

**PROCORE - FRANCE/HONG KONG JOINT RESEARCH SCHEME
COMPLETION REPORT**

Project Reference Number

F-HK11/10T

Project Title

RNA G-quadruplex sensing
(RNA 上 G 碱基形成的四连体的检测)

Particulars

| | Hong Kong team | | | | French team | | | |
|---|---|-------|--------------------------|-------|-------------------------------------|---------------|--------------------------|------------|
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| Name of Co-Investigator (if any) | English: Dr. Chung-Hang Leung Chinese: 梁重恒 | | | | Dr. Anne Bourdoncle | | | |
| Institution or Institutional affiliation | <input type="checkbox"/> | CityU | <input type="checkbox"/> | HKU | <input type="checkbox"/> | CEA | <input type="checkbox"/> | INRA |
| | <input type="checkbox"/> | CUHK | <input type="checkbox"/> | HKUST | <input type="checkbox"/> | CNRS No. | <input type="checkbox"/> | INRIA |
| | <input checked="" type="checkbox"/> | HKBU | <input type="checkbox"/> | LU | <input type="checkbox"/> | INFREMER | <input type="checkbox"/> | INSERM No. |
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| Other project team members (if any) | Ka-Ho Leung | | | | Dr. Gilmar Salgado Jun Zhou | | | |

Funding Period

| | 1 st year | 2 nd year (if applicable) |
|-----------------|----------------------|--------------------------------------|
| Start Date | 01/01/2011 | 01/01/2012 |
| Completion Date | 31/12/2011 | 31/12/2012 |

Objective(s) as per original application

1. To investigate fluorescent methods of RNA G-quadruplex detection
2. To develop organic or metal-based small molecule stabilizing ligands of G-quadruplexes
- 3...

[Please attach relevant document(s)]

i) Outline of proposed research and results obtained

The aim of this project is to develop novel fluorescent methods of detecting RNA G-quadruplexes with metal-based small molecules. The objectives outlined in this project were successfully achieved resulting in the publication of two other articles in high-impact international journals supported by this project.

G-quadruplex is a non-canonical form of DNA secondary structure observed in guanine-rich DNA. The G-quadruplex structure contains four guanines arranged in a planar arrangement, stabilized by Hoogsteen hydrogen bonding and monovalent cations such as potassium. G-quadruplexes have received much attention in the recent literature due to their existence in telomeres and in the promoter regions of oncogenes such as *c-myc*.

While there have been extensive studies on the interactions of organic ligands with G-quadruplex, there are relatively few related studies with transition metal-based G-quadruplex stabilizing compounds and molecular probes. Metal complexes typically contain organic ligands arranged around the metal centre in a precise spatial arrangement. Due to this well-defined three-dimensional structure, the interactions of metal complexes with biomolecules are highly selective.

Encouraged by these ideas, we had successfully synthesized and characterized several phosphorescent cyclometallated metal complexes bearing planar *N,N*-donor ligands as novel luminescent probes for recognizing G-quadruplex nucleic acids structures and discriminating against single-stranded and double-stranded DNA structures. These iridium(III) complexes have been characterized by ^1H , ^{13}C NMR spectroscopy and High Resolution Mass Spectroscopy. The complexes were subjected to fluorescence spectroscopy for their responses towards different conformations of nucleic acids including single-stranded DNA, double-stranded DNA and G-quadruplexes. The unusual luminescence enhancement of the complexes with G-quadruplexes is presumably due to the pi-pi stacking interaction and hydrophobic interactions of the aromatic plane of the complexes with the topology of the G-quadruplexes. The analytic performance of these complexes has been demonstrated in various projects utilizing G-quadruplexes as a transducer for the detection of biomolecular targets and heavy metal ions. The experimental findings have shown that our novel iridium(III) luminescent probes are simple, rapid and highly sensitive for the detection molecular G-quadruplexes.

