Evaluation of Porphyrus Envelope as a Novel Drug Delivery System for Photodynamic Therapy of Prostate Cancer

Erica Lin,[1,2,3] Mizuho Inai,[4] Sachiko Saito,[3] Norihiro Honda,[3,5] and Kunio Awazu[3,4,6]

[1]*Department of Applied Mathematics, Brown University, Providence, RI, U.S.A*
[2]*Nakatani RIES Program, Rice University, Houston, Texas, U.S.A.*
[3]*Graduate School of Engineering, Osaka University, Suita, Osaka, Japan*
[4]*Graduate School of Frontier Biosciences, Osaka University, Suita, Osaka, Japan*
[5]*Institute for Academic Initiatives, Osaka University, Suita, Osaka, Japan*
[6]*Global Center for Medical Engineering and Informatics, Osaka University, Suita, Osaka, Japan*

Prostate cancer is the second-leading cause of cancer-related death in men. Side effects from traditional therapies as well as increased treatment resistance have led to a search for more effective treatment modalities. Photodynamic therapy (PDT) has the potential to fulfill this need. PDT uses photosensitizers, light-sensitive drugs/dyes, which selectively accumulate in tumor cells. Light stimulation of the photosensitizers causes the formation of reactive oxygen species (i.e.: $^1O_2$) only in tumor cells, sparing normal, healthy cells. This study evaluated porphyrus envelope as a novel therapeutic agent for PDT. Porphyrus envelope is created by incorporating the protoporphyrin IX lipid (PpIX lipid), a photosensitizer, into the hemagglutinating virus of Japan envelope (HVJ-E). HVJ-E was used because it leads to immune cell recruitment and activation of anti-tumor immunity, and thus a higher therapeutic effect in deeper tissue. Additionally, HVJ-E's fragmented RNA induces apoptosis in cells via the RIG-I pathway. Direct cytotoxicity assays performed in this study found that HVJ-E alone and porphyrus envelope showed statistically similar decreases in cell survival rate in the absence of light stimulation/PDT. However, porphyrus envelope led to significantly lower tumor cell survival rate in the presence of light stimulation/PDT when compared to HVJ-E alone. The ideal irradiation wavelength (450 nm) was established using PpIX lipid's absorption spectrum. Thus, porphyrus envelope represents a novel drug delivery system that can be used to more effectively treat prostate cancer as well as enhance the treatment options of other types of cancers conducive to PDT treatment, including brain and skin cancers.

Evaluation of Porphyrus Envelope as a Novel Drug Delivery System for Photodynamic Therapy of Prostate cancer

Erica Lin,[1,2] Mizuho Inai,[3] Sachiko Saito,[4] Norihiro Honda,[4,5] and Kunio Awazu[3,4,6]

[1]Department of Applied Mathematics, Brown University, Providence, RI, U.S.A [2]Nakatani RIES Program, Rice University, Houston, Texas, U.S.A.
[3]Graduate School of Frontier Biosciences, Osaka University, Suita, Osaka, Japan [4]Graduate School of Engineering, Osaka University, Suita, Osaka, Japan
[5]Institute for Academic Initiatives, Osaka University, Suita, Osaka, Japan [6]Global Center for Medical Engineering and Informatics, Osaka University, Suita, Osaka, Japan

Contact: e_lin@brown.edu

Objective

To investigate whether porphyrus envelope can serve as a novel drug delivery system for photodynamic therapy of prostate cancer.

Background

Prostate Cancer

2nd leading cause of cancer-related death in men.

Increased Treatment Resistance + Side Effects

↓

Urgent search for more effective treatment modalities

Photodynamic Therapy (PDT)

Photodynamic therapy has shown potential to fulfill the need for better treatment options

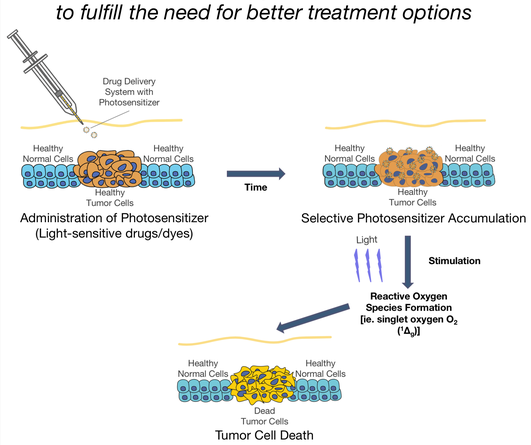


Figure 1: PDT Mechanism

Major Disadvantage:
Large dose necessary to produce consistent accumulation of photosensitizer in tumor
→Leads to unacceptable skin phototoxicity

