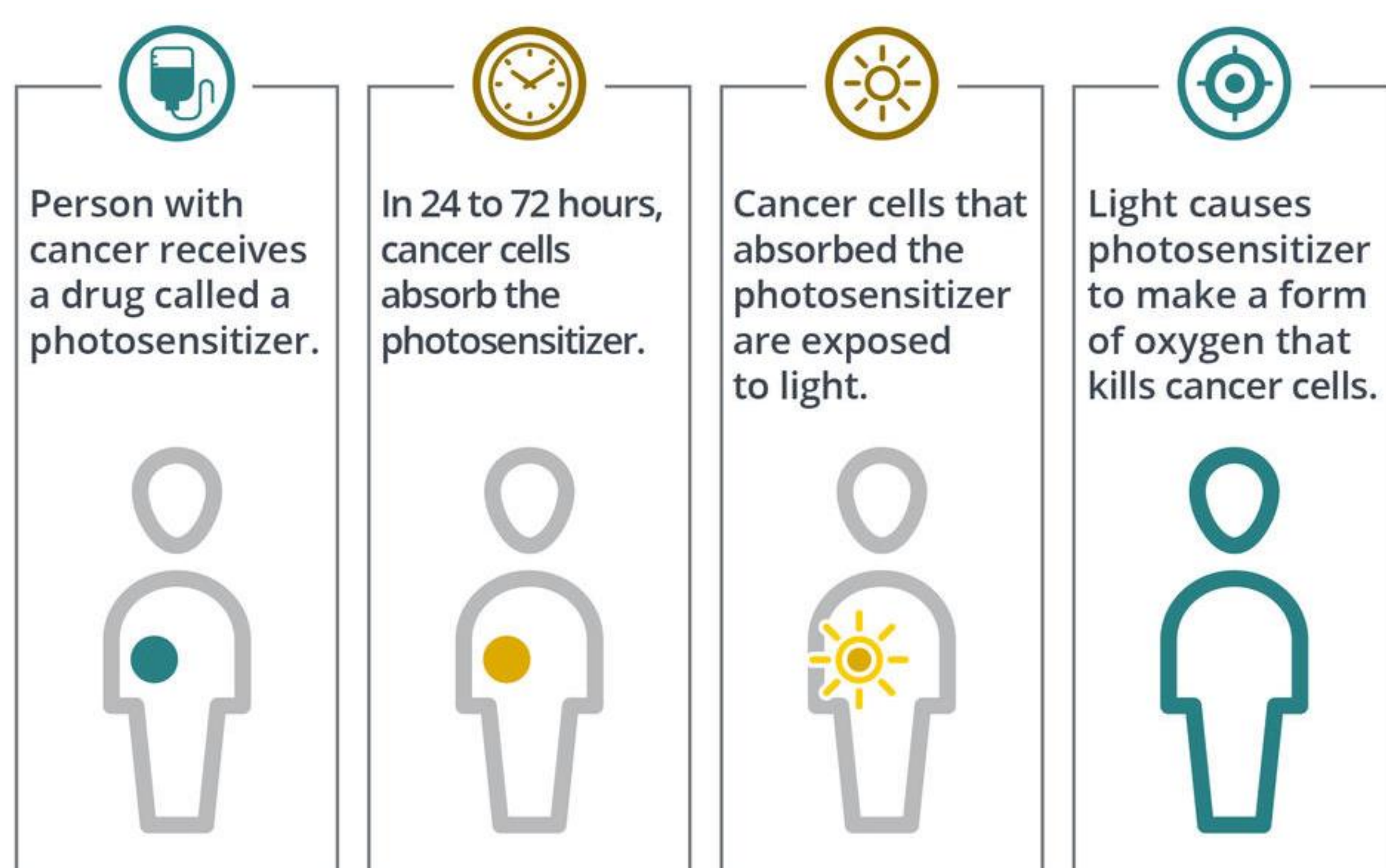


## Abstract

Photodynamic therapy (PDT) is a strategy for treating cancer with high selectivity. PDT utilizes an otherwise nontoxic prodrug called a photosensitizer (PS) that, in the presence of light and molecular oxygen, causes localized cell death. Approved PSs are generally organic tetrapyrroles that exert their PDT effects primarily through singlet oxygen and other reactive oxygen species (ROS). We have been developing next-generation metallodrug PSs that exploit different excited state configurations and alternate modes of action for photocytotoxicity. Our own TLD1433, a Ru(II) polypyridyl complex incorporating an oligothiophenyl-containing ligand, is one example and is currently in Phase 2 clinical trials (**NCT03945162**) for treating non-muscle invasive bladder cancer (NMIBC) with PDT. Herein, we report a novel class of metal-based PSs that are structurally similar to TLD1433 and highlight their light-driven PDT effects.

