JOC The Journal of Organic Chemistry

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π-Electron Manipulation of the 5,6-Dihydroxyindole/Quinone System by 3-Alkynylation: Mild Acid-Mediated Entry to (Cross)-Conjugated Scaffolds and Paradigms for Medium-Tunable Chromophores

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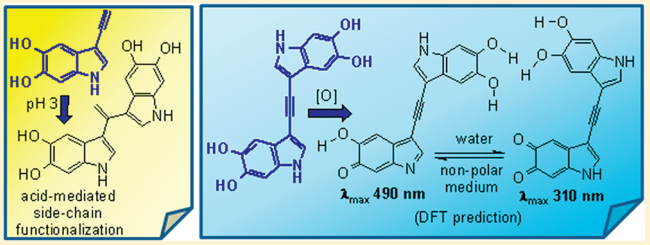
Supporting Information

ABSTRACT: 5,6-Dihydroxyindole-based systems engender increasing interest for the design and implementation of new functional aromatic scaffolds and eumelanin-like materials with tailored absorption and electronic properties. However, studies aimed at elucidating the influence of external π-conjugating groups on the redox properties and acid-induced reactivity of these highly oxidizable indolic platforms are lacking. We report herein the synthesis (as acetyl derivatives) and chemical/quantum chemical characterization of the first π-extended 5,6-dihydroxyindole derivatives, 3-ethynyl-5,6-dihydroxyindole (**1**) and 3,3′-(1,2-ethynediyl)bis-5,6-dihydroxyindole (**2**), in order to understand whether and how β extension of the enamine-like pyrrole sector affects the absorption properties, redox behavior, and protonation equilibria at both the *o*-diphenol and quinone levels. Oxidation of **1** and **2** proceeded smoothly to generate dark insoluble materials with eumelanin-like UV properties. On exposure to phosphate buffer at pH 3, **1** was rapidly converted to 3-acetyl-5,6-dihydroxyindole (**5**) and, in the presence of 5,6-dihydroxyindole, to the cross-conjugated 3,3′-ethenylidenebis-5,6-dihydroxyindole (**6**). DFT calculations on **1** and **2** and their quinones in their pristine states and after protonation provided a mechanistic frame to rationalize the unusual acid-mediated chemistry of **1** and disclosed **2**-quinone as the prototype of a novel class of medium-dependent chromophores. The ethynyl(ene) structural motif is thus proposed as the key to new tunable π-electron extended 5,6-dihydroxyindole/5,6-indolequinone paradigms for the rational design of alkyne-containing hybrid eumelanin-type polymers.



INTRODUCTION

5,6-Dihydroxyindole (DHI) and related compounds occupy a unique position among naturally occurring heterocyclic systems because of their central role in the biosynthesis of eumelanins, the black, insoluble, and heterogeneous biopolymers found in human skin, hair and eyes.[1] Chemical or enzymatic oxidation of DHI leads to the highly unstable and elusive 5,6-indolequinone which can be trapped by nucleophilic compounds.[2] This quinone triggers a complex polymerization process so far elucidated up to the early oligomer stage and commonly believed to model eumelanin build-up.[2a,3] How the growing indolic oligomers are organized into the supramolecular aggregates that form the pigment particles[4] and the detailed structural factors underlying the broad band UV−vis absorption spectrum, semiconductor-like behavior, and free-radical character of eumelanins are still open issues.[5] Parallel to biomimetic polymerization studies,[2a,3a,3b] increasing research work is currently directed to exploring the potential of DHI as a versatile platform for the design and development of bioinspired polymers with tailored properties for potential applications, e.g., in organolectronics, as light-harvesting systems, and in radioprotection.[1b,6] Several recent papers illustrate the broad range of synthetic opportunities offered by the 5,6-dihydroxyindole system,[2a,7,8] including the peculiar acid-mediated chemistry leading to benzylindolylquinoline[7a] and triazatruxene[7e] scaffolds. Unfortunately, the practical exploitation of DHI and related systems in materials science has been tempered by the synthetic difficulties encountered with manipulation of the highly oxidizable π-electron system and the severe insolubility of polymerized material, making their characterization a most challenging task. Nonetheless, the development of a flexible and versatile DHI-based reactive platform susceptible to diverse chemical transformations still remains an attractive goal, largely because of the increasing appreciation of eumelanin-like polymers as soft, biocompatible, and bioavailable materials for potential use as organic semiconductors.[1b,6d] The judicious choice of substituents and bonding patterns can tune the energetics of the conjugated polymer to stabilize quinonoid forms and to tailor the HOMO−LUMO

Received: January 31, 2011
Published: May 03, 2011

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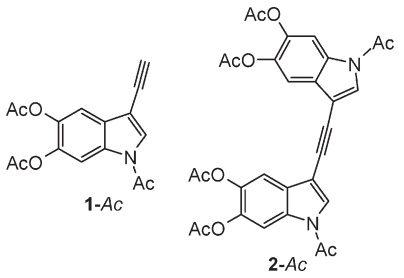
gap. In support of this approach, theoretical and experimental investigations indicated that iodine substitution at the 3-position markedly perturbs the π-electron system of 5,6-indolequinone, prolonging its life-span enough to permit its characterization.[2b] Along this line of thought, a most attractive and so far unexplored strategy for the rational manipulation of 5,6-dihydroxyindoles is inspired by the current renaissance of the chemistry of aromatic alkynes.[9] The availability of efficient protocols for extending $\pi-\pi$ conjugation has enabled the creation of linear molecular structures lacking *Z*/*E* isomerism and functioning as nanoscale "molecular wires", π-electron-based conducting organic structures, as well as self-assembling supramolecular systems.[10]

In this scenario, functionalization of the DHI system with alkynyl groups offers various theoretical and synthetic prospects. Besides providing a versatile structural motif for a broad range of conformation and conjugation opportunities, the triple bond can also undergo a rich array of chemical transformations to form other functional groups.

Herein, we report the synthesis (as acetyl derivatives) and chemical/quantum chemical characterization of 3-ethynyl-5,6-dihydroxyindole (**1**) and 3,3′-(1,2-ethynediyl)bis-5,6-dihydroxyindole (**2**), the first members of this novel class of π-extended DHI derivatives.

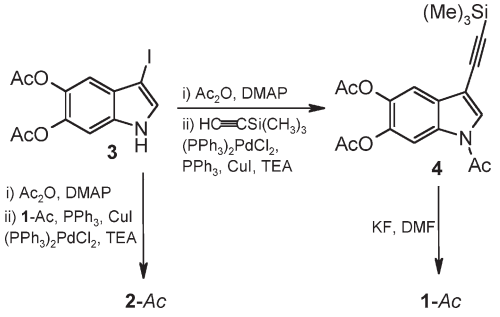


The aims of the present study were manifold: (1) to develop the synthetic chemistry of DHI derivatives possessing alkynyl substituents with special attention on compatibility of the protocols with the notorious oxidizability of the *o*-diphenolic system; (2) to probe the impact of extended π-electron conjugation and nitrogen lone pair delocalization across the triple bond on the absorption properties, redox behavior, and protonation equilibria at both the *o*-diphenol and quinone levels; and (3) to gain an insight into the fundamental aspects of the reactivity of the alkynylated 5,6-dihydroxyindole system with special reference to acid-mediated transformations, a theme of central relevance in 5,6-dihydroxyindole chemistry.[7a,e] Key issues concerning alkyne–allene tautomerism, chromophoric features, redox potential, conformation properties, and protonation equilibria were addressed by a combined computational and experimental effort. The preparation of the 3-alkynyl derivatives was intended as the first step toward the new idea of inserting ethynylene and ethynyl groups as spacers or π-extending groups of eumelanin building bocks with the view of tuning the peculiar physical and electronic properties of these unique biopolymers.

