increased upon ALG-1 O/E, and among the upregulated miRNAs, 8 were also upregulated in long-lived, stress resistant glp-1 mutants, where ALG-1 is also increased. The proteomic analysis revealed differences in proteins related to nucleotide binding, reproduction and DNA mismatch repair – all processes commonly related to aging.
CONCLUSION
Together, these results support the notion that ALG-1 expression can be dynamically modified to confer protection against oxidative stress, contributing to the general healthspan of C. elegans.
Keywords: miRNAs, Oxidative Stress, Metabolism
Supported by: FAPESP

08589 - Poster Session

EB.01 - Melanoma cell migration in response to red and near-infrared low-level light
Carolina Gouvêa de Souza Contatori [1], Mayara Santana Pinto[1], Martha Simões Ribeiro[1]
[1]CELAP, Nuclear and Energy Research Institute (São Paulo, Brazil)

INTRODUCTION
Cell migration plays an important role in tissue formation and cancer progression. In vitro scratch assay has been used for many years to study cell migration to mimic the migration of in vivo cells, and, thus, to evaluate cancer growth. Low-level red and near-infrared light (LLL) can increase normal cell migration. However, the impact of LLL on tumor cells remains unclear.
OBJECTIVES
In this work, we aimed to evaluate the effects of a single LLL dose on melanoma cell migration.
MATERIALS AND METHODS
B16F10 (murine melanoma) cells were cultivated in RPMI medium with 10% of fetal bovine serum until they reached 80% confluency. The cell line was seeded in a 6-well plate at a density of $2x10^5$ cells/well in triplicate at two different moments. A wound scratch was performed to disrupt the confluent cell monolayer with a 10 μL pipette tip. Immediately after the injury, the cells were submitted to the LLL at two distinct wavelengths (660 and 780 nm) provided by a LED and a laser, respectively, delivering 3 different energies (1.3, 3.6, and 6 J) at an irradiance of 4.2 mW/cm^2. The control group was not irradiated. Cells were photographed immediately and at 3, 12, 24, and 36 h after the scratch. The wound closure was measured using ImageJ software. To evaluate the overall migration, we calculated the areas under the curve for each group.
DISCUSSION AND RESULTS
Cells exposed to the red laser at 6 J migrated slower than control. In contrast, LLL at 780 nm promoted faster cell migration when irradiated with 3.6 J.
CONCLUSION
These results suggest that low-level LEDs at 660 nm could prevent melanoma progression in higher energies. However, 780 nm should be avoided at middle energies.
Keywords: Melanoma, Photobiomodulation therapy, Scratch-wound assay
Supported by: CNPq and CAPES

08700 - Poster Session

EB.02 - Mechanisms of membrane protection by deuterated PUFA
Márcia Silvana Freire Franco [1]
[1]Departamento de Bioquímica, Universidade de São Paulo - Instituto de Química (São Paulo, Brasil), [2]Departamento de Física Aplicada, Universidade de São Paulo - Instituto de Física (São Paulo, Brasil), [3]Drug Discovery, Retrotope (Califórnia , United States)

INTRODUCTION
Polyunsaturated fatty acids (PUFAs) constitute one of the most abundant and important components found in membrane bilayers. PUFAs stabilize protein complexes and modulate membrane properties, ensuring homeostasis of organelles and cells. However, PUFAs are highly susceptible to oxidative damage through lipid peroxidation (LPO) chain reaction which triggers atherosclerosis, cancer and neurodegenerative diseases. Selective hydrogen replacement of bis-allylic PUFA hydrogens by deuterium offers protection against LPO, but the protection mechanism is not fully understood.
OBJECTIVES
To understand the protection mechanism by deuterium substitution, Giant Unilamellar Vesicles (GUV) were prepared with H-Lin-PC, in the presence of small amounts of D-PUFAs.
MATERIALS AND METHODS
We analyzed photo-induced oxidation in the presence of 1μM of Al(III) Phthalocyanine tetrasulfonic acid chloride. The initial steps of the membrane oxidation, which consists of lipid hydroperoxidation by singlet oxygen, are characterized by fluctuations and area expansion of the GUVs. Membrane permeabilization results from further oxidation steps, forming lipid truncates aldehyds.
DISCUSSION AND RESULTS
We show that the presence of 20% of D-PUFA in the 80%of H-Lin-PC matrix of vesicles, prevents substantially the fluctuation/area increase, and the loss of contrast. The presence of tocopherol, following the same proportion of D2-PUFA-PC in H-Lin-PC, is effective in preventing the formation of pores/membrane permeabilization, however it does not inhibit the formation of hydroperoxides, resulting in area fluctuation and increase.
CONCLUSION
These findings demonstrate that a small proportion of D-PUFAs is sufficient for the protection of both contact-dependent and contact-independent oxidation processes. Deuterium reinforced lipids offer membrane protection and the relief of the oxidative stress, mitigating several diseases.
Keywords: Lipid peroxidation, Giant Unilamellar vesicles, Deuterium reinforced lipids
Supported by: FUSP - RETROTOPE