## Photodynamic Therapy

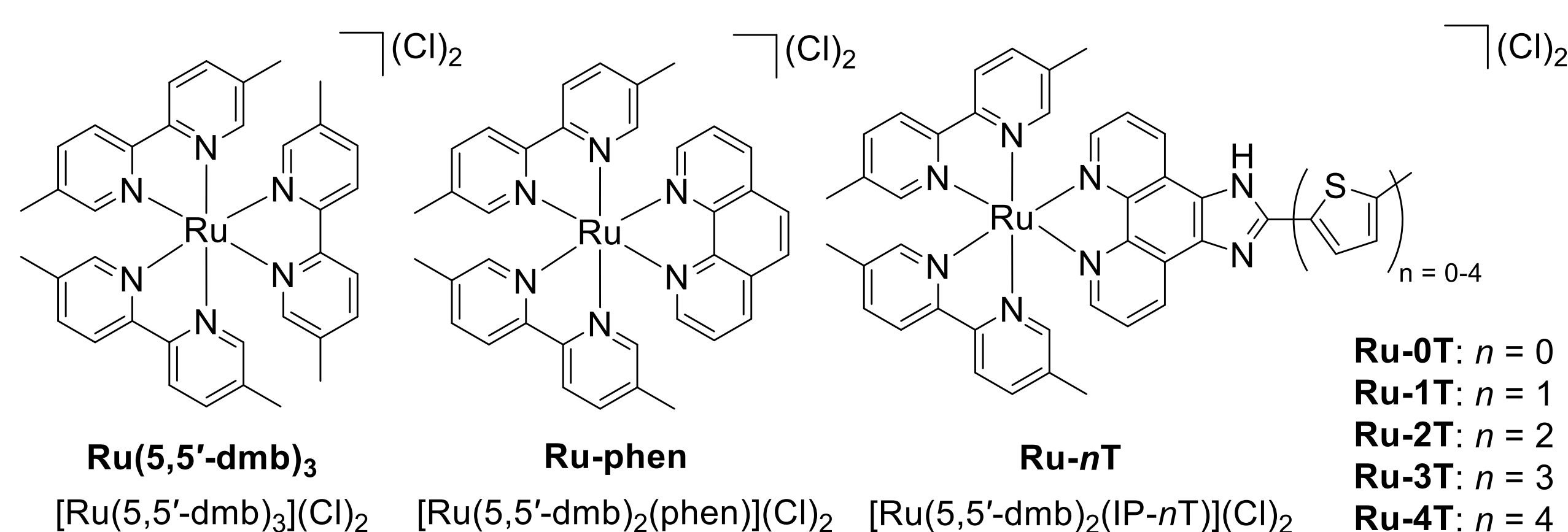


[cancer.gov/about-cancer/treatment/types/photodynamic-therapy](https://cancer.gov/about-cancer/treatment/types/photodynamic-therapy)