RESULTS AND DISCUSSION

Study Rationale and Product Synthesis. In designing alkynylated 5,6-dihydroxyindoles, the site of substitution was anticipated to be critical for directing control toward the catechol and/or pyrrole sectors of the π-electron platform. In particular, installation of the triple bond onto the 3-position was expected to impact significantly on the electronic makeup of the enamine-like

Scheme 1. Synthesis of 1-*Ac* and 2-*Ac*



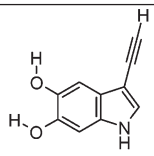
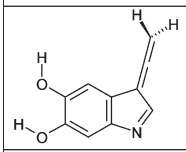
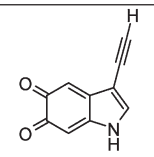
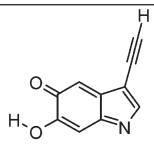
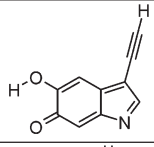
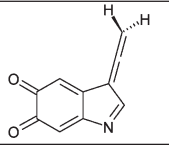
moiety. The expected partial isolation of the enamine sector from the *o*-dioxygenated system, especially in its oxidized *o*-quinone level, would lead to splitting of the electron donation pathway from the nitrogen into two divergent channels. Important issues raised by the 3-alkynylated derivatives were (a) whether and how π-electron delocalization by an alkynyl group perturbs *o*-diphenol/*o*-quinone redox conversion and reactivity; (b) whether insertion of a triple bond bridge as in **2**, alleviating steric influences within the biindole motif, favors or tunes out oxidation-dependent electronic communication between units; (c) whether the 3-alkynyl functionality, which benefits from π-electron donation from the pyrrole sector, offers opportunities to expand the current repertoire of synthetic 5,6-dihydroxyindole chemistry. The relative impact of the 3-alkynyl group on the parent and oxidized quinonoid forms of **1** and **2** is of considerable relevance if one has to interpret and predict the electronic properties and chemical behavior of alkynyl-containing eumelanin-type polymers. Synthetic entries to **1** and **2** required protection of the phenolic groups due to the notorious instability of the 5,6-dihydroxyindole system to oxidation. Accordingly, DHI was protected by an established acetylation methodology,[11] since acetyl groups were easily removable in situ prior to reaction. For both compounds 5,6-diacetoxy-3-iodoindole (**3**)[2b] was selected as the starting material (Scheme 1).

Because of the high electron density on the 3-position, introduction of the ethynyl group by Sonogashira coupling was unsuccessful under commonly used conditions.[9,11] However *N*-acetylation of **3** with acetic anhydride/*N*,*N*-dimethylaminopyridine (DMAP) prior to in situ Sonogashira reaction with trimethylsilylacetylene allowed us to circumvent these difficulties, leading to the desired **4** in satisfactory yield. Desilylation of **4** then led to **1**-*Ac* in moderate overall yield. Sonogashira coupling of **1**-*Ac* with a unit of **3** then gave **2**-*Ac*.

Oxidation Behavior. For oxidation studies, compounds **1**-*Ac* and **2**-*Ac* were completely deacetylated by a previously established procedure.[11] Oxidation of **1** and **2** was then carried out in comparison with the parent DHI using two oxidizing systems typically used in melanin chemistry, namely Ni^{2+}/O_2 in HEPES buffer, pH 7.4, or horseradish peroxidase (HRP)/H_2O_2 in phosphate buffer, pH 7.4. Exposure of **1** to the above oxidizing systems resulted in the generation in both cases of a dark precipitate resembling DHI eumelanin. The UV–vis spectrum of a suspension of the precipitate from the HRP/H_2O_2 oxidation in phosphate buffer at pH 7 showed a broad maximum at 312 nm and a broad band absorption throughout the entire visible range (Supporting Information). These spectral features did not change over a time scale of hours, denoting stable components. A similar spectrum was obtained from a suspension of the precipitate

dx.doi.org/10.1021/jo200232t |*J. Org. Chem.* 2011, 76, 4457–4466

Table 1. Structures, Relative Free Energies, Transition Wavelengths, and Oscillator Strengths of the Most Stable Conformers/Tautomers of the Reduced and Two-Electron Oxidized Forms of 1[b]

Conformer/ tautomer	ΔG_{vac} / kcal/mol	ΔG_{wat} / kcal/mol	$\lambda_{max,\,vac}$ / nm (f)[a]	$\lambda_{max,\,wat}$ / nm (f)[a]
Reduced forms (QH_2)				
	**0.0**	**0.0**	280 (0.12), 273 (0.12)	281 (0.16), 274 (0.17)
	3.5	4.5	393 (0.02), 311 (0.12), 255 (0.52)	389 (0.03), 311 (0.18), 260 (0.64)
Two-electron oxidized forms (Q)				
	1.5	**0.0**	645 (0.03), 268 (0.18), 250 (0.13)	788 (0.02), 373 (0.03), 274 (0.29), 259 (0.19)
	5.4	11.5	940 (0.02), 300 (0.11)	934 (0.02), 302 (0.19), 251 (0.38)
	**0.0**	6.0	726 (0.04), 366 (0.18), 290 (0.09), 255 (0.12)	736 (0.05), 375 (0.26), 292 (0.12), 257 (0.17)
	0.4	2.0	462 (0.02), 322 (0.16), 260 (0.11), 247 (0.58), 223 (0.03)	524 (0.02), 317 (0.17), 279 (0.17), 262 (0.74), 223 (0.04)

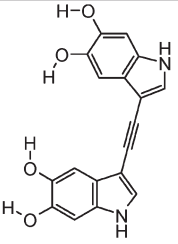
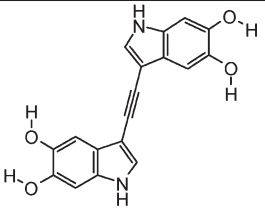
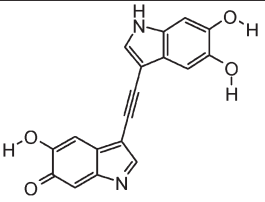
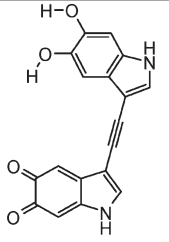
[a] Only transitions lying above 250 nm and with oscillator strengths larger than 0.02 are listed. [b] Structural parameters and electronic excitations were modeled at the PBE0/6-31+G(d,p) and TDPBE0/6-311++G(2d,2p) level, respectively, either in vacuo or in water (PCM).

in DMSO (Supporting Information). In both solvents, the absorption profiles were similar to those of typical eumelanins, supporting the polymeric nature of the material.[5,8] On this ground, it can be concluded that the absorption profile of the precipitate, although reflecting the features of the more soluble component(s), is representative of the whole material, as is the case with most eumelanins.[3g,8] TLC and HPLC analysis revealed the complete consumption of the starting material and the generation of a very complex mixture of products giving poor chromatograms (for representative TLC chromatograms see the Supporting Information). All attempts to isolate the main oxidation products by the usual reduction/extraction protocol for isolation of DHI oligomers[2a] met with failure.[12]

Oxidation of **2** under the above conditions proceeded at a slower rate to give likewise a dark brown material without isolable intermediates. The marked insolubility of the material prevented recording of meaningful UV−vis spectra from suspensions in neutral phosphate buffer or in DMSO (Supporting Information). Whereas the spectrum taken as suspension in phosphate buffer, pH 7, was featureless throughout the entire UV−vis range, the absorption profile in DMSO revealed solubilization of trace UV-absorbing components which, because of the lack of visible absorption and their selective solubility in the organic solvent, cannot be taken to be representative of the whole dark precipitate. Clearly, employment of the dark precipitates as functional materials requires that the insolubility problems are overcome. The FT-IR spectra of the dark insoluble materials from **1** and **2** resembled that of a DHI eumelanin (Supporting Information), displaying moreover detectable bands for the alkynyl groups (weak broad C≡C stretch bands at 2070 and 2160 cm^{-1} for **1**- and **2**-derived materials, respectively). A more detailed structural characterization of the dark eumelanin-like polymers from **1** and **2** was not pursued further due to the insolubility of the materials, preventing meaningful mass spectrometric analysis (MALDI) and size-exclusion chromatography, and the inadequacy of current analytical methodologies based on chemical degradation.[13]

Despite the disappointing, though not unexpected, failure to isolate oligomer products, the results of these oxidation experiments were informative enough as they demonstrated that 3-alkynyl-5,6-dihydroxyindoles **1** and **2** can undergo oxidative polymerization to afford dark eumelanin-like polymers. This finding was

Table 2. Structures, Inter-ring Torsion Angles, Transition Wavelengths, And Oscillator Strengths of the Most Stable Conformers/Tautomers of the Reduced and Two-Electron Oxidized Forms of 2[b]

Most stable conformer/tautomer	Medium	C2-C3-C3'-C2' torsion angle	λ_{max} / nm (f)[a]
Reduced forms (QH_2)			
	vacuo	77°	294 (0.11), 282 (0.09), 279 (0.08), 274 (0.20), 272 (0.06)
	water	160°	319 (0.48), 281 (0.75), 277 (0.09)
Two-electron oxidized forms (Q)			
	vacuo	0°	775 (0.18), 557 (0.05), 491 (0.26), 368 (0.09), 306 (0.22), 286 (0.22), 275 (0.52), 273 (0.06), 258 (0.03)
	water	13°	316 (0.78), 295 (0.04), 290 (0.04), 282 (0.16), 270 (0.15), 267 (0.16)

[a] Only transitions lying above 250 nm and with oscillator strengths larger than 0.02 are listed. [b] Structural parameters and electronic excitations were modeled at the PBE0/6-31+G(d,p) and TDPBE0/6-311++G(2d,2p) level, respectively, either in vacuo or in water (PCM).

not obvious given substitution and extended electron delocalization at C3, two factors that were expected to lower the oxidation potential of the DHI system and, concomitantly, to hinder or prevent oxidative coupling at the reactive pyrrole sector. On the contrary, these results would encourage further pursuit of rationally engineered alkynyl-containing eumelanins for structure−property-function purposes.

