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A metal-free cascade reaction was developed for the synthesis of indolo[1,2-a]quinoline derivatives from arylsulfonyl hydrazides and 1-(2-(arylethynyl)phenyl)indolets in the presence of TBAI/TBHP. Impressively, these products exhibit excellent fluorescence properties, which is promising for cell imaging.

N-Heterocycles are structural elements of natural products, drugs and functional materials.¹ Among them, indolo[1,2-a]quinolines have unique nitrogen-containing tetracyclic scaffolds which are widely spread in many bioactive pharmaceuticals and organic semiconductors.² However, only limited synthetic methods toward indolo[1,2-a]quinolines have been reported.³ For example, in 2007, Lautens *et al.* first developed an elegant Pd-catalyzed retro-Diels-Alder strategy to access indolo[1,2-a]quinoline (Scheme 1a).^{3f} In 2011, Verma and co-workers developed an iodine-mediated electrophilic cyclization to access iodo-substituted indolo[1,2-a]quinolines (Scheme 1b).^{3d} One year later in 2012, Verma's group further developed a Pd-catalyzed Sonogashira coupling conjoined C–H activation strategy for the preparation of indolo[1,2-a]quinolines (Scheme 1c).^{3c} Despite these significant advances, the development of straightforward and efficient synthetic methodologies for the synthesis of diverse functionalized indolo[1,2-a]quinolines, especially those that cannot be directly prepared *via* the previous reported strategies, has been enthusiastically pursued and highly desired.

Sulfones not only are versatile synthetic intermediates, but also exhibit a wide range of physical, chemical and biological activities.⁴ The preparation of sulfonated N-heterocycles has

Metal-free sulfonyl radical-initiated cascade cyclization to access sulfonated indolo[1,2-a]-quinolines†

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been one of the most challenging tasks which have gained increasing attention in recent years.⁵ Over the past decade, radical cascade cyclization reactions have emerged as an enabling platform to access a variety of sulfonyl-substituted heterocycles and carbocycles *via* sulfonyl radical-initiated cascade cyclization reactions.⁶ Those cascade cyclization reactions were able to incorporate the biologically valuable sulfonyl group into the cyclic ring construction within a one-step reaction, showing remarkable atom-/step-economy.⁷ However, it is especially worth mentioning here that the practical and efficient strategy for incorporating sulfonyl groups into indolo[1,2-a]quinolines has not been well established. In radical cascade cyclization reactions, one of the most prominent research objectives is developing new radical partners.⁸ As part of our continuing efforts in the development of convenient radical-initiated reactions,⁹ we herein disclose a novel and efficient sulfonyl radical-initiated cascade cyclization strategy, by which, a wide range of indolo[1,2-a]quinolines with an arylsulfonyl group attached at the 5-position and an aryl group at the 6-position were prepared *via* reaction of 1-(2-(arylethynyl)phenyl)indolets (1) with arylsulfonyl hydrazides (2) in the presence of TBAI/TBHP under mild reaction conditions (Scheme 1d). To the best of our knowledge, this is the first example to construct indolo[1,2-a]quinolines *via* radical cascade cyclization reactions. This method features metal-free and mild conditions, providing a novel and efficient procedure to access indolo[1,2-a]quinolines.

We initiated the study by establishing optimal experimental conditions using the model reaction of 3-methyl-1-(2-(phenylethynyl)phenyl)indole (1a) with TsNHNH₂ (2a), as summarized in Table S1 (ESI†). After extensive experimentation, the optimized reaction conditions were established as follows: 1a (0.5 mmol), 2a (1 mmol), TBAI (10 mol%) and TBHP (3 equiv.) were mixed in MeOH at 65 °C for 8 h.

With the optimized reaction conditions established, the substrate scope was then explored by examining various 1-(2-(arylethynyl)phenyl)indolets (1) and sulfonyl hydrazides (2), as illustrated in Table 1. As can be seen, a group of phenylsulfonyl hydrazides bearing electron-withdrawing groups (–F, –Cl, –Br, –CF₃, –CN)

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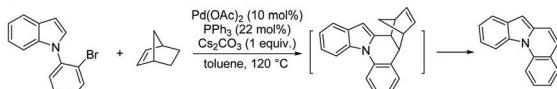
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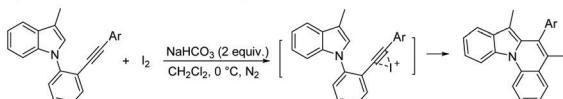
† Electronic supplementary information (ESI) available. CCDC 1850740 and 1947659. For ESI and crystallographic data in CIF or other electronic format see DOI: 10.1039/c9cc06924k