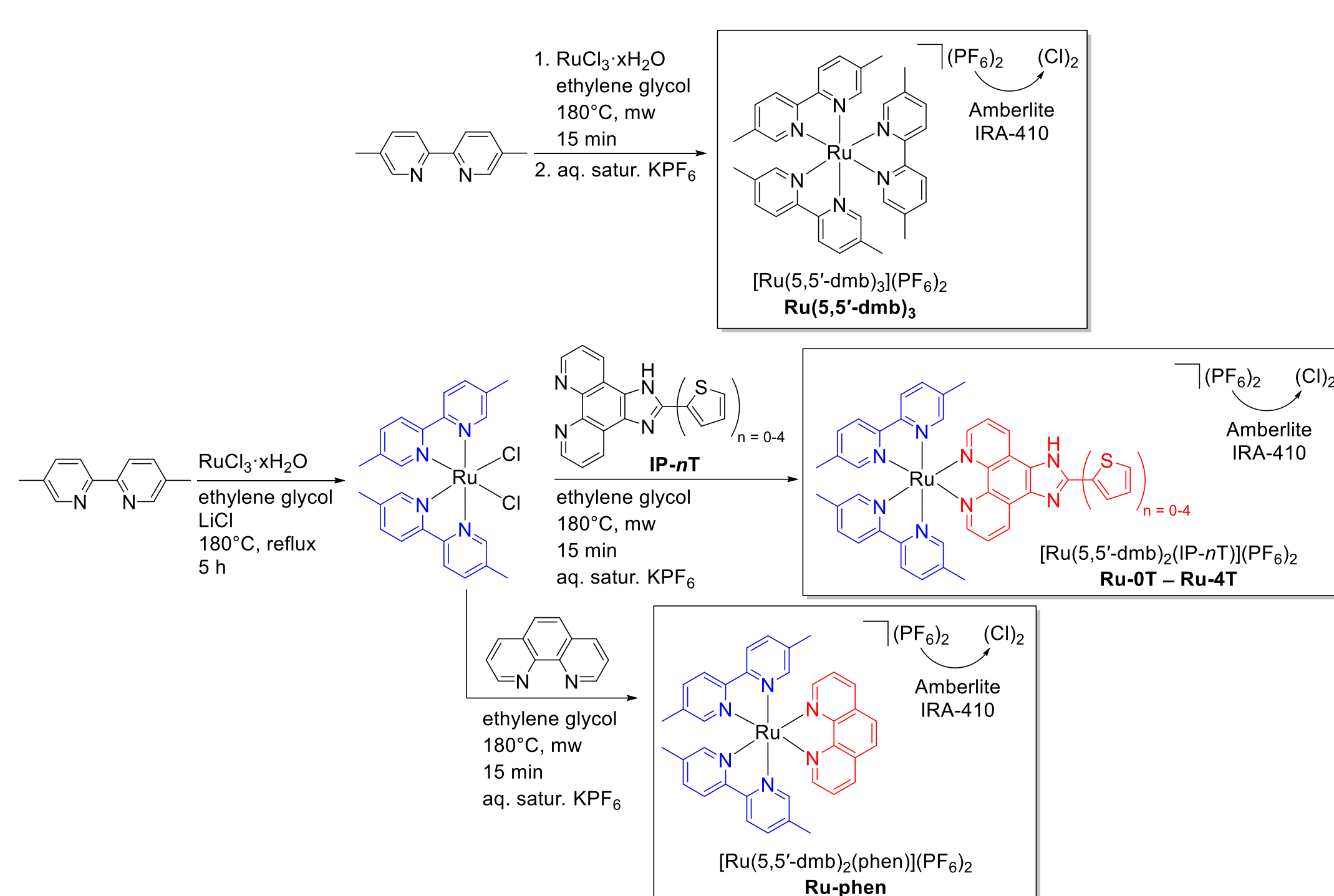
## Objectives

- Synthesize, characterize, and evaluate a new class of Ru(II) polypyridyl oligothiophenyl complexes.
- Determine the effect of oligothiophene chain length and coligand identity on biological activity.

