List of RGJ advisors 2023/2024

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Keywords: Fluorenes; DNA binding; photophysical properties; fluorescence spectrometry

Summary of research: In this work, a novel series of fluorene derivatives will be synthesized and characterized. The steady-state UV absorption, fluorescence spectroscopy including time-resolved fluorescence, will be investigated to explain the mode, mechanism, affinity and energetics of binding of the fluorene derivatives and ct-DNA, which could make them potential anti-cancer drugs. The cytotoxicity against cancer cell lines will be also investigated, and molecular docking will be performed to support the binding mode as well as the relative binding energy of the fluorene derivatives and ct-DNA.

การกรอกรายละเอียดในแบบฟอร์มนี้ ต้องดำเนินการให้ครบถ้วนตามความเป็นจริง หากตรวจสอบพบว่ามีการปกปิดหรือเป็นเท็จ วช. ขอสงวนสิทธิ์ที่จะไม่พิจารณาสนับสนุนและจะเป็นผู้ไม่มีสิทธิ์รับทุน วช. เป็นเวลา ๓ ปี

แบบเสนอโครงการวิจัย (Research Project)
ประกอบการเสนอขอทุนอุดหนุนการวิจัยของสำนักงานการวิจัยแห่งชาติ (วช.)
โครงการปริญญาเอกกาญจนาภิเษก (คปก.) ภายใต้ความร่วมมือไตรภาคีไทย-สวีเดน
ประจำปีงบประมาณ ๒๕๖๗

๑. ชื่อโครงการวิจัย การศึกษาฟิสิกส์เชิงแสงของการจับกันของดีเอ็นเอกับอนุพันธ์ฟลูออรีนที่มีไพคอนจูเกต
(Photophysical studies of binding DNA and pi-conjugated fluorene derivatives)

๒. ชื่อ-สกุล อาจารย์ที่ปรึกษา รองศาสตราจารย์ ดร. ธิตินันท์ กาพย์เกิด
(Associate Professor Thitinun Karpkrid, Ph.D)
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๓. กลุ่มสาขาวิทยาศาสตร์พื้นฐานที่สมัคร (เลือกเพียง ๑ กลุ่ม)

☐ ชีววิทยา (Biology) ☑ เคมี (Chemistry)
☐ ฟิสิกส์ (Physics) ☐ คณิตศาสตร์ (Mathematics)

๔. ผู้ใช้ประโยชน์ (Research stakeholders) (กรณีมีความร่วมมือฯ) เช่น ความร่วมมือของหน่วยงานภาครัฐ (เช่น กระทรวง กรม)/เอกชนที่ร่วมสนับสนุนทุนวิจัย เช่น MOU เป็นต้น

☑ มี I (Thitinun Karpkird) have been collaborated with Prof. Bo Albinsson, Department of Physical Chemistry, Chalmers University of Technology, Gothenburg, Sweden for almost two decades since I was a Ph.D. student under RGJ program and did a part of Ph.D. dissertation at Prof. Albinsson's Lab at Chalmers during 2005-2006. During five past years, I have sent my graduated students to do short-term projects which were a part of their thesis at Chalmers with Prof. Albinsson's group. As well as I just visited Prof. Albinsson lab last May (May 2023) and did a small project using fluorescence spectrometry technics.

☐ ไม่มี

๕. คำสำคัญ (Keyword) ของโครงการ

Fluorenes; DNA binding; photophysical properties; fluorescence spectrometry

๖. ความสำคัญและที่มาของปัญหาที่ทำการวิจัย (Problem statement and significance of research)

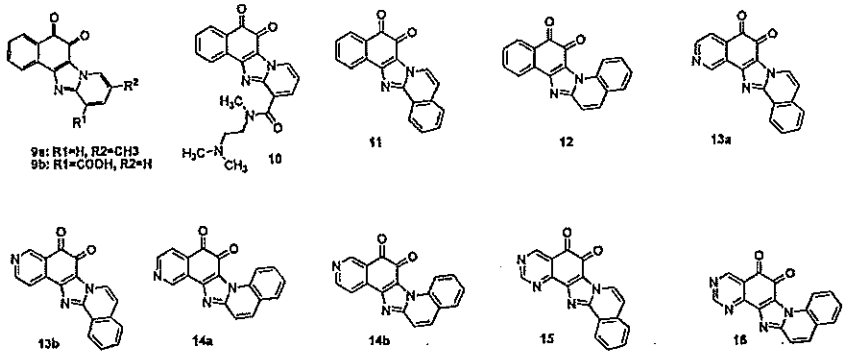
π-Conjugate fluorene is a rigid, planar molecule containing delocalize π-electron throughout the structure. Fluorene can be utilized as a spacer via aromatic coupling, which

could be helpful for fluorescence emission properties. Many of the tetra- and pentacyclic fluorene derivatives have been potent DNA intercalating agents. The modification of fluorene structures can increase the activity of the intercalating structures by interaction with the phosphate backbone of DNA, hydroxyl groups could also interact via H-bridges. Normally the stabilization of DNA-drug binding proceeds through three different modes of non-covalent interaction: electrostatic interaction, intercalative and groove binding modes. The alteration affecting the DNA structure can lead to its dysfunction and are linked with risk of complex diseases. Therefore, novel compounds have been developed to enhance the anti-cancer efficiency, achieve specific binding with DNA binding sites, and be less toxic to the normal cells. Studying the binding mode of DNA target drugs is important to control many diseases by improvising systematic strategies in drug designing. The study of photophysical properties of drugs interactions with biomacromolecules is important and becomes interesting when studying certain biological targets.