To ease interpretation of the oxidation chemistry of **1** and **2**, a systematic density functional theory (DFT) investigation of tautomerism and redox equilibria in **1** and **2** was undertaken. Data in Table 1 show that both in vacuo and in water (polarizable continuum model, PCM[14]) the ethynyl form of **1** is more stable than the allene tautomer. A similar trend is observed in the case of **1**-quinone, where two different alkyne-containing species, the quinonemethide and the *o*-quinone tautomers of 5,6-indolequinone, are the most stable in vacuo and in water, respectively. However, in this case the free energy separations among the tautomers tend to be smaller (e.g., in vacuo the quinone and allene forms are within 1.5 kcal/mol from the quinonemethide). An exploration extended to other solvents (Supporting Information) showed that in heptane the *o*-quinone and the allene forms of **1**-quinone become essentially isoenergetic, while in acetone the ordering of the tautomers is predicted to be quite similar to that in water.

Because of the relevance of 5,6-indolequinone absorption features to eumelanin optical properties and the notorious difficulties to detect quinone intermediates during DHI polymerization,[2b,3c] the influence of the conjugated triple bond on the 5,6-indolequinone chromophore was investigated by estimating the UV−vis spectrum of the most stable tautomers of **1**-quinone by a TDDFT approach; calculations were performed both in vacuo and in water (PCM). For comparison, the spectral features of the reduced forms were also estimated. Satisfactory spectral simulations for 5,6-indolequinone tautomers and derivatives have recently been obtained by this approach.[2b,3c,15] The results in Table 1 indicated a negligible influence of the 3-ethynyl group on the 5,6-dihydroxyindole chromophore (for DHI in EtOH, λ_{max} = 275, 302 nm).[2a] However, calculated maxima for the most

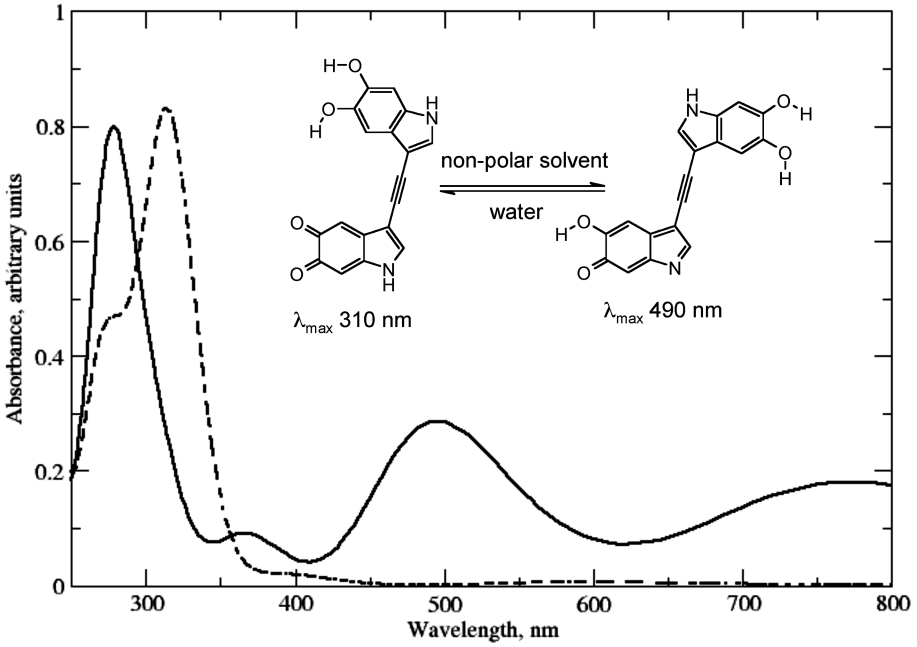


Figure 1. Computed UV spectra of 2-quinone tautomers: solid line, quinonemethide, *anti*, C_s (most stable species in vacuo); dashed line, quinone, *syn* (most stable species in water). (Inset) 2-Quinone as a theoretically predicted prototype of novel chromophores: solvent- and conformation-dependent effects on the visible absorption properties.

stable tautomers of **1**-quinone disclosed a significant interaction of the triple bond with the π-electron systems of 5,6-indolequinone (transition wavelengths computed in vacuo, 289, 259, 237 nm)[15a] and its quinonemethide tautomer (318, 291, 236 nm),[15a] resulting in the latter case in a ca. 50 nm bathochromic shift. Determination of the free energy changes for the most stable species in the equation: **1** + DHI-Q → **1**-Q + DHI gave ΔG = 0.3 kcal/mol in vacuo and 2.3 kcal/mol in water, indicating that the 3-alkynyl substituent stabilizes the 5,6-dihydroxyindole system toward oxidation. Investigation of the main tautomers of the reduced (QH_2) and two-electron oxidized (Q) forms of **2** gave the results reported in Table 2 (for more details, see the Supporting Information).

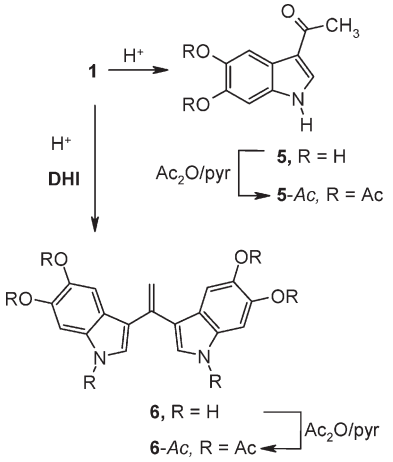
The alkyne tautomer in a twisted conformation proved to be the most stable structure for the reduced form. However, rotation of the aromatic substituents around the connecting triple bond is characterized by very small barriers, especially in water. Notably, the alkyne form is still the most stable tautomer for the corresponding two-electron oxidation product. For this latter, localized quinonemethide and *o*-quinone structures with planar conformations were predicted to be the most stable in vacuo and in water, respectively, whereas allene-containing structures were disfavored by several kcal/mol. While the computed rotational barriers remain quite low for the *o*-quinone form, they increase significantly in the quinonemethide (3.0 kcal/mol in vacuo, and 4.6 kcal/mol in water; several relaxed potential energy scans are reported in the Supporting Information). Computational explorations extended to other solvents showed that in heptane the quinonemethide tautomer remains the most stable one, as in vacuo, while in acetone the *o*-quinone form is predicted to be marginally more stable (by less than 1 kcal/mol; see the Supporting Information for details). Since compound **2** and its oxidized forms were conceived as prototypes of novel molecular building blocks for eumelanin-type polymers, the absorption spectra of the most stable tautomers of **2**-quinone were computed (Table 2). Calculations indicated for the quinonemethide tautomer of **2**-quinone an intense band in the visible range centered around 490 nm, which was missing in the 5,6-indolequinone tautomer of **2**-quinone (Figure 1). On this basis, a significant solvatochromism was predicted for **2**-quinone due to the medium-controllable shift between the 5,6-indolequinone and the quinonemethide tautomers (Figure 1, inset). In the latter, the efficient delocalization of the nitrogen lone pair from the 5,6-dihydroxyindole ring (the donor D) to the quinonemethide system of the oxidized unit (the acceptor A) through the π-bridging triple bond would allow for the development of a D-π-A-like system within the coplanar *syn* or *anti* conformations. On this basis, **2**-quinone would emerge as a prototype of a novel controllable push−pull electronic system.