## Target Structures

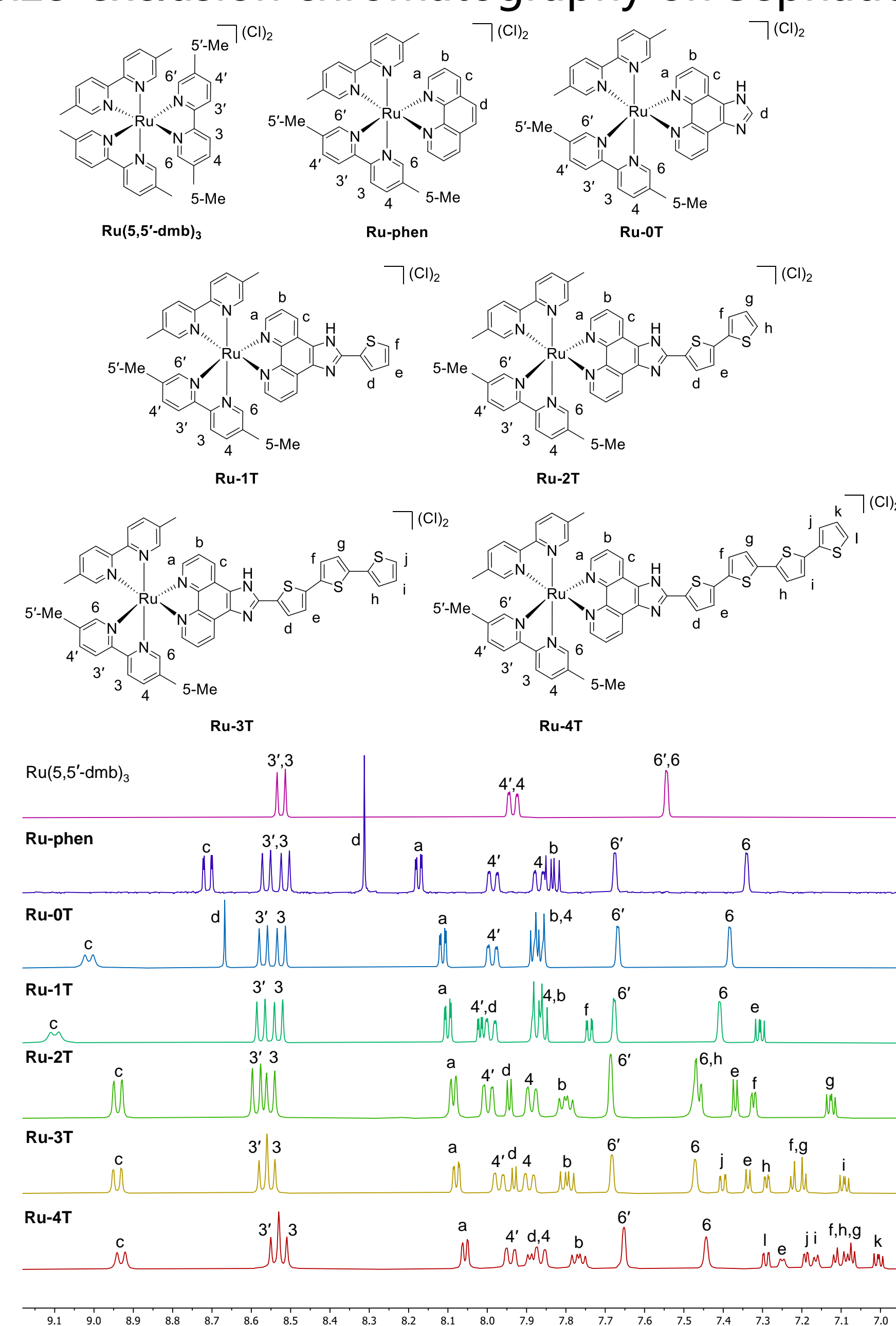


## Synthesis and Characterization



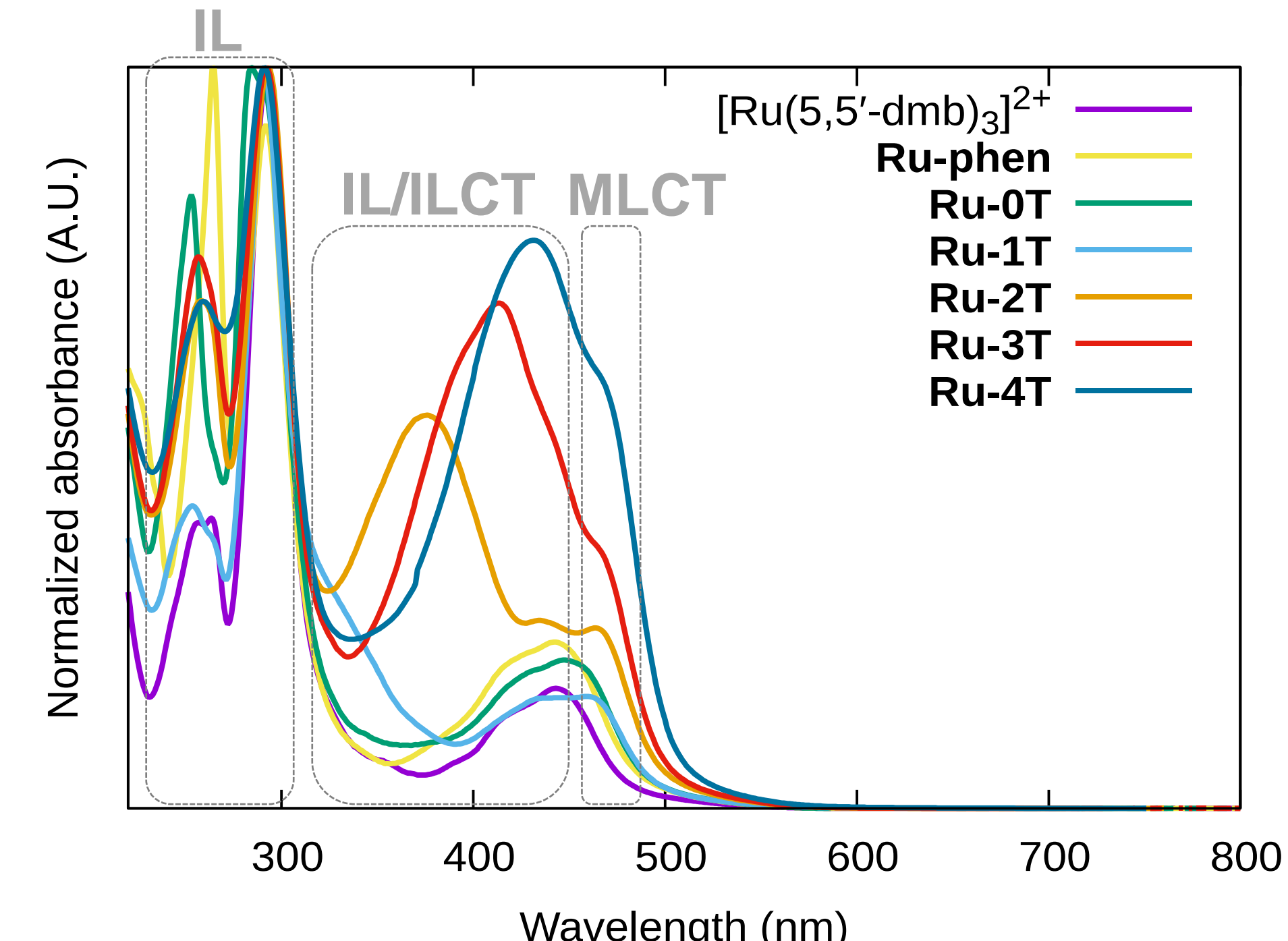
### Microwave-assisted synthesis of target complexes.

The  $Ru(5,5'-dmb)_2(Cl)_2$  precursor, phen and different IP- $nT$  ligands were heated at 180°C using microwave irradiation for 15 minutes.  $Ru(5,5'-dmb)_3$  was also synthesized by reacting  $RuCl_3 \cdot xH_2O$  and 5,5'-dmb under similar conditions. Crude products were isolated as  $PF_6^-$  salts and purified using silica gel column chromatography, followed by conversion to their corresponding  $Cl^-$  salt via anion exchange on Amberlite. Finally, the  $Cl^-$  salts were further purified using size-exclusion chromatography on Sephadex.



**Characterization by <sup>1</sup>H NMR.** Assignment of all <sup>1</sup>H signals was confirmed by 2D COSY (<sup>1</sup>H-<sup>1</sup>H) NMR. <sup>19</sup>F NMR was performed after purification to confirm that all  $PF_6^-$  ions were removed.