๗. ทฤษฎี/สมมุติฐานของโครงการ (Hypothesis)

To understanding the potential of DNA binding mechanism of π-conjugated fluorene derivatives and enhance their anti-cancer potential, comprehensive photophysical and biophysical methods have conducted. As fluorene-base compounds are π-conjugated structures, rigid and planar, they have ability to intercalate between the base pairs of DNA. These binding mechanisms can be studied using absorption and fluorescence techniques. The different substituents connected to fluorene moiety may affect the interaction with DNA via H-bond or ionic interaction.

In this work, a novel series of fluorene derivatives **9-16** will be synthesized. The steady-state UV absorption, fluorescence spectroscopy including time-resolved fluorescence, will be investigated to explain the mode, mechanism, affinity and energetics of binding of the fluorene derivatives and ct-DNA, which could make them potential anti-cancer drugs. The cytotoxicity against cancer cell lines will be also investigated, and molecular docking will be performed to support the binding mode as well as the relative binding energy of the fluorene derivatives and ct-DNA.



๘. วัตถุประสงค์ของโครงการ (Objectives)

(1) To synthesize a novel series of benzo[a]fluorene-5,6-diones and azabenzo[a]fluorene-5,6-diones (compound 9-16).

(2) To study the relationship between fluorene derivative structures and binding mechanisms, binding affinity as well as binding energy of fluorene derivatives to ct-DNA using UV-Vis absorption and fluorescence spectroscopy methods.

(3) To investigate the relationship between fluorene derivative structures to anti-cancer affinity by evaluating cytotoxicity against cancer cell lines and employing the molecular docking method.

๙. การทบทวนวรรณกรรม/ผลงานวิจัยที่เกี่ยวข้อง (Literature Review)

Hyun-Jung *et. al.* synthesized azabenzo[a]fluorene and investigated their cytotoxicity [1]. They found that it could exhibit strongly cytotoxicity against several cancer cell lines including A539 (lung carcinoma), SKOV3 (ovarian carcinoma), SK-MEL-2(melanoma), XF498(CNS) and HCT15 (colon carcinoma). Moreover, our group has previous reported a new series of substituted tri-/tetraazabenzo[3,2-a] fluorene-5,6-diones and oxime derivatives have been synthesized. The pyrimidine chemophore showed antiproliferative activities similar to mitoxantrone and doxorubicin.[2] Many literatures have reported the promising use of UV absorption and fluorescent technics to explain the mode of DNA binding of drugs. The UV absorption and fluorescence data showed that the fluorene-based imines interact with DNA via intercalation [3]. Porphyrin-containing fluorenyl units have a greater affinity for binding to DNA via the minor groove mode.[4] Fluorinated quinoxalines derviatives were selectively DNA binding in minor groove mode.[5] Benzoxazole derivatives bind ctDNA via intercalation mode.[6] Moreover, the DNA binding mode of camptothecin, an anti-tumor alkaloid drug, involved the hydrogen bonding and van der Waals interactions which confirm the groove binding mode.[7]

๑๐.ระเบียบวิธีวิจัย (Methodology)

(1) Synthesis of fluorene derivatives

Compound **9-16** will be synthesized by using the cyclization followed the previous report with some modification [2]

(2) Photophysical studies of benzofluorene derivative and DNA

2.1) Steady state absorption and fluorescence spectroscopy studies: to determine fluorescence quantum yield, binding affinities and Thermodynamic behavior

2.2) Fluorescence lifetime studies: to determine quenching constant, binding constant and binding sites

(3) *Cytotoxicity study*: cytotoxicity of benzofluorene derivatives will be studied against A549

(4) *Molecular docking study*: Binding investigation of benzofluorene derivatives to DNA will be performed using molecular docking with Autodock program.

๑๑.ขอบเขตของการวิจัย (Scope of the study)

Part I: Benzofluorene with different substitutions (9-16) will be synthesized and characterized by using NMR, IR and mass spectroscopy. The steady state absorption and fluorescence emission of fluorenes-DNA complexes will be studied in various organic solvents to determine the Stoke shift and fluorescence quantum yield. This part will be conducted at Department of Chemistry, Kasetsart University, Bangkok Thailand,

Part II: Advance fluorescence measurements, including anisotropy fluorescence and time-resolved fluorescence spectroscopy of the benzofluorenes derivatives and DNA binding, will be studied to determine the quenching constant, binding constant and thermodynamic properties. This part will be done at Chalmers, Sweden under supervisor of Prof. Bo Albinsson.

Part III and IV: Cytotoxicity studies against cancer cell line A549 will be conducted at the Department of Biochemistry, Kasetsart University. Molecular docking will be performed to support and explain the binding affinity of fluorene-DNA complexes at Department of Chemistry, Kasetsart University.

๑๒.ผลผลิต (Output) ผลลัพธ์ (Outcome) และ ผลกระทบ (Impact) ที่คาดว่าจะได้จากการวิจัย

Output: publications, more graduate students trained

Outcome: development/method of using fluorescence method to propose the efficiency of drugs for cancer therapy.

Impact: development of a new series of candidate drugs

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กรุณาจัดทำเป็นภาษาอังกฤษ เนื่องจากฝ่ายสวีเดนจะเป็นผู้พิจารณาข้อเสนอ
ความยาวไม่เกิน ๔ หน้ากระดาษเอ ๔ โดยใช้อักษร TH Sarabun ขนาด ๑๖