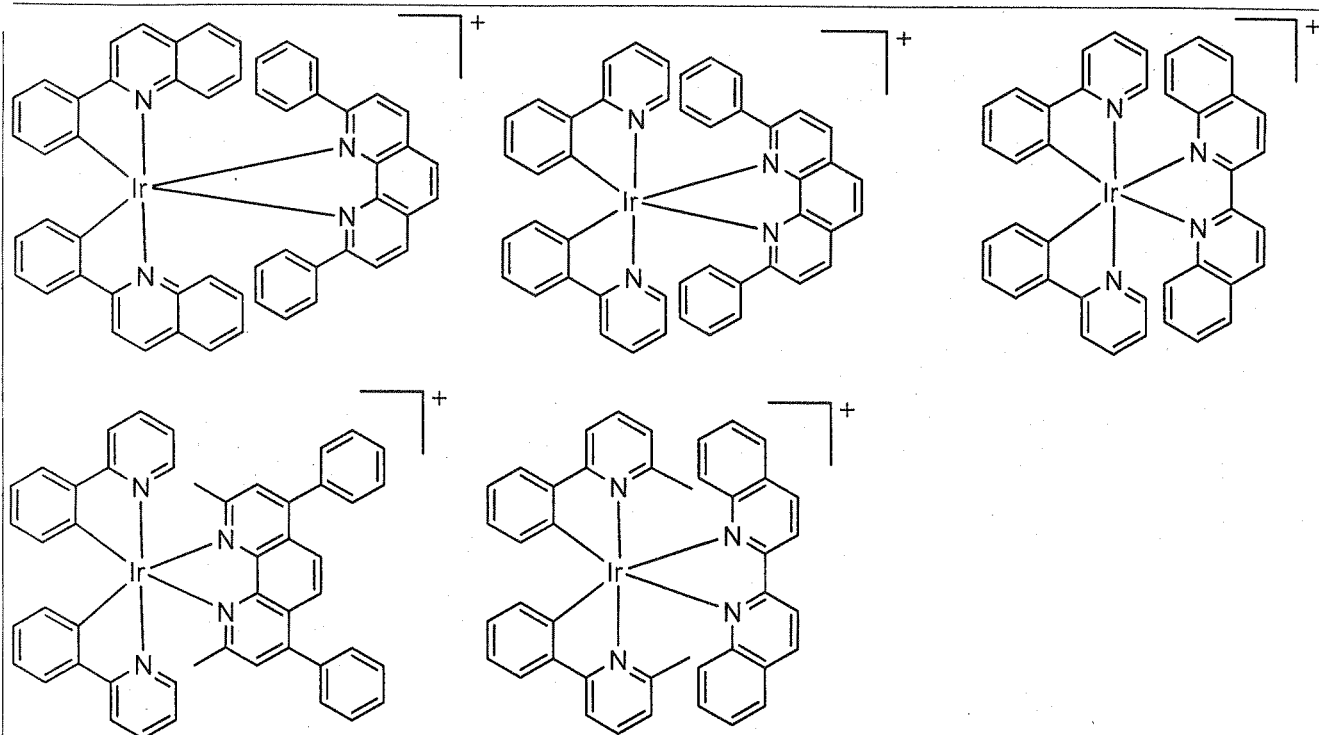


Figure 1. Selected chemical structures of the novel G-quadruplex DNA selective luminescent probes based on iridium(III) metal center.