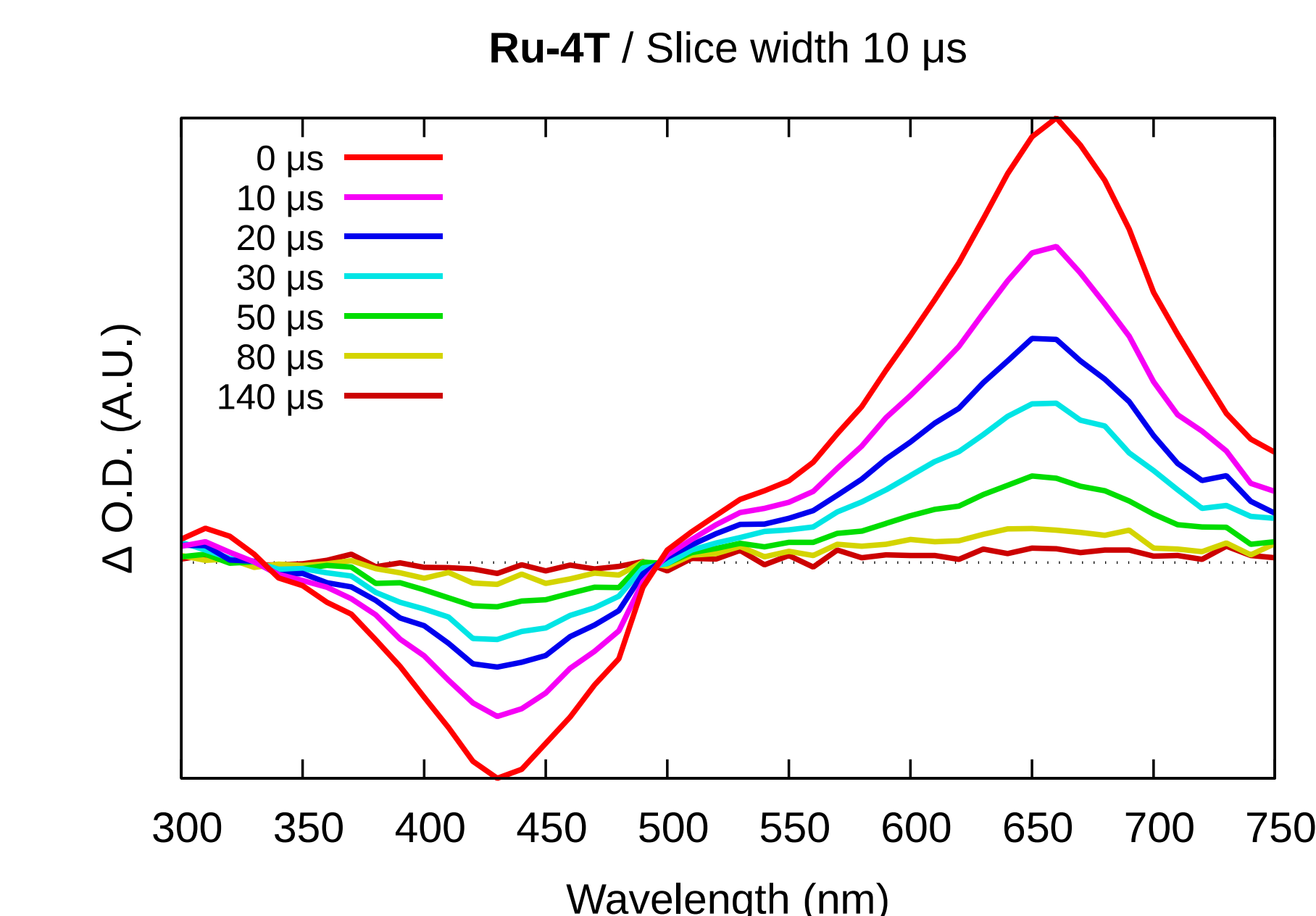