Previous works for the construction of indolo[1,2-a]quinolines

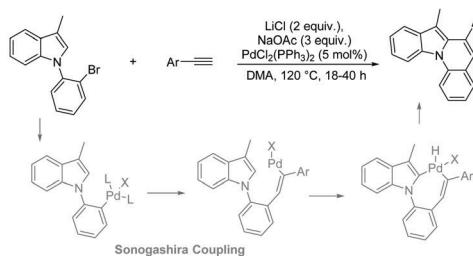
a) Synthesis of Indolo[1,2-a]quinolines via Retro-Diels-Alder



b) Iodine-mediated Electrophilic Cyclization

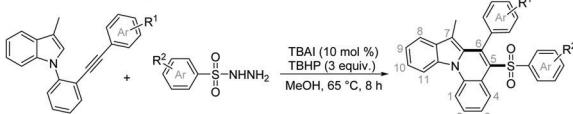


c) Sonogashira Coupling Conjoined C-H Activation



This work

d) Radical Cascade Cyclization



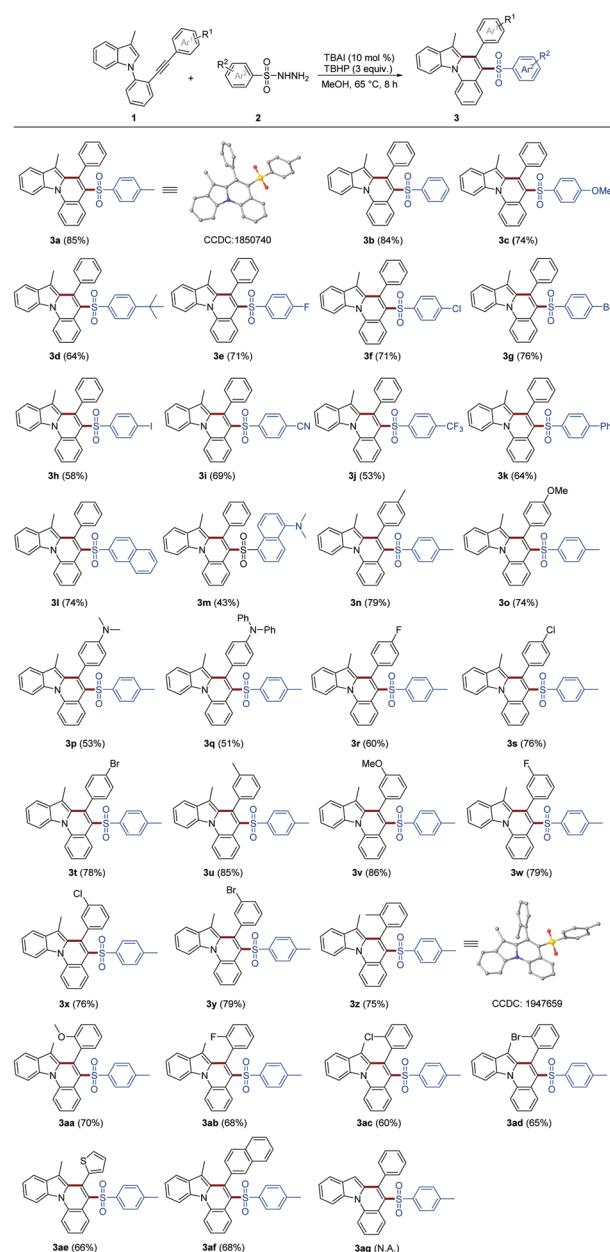
Eminent Synthetic Advantages:

- Simple one-pot procedure without tedious later modifications
- Metal-free and open air reaction conditions
- High atom-step-economy

Scheme 1 Comparison with previous works.