A requisite for the implementation of switchable chromophoric systems based on **2**, currently underway in our laboratories, is the inhibition of oxidative polymerization (melanization) processes through proper substitution at the reactive DHI positions. Comparison of the free energies for the equation **2** + DHI-Q → **2**-Q + DHI gave ΔG = 0.2 kcal/mol in vacuo and 0.6 kcal/mol in water, confirming the modest stabilizing effect of the triple bond on DHI oxidation.

Acid-Mediated Chemistry. Preliminary experiments showed that **1** is unstable throughout the entire acidic pH range and undergoes a remarkably rapid decay even when left in mildly acidic aqueous medium. Complete consumption of compound **1** was observed after ca. 10 min (HPLC and TLC evidence) in 0.01 M phosphate buffer at pH 3, whereas under the same conditions the parent DHI was converted only to a small extent (<10%). Acid-induced consumption of **1** was also observed in 0.05 M acetate buffer at pH 4 and in 0.1 M HCl. In 0.01 M phosphate buffer at pH 7 or 12, compound **1** proved to be stable for more than 1 h, provided that oxygen was rigorously excluded. Since in 0.1 M HCl the parent DHI decayed at much faster rate giving typical acid-induced dimerization and oligomerization

dx.doi.org/10.1021/jo200232t |*J. Org. Chem.* 2011, 76, 4457–4466

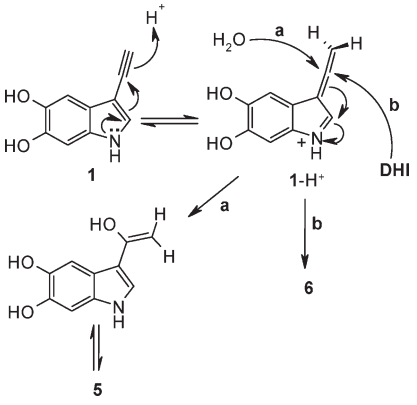
Scheme 2. Reactivity of 1 under Acidic Conditions



products, as previously reported,[7a] 0.01 M phosphate buffer at pH 3 was selected as a convenient medium for product investigation to avoid undesired side reactions when DHI was added as cosubstrate (see below). TLC analysis of the ethyl acetate extractable fraction revealed the formation under such conditions of a main product which was isolated after acetylation and subjected to spectral analysis. The ESI(+) mass spectrum indicated a pseudomolecular ion peak ($[M + H]^+$) at m/z 276 whereas the ^{1}H NMR spectrum displayed a singlet in the aliphatic proton region at δ 2.39 (3H) correlating in the ^{1}H,^{13}C HMBC spectrum with a carbon signal at δ 195.1 indicating an acetyl group. On this basis, the compound was readily identified as 5,6-diacetoxy-3-acetylindole (**5**-Ac) (Scheme 2).

To the best of our knowledge, alkyne−ketone conversions under very mild acidic conditions and in the absence of added catalysts are unprecedented. In marked contrast to **1**, compound **2** did not exhibit significant reactivity in acidic buffer. On this basis we returned to compound **1** to assess the scope of the acid-mediated chemistry for synthetic transformations. It seemed of interest, in particular, to investigate the susceptibility of the triple bond to nucleophilic attack by DHI under acidic conditions, since the availability of cross-conjugated, ethylidene-spaced bisindoles was desirable for preparing novel molecular scaffolds and to probe π-conjugation effects under polymerization conditions. When compound **1** was reacted in 0.1 M phosphate buffer, pH 3, in the presence of 1.5 molar equiv of DHI, the starting material was rapidly consumed and a main product was detected after a few minutes by TLC. This product was isolated after acetylation and was identified as the desired 3,3′-ethenylidene-bis-5,6-diacetoxyindole (**6**-*Ac*) (Scheme 2). The structural assignment was supported by complete spectral characterization, including (a) the ESI(+) mass spectrum, showing a pseudomolecular ion peak ($[M + Na]^+$) at m/z 597; (b) the ^{1}H NMR spectrum, showing a 2H singlet at δ 5.74 and the resonances for three protons framed into a 3-substituted indole ring; (c) the presence of 10 sp^2 carbon resonances (excluding the acetyl groups), consistent with the high molecular symmetry; and (d) a distinct one-bond correlation in the ^{1}H,^{13}C HSQC spectrum of the singlet at δ 5.74 with a carbon resonance at δ 115.8, compatible with an olefinic methylene group. The isolation of **6** under mild acid conditions would suggest an expeditious approach to the cross-conjugated 3,3′-ethenylidenebisindole skeleton for which there is no precedent in the chemistry of ethynylindoles.[16]

Scheme 3. Proposed Mechanisms for the Acid-Induced Conversion of 1 to 5 and 6



It appears that the indole nitrogen exerts a strong activating effect on the triple-bond-directing protonation toward the β position of the ethynyl moiety. Formation of the 3-acetyl derivative **5** conceivably proceeds via an alkyne hydration pathway, which would be triggered by initial protonation of triple bond followed by nucleophilic addition of water, restoring the aromaticity of the pyrrole moiety, and tautomerism (Scheme 3).

Product **6** would result from nucleophilic attack of DHI to the 3-position of the protonated intermediate **1**-H$^+$. Operation of an alternate mechanism involving nucleophilic attack of DHI to **5** followed by dehydration was ruled out by the observation that **5** was inert to acids in the presence of DHI and that no trace of **5** was detected in the reaction mixture of **1** and DHI. This latter observation, though not conclusive, would suggest that route b in Scheme 3 competes successfully with route a, on account of a significant nucleophilicity of DHI even in acidic medium. It is known that DHI reacts preferentially in acids via the 3-position, although at neutral or slightly alkaline pH the influence of the 6-OH group directs attack through the 2-position.[7d] In the case of **2**, failure of acids to induce reactivity can be imputed to the inability of the protonated intermediate from **2** to evolve to products. It is possible that this is due to the electron-donating effects of the second indole ring decreasing electrophilic reactivity toward nucleophiles. To verify the mechanism proposed in Scheme 3, the most stable protonated forms of DHI and **1** were investigated by DFT. Data in Figure 2 show that the allene is indeed the most stable protonated form of **1** and **1**-quinone, though in the latter case the *O*-5-protonated alkyne is close in energy, differing only by 0.2 kcal/mol.

Overall, these calculations would support the mechanism proposed in Scheme 3 for the acid-mediated chemistry of **1** and formation of **5** and **6**. In Scheme 3 it is proposed that the most stable cation derived from **1** is present on the reaction pathway. Although other protonated forms of **1** may be involved, only the stable cation derived from β protonation of the ethynyl moiety would direct nucleophilic addition to the α position. Further computational explorations (see the Supporting Information) showed that, once protonation of **1** has taken place as described above, nucleophilic attack to the α position should occur easily; thus, a barrier of ca. 6 kcal/mol is predicted for the reaction step in which attack of water to **1**-H$^+$ leads to the *O*-protonated enolic form of **5**. Protonation of DHI is favored at the 3-position, consistent with previous experimental observations, but protonation of 5,6-indolequinone is directed mainly to the *O*-5 center as

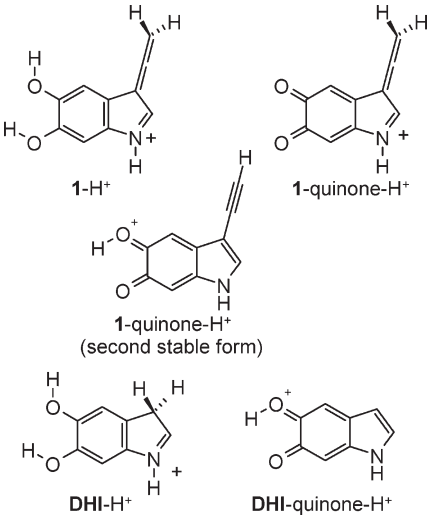


Figure 2. Structures of the main conformers/tautomers of the reduced (QH_2) and two-electron oxidized (Q) protonated forms of 1 and DHI, computed in water (PCM) at the PBE0/6-31+G(d,p) level.

a result of long-range activation by the NH group. Comparison of the protonation free energies for the most stable species of **1** and DHI, both in the reduced (QH_2) and two-electron oxidized (Q) stages, suggested a thermodynamically favored protonation for **1**. Thus, for the equation **1** + DHI-H$^+$ → **1**-H$^+$ + DHI, $\Delta G = -5.7$ kcal/mol (in water). This indicates that **1** is about 10000 times more basic than DHI. Interestingly, an opposite behavior is noted when the same equilibrium is considered at the two-electron (quinone) oxidation stage. Thus, for **1**-quinone + DHI-quinone-H$^+$ → **1**-quinone-H$^+$ + DHI-quinone, ΔG = 1.7 kcal/mol. These results consistently supported the marked reactivity of **1** in acids and disclosed the dichotomous influence of the triple bond at C3 on the acid-induced equilibria, activating the reduced form toward protonation but deactivating the corresponding quinone.