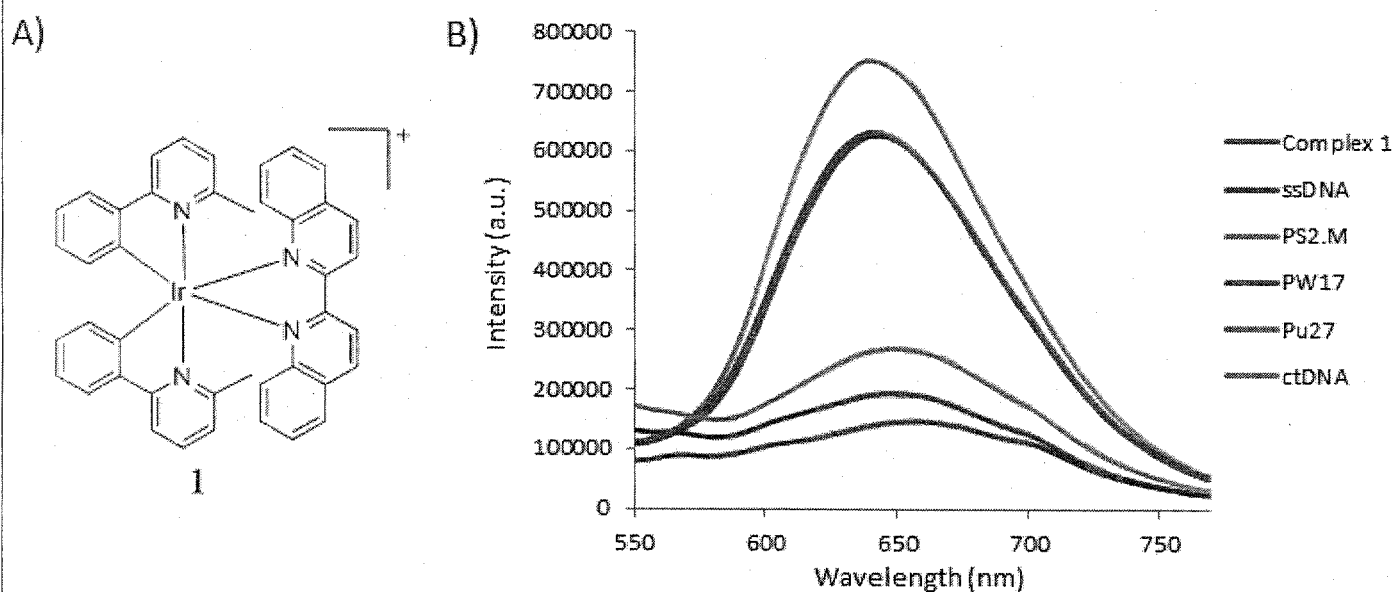


Figure 2. A) One of the novel G-quadruplex selective luminescent probes synthesized B) Luminescence response of complex 1 (1 μ M) in 50 mM Tris buffer (pH 7.0) in the presence of: 25 μ M ct DNA (double-stranded DNA); 25 μ M ss DNA (single-stranded DNA); 25 μ M PS2.M (G-quadruplex); 25 μ M PW17 (G-quadruplex); 25 μ M Pu27 (G-quadruplex).

ii) Significance of research results

The unusual enhanced luminescence response of the novel iridium(III) complexes synthesized is presumably due to the pi-pi stacking interaction and hydrophobic interactions with the topology of the G-quadruplexes. This interesting property of these novel complexes represent an attractive molecular structure for further development of more potent G-quadruplex specific luminescent probes. By derivatizing the donor coligands with appended G-quadruplex groove binding moieties, which are capable of binding to the grooves of the G-quadruplexes, more G-quadruplex selective probes and oncogenic RNA/DNA G-quadruplexes stabilizers could be developed.

iii) Research output

- 1) **D.-L. Ma**, H.-Z. He, V. P.-Y. Ma, D. S.-H. Chan, K.-H. Leung, H.-J. Zhong, L.-H. Lu, **J.-L. Mergny**, C.-H. Leung. "Label-free sensing of pH and silver nanoparticles using an "OR" logic gate" *Anal. Chim. Acta*, **2012**, 733, 78.
- 2) H.-Z. He, H. Yang, D. S.-H. Chan, C.-H. Leung, **J. Zhou**, **A. Bourdoncle**, **J.-L. Mergny**, **D.-L. Ma**. "Label-free detection of picomolar lead(II) ions in aqueous solution using a metal-based luminescent switch-on probe" *Biosensors and Bioelectronics*, DOI: 10.1016/j.bios.2012.08.060.

iv) Potential for or impact on further research collaboration

The interesting property of the rationally designed novel iridium(III) complexes as G-quadruplex luminescent probes represent an attractive molecular structure for further development of more potent G-quadruplex specific luminescent probes and stabilizers. By derivatizing the donor coligands with appended G-quadruplex groove binding moieties, which are capable of binding to the grooves of the G-quadruplexes, more G-quadruplex selective probes and oncogenic RNA/DNA G-quadruplexes stabilizers could be developed. It is expected that an in silico "fragment-based molecular docking" would serve as a key element in identifying the potential groove binding small motif and thus play an important role in the design of novel iridium(III) complexes as highly specific RNA G-quadruplex stabilizers and luminescent probes for future collaborations.