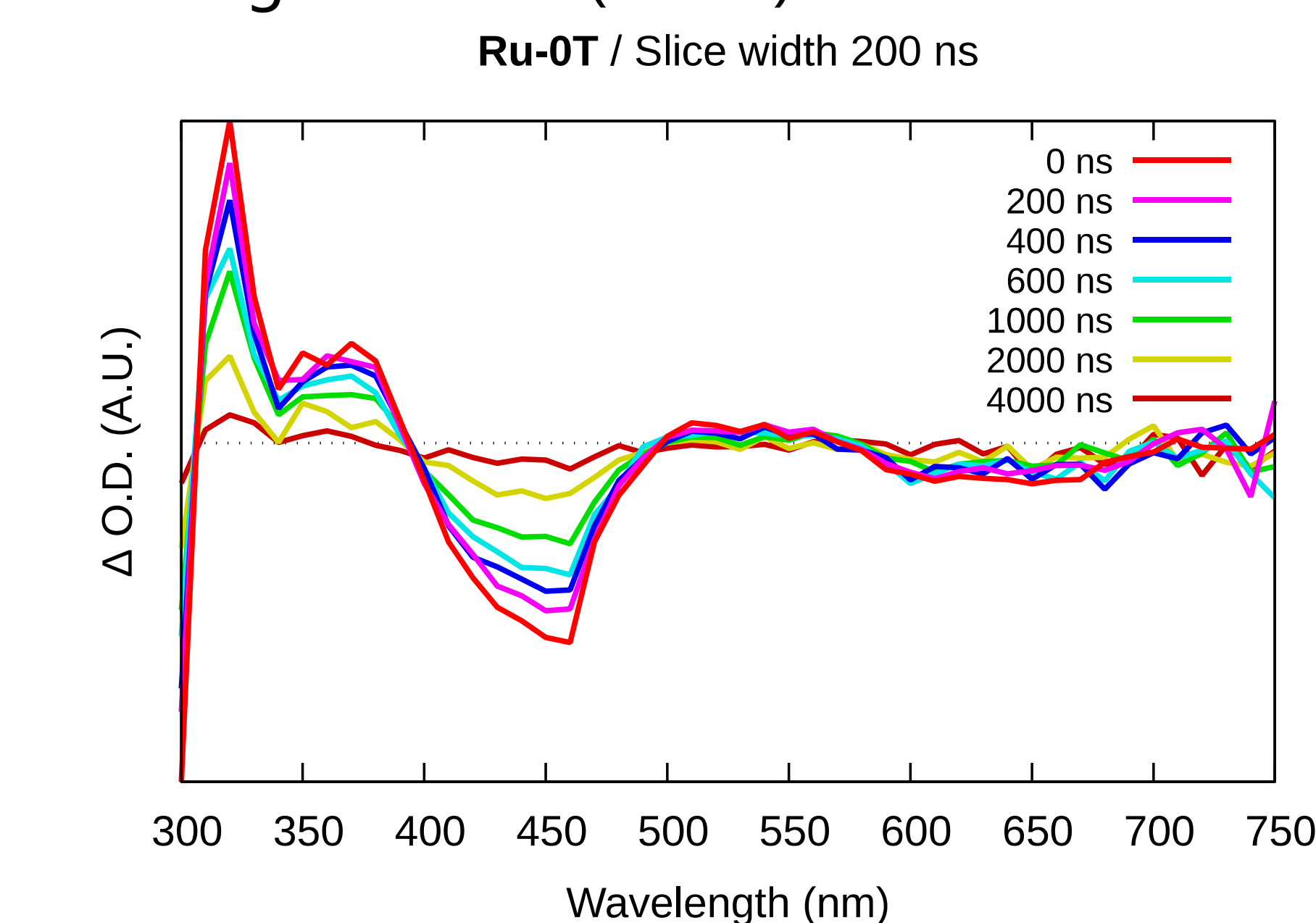
## Photophysical Characterization