CONCLUSIONS AND PERSPECTIVES

Indoles occupy a privileged position in natural products, pharmaceuticals, and material sciences. As a consequence, methods to synthesize, functionalize, and control the properties of these heterocycles are of utmost importance in organic chemistry both for academic and application purposes. Herein, we have successfully developed the chemistry of the 3-alkynyl-5,6-dihydroxyindoles **1** and **2**, useful leads for a new class of DHI derivatives endowed with external π-conjugated groups amenable to manipulation under oxidative or mildly acidic conditions in completely aqueous environments. The main conceptual and practical outcomes of the study can be summarized as follows: (1) the elucidation of the efficient electron-releasing effect from the enamine-type nitrogen to the alkynyl sector, which was rationalized by DFT calculations and attested by the fast conversion of **1** to the cross-conjugated biindole derivative **6** under mild acidic conditions, an unprecedented transformation in indole chemistry, and (2) the theoretical prediction and rationalization of a medium- and conformation-dependent chromophoric motif from **2**, which might be the paradigm for novel switchable D-π-A electronic systems. A key requisite of the new chromophore is that one indole unit must be in the quinone state, implying chromophore activation by oxidative "turn on" of **2**.

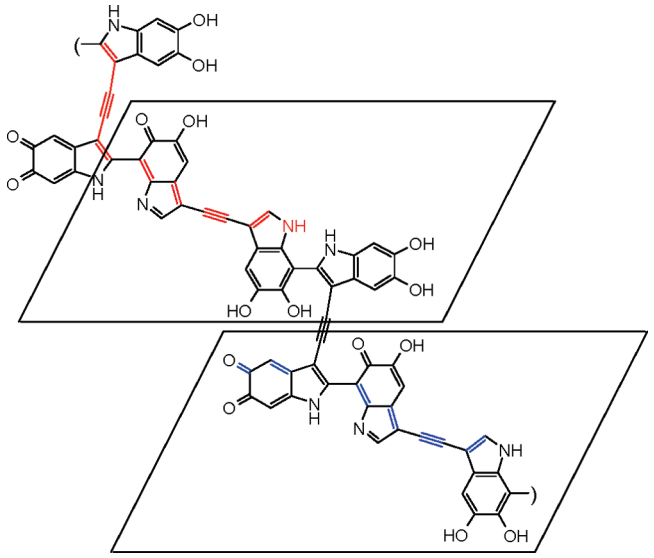


Figure 3. Schematic representation of interunit interactions in theoretically designed ethynylene-containing hybrid eumelanin polymers based on **2**. π-Electron delocalization over extended conjugated sectors and free access to locally planar conformations enabling efficient stacking interactions are highlighted.

Conformational equilibria and medium-dependent tautomerism would then govern the switch from UV to visible absorption, reflecting a 180 nm shift predicted for accessibility of an extended push−pull communication between the electron-donating ethynylene function and the *o*-dioxygenated functionality in the quinone methide moiety. These results would also underscore the potential of the alkynyl structure for probing the π-orbital system of DHI and for implementing tunable π-conjugated polymers whose properties can be externally controlled through protonation and/or alteration of the redox state perturbing the local aromatic character within π-bridged units. The idea developed herein of tuning electron properties of melanin building blocks through alkynyl spacers and to probe the ability of rigid triple bond bridges to mediate interunit electronic communications by relieving steric inhibition to conformational mobility (Figure 3) has manifold conceptual and practical implications. Understanding how these variables impact intramolecular and intermolecular electron conductivity and stacked interactions in supramolecular assemblies will allow us to engineer and evaluate a range of innovative eumelanin-inspired functional polymers and scaffolds of possible technological relevance.

EXPERIMENTAL SECTION

Materials and Methods. All commercially available reagents were used as received. Anhydrous solvents were purchased from commercial sources and withdrawn from the container by a syringe under a slight positive pressure of argon. All the solvents were of analytical grade quality. Compounds **3** and 5,6-dihydroxyindole (DHI) were prepared according to reported procedures.[2b,17] HEPES buffer (0.1 M), pH 7.4, and 0.1 M phosphate buffers, pH 7.4 and 3, were treated with Chelex-100 resin before use to remove transition-metal contaminants. UV and FT-IR spectra were performed using a diode array and an FT-IR spectrophotometer, respectively. NMR spectra were recorded with 200 and 400 MHz instruments. ^{1}H and ^{13}C NMR spectra were recorded in $CDCl_3$ using TMS as the internal standard. ^{1}H,^{13}C HSQC-DEPT and ^{1}H,^{13}C HMBC experiments were run at 400.1 MHz using standard pulse

dx.doi.org/10.1021/jo200232t |*J. Org. Chem.* 2011, 76, 4457–4466

programs. Mass spectra were registered in the electrospray ionization−positive-ion (ESI+) mode. ESI analyses were performed with the cone and the fragmentator voltages set at 4 kV and 80 V, respectively; nitrogen was used as carrier gas at a flow of 8 mL/min, and the nebulizer pressure was set at 50 psi. High-resolution mass spectra were registered in the electrospray ionization−positive-ion (ESI+) mode. Analytical and preparative TLC analyses were performed on F_{254} silica gel plates (0.25 and 0.5 mm, respectively). Liquid chromatography was performed on silica gel (60−230 mesh). HPLC analyses were carried out on an HPLC apparatus coupled with a UV−vis detector set at 280 nm. Analytical HPLC runs were performed on an octadecylsilane-coated column (Sphereclone C18-4.6 × 250 mm, 5 μm) using the following elution conditions: water/acetonitrile 40:60 (v/v); flow rates of 1 mL/min were used.

Computational Methods. All calculations were performed with the Gaussian package of programs.[18] Geometry optimizations and frequency calculations were carried out at the DFT level of theory using the PBE0 functional with the 6-31+G(d,p) basis set.[19] The PBE0 (also referred to as PBE1PBE) is a hybrid functional obtained by combining a predetermined amount of exact exchange with the Perdew−Burke−Ernzerhof exchange and correlation functionals.[20] In a series of calculations, the influence of the aqueous medium was accounted for by the polarizable continuum model (PCM);[14] a scaled van der Waals cavity based on universal force field (UFF) radii[21] was used, and polarization charges were modeled by spherical Gaussian functions.[22] For comparison, single-point energies in water were also evaluated by the SMD solvation model.[23] Electronic absorption spectra of significant tautomers were estimated on the basis of excitation energy calculations using the TDDFT approach.[24] The PBE0 functional with the large 6-311++G(2d,2p) basis set was used for TDDFT computations. As the absorption process presents a short characteristic time, only the electronic distribution of the solvent can adapt to the excited state electronic structure; therefore, the appropriate nonequilibrium formulation of the PCM[25] was employed. To produce graphs of computed UV−vis spectra, transitions above 220 nm were selected, and an arbitrary Gaussian line width of 20 nm was imposed. To produce graphs, transitions below 5.6 eV were selected, and an arbitrary Gaussian line width of 0.25 eV was imposed; the spectra were finally converted to a wavelength scale.

Synthesis of 1-Acetyl-5,6-diacetoxy-3-(trimethylsilylethynyl)-indole (4). A solution of **3** (700 mg, 2.0 mmol) in toluene (16.2 mL) was treated under stirring and an argon atmosphere with acetic anhydride (840 μL) and *N*,*N*-dimethylaminopyridine (210 mg, 1.7 mmol). After 5 min, triethylamine (16.2 mL), PPh_3 (51.3 mg, 0.2 mmol), CuI (37.8 mg, 0.2 mmol), $(PPh_3)_2PdCl_2$ (68.4 mg, 0.1 mmol), and trimethylsilylacetylene (1.4 mL, 10 mmol) were added to the reaction mixture, and the temperature was raised to 60 °C. After 30 min, the reaction mixture was extracted with a 10% water solution of NH_4Cl and toluene. The organic layers were collected, dried over anhydrous sodium sulfate, and evaporated under reduced pressure, and the residue was fractionated on silica gel (eluant: cyclohexane/ethyl acetate 8:2) to afford pure **4** (593 mg, 80%, R_f = 0.56 eluant: cyclohexane/ethyl acetate 1:1 (v/v)).