Porphyrus Envelope (PE)

Hemagluttinating Virus of Japan (HVJ-E)

- Recruitment of immune cells and activation of anti-tumor immunity
  → Higher therapeutic effect in deeper tissue
- Fragmented RNA induces cell death via RIG-I pathway

+

PpIX Photosensitizer

- Derived from natural *heme* precursor
- Fast excretion and low toxicity

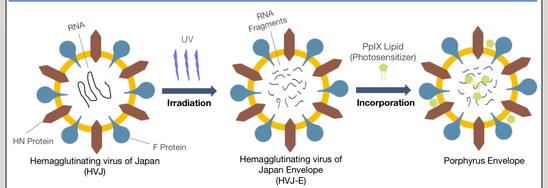
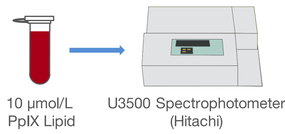


Figure 2: Synthesis of PE

Materials & Methods

PpIX Absorption Spectrum

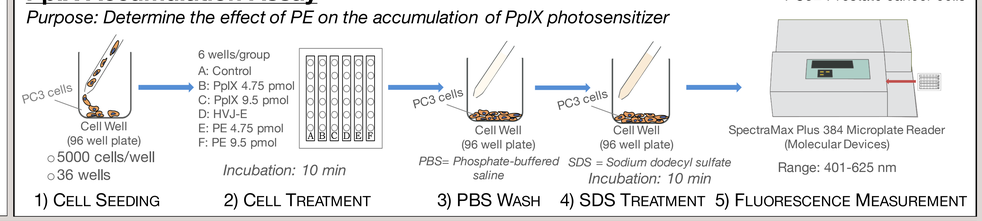
Purpose: Determine ideal irradiation wavelength



PpIX Accumulation Assay

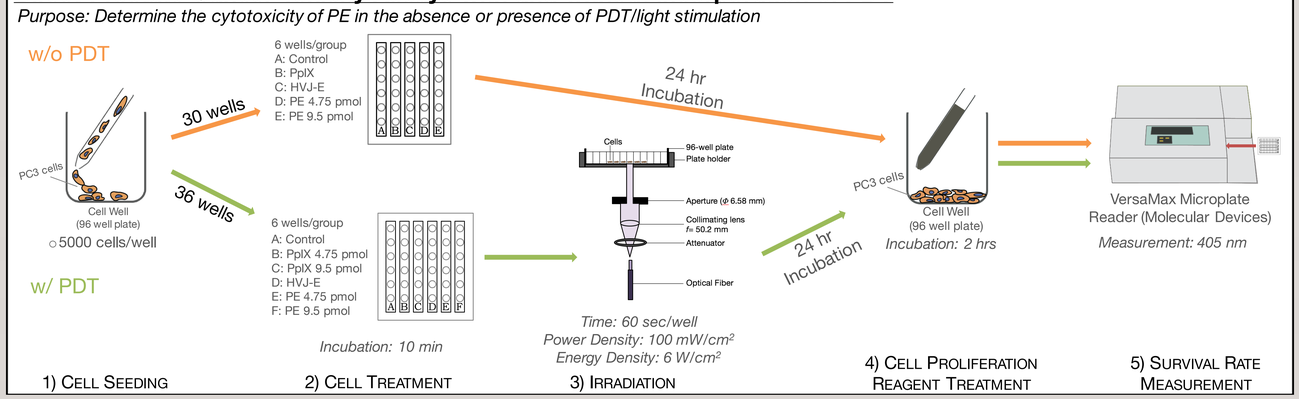
Purpose: Determine the effect of PE on the accumulation of PpIX photosensitizer

*PC3= Prostate cancer cells



Direct Prostate Cancer Toxicity Assays with and without incorporation of PDT

Purpose: Determine the cytotoxicity of PE in the absence or presence of PDT/light stimulation



Results

PpIX absorption spectrum indicates maximum absorption at 405 nm

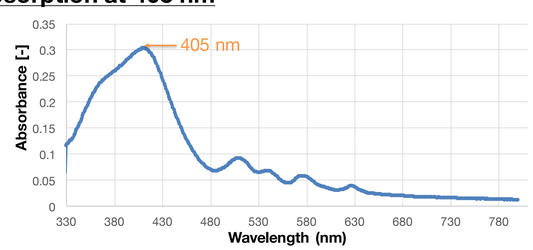


Figure 3: PpIX Absorption Spectrum

PpIX accumulation assay shows porphyrus envelope leads to higher PpIX accumulation

