



Rational design of two-photon absorbing dicyanomethylene-4H-chromene derivatives and their application in bioimaging



Shuyi Chen^{a,1}, Mingzhu Zhang^{a,1}, Chuanzhen Zhu^a, Huanxuan Lu^a, Meng Zhao^a, Xiaohu Tian^b, Qiong Zhang^a, Senio Campos De Souza^c, Fang Rong^b, Hongping Zhou^a, Jieying Wu^{a,**}, Yupeng Tian^{a,*}

^a Department of Chemistry, Key Laboratory of Functional Inorganic Materials Chemistry of Anhui Province, Anhui University, Hefei 230601, China

^b School of Life Science, Anhui University, Hefei 230039, China

^c Department of Chemistry, University College London, London WC1H 0AJ, UK

ARTICLE INFO

Article history:

Received 9 August 2017

Received in revised form

9 September 2017

Accepted 14 September 2017

Available online 21 September 2017

Keywords:

DCM derivatives

Large Stokes-shift

Two-photon absorbing

Biological imaging

Minimal autofluorescence

ABSTRACT

In vitro and *in vivo* fluorescence imaging of cells and animals plays a significant role in studying biological systems. However, autofluorescence becomes a serious issue *in vivo* imaging under excitation at UV–vis wavelengths where biological molecules cause interference with the chromophore. In order to address this issue, a novel class of dicyanomethylene-4H-chromene (DCM) derivatives (**PCM**, **PCV**, **BPCM** and **BPCV**) that can be excited at ~900 nm under two-photon excitation conditions and emit in the red wavelength region (≥ 600 nm) have been developed. Particularly, one of the DCM derivatives (**PCM**) shows several advantages, including remarkable two-photon absorption (2PA) properties in the near-infrared (NIR) region, superior photostability, large Stokes-shift and pronounced solvent-sensitive emission behavior, which is favorable for *in vivo* fluorescence labelling with a high signal-to-noise ratio.

© 2017 Elsevier Ltd. All rights reserved.

1. Introduction

Utilizing optical chromophores as imaging agents has become one of the most powerful tools for non-invasive visualization of biological processes. These chromophores are easy to use, cost-effective, noninvasive, highly sensitive detection and able to visualize living systems at a subcellular level [1–3]. Accordingly, many high-performance chromophores have been explored for such purpose [4–9].

In conventional one-photon confocal microscopy (1PM) light scattering is a serious issue; therefore, during the last two decades, two-photon microscopy (2PM) based on nonlinear interaction between light and matter has emerged as an important tool for imaging biological species [10,11]. Near-infrared (NIR) fluorescence channel under two-photon excitation conditions (650–950 nm) allows for deeper penetration and minimal absorption through

highly scattering living species media, decreased photodamage as well as minimal photobleaching to living species [12,13]. Furthermore, since autofluorescence is mainly caused by two-photon emission in the green region of intrinsic biomolecules in biological samples such as NADH, riboflavins, retinol and folic acid [14,15]. Two-photon excitation of chromophores in the wavelength range of 650–950 nm and collection of emission light in the red wavelength region (around 600 nm) could greatly suppress autofluorescence from these intrinsic biomolecules. These promising features of 2PM have motivated chemists to search for two-photon absorbing (2PA) chromophores with desirable photophysical properties for the detection and imaging of biological processes in living species [16,17].

Recently, a myriad of dicyanomethylene-4H-chromenes (DCM), as well as its derivatives have been developed as one-photon fluorescent sensors, photovoltaic sensitizers and logic gates [18–20]. However, there are only few DCM derivatives with 2PA developed in recent years. Although Wu et al. reported the DCM-GA as a g-glutamyltranspeptidase two-photon fluorescent sensor [21], the maximum value of 2PA cross-section is 150 GM at 820 nm. Dong et al. reported the TP-NIR as a cysteine two-photon fluorescent sensor [22], the maximum value of its 2PA cross-section is only

* Corresponding author.

** Corresponding author.

E-mail addresses: jywu1957@163.com (J. Wu), yptian@ahu.edu.cn (Y. Tian).

¹ Co-first authors.

16 GM at 830 nm. Therefore, chromophores with high 2PA cross-section values are still in high demand for specific bioimaging applications. In general, DCM is composed of an electron-withdrawing group (EWG), a π -conjugated linker and an electron-donating group (EDG), contributing as a typical push–pull system.

To our knowledge, extending the π -conjugated frameworks or optimizing the combination of an electron donor and acceptor would be the preferred approach for the rational molecular design of two-photon fluorescent materials with relatively large 2PA cross-section [23–30]. In this work, we focus on extending the π -conjugated system by increasing the number of branches of DCM to obtain a novel class of DCM derivatives with excellent two-photon fluorescent properties (the maximum value of 2PA cross-section over 272.16 GM at ~900 nm). Meanwhile, we substituted acceptors of DCM analogues by replacing the ethyl cyanoacetate group to improve cell membrane permeability and biocompatibility. The influence of different electron donor on photophysical properties also has been systemically investigated. Linear optical properties and nonlinear optical properties were described as the basis for potential applications in two-photon fluorescence imaging. Relying on the comprehensive studies, **PCM** and **BPCV** (Scheme 1) were selected for bioimaging study using 2PM. **PCM** was successfully applied for labelling cancer cells and zebrafish with minimal autofluorescence and deeper penetration (>300 μm). The results suggested that **PCM** could potentially be applied as 2PA probes in biological system.

