NO_2$ [M+Na]: 320.1626. Found: 320.1625.

5.2.19. 2-(3-Cyano-4-{(*E*)-2-[4-(3-ethoxypropyl)-1,2,3,4-tetrahydrobenzo[*f*]quinolin-8-yl]-vinyl}-5,5-dimethyl-5*H*-furan-2-ylidene)-malononitrile (30). Following the procedure described for compound **8**, 1-(3-ethoxypropyl)-1,2,3,4-tetrahydrobenzo[*g*]quinoline-7-carbaldehyde **29**

(0.15 g, 0.50 mmol) was reacted with 2-dicyanomethylen-3-cyano-4,5,5-trimethyl-2,5-dihydrofuran **3** (0.07 g, 0.74 mmol) in pyridine (20 ml) to give the product as blue crystals (0.12 g, 50%). DSC, mp: 198 °C; UV–vis (CH_2Cl_2): λ_{max}=627 nm, ε_{max}=4.42×10^4 l cm^{-1} mol^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$): 7.88 (d, *J*=2.0 Hz, 1H), 7.78 (d, *J*=16.0 Hz, 1H), 7.73 (d, *J*=9.2 Hz, 1H), 7.65 (m, 2H), 7.18 (d, *J*=9.2 Hz, 1H), 7.01 (d, *J*=16.0 Hz, 1H), 3.60 (t, 2H), 3.52 (m, 4H), 3.45 (t, 2H), 3.07 (t, 2H), 2.15 (m, 2H), 1.93 (m, 2H), 1.83 (s, 6H), 1.27 (t, *J*=14.0 Hz, 3H); ^{13}C NMR ($CDCl_3$): 175.9, 173.9, 148.4, 146.5, 135.5, 134.4, 129.7, 126.4, 125.7, 123.7, 122.8, 115.6, 112.8, 112.3, 111.55, 111.49, 111.07, 97.1, 96.4, 67.5, 66.4, 49.4, 48.8, 27.7, 26.7, 23.5, 21.5, 15.3; IR (cm^{-1}): 2927, 2853, 2220, 1560. HRMS *m/z* calcd for $C_{30}H_{30}N_4O_2$ [M+Na]: 501.2266. Found: 501.2268.

5.2.20. *N*,*N*-Dibutylnaphthylamine (31) and *N*-butylnaphthylamine (32).[34] A mixture of 1-naphthylamine (14.3 g, 100 mmol), 1-bromobutane (30 g, 220 mmol), potassium carbonate (55 g, 400 mmol), and ethanol (300 ml) was refluxed for 48 h. The mixture was filtered and the filtrate was extracted with ether, washed with brine, and dried over magnesium sulfate. The solvent was removed and the residue was purified by flash chromatography to give the disubstituted product as a clear liquid (8.0 g, 31%). ^{1}H NMR (400 MHz, $CDCl_3$): 8.31 (d, *J*=7.2 Hz, 1H), 7.78 (d, *J*=8.0 Hz, 1H), 7.51 (d, *J*=8.0 Hz, 1H), 7.42 (m, 2H), 7.38 (t, 1H), 7.13 (d, *J*=6.4 Hz, 1H), 3.11 (t, *J*=14.8 Hz, 4H), 1.46 (m, 4H), 1.30 (m, 4H), 0.83 (t, *J*=14.8 Hz, 6H); ^{13}C NMR ($CDCl_3$): 148.8, 135.0, 131.3, 128.2, 125.7, 125.6, 125.1, 124.4, 123.2, 118.0, 54.2, 29.4, 20.6, 14.1. The mono-substituted product was also obtained from the later fractions (12.0 g, 61%). ^{1}H NMR (400 MHz, $CDCl_3$): 7.84 (m, 2H), 7.47 (m, 2H), 7.40 (m, 1H), 7.27 (d, *J*=9.2 Hz, 1H), 6.66 (d, *J*=7.6 Hz), 4.34 (s, 1H), 3.32 (t, *J*=14.0 Hz, 2H), 1.81 (m, 2H), 1.58 (m, 2H), 1.06 (t, *J*=14.8 Hz, 3H); ^{13}C NMR ($CDCl_3$): 144.0, 134.6, 128.9, 127.0, 125.9, 124.8, 123.7, 120.1, 117.3, 104.4, 44.1, 31.8, 20.8, 14.3.