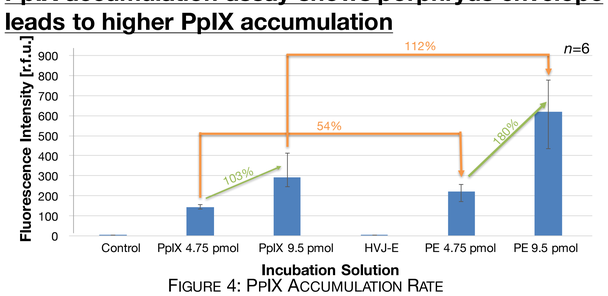


Figure 4: PpIX Accumulation Rate

In absence of light stimulation/PDT, porphyrus envelope retains similar prostate cancer (PC3) toxicity in comparison to HVJ-E alone

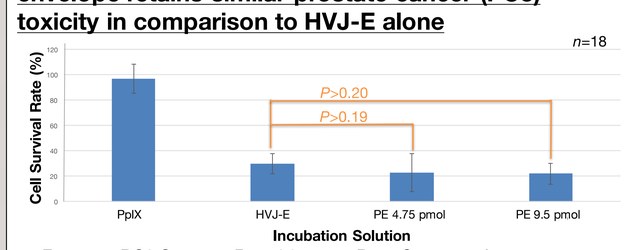


Figure 5: PC3 Survival Rate 24 hours Post-Solution Administration

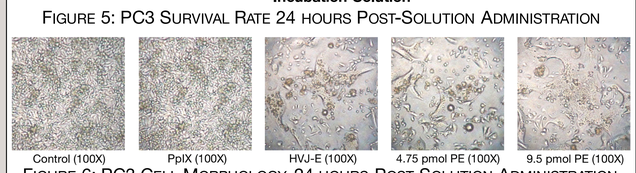


Figure 6: PC3 Cell Morphology 24 hours Post-Solution Administration

In presence of light stimulation/PDT, porphyrus envelope leads to significantly higher prostate cancer (PC3) toxicity in comparison to HVJ-E alone

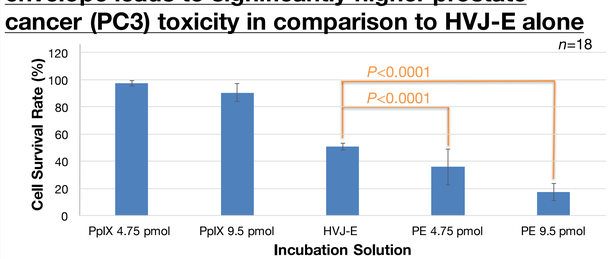


Figure 7: PC3 Survival Rate 24 hours Post-PDT

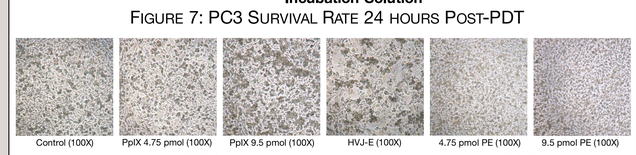


Figure 8: PC3 Cell Morphology 24 hours Post-PDT

Conclusions

Porphyrus envelope represents a novel drug delivery system that can be used to more effectively treat prostate cancer.

Impact

Experimentally Established Mechanism:

Administration of PE to *in vitro* system → **HVJ-E portion of PE induced cell death and led to greater accumulation of PpIX in tumor cells** → **Increased tumor cell death upon PDT exposure**

Impact of this study:

Use of PE as novel drug delivery system → **Selective targeting of tumor cells** → **More effective prostate cancer treatment and enhanced options for other cancers (ie. brain, skin)**

Future Research

- Continued investigation of optimal PpIX concentration
  — Minimum amount of PpIX lipid needed for creation of effective PE
- Evaluation of PpIX accumulation in healthy prostate tissue vs. prostate cancer cells
- Spheroid and *in vivo* studies
  — Comparison to *in vitro* results
  — Identification of optimal PE dosage for use in clinical settings

Selected References

Kawaguchi, Y. et al., *Efficient eradication of hormone-resistant human prostate cancers by inactivated Sendai virus particle*. Int J Cancer, 2009. 124(10): 2478-87.

Matsushima-miyagi, T. et al., *TRAIL and Noxa are selectively Upregulated in Prostate Cancer Cells Downstream of the RIG-I/MAVS Signaling Pathway by Nonreplicating Sendai Virus Particles*. Clin Cancer Res, 2012. 8(22): 6271-83.

Acknowledgements

This research was conducted as part of the Nakatani RIES: Research & International Experience for Students program generously funded by the Nakatani Foundation. For information on the Nakatani RIES program, please visit http://nakatani-ries.rice.edu/. Further research inquiries should be directed to the first author of this poster.