Organic Light-Emitting Diodes as Wearable Light Sources for Antiparasitic Photodynamic Therapy

Fernanda V. Cabral[1*], Cheng Lian[2], Saydulla Persheyev[2], Ifor D.W. Samuel[2], Martha S. Ribeiro[1]
[1] *Nuclear and Energy Research Institute – IPEN-CNEN/SP, Av. Prof. Lineu Prestes, 2242, 05508-000, São Paulo, SP, Brazil*
[2] Organic Semiconductor Centre, School of Physics and Astronomy, University of St Andrews, St Andrews KY16 9SS, UK
** fernanda.cabral@alumni.usp.br*

Abstract: Herein, we used a photosensitizing agent and red organic light-emitting diodes (OLEDs) against two strains of *Leishmania amazonensis* amastigotes, including a drug-resistant line. OLEDs are promising wearable light sources for photodynamic therapy of cutaneous leishmaniasis. © 2022 The Author(s)

1. Introduction

Flexible organic light-emitting diodes (OLEDs) hold promising wearable applications since they are thinner and weigh less than LEDs and can be easily adapted to human skin [1]. Thus, OLEDs are ideal for photodynamic therapy (PDT) by light bandages for ambulatory care [2]. Herein, we explored the use of OLEDs to inactivate wild-type (WT) and miltefosine-resistant (MF) amastigotes of *Leishmania amazonensis*. Indeed, Leishmania protozoa are dimorphic parasites that live extra (promastigotes) and intracellular (amastigotes) [3]. Thus, amastigotes are the parasite forms that persist in the infected host [4]. We also evaluated cytotoxicity on mammalian cells to measure the selectivity index (SI), once parasites dwell inside macrophages.

2. Material and Methods

Firstly, miltefosine cytotoxicity on macrophages was performed by the addition of serial dilutions of the drug (0-500 µM) for 48 h. For the PDT cytotoxicity, 1,9-dimethyl-methylene blue (DMMB, 0-3000 nM) was incubated for 10 min to allow the photosensitizer uptake. Then, cells were irradiated using a red OLED (671 ± 140 nm) at an irradiance of 6.5 mW/cm^2. Cell viability was measured by MTT assay. Thereafter, intracellular parasites were treated with either miltefosine (0-50 µM) or DMMB (0-750 nM)-PDT with OLEDs delivering 8 J/cm^2 for approximately 20 min. Following 48 h, parasites were quantified using optical microscopy. Results were determined by counting 100 cells per coverslip and expressed as the infection index, calculated by the percentage of infected macrophages x mean amastigotes number per macrophage.

3. Results

Our results showed that DMMB-PDT effectively reduced the number of intracellular parasites in both strains of *L. amazonensis* without promoting cytotoxic effects on mammalian cells, whereas miltefosine was able to kill only the WT cell line (Figs. 1 and 2).

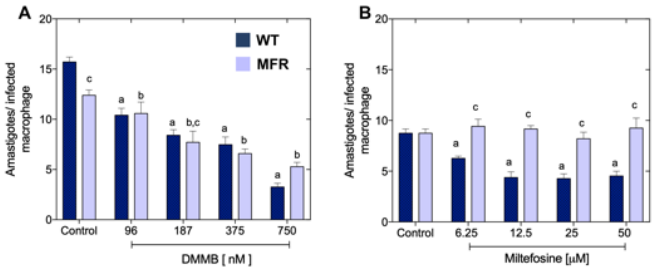

Fig. 1. **(A)** DMMB-PDT (0-750 nM) at 8 J/cm^2 and (B) miltefosine (0 to 50 µM) on intracellular amastigotes of *L. amazonensis* WT and MFR. "a" represents statistically significant differences between WT control and treated groups. "b" denotes statistically significant differences between MFR control and treated groups. "c" denotes statistically significant differences between PDT WT and PDT MFR. ($p < 0.05$).

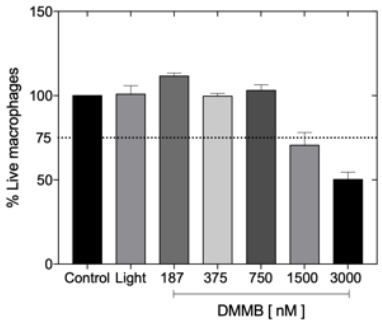

Fig.2. Macrophages treated with increasing concentrations of DMMB at a radiant exposure of 8 J/cm^2. Mean values ± SEM.

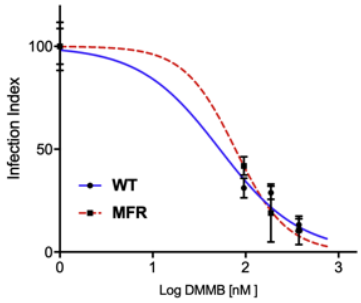

Fig. 3. Infection index of WT and MFR *L. amazonensis* treated with DMMB-PDT at 8 J/cm^2. Mean values ± SEM. The half-maximal effective concentration (EC_{50}) of DMMB is nearly 52 and 77 nM for the WT and MFR, respectively.

The selectivity index (SI), i.e., the ratio that measures the window between macrophages cytotoxicity (Fig. 2) and antiparasitic activity of the treatment (Fig. 3) is shown in Table 1. For WT *L. amazonensis*, PDT SI is about 3 times higher than that for miltefosine. For resistant amastigotes, PDT SI is more than 15-fold higher compared to miltefosine. These data suggest DMMB-PDT is a safe antiparasitic therapy.

Table 1. Selectivity index of intracellular amastigotes of WT and MFR *L. amazonensis* treated with miltefosine and DMMB-PDT at 8 J/cm^2.

	Miltefosine WT	Miltefosine MFR	PDT WT	PDT MFR
Selectivity index	7.76	> 0.92	22.59	15.32

4. Conclusion

These findings demonstrate the great potential of OLEDs as new wearable light sources for ambulatory treatment of cutaneous leishmaniasis by DMMB-PDT. Besides, DMMB-PDT could be used to fight resistant parasites.

5. Acknowledgments

The authors acknowledge financial support from CNEN, CAPES, and CNPq.

6. References

[1] Lian C, Piksa M, Yoshida K, et al. Flexible organic light-emitting diodes for antimicrobial photodynamic therapy. *npj Flex Electron* 2019; **3**(1): 1-6.
[2] Cabral FV, Lian C, Persheyev S, Smith TK, Ribeiro MS, Samuel IDW. Organic light-emitting diodes as an innovative approach for treating cutaneous leishmaniasis. Adv Mat Technol 2021, **6**:2100395.
[3] Burza S, Croft SL, Boelaert M. Leishmaniasis. *Lancet* 2018; **392**:10151.
[4] Ponte-Sucre A, Gamarro F, Dujardin JC, et al. Drug resistance and treatment failure in leishmaniasis: A 21st century challenge. *PLoS Negl Trop Dis* 2017; **11**(12): e0006052.