

Synthesis of squaraine dyes with potential application in protein detection by fluorescence spectroscopy

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Keywords: squaraine dyes, protein detection, fluorescence

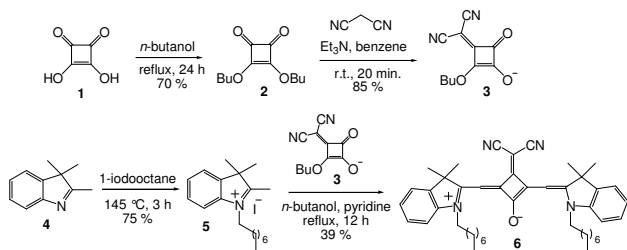
INTRODUCTION

Squaraines are interesting organic molecules due to the strong absorption in the visible region ($\epsilon \sim 10^5 \text{ M}^{-1} \cdot \text{cm}^{-1}$), intense fluorescence emission and good photoconductivity presented in these dyes.¹ These features make them useful in a variety of applications in the area of imaging, nonlinear optics, photodynamic therapy and photovoltaic cells.² Furthermore, squaraine dyes are suitable for fluorescence detection of proteins at long-wavelength excitation and labels in biological assays due their unique photochemical and photophysical properties namely light absorption in the visible red and near-infrared regions.³

This work describes the synthesis of squaraine dyes prepared by the condensation of heteroaromatic compounds with a dialkyl ester derived from squaric acid in a one-step reaction.

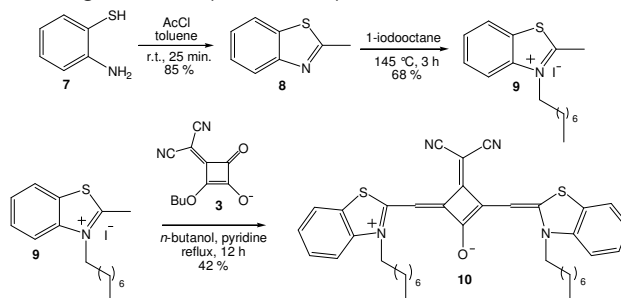
RESULTS AND DISCUSSION

Symmetrical squaraine **6** was synthesized as shown in Scheme 1. The reaction of squaric acid (**1**) with excess of *n*-butanol afforded the dibutyl ester (**2**), which was subsequently treated with 1.0 equivalent of malononitrile to give the intermediate (**3**) in 85% yield. Then, 2,3,3-trimethylindolenine (**4**) was converted in the quaternary ammonium salt **5** by coupling of **4** with excess of iodoctane in 75% yield. The subsequent base-catalyzed condensation of 2.0 equivalents of 3*H*-indolium salt **5** and 1.0 equivalent of **3** under reflux in a *n*-butanol:pyridine mixture, afforded the symmetrical squaraine dye **6** in 39% yield.



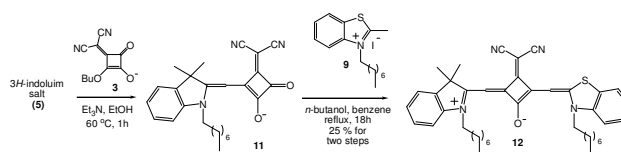
Scheme 1. Synthesis of squaraine 6

Squaraine dye **10** was obtained by a similar synthetic pathway using 2-aminothiophenol (**7**) as starting material (Scheme 2).



Scheme 2. Synthesis of squaraine 10

Condensation reaction of iodide **5** with dicyanomethylene **3** in the presence of triethylamine, afforded the intermediate mono-squaraine **11** (Scheme 3). Subsequent reflux of crude product containing mainly **11** with ammonium salt **7** in a *n*-butanol:benzene mixture led to the unsymmetrical squaraine dye **12** in 25% yield from quaternized indolenine **5**.



Scheme 3. Synthesis of squaraine 12

CONCLUSION

The dyes **6**, **10** and **12** were synthesized by the base-catalysed condensation of heterocyclic ammonium salts with dicyanomethylene **3**. These compounds exhibit interesting photophysical properties for application in protein detection by fluorescence spectroscopy.

ACKNOWLEDGEMENTS

INDI-Saúde and CNPq for financial support.

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