08905 - Poster Session

EB.03 - Effects of photodynamic inactivation mediated by Zn(II) porphyrin on promastigote and amastigote forms of Leishmania amazonensis
Tiago Henrique dos Santos Souza [1,5], Camila Andrade[1], Fernanda Cabral[2], José Sarmento-Neto[3], Júlio Rebouças [3], Beate Santos[4], Martha Ribeiro[2], Regina Figueiredo[5], Adriana Fontes[1]
[1]Departamento de Biofísica e Radiobiologia, Universidade Federal de Pernambuco (Pernambuco, Brazil), [2]Centro de Lasers e Aplicações, Instituto de Pesquisas Energéticas e Nucleares (São Paulo, Brazil), [3]Departamento de Química, Universidade Federal da Paraíba (Paraíba, Brazil), [4]Departamento de Ciências Farmacêuticas, Universidade Federal de Pernambuco (Pernambuco, Brazil), [5]Instituto Aggeu Magalhães, Fundação Oswaldo Cruz (Pernambuco, Brazil)

INTRODUCTION
Photodynamic inactivation (PDI) has been attracting attention as an innovative technology to treat topical diseases, such as cutaneous leishmaniasis (CL) and infections caused by multidrug-resistant microorganisms. Zn(II) *meso*-tetrakis(*N*-n-hexylpyridinium-2-yl)porphyrin ($ZnTnHex\text{-}2\text{-}PyP^{4+}$) is a lipophilic water-soluble Zn(II) porphyrin with improved photophysical properties, high chemical stability, and cationic/amphiphilic character that can enhance its interaction with cells.

OBJECTIVES
Thus, this study aimed to investigate the PDI effects mediated by ZnTnHex-2-PyP $^{4+}$ on *Leishmania amazonensis*.
MATERIALS AND METHODS
Confocal fluorescence microscopy was explored to study the interaction of ZnTnHex-2-PyP $^{4+}$ with promastigotes. The PDI action was analyzed by cell membrane integrity, mitochondrial membrane potential ($\Delta\Psi m$), and cell morphology. Promastigotes were incubated with ZnTnHex-2-PyP $^{4+}$ for 5 min at 0.62 and 1.25 μM and irradiated by a LED (410 nm) for 1 or 3 min (2.3 and 3.4 J/cm ^{2}, respectively). PDI on amastigotes and the cytotoxicity on macrophages were also analyzed (3.4 J/cm ^{2}).
DISCUSSION AND RESULTS
Fluorescence microscopy revealed that parasites efficiently uptake ZnTnHex-2-PyP $^{4+}$ and displayed a punctate labeling pattern along with the cytoplasm. An intense $\Delta\Psi m$ depolarization was also observed, which in association with microscopy results, suggests that ZnTnHex-2-PyP $^{4+}$ may accumulate in the mitochondrion, or other well-defined structures close to it. Moreover, ZnTnHex-2-PyP $^{4+}$ at concentration as low as 0.62 μM led to the immediate inactivation of >95% of promastigotes, regardless of the light dose used. Loss of the fusiform shape and plasma membrane wrinkling were also observed. After a single treatment session in amastigotes, PDI led to a reduction of 70% in the infection index. No considerable toxicity was observed on mammalian cells.
CONCLUSION
Thus, PDI of *Leishmania* parasites showed *in vitro* efficiency at a submicromolar concentration of ZnTnHex-2-PyP $^{4+}$, with short pre-incubation and irradiation times. The results encourage further studies in CL pre-clinical assays and PDI of other microorganisms.
Keywords: Cutaneous leishmaniasis, photodynamic therapy, ZnTnHex-2-PyP4+
Supported by: CAPES, CNPq, FACEPE, FAPESP, IAM/FIOCRUZ, FINEP and INCT-INFo.