4: ^{1}H NMR (200 MHz, $CDCl_3$) δ (ppm) 0.27 (3 × 3H, s, $Si(CH_3)_3$), 2.33 (2 × 3H, s, 2 × $OCOCH_3$), 2.60 (3H, s, $NCOCH_3$), 7.44 (1H, s, H-4), 7.63 (1H, s, H-2), 8.29 (1H, s, H-7); ^{13}C NMR (50 MHz, $CDCl_3$) δ (ppm) 0.05 (−$Si(CH_3)_3$), 20.6 (2 × $OCOCH_3$), 23.5 ($NCOCH_3$), 95.3 (−*C*≡CSi$(CH_3)_3$), 99.6 (−C≡*C*Si$(CH_3)_3$), 105.2 (C-3), 111.9 (C-7), 114.0 (C-4), 128.4 (C-9), 129.5 (C-2), 132.1 (C-8), 139.6 (C-5), 140.8 (C-6), 167.9 ($NCOCH_3$), 168.6 (2 × $OCOCH_3$); HRMS (ESI) *m*/*z* $C_{19}H_{22}NO_5Si$ $[M + H]^+$ calcd 372.1267, found 372.1270.

Synthesis of 1-Acetyl-5,6-diacetoxy-3-ethynylindole (1-*Ac*). A solution of **4** (600 mg, 1.6 mmol) in DMF (14.8 mL) was treated with KF (139.2 mg, 2.20 mmol) under vigorous stirring. After 1 h, the reaction mixture was extracted with water and toluene. The organic layers were collected, dried over anhydrous sodium sulfate and evaporated under reduced pressure to afford pure **1**-*Ac* (344 mg, 72%, R_f = 0.68 eluant: chloroform/ethyl acetate 1:1 (v/v)).

1-*Ac*: UV−vis (CH_3OH) λ 245 (sh), 278, 294, 304 nm; ^{1}H NMR (200 MHz, $CDCl_3$) δ (ppm) 2.32 (2 × 3H, s, 2 × $OCOCH_3$), 2.61 (3H, s, $NCOCH_3$), 3.25 (1H, s, −C≡CH), 7.49 (1H, s, H-4), 7.65 (1H, s, H-2), 8.29 (1H, s, H-7); ^{13}C NMR (50 MHz, $CDCl_3$) δ (ppm) 20.6 (2 × $OCOCH_3$), 23.6 ($NCOCH_3$), 74.5 (−*C*≡CH), 81.8 (−C≡*C*H), 104.0 (C-3), 111.9 (C-7), 113.9 (C-4), 128.3 (C-9), 129.9 (C-2), 132.0 (C-8), 139.6 (C-5), 140.8 (C-6), 167.8 ($NCOCH_3$), 168.5 (2 × $OCOCH_3$); HRMS (ESI) *m*/*z* $C_{16}H_{14}NO_5$ $[M + H]^+$ calcd 300.0872, found 300.0869.

Synthesis of 3,3′-(1,2-ethynediyl)bis-5,6-dihydroxyindole (2-*Ac*). A solution of **3** (400 mg, 1.12 mmol) in toluene (9.8 mL) was treated in an argon atmosphere under stirring with acetic anhydride (484 μL) and *N*,*N*- dimethylaminopyridine (122 mg, 1 mmol). After 5 min, triethylamine (9.8 mL), PPh_3 (29.2 mg, 0.11 mmol), CuI (22 mg, 0.11 mmol), $(PPh_3)_2PdCl_2$ (40 mg, 0.056 mmol), and **1**-*Ac* (334 mg, 1.12 mmol) were added to the reaction mixture that was then heated to 60 °C. After 30 min, the reaction mixture was filtered and the solid washed with toluene and extracted with chloroform and a 10% water solution of NH_4Cl. The organic layers were collected, dried over anhydrous sodium sulfate, and evaporated under reduced pressure to afford pure **2**-*Ac* (608 mg, 87%, R_f = 0.82 eluant: cyclohexane/ethyl acetate 2:8 (v/v)).

2-*Ac*: ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 2.33 (4 × 3H, s, 4 × $OCOCH_3$), 2.65 (2 × 3H, s, 2 × $NCOCH_3$), 7.55 (2 × 1H, s, H-4, H-4″), 7.71 (2 × 1H, s, H-2, H-2″), 8.33 (2 × 1H, s, H-7, H-7″); ^{13}C NMR (50 MHz, $CDCl_3$) δ (ppm) 20.6 (4 × $OCOCH_3$), 23.6 (2 × $NCOCH_3$), 84.4 (−*C*≡C−), 104.7 (C-3, C-3″), 112.0 (C-7, C-7″), 114.0 (C-4, C-4″), 128.3 (C-9, C-9″), 129.2 (C-2, C-2″), 132.2 (C-8, C-8″), 139.7 (C-5, C-5”), 140.9 (C-6, C-6″), 167.9 (2 × $NCOCH_3$), 168.6 (4 × $OCOCH_3$); ESI(+)-MS *m*/*z* 573 ($[M + H]^+$); HRMS (ESI) *m*/*z* $C_{30}H_{25}N_2O_{10}$ $[M + H]^+$ calcd 573.1509, found 573.1512.

Oxidation Reaction of Alkynyl Derivatives: General Procedure. A 0.01 M solution of **1**-*Ac* or **2**-*Ac* in methanol was degassed with an argon flux and then treated with sodium *tert*-butoxide (2 molar equiv in the case of **1**-*Ac*, 4 molar equiv in the case of **2**-*Ac*). After 10 min, the reaction mixture was acidified to pH 3−4 with acetic acid and treated as follows:

- In a first group of experiments, the reaction mixture was taken in 0.1 M HEPES buffer, pH 7.4 (3.5×10^{-3} M), and treated under vigorous stirring with 2 molar equiv of $NiSO_4 \cdot 7H_2O$ under an oxygen flux.
- In a second group of experiments, the reaction mixture was taken in 0.1 M phosphate buffer, pH 7.4 (2×10^{-3} M), and treated with horseradish peroxidase (20 U/mL) and H_2O_2 (2 molar equiv in the case of **1**-*Ac* and 4 molar equiv in the case of **2**-*Ac*).

At regular time intervals, aliquots of the reaction mixtures were reduced with sodium dithionite, acidified to pH 3−4 with 3 M HCl, and extracted with ethyl acetate. The organic layers were collected, dried over anhydrous sodium sulfate, evaporated under reduced pressure, and acetylated with acetic anhydride and pyridine. The resulting mixtures were analyzed by TLC (eluant: chloroform/ethyl acetate 1:1 (v/v) for **1**-*Ac*; cyclohexane/ethyl acetate 2:8 (v/v) for **2**-*Ac*) and HPLC. In separate experiments, the oxidation reactions were carried out with the horseradish peroxidase/H_2O_2 system as described previously for a time period of 2 h. Then the reaction mixtures were centrifuged (5000 rpm) for 30 min at 4 °C; the supernatant was discarded, and the precipitate was washed twice with water, dried under reduced pressure, and analyzed by UV−vis and FT-IR spectroscopy.

Acid-Mediated Chemistry of 1-*Ac* and 2-*Ac*: General Procedure. The appropriate compound was dissolved in methanol (0.022 M) and deacetylated with sodium *tert*-butoxide as reported above. After 10 min, the reaction mixtures were acidified to pH 3−4 with acetic acid and taken up in 0.1 M phosphate buffer, pH 3 (0.01 M final concentration). At regular time intervals, aliquots of the reaction mixtures were extracted

with ethyl acetate and the organic layers collected, dried over anhydrous sodium sulfate, and evaporated under reduced pressure. The residues were treated with acetic anhydride (1 mL) and pyridine (50 μL) and the acetylated mixtures analyzed by TLC (eluant: chloroform/ethyl acetate 1:1 (v/v), for **1**-*Ac*, cyclohexane/ethyl acetate 2:8 (v/v) for **2**-*Ac*). In separate experiments, the reaction was carried out as described before by using 0.1 M HCl, 0.05 M acetate buffer pH 4, and 0.01 M phosphate buffer pH 7 or 12 instead of 0.01 M phosphate buffer pH 3.