and electron-donating substituents ($-Me$, $-OMe$, $-N(Me)_2$, $-tBu$, $-Ph$) at the *para*-position of the phenyl group, reacted smoothly with **1a**, affording the corresponding products **3a–k** in moderate to excellent yields (53–85%). No obvious electronic effects were observed in those cases (**3a–k**). Further screening indicated that two naphthalene sulfonylhydrazides were also good at reacting with **1a**, rendering **3l–m** in moderate to good yields. Besides that, various 1-(2-(phenylethynyl)phenyl)indoles (**1**) bearing different substituents on Ar^1 , including electron-withdrawing groups ($-F$, $-Cl$, $-Br$) and electron-donating substituents ($-Me$, $-OMe$, $-N(Me)_2$, $-N(Ph)_2$), were also chosen to react with $TsNHNH_2$ (**2a**), giving target **3n–3ad** in moderate to excellent yields. No obvious electronic effects were observed from those cases (**3n–3ad**) as well. In addition, two starting reactants (**1**) containing thiophene and naphthalene rings (Ar^1) could react with $TsNHNH_2$ smoothly, leading to the formation of **3ae–3af** in satisfactory yields, respectively. Finally, the substrate without a methyl group at the 3-position of the indole failed to afford the desired product **3ag** but produced a complex mixture. All the newly synthesized sulfonated indolo[1,2-a]quinolines are new compounds, and the structures of **3a** and **3z** were confirmed by X-ray crystallography (hydrogen atoms have been omitted for clarity).

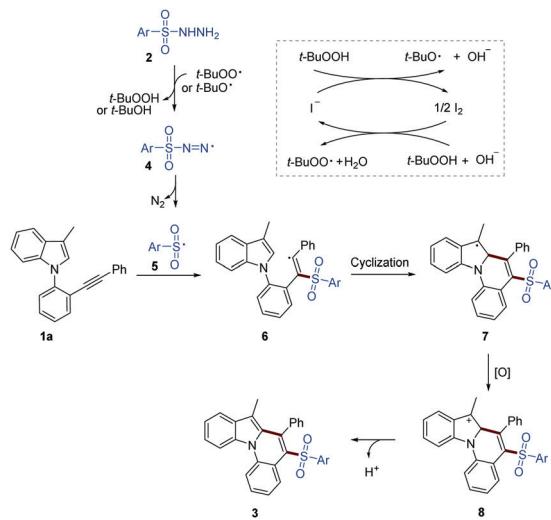
A plausible mechanism is proposed as shown in Scheme 2. Initially, TBHP reacts with the iodide anion from TBAI, generating $t-BuO^\bullet$ as well as $t-BuOO^\bullet$ radicals.¹⁰ Then, successive H-abstraction of sulfonyl hydrazide **2** by the resultant radicals affords

Table 1 Synthesis of sulfonyl substituted indolo[1,2-a]quinolines^a

^a Reaction conditions: **1** (0.5 mmol), **2** (1 mmol), TBAI (10 mol%), TBHP (3 equiv.), MeOH (10 mL) at 65 °C for 8 h. TBHP = *tert*-butyl hydroperoxide (70% aqueous solution). Isolated yields are given. N.A. = not analysed.

sulfonyldiazene radical **4**, which subsequently yields sulfonyl radical **5** with the release of N_2 . Then, the regioselective addition of sulfonyl radical **5** to the carbon–carbon triple bond of **1a** forms alkenyl radical **6**, which subsequently undergoes an intramolecular cyclization giving radical **7**. Then, **7** is quickly oxidized to carbocation intermediate **8**. Finally, a rapid deprotonation of carbocation **8** regenerated the aromatic ring to give the final product **3**.

Additional control experiments were then carried out, giving substantial support to the proposed reaction mechanism. As can

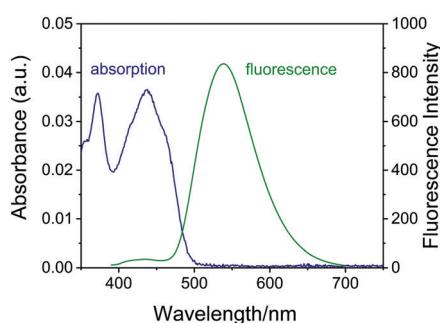
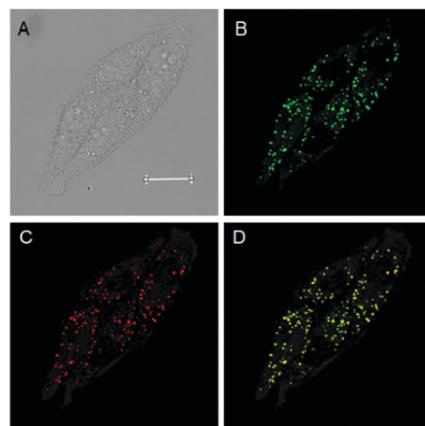