08549 - Poster Session

EB.04 - Blue light supports the aging of the skin of Swiss mice
Isabella Favarato Benedito[1], Nayara Zanetti Comério[1], Lucas G. Rabello[2], Clairton Marcolongo-Pereira[1], Maurício Lawrence Freitas[1], Maurício S. Baptista[3], **Orlando Chiarelli-Neto [1]**
[1]Faculdade de Medicina, Centro Universitário do Espírito Santo (Espirtio Santo, Brasil), [2]Faculdade de Medicina, Escola Superior de Ciências da Santa Casa de Misericórdia (Espirtio Santo, Brasil), [3]Departamento de Bioquímica , Instituto de Química, Universidade de São Paulo (São Paulo, Brasil)

INTRODUCTION
Solar irradiation causes skin aging by generating biomolecular damage and oxidized structure accumulations. Although the effects of ultraviolet (UV) light are well characterized, little is known about the effects of visible light. In vitro studies showed that UV-A and blue light stimulate redox processes with the generation of reactive oxygen species (ROS) and nitrogen (RNS) damaging skin cells. However, there are no in vivo studies that show the consequences of blue light on the skin and its association with UV-A. In addition there is no evidence of factors that block or even minimize damage.
OBJECTIVES
The objective here was to verify the effects of blue light (465nm) and in synergy with UV-A (365nm) in the skin of Swiss mice.
MATERIALS AND METHODS
Animal experimentation was approved by the Animal Use Ethics Committee (CEUA – 613296), with seven groups (G) (8 animals/group) divided into G0 (control group), G1 (UV-A), G2 (blue light) and G3 (UV-A+blue light). Groups G4, G5 and G6 followed the same order, but were added sunscreen (PP Photo Ultra ISDIN active unify 99) before each irradiation cycle in BlackBox Smart equipment (BioLambda – SP) with UV-A and blue doses, 20J/cm² and 100J/cm² respectively. All animals underwent dorsal trichotomy before irradiation, with subsequent photographic recording and biopsy in 3 times: zero, fifth and tenth irradiation. Statistics was performed by Origin 8.0.
DISCUSSION AND RESULTS
Through macro and microscopic analysis (HE technique at 40x magnification) it was noted that groups irradiated without PP, showed greater hyperpigmentation of the skin, indicative of melanogenesis. G3 stands out, in which 50% of the animals showed hyperkeratosis and 12% elastosis. Furthermore, accumulations were observed in G2 (blue light), suggestive of lipofuscin or hemosiderin.
CONCLUSION
Therefore, blue light was more harmful and accelerated the aging process, especially when associated with UV-A.
Keywords: Blue light, photoaging, Swiss mice
Supported by: Fundação de Amparo a Pesquisa e Inovação do Espírito Santo - FAPES

08669 - Poster Session

EB.05 - Sun-induced modifications on collagen molecular structure explored by fluorescence spectroscopy and advanced microscopy techniques
Giuseppe De Luca[1], Giuseppe Sancataldo[2], Valeria Vetri[2]
[1]Department of Biological, Chemical and Pharmaceutical Sciences and Technologies, University of Palermo (Exterior, Italy), [2]Department of Physics and Chemistry - Emilio Segrè, University of Palermo (Exterior, Italy)

INTRODUCTION
UV light is known to induce dangerous modifications on various biological molecule, which are linked to onset of pathological conditions. Since collagen is a structural protein ubiquitous in all human body, exploring the modifications induced by sun radiation and particularly focusing on UV range, is of crucial interest.
OBJECTIVES
We here present an experimental study based on steady state fluorescence spectroscopy and time resolved non-linear optical imaging aimed at characterizing collagen modifications at molecular level.
MATERIALS AND METHODS
Collagen was exposed to single wavelength UV lamp radiation (270 nm) and to solar simulator which emits a radiation with similar characteristic to the one Sun produces in term of spectrum and power.
DISCUSSION AND RESULTS
The results were a reduction of auto-fluorescence signal, attributed to the aromatic residues and to cross-links, and a decrease of the turbidity of samples, this highlighting an overall loss of supramolecular structure, associated to a solubility increase. CD spectra also reveal structural modifications which we investigate more in depth using 8-Anilino-1-naphthalenesulfonic acid (ANS). ANS is a fluorescent dye known to bind to collagen fibers and whose fluorescence is affected by its molecular environment, and in particular its hydrophobicity. We carried out fluorescence lifetime imaging (FLIM) measurements of ANS-stained samples, analyzed by phasor approach to observe alterations in the molecular structure of collagen triple helix. As already hypothesized from ANS steady state fluorescence measurement, FLIM analysis confirm a reduction of binding sites affinity, showing a high grade of lifetime distributions heterogeneity and an overall reduction of the ANS fluorescence lifetime.
CONCLUSION
Moreover, since we observe that irradiated collagen isn't able to self-organize in fibers, we conclude that solar radiation degrades the hydrophobic terminals of collagen triple helix, vital for its fibrillogenesis, and that ANS provide a useful and simple tool to investigate them.
Keywords: Collagen, UV-light, FLIM