5.2.21. 4-Bromo-1-*N*,*N*-dibutylnaphthylamine (33). Following the procedure described for compound **8**, 4-*N*,*N*-dibutylnaphthyl-1-amine **31** (1.0 g, 3.9 mmol) in dichloromethane (20 ml) was reacted with 2,4,4,6-tetrabromo-2,5-cyclohexadien-1-one (1.6 g, 3.9 mmol) to give the brominated product as a yellow oil (1.2 g, 92%). ^{1}H NMR (400 MHz, $CDCl_3$): 8.37 (dd, *J*=9.6 Hz, 1H), 8.22 (dd, *J*=8.4 Hz, 1H), 7.71 (d, *J*=8.0 Hz, 1H), 7.56 (m, 2H), 7.05 (d, *J*=8.0 Hz, 1H), 3.13 (t, *J*=14.8 Hz, 4H), 1.49 (m, 4H), 1.31 (m, 4H), 0.87 (t, *J*=14.8 Hz, 6H); ^{13}C NMR ($CDCl_3$): 148.7, 134.2, 132.9, 129.4, 127.4, 127.1, 125.8, 124.7, 118.6, 116.9, 54.0, 29.2, 20.5, 14.0; IR (cm^{-1}): 2923, 2854, 1459. HRMS *m/z* calcd for $C_{18}H_{24}BrN$ [M+H]: 334.1170. Found: 334.1165.

5.2.22. 4-*N*,*N*-Dibutyl-1-naphthaldehyde (34). Following the procedure described for compound **29**, 4-bromo-*N*,*N*-dibutylnaphthylamine **33** (1.5 g, 4.5 mmol) was reacted with 2.5 M of *n*-BuLi in hexane (2.5 ml, 6.3 mmol) and anhydrous DMF (1.7 g, 22.5 mmol) to give the product as a light yellow liquid (0.7 g, 55%). ^{1}H NMR (400 MHz, $CDCl_3$): 10.23 (s, 1H), 9.32 (d, 1H), 8.23 (dd, *J*=8.4, 0.8 Hz, 1H), 7.88 (d, *J*=8.0 Hz, 1H), 7.65 (m, 1H), 7.55 (m, 1H), 7.14 (d, *J*=8.0 Hz, 1H), 3.34 (t, *J*=14.8 Hz, 4H), 1.59 (m, 4H), 1.30 (m, 4H), 0.89 (t, *J*=14.8 Hz, 6H); ^{13}C NMR ($CDCl_3$): 192.3, 155.72, 138.3, 128.7, 125.7, 125.5, 125.0, 114.9, 53.1, 29.2, 20.4, 13.9; IR (cm^{-1}): 2930, 2862, 1665, 1460. HRMS *m/z* calcd for $C_{19}H_{25}NO$ [M+Na]: 306.1834. Found: 306.1840.