Isolation of 3-Acetyl-5,6-diacetoxyindole (5-*Ac*). A solution of **1**-*Ac* (100 mg, 0.34 mmol) in methanol (15 mL), previously degassed with an argon flux for 10 min, was treated under stirring with sodium *tert*-butoxide (68 mg, 0.68 mmol). After 10 min, the reaction mixture was acidified to pH 3−4 with acetic acid and taken up in 0.1 M phosphate buffer, pH 3 (33 mL). After 10 min, the reaction mixture was extracted with ethyl acetate, and the organic layers were collected, dried over anhydrous sodium sulfate, and evaporated under reduced pressure. The residue was then treated with acetic anhydride (2 mL) and pyridine (100 μL). The acetylated mixture was evaporated under reduced pressure and the residue fractionated on silica gel (eluant: chloroform/ethyl acetate 4:6 (v/v)) to afford pure **5**-*Ac* (20 mg, 21%, R_f = 0.33 eluant: chloroform/ethyl acetate 4:6 (v/v)).

5-*Ac*: ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 2.35 (2 × 3H, s, $OCOCH_3$), 2.38 (3H, s, $COCH_3$), 7.04 (1H, s, H-7), 7.48 (1H, d J = 2.8 Hz, H-2), 7.97 (1H, s, H-4), 8.84 (1H, bs, NH); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 20.6 ($OCOCH_3$), 20.8 ($OCOCH_3$), 27.0 ($COCH_3$), 106.7 (C-7), 115.8 (C-4), 133.5 (C-3), 133.9 (C-2), 134.6 (C-9), 137.7 (C-8), 138.5 (C-5), 139.3 (C-6), 170.0 ($OCOCH_3$), 170.2 ($OCOCH_3$), 193.5 ($COCH_3$); ESI (+)-MS m/z 276 ($[M + H]^+$); HRMS (ESI) m/z $C_{14}H_{14}NO_5$ $[M + H]^+$ calcd 276.0872, found 276.0870.

Isolation of 3,3′-Ethenylidenebis-5,6-diacetoxyindole (6-*Ac*). Compound **1**-*Ac* (100 mg, 0.34 mmol) was deacetylated and taken up in 0.1 M phosphate buffer, pH 3, as described above and then treated with DHI (74 mg, 0.5 mmol) under vigorous stirring. After 10 min, the reaction mixture was extracted with ethyl acetate and the organic layers were collected, dried over anhydrous sodium sulfate, and evaporated under reduced pressure. The residue was then treated with acetic anhydride (2 mL) and pyridine (100 μL). The acetylated mixture was evaporated under reduced pressure and the residue fractionated on silica gel (eluant: chloroform/ethyl acetate 1:1 (v/v)) to afford pure **6**-*Ac* (36 mg, 18%, R_f = 0.66 eluant: chloroform/ethyl acetate 1:1 (v/v)).

6-*Ac*: ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 2.29 (2 × 3H, s, 2 × $OCOCH_3$), 2.34 (2 × 3H, s, 2 × $OCOCH_3$), 2.57 (2 × 3H, s, 2 × $NCOCH_3$), 5.74 (2H, s, $-C{=}CH_2$), 7.39 (2 × 2H, s, H-2, H-2″, H-4, H-4″), 8.40 (2 × 1H, s, H-7, H-7″); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm) 20.9 (4 × $OCOCH_3$), 23.7 (2 × $NCOCH_3$), 111.6 (C-7, C-7″), 114.3 (C-4, C-4″), 116.3 ($=CH_2$), 122.4 (C-3, C-3″, $C{=}CH_2$), 125.1 (C-2, C-2″), 126.3 (C-9, C-9″), 133.1 (C-8, C-8″), 139.4 (C-6, C-6″), 139.9 (C-5, C-5″), 168.3 ($NCOCH_3$), 168.4 ($OCOCH_3$); ESI(+)-MS m/z 597 ($[M + Na]^+$); HRMS (ESI) m/z $C_{30}H_{26}NaN_2O_{10}$ $[M + Na]^+$ calcd 597.1485, found 597.1483.

■ ASSOCIATED CONTENT

Supporting Information. Mono- and bidimensional NMR spectra for compounds 1-*Ac*, 2-*Ac*, 4, 5-*Ac*, and 6-*Ac* are provided as well as UV−vis and FT-IR spectra of polymers from **1** and **2** and the results of computational calculations carried out on **1**, **2**, and DHI. This material is available free of charge via the Internet at http://pubs.acs.org.

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■ ACKNOWLEDGMENT

We acknowledge financial support by MIUR (Italy), PRIN 2008 project. This work is in the frame of the EuMelaNet program (http://www.espcr.org/eumelanet/).

■ REFERENCES

(1) (a) Simon, J. D.; Peles, D. N. *Acc. Chem. Res.* **2010**, *43*, 1452–1460. (b) d'Ischia, M.; Napolitano, A.; Pezzella, A.; Meredith, P.; Sarna, T. *Angew. Chem., Int. Ed.* **2009**, *48*, 3914–3921. (c) Simon, J. D.; Peles, D.; Wakamatsu, K.; Ito, S. *Pigment Cell Melanoma Res.* **2009**, *22*, 563–579.

(2) (a) d'Ischia, M.; Napolitano, A.; Pezzella, A.; Land, E. J.; Ramsden, C. A.; Riley, P. A. *Adv. Heterocycl. Chem.* **2005**, *89*, 1–63. (b) Pezzella, A.; Crescenzi, O.; Natangelo, A.; Panzella, L.; Napolitano, A.; Navaratnam, S.; Edge, R.; Land, E. J.; Barone, V.; d'Ischia, M. *J. Org. Chem.* **2007**, *72*, 1595–1603. (c) Napolitano, A.; Palumbo, A.; d'Ischia, M.; Prota, G. *J. Med. Chem.* **1996**, *39*, 5192–5201.

(3) (a) Pezzella, A.; Panzella, L.; Natangelo, A.; Arzillo, M.; Napolitano, A.; d'Ischia, M. *J. Org. Chem.* **2007**, *72*, 9225–9230. (b) Panzella, L.; Pezzella, A.; Napolitano, A.; d'Ischia, M. *Org. Lett.* **2007**, *9*, 1411–1414. (c) Pezzella, A.; Panzella, L.; Crescenzi, O.; Napolitano, A.; Navaratnam, S.; Edge, R.; Land, E. J.; Barone, V.; d'Ischia, M. *J. Am. Chem. Soc.* **2006**, *128*, 15490–11221. (d) d'Ischia, M.; Crescenzi, O.; Pezzella, A.; Arzillo, M.; Panzella, L.; Napolitano, A.; Barone, V. *Photochem. Photobiol.* **2008**, *84*, 600–607. (e) Okuda, H.; Wakamatsu, K.; Ito, S.; Sota, T. *J. Phys. Chem. A* **2008**, *112*, 11213–11222. (f) Okuda, H.; Wakamatsu, K.; Ito, S.; Sota, T. *Chem. Phys. Lett.* **2010**, *490*, 226–229. (g) Pezzella, A.; d'Ischia, M.; Napolitano, A.; Palumbo, A.; Prota, G. *Tetrahedron* **1997**, *53*, 8281–8286.

(4) Watt, A. A. R.; Bothma, J. P.; Meredith, P. *Soft Matter* **2009**, *5*, 3754–3760.

(5) Meredith, P.; Sarna, T. *Pigment Cell Res.* **2006**, *19*, 572–594.

(6) (a) Tran, M. L.; Powell, B. J.; Meredith, P. *Biophys. J.* **2006**, *90*, 743. (b) Schweitzer, A. D.; Howell, R. C.; Jiang, Z.; Bryan, R. A.; Gerfen, G.; Chen, C.-C.; Mah, D.; Cahill, S.; Casadevall, A.; Dadachova, E. *PLoS ONE* **2009**, *4*, e7229. (c) Ambrico, M.; Cardone, A.; Ligonzo, T.; Augelli, V.; Ambrico, P. F.; Cicco, S.; Farinola, G. M.; Filannino, M.; Perna, G.; Capozzi, V. *Org. Electron.* **2010**, *11*, 1809–1814. (d) Bothma, J. P.; de Boor, J.; Divakar, U.; Schwenu, P. E.; Meredith, P. *Adv. Mater.* **2008**, *20*, 3539–3542.