### Steady-State Absorption Properties. UV-Vis

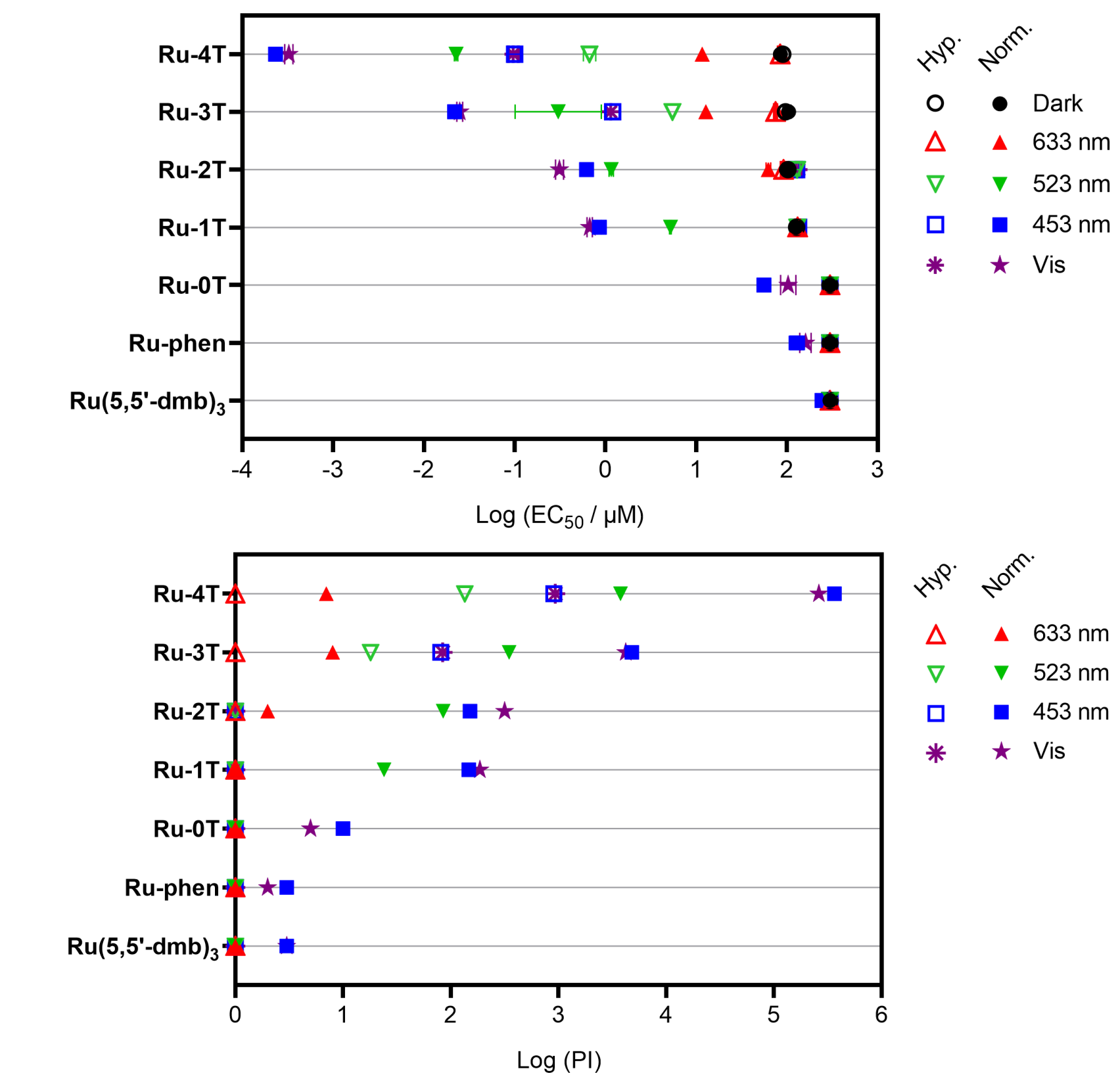
absorption profiles major features:

- <300 nm intraligand (IL)  $\pi\pi^*$  transitions on coligands and phen/IP
- 330-450 nm IL or ILCT  $\pi\pi^*$  involve  $nT$  of the IP- $nT$ . With increasing  $n$ :  $\uparrow$  intensity, red shift  $\rightarrow$
- $\sim 470$  nm  $Ru(d\pi) \rightarrow phen(\pi^*)$  metal-ligand charge transfer (MLCT) transitions



**Excited-State Absorption Properties.** The transient absorption (TA) spectra of Ru-0T and Ru-4T are compared above. Ground state bleach centred at 450 nm - depopulation of the <sup>1</sup>MLCT state. The ESA  $\sim 660$  nm is due to the <sup>3</sup>ILCT state.

## Photobiological Evaluation



**Light-triggered activity in SKMEL-28 cancer cells.** All of the complexes were relatively nontoxic in the dark ( $EC_{50} > 100 \mu M$ ). The light  $EC_{50}$  values decreased systematically with  $n$ , reaching low nM and a phototherapeutic index (PI) as large as  $4 \times 10^5$  with Ru-4T in normoxia. Ru-4T was a notable hypoxia-active compound, with PIs >100 down to 1% hypoxia.

## Conclusions

- A new family of Ru(II) polypyridyl oligothiophene based complexes has been synthesized, characterized, and evaluated for photobiological activity against cancer cells.
- The <sup>3</sup>ILCT state is key to achieving high photobiological activity.
- Increasing the number of thiophenes energetically positions the <sup>3</sup>ILCT state to be accessible upon photoexcitation.

## References

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