5.2.23. 1-(3-Cyano-2-dicyanomethylen-5,5-dimethyl-2,5-dihydrofuran-4-yl)-2-{2-[*N*,*N*-dibutylaminonaphthyl]}-ethane (35). Following the procedure described for compound **8**, 4-*N*,*N*-dibutylnaphthaldehyde **34** (0.5 g, 1.77 mmol) was reacted with 2-dicyanomethylen-3-cyano-4,5,5-trimethyl-2,5-dihydrofuran **3** (0.32 g, 1.60 mmol) in pyridine (20 ml) to give the product as blue crystals (0.45 g, 55%). UV–vis (CH_2Cl_2): λ_{max}=583 nm, ε_{max}=2.22×10^4 l cm^{-1} mol^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$): 8.68 (d, *J*=16.0 Hz, 1H), 8.21 (d, *J*=8.0 Hz, 1H), 8.11 (d, *J*=8.4 Hz, 1H), 8.03 (d, *J*=8.4 Hz, 1H), 7.66 (m, 1H), 7.56 (m, 1H), 7.14 (d, *J*=8.4 Hz, 1H), 7.03 (d, *J*=16.0 Hz, 1H), 3.40 (t, *J*=14.8 Hz, 3H), 1.87 (s, 6H), 1.58 (m, 4H), 1.33 (m, 4H), 0.89 (t, *J*=14.4 Hz, 6H); ^{13}C NMR ($CDCl_3$): 176.0, 174.0, 155.6, 143.8, 133.8, 129.0, 128.3, 127.7, 126.0, 125.6, 123.6, 122.6, 116.1, 112.5, 112.3, 111.4, 111.4, 97.2, 53.2, 29.4, 26.8, 20.4, 13.9. IR (cm^{-1}): 2957, 2931, 2871, 2216, 2203, 1539. Anal. Calcd for $C_{30}H_{32}N_4O$: C, 77.55; H, 6.94; N, 12.06. Found: C, 77.29; H, 7.33; N, 12.34.

5.2.24. 1-Butyl-1,2,3,4-tetrahydrobenzo[*h*]quinoline (37). Following the procedure described for compound **22**, 2-*N*-butylnaphthylamine (1.1 g, 7.4 mmol), sodium carbonate (3.0 g, 28 mmol), and 1,3-bromochloropropane (15.0 g, 95.5 mmol) were reacted together to give a crude liquid as a mixture of the desired product and 1,3-bromochloropropane, which is used in the next step without purification.

5.2.25. 1-Butyl-1,2,3,4-tetrahydrobenzo[*h*]quinoline-6-carbaldehyde (38). Following the procedure described for compound **6**, the crude 1-butyl-1,2,3,4-tetrahydrobenzo[*h*]-quinoline **37** was reacted with phosphorous oxychloride and DMF to give the product as a slight yellow liquid (0.7 g, 36% in two steps). ^{1}H NMR (400 MHz, $CDCl_3$): 10.15 (s, 1H), 9.29 (d, *J*=8.4 Hz, 1H), 8.05 (d, *J*=8.4 Hz, 1H), 7.60 (m, 2H), 7.51 (m, 1H), 3.32 (t, *J*=10.4 Hz, 2H), 3.19 (t, *J*=16.4 Hz, 2H), 2.95 (t, *J*=12.8 Hz, 2H), 1.91 (m, 4H), 1.35 (m, 2H), 1.00 (t, *J*=14.8 Hz); ^{13}C NMR ($CDCl_3$): 192.1, 152.3, 140.4, 131.4, 127.8, 125.4, 125.3, 124.2, 122.8, 56.9, 47.9, 31.1, 28.1, 20.2, 18.4, 14.0; IR (cm^{-1}): 2956, 2929, 2861, 1670, 1507, 1460. HRMS *m/z* calcd for $C_{18}H_{21}NO$ [M+Na]: 290.1521. Found: 290.1523.

5.2.26. 6-(3-Dicyanomethylen-2-cyano-5,5-dimethyl-3,5-dihydrofuryl)-1-butyl-1,2,3,4-tetrahydrobenzo[*h*]quinoline (39). Following the procedure described for compound **8**, 1-butyl-1,2,3,4-tetrahydrobenzo[*h*]quinoline-6-carbaldehyde (0.1 g, 0.37 mmol) and 2-dicyanomethylen-3-cyano-4,5,5-trimethyl-2,5-dihydrofuran (0.07 g, 0.74 mmol) were reacted in pyridine (20 ml) to give the product as blue crystals (0.1 g, 60%). DSC, mp: 222 °C (decomposes upon melting); UV–vis (CH_2Cl_2): λ_{max}=656 nm, ε_{max}=3.75×10^4 l cm^{-1} mol^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): 8.64 (d, *J*=16.0 Hz, 1H), 8.06 (m, 2H), 7.77 (s, 1H), 7.60 (m, 1H),