2. Results and discussion

2.1. Crystal structure description and DFT calculations

The single crystals of **PCM**, suitable for X-ray diffraction analysis, were obtained via the method of slow evaporation from pure CH_2Cl_2 solution at room temperature. The molecular structure of **PCM** was shown in Fig. 1. The relevant crystal data and structural parameters were summarized in Table S1. The selected bond distances and angles were listed in Table S2. Single crystal X-ray diffraction analysis revealed that **PCM** crystallize in the monoclinic $P2_1/n$ space group. As shown in Fig. 1, the bond length of C (9)–C (10) and C (19)–C (20) (1.319 and 1.318 Å) indicated the formation of a double-bond connecting the donor and acceptor moieties. Herein, the double-bond in **PCM** maintains a *trans*-conformation, with torsion angles (178.39° , C7–C9–C10–C11; 178.80° ,

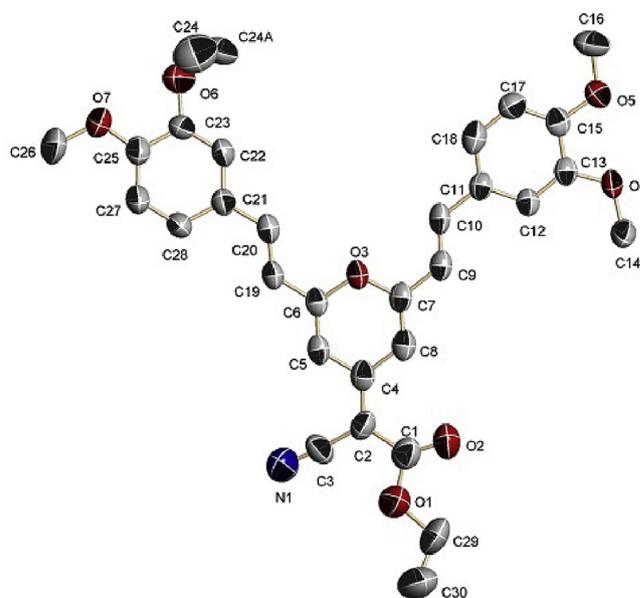
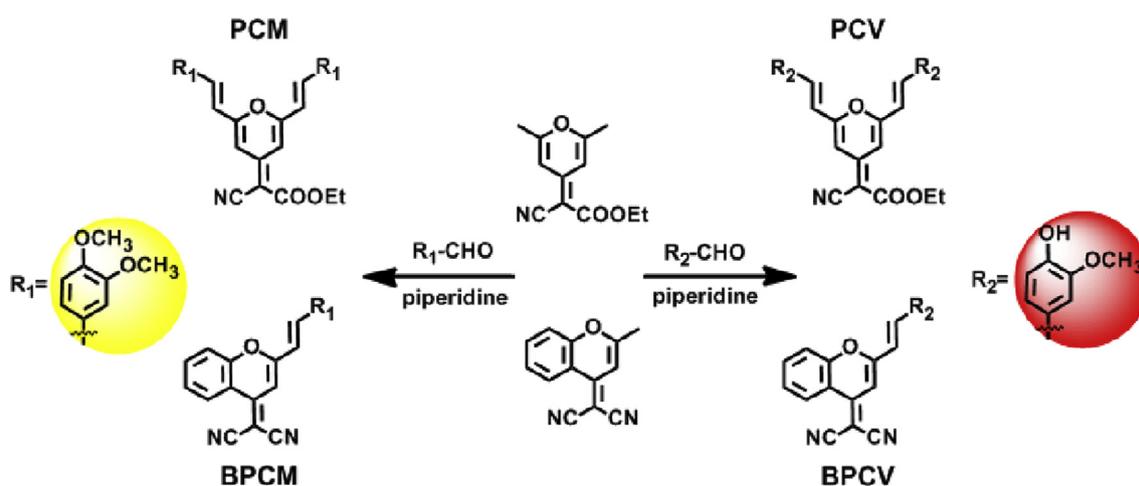


Fig. 1. The ORTEP structure of **PCM** with atomic labeling scheme (with 50% thermal ellipsoid probability); all hydrogen atoms are omitted for clarity.

C6–C19–C20–C21) and the small dihedral angles (6.13° and 9.22°) between the aromatic ring and pyran unit. The molecular conformation was almost planar in the ground state, which corresponds to the maximum conjugation between donor and acceptor.

Fig. 2 showed straightforward representations of the electron density distribution. Orbital analysis suggested that the highest occupied molecular orbital (HOMO) was comprised of aromatic ring with a smaller contribution from conjugated π -bridge. Moreover, the lowest unoccupied molecular orbital (LUMO) distribution was localized in the pyran unit with abundant conjugated π -bridge. Hereby, there were relatively strong π -donor interactions between the aromatic ring and the pyran center, and the electron density in each molecule was very similar. As shown in Table S3, the TD-DFT excited state calculations of all the chromophores gave two transitions, respectively. As indicated in Fig. 2 and Table S3, the lowest-energy excitation bands of all the chromophores (422–460 nm) were assigned as the ICT transition [$\pi(\text{aromatic ring}) \rightarrow \pi^*(\text{CH}=\text{CH})$] mixed with the [$\pi(\text{aromatic ring}) \rightarrow \pi^*(\text{pyran unit})$] due to the H - 1 \rightarrow L



Scheme 1. Synthetic route of all the chromophores.

Download English Version:

<https://daneshyari.com/en/article/4765736>

Download Persian Version:

<https://daneshyari.com/article/4765736>

[Daneshyari.com](https://daneshyari.com)