Scheme 2 Proposed mechanism.

be seen from Scheme S1 (ESI[†]), when the model reaction was performed in the presence of (2,2,6,6-tetramethylpiperidin-1-yl)oxidanyl (TEMPO) or 2,6-di-*tert*-butyl-4-methylphenol (BHT), two widely used radical scavengers, no or little desired product 3 was obtained, suggesting that the reaction might experience a radical process. In particular, when the model reaction was performed in the presence of BHT (Scheme S1b, ESI[†]), we successfully isolated product 9, evidencing that tosyl radical (Ts^\bullet) was indeed produced from TsNHNNH_2 and then trapped by BHT.

The large π -conjugated systems give the potential for these synthetic compounds to possess good fluorescence properties. Therefore, compound 3z was selected to carefully investigate the photo-physical properties. UV/vis and fluorescence spectra were thoroughly recorded in CH_2Cl_2 at room temperature. As can be seen in Fig. 1, a strong absorption for compound 3z at 300–450 nm and excellent fluorescence at 525 nm can be observed. There is almost no overlap between the fluorescence spectrum and absorption spectrum, exhibiting large Stokes shifts.

Lipid droplets (LDs) play important roles in a number of physiological processes, such as the construction and maintenance of membranes, regulations of the storage and metabolism of neutral lipids, signal transduction and protein degradation *etc.*¹¹

Fig. 1 UV-vis absorption and fluorescence spectra of 3z ($4 \mu\text{mol L}^{-1}$) in CH_2Cl_2 .Fig. 2 Confocal images of HepG-2 cells co-stained with $5 \mu\text{mol L}^{-1}$ 3z and $2 \mu\text{mol L}^{-1}$ Nile Red for 30 min. (A) Bright-field image; (B) confocal image from 3z on channel 1 (405 nm, 450–550 nm); (C) confocal image from Nile Red on channel 2 (488 nm, 600–650 nm); (D) the overlay of B and C. Scale bar = $20 \mu\text{m}$.

Therefore, the specific imaging of LDs has attracted widespread attention in recent years. In light of the excellent fluorescence property of 3z, further attempts were carried out to examine its fluorescence activity in living cells. When fluorophores are used for living cells imaging, cytotoxicity must be controlled. The cytotoxicity of 3z to HepG-2 cells was initially examined using a 3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2-*H*-tetrazolium bromide (MTT) assay. The results showed even when HepG-2 cells were treated with 3z at $30 \mu\text{mol L}^{-1}$, the cell viability was still higher than 80%, demonstrating the low toxicity of 3z to living cells (Fig. S1, ESI[†]). Afterward, HepG-2 cells were co-incubated with 3z ($5 \mu\text{mol L}^{-1}$) and the commercially available lipid probe Nile Red ($2 \mu\text{mol L}^{-1}$) for 30 minutes at 37°C . As can be seen in Fig. 2, compound 3z showed a group of clearly distinguishable green fluorescence dots within the cells under excitation at 405 nm (Fig. 2B), while Nile Red showed a group of similarly distributed red fluorescence dots under excitation at 488 nm (Fig. 2C). The merged image (Fig. 2D) revealed that the fluorescence signals from 3z overlap nicely with those from Nile Red. The Pearson correlation coefficient value of 3z and Nile Red in the image was determined as 0.92, showing that 3z is well qualified as a LDs-targeted fluorescence probe.

In conclusion, we have developed a novel and efficient sulfonyl radical-initiated cascade cyclization strategy, by which a wide range of indolo[1,2-*a*]quinolines with an arylsulfonyl group attached at the 5-position and an aryl group at the 6-position were prepared for the first time, *via* reaction of 1-(2-(arylethynyl)phenyl)indoles with arylsulfonyl hydrazides in the presence of TBAI/TBHP in MeOH at 65°C for 8 h. Impressively, the synthetic compounds exhibit excellent fluorescence properties, and cell imaging experiments were conducted to show the application in cell organelle imaging. Further research on the fluorescence properties and applications of these synthetic compounds is currently ongoing in our laboratory.

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Conflicts of interest

There are no conflicts to declare.

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