7.50 (m, 1H), 6.97 (d, J=15.6 Hz, 1H), 3.43 (t, J=10.8 Hz, 2H), 3.38 (t, J=15.6 Hz, 2H), 2.98 (t, J=12.4 Hz, 2H), 2.04 (m, 2H), 1.87 (s, 6H), 1.39 (m, 2H), 0.99 (t, J=14.8 Hz, 3H); ^{13}C NMR (DMSO): 173.8, 152.8, 143.5, 133.2, 129.3, 127.6, 127.1, 125.4, 125.0, 124.2, 122.4, 121.8, 111.3, 96.7, 57.3, 48.9, 30.9, 28.4, 26.7, 20.1, 19.7, 13.7; IR (cm^{-1}): 2932, 2867, 2218, 1485. Anal. Calcd for $C_{29}H_{28}N_4O$: C, 77.65; H, 6.29; N, 12.49. Found: C, 77.48; H, 6.25; N, 12.78.

5.3. X-ray crystallographic data

X-ray crystallography was performed by mounting each crystal onto a thin glass fiber from a pool of Fluorolube™ and immediately placing it under a liquid N_2 stream, on a Bruker AXS diffractometer. The radiation used was graphite monochromatized Mo Kα radiation (λ=0.7107 Å). The lattice parameters were optimized from a least-square calculations on carefully centered reflections. Lattice determination, data collection, structure refinement, scaling, and data reduction were carried out using APEX2 version 1.0-27 software package. Each structure was solved using direct methods. This procedure yielded a number of C, N, and O atoms. Subsequent Fourier synthesis yielded the remaining atom positions. The hydrogen atoms were fixed in positions of ideal geometry and refined within the XSHELL software. These idealized hydrogen atoms had their isotropic temperature factors fixed at 1.2 or 1.5 times the equivalent isotropic U of the C atoms to which they were bonded. The final refinement of each compound included anisotropic thermal parameters on all nonhydrogen atoms.

5.3.1. Crystal data of 16. $C_{22}H_{20}N_4O$, M_w=356.42 g mol^{-1}, crystal dimensions 0.21×0.14×0.13 mm, monoclinic, space group $P2(1)/c$, a=11.048(3), b=9.683(3), c=17.080(5) Å, β=98.456(5)°; V=1807.4(9) Å^3, Z=4, ρ_{calcd}=1.310 g cm^{-3}, Bruker SMART APEX II diffractometer, 1.87<θ<22.78, Mo Kα radiation (λ=0.71073 Å), ω scans, T=105(2) K; of 11176 measured reflections, 2442 were independent and 1430 observed with $I>2\sigma(I)$, $-11<h<12$, $-10<k<10$, $-18<l<18$; R_1=0.0418, wR_2=0.1041, GOF=0.880 for 247 parameters, $\Delta\rho_{max}$=0.213 eÅ^{-3}. The structure was solved by direct methods (SHELXS-97) and refined by full-matrix least-square procedures (SHELXL-97), Lorentzian and polarization corrections and absorption correction SADABS were applied, μ=0.083 mm^{-1}.

5.3.2. Crystal data of 17. $C_{25}H_{28}N_4O$, M_w=400.51 g mol^{-1}, crystal dimensions 0.44×0.31×0.08 mm, triclinic, space group P-1, a=7.7832(12), b=9.7606(15), c=14.830(2) Å, α=78.033(3)°, β=83.752(3)°; γ=85.128(3)°, V=1093.4(3) Å^3, Z=2, ρ_{calcd}=1.217 g cm^{-3}, Bruker SMART APEX II diffractometer, 1.87<θ<25.05, Mo Kα radiation (λ=0.71073 Å), ω scans, T=100(2) K; of 8787 measured reflections, 3854 were independent and 2241 observed with $I>2\sigma(I)$, $-9<h<9$, $-11<k<11$, $-17<l<17$; R_1=0.0987, wR_2=0.2718, GOF=1.834 for 247 parameters, $\Delta\rho_{max}$=0.856 eÅ^{-3}. The structure was solved by direct methods (SHELXS-97) and refined by full-matrix least-square procedures (SHELXL-97), Lorentzian and polarization corrections and absorption correction SADABS were applied, μ=0.076 mm^{-1}.