(7) (a) Panzella, L.; Pezzella, A.; Arzillo, M.; Manini, P.; Napolitano, A.; d'Ischia, M. *Tetrahedron* **2009**, *65*, 2032–2036. (b) Arzillo, M.; Pezzella, A.; Crescenzi, O.; Napolitano, A.; Land, E. J.; Barone, V.; d'Ischia, M. *Org. Lett.* **2010**, *12*, 3250–3253. (c) Pezzella, A.; Palma, A.; Iadonisi, A.; Napolitano, A.; d'Ischia, M. *Tetrahedron Lett.* **2007**, *48*, 3883–3886. (d) Manini, P.; Pezzella, A.; Panzella, L.; Napolitano, A.; d'Ischia, M. *Tetrahedron* **2005**, *61*, 4075–4080. (e) Manini, P.; d'Ischia, M.; Milosa, M.; Prota, G. *J. Org. Chem.* **1998**, *63*, 7002–7008.

(8) Pezzella, A.; Iadonisi, A.; Valerio, S.; Panzella, L.; Napolitano, A.; Adinolfi, M.; d'Ischia, M. *J. Am. Chem. Soc.* **2009**, *131*, 15270–15275.

(9) (a) Sonogashira, K.; Tohda, Y.; Hagihara, N. *Tetrahedron Lett.* **1975**, *16*, 4467–4470. (b) Chinchilla, R.; Najera, C. *Chem. Rev.* **2007**, *107*, 874–922.

(10) (a) Wang, C.; Bryce, M. R.; Gigon, J.; Ashwell, G. J.; Grace, I.; Lambert, C. J. *J. Org. Chem.* **2008**, *73*, 4810–4818. (b) Hauck, M.; Schönhaber, J.; Zucchero, A. J.; Hardcastle, K. I.; Müller, J. J.; Bunz, U. H. F. *J. Org. Chem.* **2007**, *72*, 6714–6725. (c) Ma, C.; Lo, A.; Abdolmaleki, A.; MacLachlan, M. J. *Org. Lett.* **2004**, *6*, 3841–3844. (d) Hsu, H.-F.; Lin, M.-C.; Lin, W.-C.; Lai, Y.-H.; Lin, S.-Y. *Chem. Mater.* **2003**, *15*, 2115–2118. (e) Silvestri, F.; Marrocchi, A.; Seri, M.; Kim, C.; Marks, T. J.; Facchetti, A.; Taticchi, A. *J. Am. Chem. Soc.* **2010**, *132*, 6108–6123. (f) Wang, C.; Palsson, L.-O.; Batsanov, A. S.; Bryce, M. R. *J. Am. Chem. Soc.* **2006**, *128*, 3789–3799.

(11) (a) Capelli, L.; Manini, P.; Pezzella, A.; Napolitano, A.; d'Ischia, M. *J. Org. Chem.* **2009**, *74*, 7191–7194. (b) Capelli, L.; Manini, P.; Pezzella, A.; d'Ischia, M. *Org. Biomol. Chem.* **2010**, *8*, 4243–4245.

(12) Failure to isolate intermediate oxidation products of **1** and **2** was attributed to the insolubility of the resulting oligomers, causing extraction only in trace amounts into organic solvents under the workup conditions preventing extraction into organic solvents under the usual workup conditions.

(13) (a) Panzella, L.; Manini, P.; Monfrecola, G.; d'Ischia, M.; Napolitano, A. *Pigment Cell Res.* **2006**, *20*, 128–133. (b) Ito, S.; Wakamatsu, K. *Pigment Cell Res.* **2003**, *16*, 523–531.

(14) (a) Miertus, S.; Scrocco, E.; Tomasi *J. Chem. Phys.* **1981**, *55*, 117–129. (b) Cossi, M.; Scalmani, G.; Rega, N.; Barone, V. *J. Chem. Phys.* **2002**, *117*, 43–54. (c) Scalmani, G.; Barone, V.; Kudin, K. N.; Pomelli, C. S.; Scuseria, G. E.; Frisch, M. J. *Theor. Chem. Acc.* **2004**, *111*, 90–100. (d) Tomasi, J.; Mennucci, B.; Cammi, R. *Chem. Rev.* **2005**, *105*, 2999–3093.

(15) (a) Il'ichev, Y. V.; Simon, J. D. *J. Phys. Chem. B* **2003**, *107*, 7162–7171. (b) Pezzella, A.; Panzella, L.; Crescenzi, O.; Napolitano, A.; Navaratnam, S.; Edge, R.; Land, E. J.; Barone, V.; d'Ischia, M. *J. Org. Chem.* **2009**, *74*, 3727–3734. (c) Stark, K. B.; Gallas, J. M.; Zajac, G. W.; Eisner, M.; Golab, J. T. *J. Phys. Chem. B* **2003**, *107*, 3061–3067.

(16) Tarshits, D. L.; Przhiyalgovskaya, N. M.; Buyanov, V. N.; Tarasov, S. Y. *Chem. Heterocycl. Compd.* **2009**, *45*, 501–523.

(17) Edge, R.; d'Ischia, M.; Land, E. J.; Napolitano, A.; Navaratnam, S.; Panzella, L.; Pezzella, A.; Ramsden, C. A.; Riley, P. A. *Pigment Cell Res.* **2006**, *19*, 443–450.

(18) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Scalmani, G.; Barone, V.; Mennucci, B.; Petersson, G. A.; Nakatsuji, H.; Caricato, M.; Li, X.; Hratchian, H. P.; Izmaylov, A. F.; Bloino, J.; Zheng, G.; Sonnenberg, J. L.; Hada, M.; Ehara, M.; Toyota, K.; Fukuda, R.; Hasegawa, J.; Ishida, M.; Nakajima, T.; Honda, Y.; Kitao, O.; Nakai, H.; Vreven, T.; Montgomery, J. A., Jr.; Peralta, J. E.; Ogliaro, F.; Bearpark, M.; Heyd, J. J.; Brothers, E.; Kudin, K. N.; Staroverov, V. N.; Kobayashi, R.; Normand, J.; Raghavachari, K.; Rendell, A.; Burant, J. C.; Iyengar, S. S.; Tomasi, J.; Cossi, M.; Rega, N.; Millam, J. M.; Klene, M.; Knox, J. E.; Cross, J. B.; Bakken, V.; Adamo, C.; Jaramillo, J.; Gomperts, R.; Stratmann, R. E.; Yazyev, O.; Austin, A. J.; Cammi, R.; Pomelli, C.; Ochterski, J. W.; Martin, R. L.; Morokuma, K.; Zakrzewski, V. G.; Voth, G. A.; Salvador, P.; Dannenberg, J. J.; Dapprich, S.; Daniels, A. D.; Farkas, O.; Foresman, J. B.; Ortiz, J. V.; Cioslowski, J.; Fox, D. J. *Gaussian 09, Revision A.02*; Gaussian, Inc.: Wallingford, CT, 2009.

(19) (a) Francl, M. M.; Petro, W. J.; Hehre, W. J. S.; Binkley, J.; Gordon, M. S.; DeFrees, D. J.; Pople, J. A. *J. Chem. Phys.* **1982**, *77*, 3654–3665.(b) For a general introduction to basis sets, see: Foresman, J. B.; Frisch, A. *Exploring Chemistry with Electronic Structure Methods*, 2nd ed.; Gaussian, Inc.: Pittsburgh, PA, 1996.

(20) Adamo, C.; Barone, V. *J. Chem. Phys.* **1999**, *110*, 6158–6170.

(21) Rappé, A. K.; Casewit, C. J.; Colwell, K. S.; Goddard, W. A., III; Skiff, W. M. *J. Am. Chem. Soc.* **1992**, *114*, 10024–10035.

(22) (a) York, D. A.; Karplus, M. *J. Phys. Chem. A* **1999**, *103*, 11060–11079. (b) Scalmani, G.; Frisch, M. J. *J. Chem. Phys.* **2009**, *132*, 114110.

(23) Marenich, A. V.; Cramer, C. J.; Truhlar, D. G. *J. Phys. Chem. B* **2009**, *113*, 6378–6396.

(24) (a) Stratmann, R. E.; Scuseria, G. E.; Frisch, M. J. *J. Chem. Phys.* **1998**, *109*, 8218–8224. (b) Bauernschmitt, R.; Ahlrichs, R. *Chem. Phys. Lett.* **1996**, *256*, 454–464. (c) Casida, M. E.; Jamorski, C.; Casida, K. C.; Salahub, D. R. *J. Chem. Phys.* **1998**, *108*, 4439–4449. (d) Adamo, C.; Scuseria, G. E.; Barone, V. *J. Chem. Phys.* **1999**, *111*, 2889–2899.

(25) Cossi, M.; Barone, V. *J. Chem. Phys.* **2001**, *115*, 4708–4717.