5.3.3. Crystal data of 30. $C_{30}H_{30}N_4O_2$, M_w=478.58 g mol^{-1}, crystal dimensions 0.58×0.36×0.15 mm, triclinic, space group P-1, a=9.484(2), b=10.296(3), c=14.516(4) Å, α=83.188(4)°, β=73.346(4)°; γ=70.007(4)°, V=1275.8(6) Å^3, Z=2, ρ_{calcd}=1.246 g cm^{-3}, Bruker SMART APEX II diffractometer, 2.11<θ<25.05, Mo Kα radiation (λ=0.71073 Å), ω scans, T=100(2) K; of 10066 measured reflections, 4514 were independent and 3211 observed with $I>2\sigma(I)$, $-11<h<11$, $-12<k<12$, $-17<l<17$; R_1=0.0413, wR_2=0.1078, GOF=0.732 for 328 parameters, $\Delta\rho_{max}$=0.221 eÅ^{-3}. The structure was solved by direct methods (SHELXS-97) and refined by full-matrix least-squares procedures (SHELXL-97), Lorentzian and polarization corrections and absorption correction SADABS were applied, μ=0.079 mm^{-1}.

Crystallographic data (excluding structural factors) for the structures in this paper have been deposited at the Cambridge Crystallographic Data Centre as supplementary publication nos. CCDC 618492 for **16**,CCDC 618493 for **17**, and CCDC 623895 for **30**. Copies of the data can be obtained, free of charge, on application to CCDC, 12 Union Road, Cambridge CB2 1EZ, UK (fax: +44 1223 336033 or e-mail: deposit@ccdc.cam.ac.uk).

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References and notes

1. Moerner, W. E. *Trends Anal. Chem.* **2003**, *22*, 544–548.
2. Doroshenko, A. O.; Bilokin, M. D.; Pivovarenko, V. G. *J. Photochem. Photobiol., A* **2004**, *163*, 95–102.
3. Arunkumar, E.; Forbes, C. C.; Noll, B. C.; Smith, B. D. *J. Am. Chem. Soc.* **2005**, *127*, 3288–3289.
4. Kovalska, V. B.; Kryvorotenko, D. V.; Balanda, A. O.; Losytskyy, M. Y.; Tokar, V. P.; Yarmoluk, S. M. *Dyes Pigments* **2005**, *67*, 47–54.
5. Willets, K. A.; Ostroverkhova, O.; Hess, S.; He, M.; Twieg, R. J.; Moerner, W. E. *Proc. SPIE-Int. Soc. Opt. Eng.* **2003**, *5222*, 150–157.
6. Willets, K. A.; Ostroverkhova, O.; He, M.; Twieg, R. J.; Moerner, W. E. *J. Am. Chem. Soc.* **2003**, *125*, 1174–1175.
7. Schuck, P. J.; Willets, K. A.; Fromm, D. P.; Twieg, R. J.; Moerner, W. E. *Chem. Phys.* **2005**, *318*, 7–11.
8. Nishimura, S. Y.; Lord, S. J.; Klein, L. O.; Willets, K. A.; He, M.; Lu, Z. K.; Twieg, R. J.; Moerner, W. E. *J. Phys. Chem. B* **2006**, *110*, 8151–8157.
9. Willets, K. A.; Nishimura, S. Y.; Schuck, P. J.; Twieg, R. J.; Moerner, W. E. *Acc. Chem. Res.* **2005**, *38*, 549–556.
10. Valeur, B. *Molecular Fluorescence: Principles and Applications*; Wiley-VCH: Weinheim, 2002.
11. Willets, K. A.; Callis, P. R.; Moerner, W. E. *J. Phys. Chem. B* **2004**, *108*, 10465–10473.
12. Melikian, G.; Rouessac, F. P.; Alexandre, C. *Synth. Commun.* **1995**, *25*, 3045–3051.
13. Gubler, U.; He, M.; Wright, D.; Roh, Y.; Twieg, R.; Moerner, W. E. *Adv. Mater.* **2002**, *14*, 313–317.

14. Zhang, C.; Dalton, L. R.; Oh, M. C.; Zhang, H.; Steier, W. H. *Chem. Mater.* **2001**, *13*, 3043–3050.
15. Zhang, C.; Wang, C. G.; Dalton, L. R.; Zhang, H.; Steier, W. H. *Macromolecules* **2001**, *34*, 253–261.
16. You, W.; Hou, Z. J.; Yu, L. P. *Adv. Mater.* **2004**, *16*, 356.
17. Ostroverkhova, O.; He, M.; Twieg, R. J.; Moerner, W. E. *ChemPhyschem* **2003**, *4*, 732–744.
18. Jung, B. J.; Lee, J. I.; Chu, H. Y.; Do, L. M.; Lee, J.; Shim, H. K. *J. Mater. Chem.* **2005**, *15*, 2470–2475.
19. Cai, G. L.; Bozhkova, N.; Odingo, J.; Berova, N.; Nakanishi, K. *J. Am. Chem. Soc.* **1993**, *115*, 7192–7198.
20. Hou, Z. J.; You, W.; Yu, L. P. *Appl. Phys. Lett.* **2004**, *85*, 5221–5223.
21. Guinot, S. G. R.; Hepworth, J. D.; Wainwright, M. *J. Chem. Soc., Perkin Trans. 2* **1998**, 297–303.
22. Katayama, H.; Abe, E.; Kaneko, K. *J. Heterocycl. Chem.* **1982**, *19*, 925–926.
23. Saeki, S. *Chem. Pharm. Bull.* **1961**, *9*, 226–233.
24. Lobo, B. C.; Abelt, C. J. *J. Phys. Chem. A* **2003**, *107*, 10938–10943.
25. Moerner, W. E.; Fromm, D. P. *Rev. Sci. Instrum.* **2003**, *74*, 3597–3619.
26. Enderlein, J. *Opt. Lett.* **2000**, *25*, 634–636.
27. Soper, S. A.; Nutter, H. L.; Keller, R. A.; Davis, L. M.; Shera, E. B. *Photochem. Photobiol.* **1993**, *57*, 972–977.
28. March, J. *Advanced Organic Chemistry: Reactions, Mechanisms and Structure*, 4th ed.; Wiley: New York, NY, 1992.
29. Newman, M. S.; Galt, R. H. E. *J. Org. Chem.* **1960**, *25*, 214–215.
30. Lakowicz, J. R. *Principles of Fluorescence Spectroscopy*, 2nd ed.; Kluwer Academic/Plenum: New York, NY, 1999.
31. Katayama, H.; Ohkoshi, M.; Kaneko, K. *Chem. Pharm. Bull.* **1984**, *32*, 1770–1779.
32. Sanguinet, L.; Twieg, R. J.; Wiggers, G.; Mao, G. L.; Singer, K. D.; Petschek, R. G. *Tetrahedron Lett.* **2005**, *46*, 5121–5125.
33. Picard, J. P.; Azizelyusufi, A.; Calas, R.; Dunogues, J.; Duffaut, N. *Organometallics* **1984**, *11*, 1660–1665.
34. Voronin, M. V.; Nasibulin, A. A.; Klyuyev, M. V. *Pet. Chem. USSR* **1997**, *6*, 508–514.