



**FloraGLO**<sup>TM</sup>  
LUTEIN  
TOPICAL



## PROTECTIVE ACTION OF FLORAGLO<sup>TM</sup> LUTEIN AGAINST BLUE LIGHT ON HUMAN SKIN CELL LINES

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Blue light is the portion of the electromagnetic spectrum in the visible region with wavelengths ranging from 400-500nm that has adverse effects on a variety of skin cell lines. The purpose of the study was to identify the protective effect of FloraGLO<sup>TM</sup> Lutein Topical against the damaging effects of blue light on skin, due to its high absorptive capacity in blue light region. We investigated the effect of blue light in human epidermal keratinocytes (HEK) and human dermal fibroblast cells (HDF) on: cells viability, cell proliferation and the generation of reactive oxygen species.

### MATERIALS AND METHODS

#### Cell viability and proliferation assays - Shielding of blue light

**Design:** The effect of blue light on keratinocytes and fibroblast cells and the blue light shielding capabilities of FloraGLO Lutein was investigated using the CellTiter 96<sup>®</sup> AQueous One Solution reagent (MTS) assay by measuring viability and proliferation of metabolically active cells. Cells were grown to confluence in 96-microwell plates with a uniform number of cells in each well.

#### Treatments:

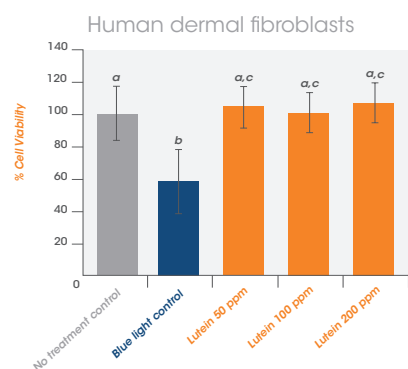
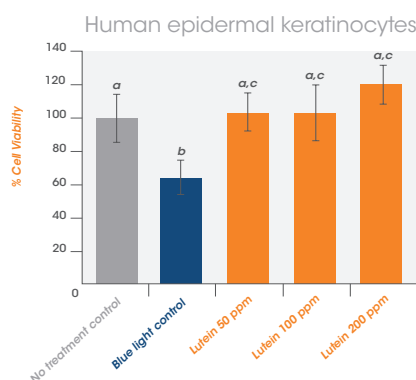
- No treatment control: cells not exposed to blue light at room temperature
- Blue light control: cells exposed to blue light without treatment of lutein
- FloraGLO Lutein solutions: 50, 100 and 200 ppm, in sterile DMSO (dimethyl sulfoxide)

**Test:** The microplates containing HEK and HDF cells were covered with a flat bottom sterile plate containing FloraGLO Lutein solutions prepared at concentrations of 50, 100 and 200 ppm and were directly exposed to a blue LED light source (476 nm, 1900 lux) positioned 30 cm above the culture plate for 9 hours. At the end of the 9 hour treatment period, cell viability assays were performed using the CellTiter 96<sup>®</sup> AQueous One Solution reagent (MTS) assay.

#### Results:

##### The % cell viability for different treatments in response to exposure of blue light

(The data points represented are the average of  $n = 6 \pm S.D$ )



P<0.05

Means with same letters are not statistically significant from each other

After 9 hours of exposure to blue light, the test showed a significant reduction in cell viability, 37-42%, compared to the no treatment control. However, when HEK and HDF cells were exposed to blue light in presence of FloraGLO Lutein at different concentration (50, 100 and 200 ppm), the cell proliferation was improved by 35-45 % compared to the blue light control, indicating that the FloraGLO Lutein solutions shielded blue light effectively.

**FloraGLO Lutein Topical is able to reduce the damage caused by blue light in both the cell lines.**

CellTiter 96<sup>®</sup> is a registered trademark of Promega Corporation

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## Reactive Oxygen Species (ROS) Assay

**Design:** Intracellular reactive oxygen species (ROS) activity and oxidative stress was measured using Oxiselect™ Intracellular ROS Assay kit, which measures hydroxyl, peroxy and other free radicals in cells, for different treatments in response to exposure to blue light.

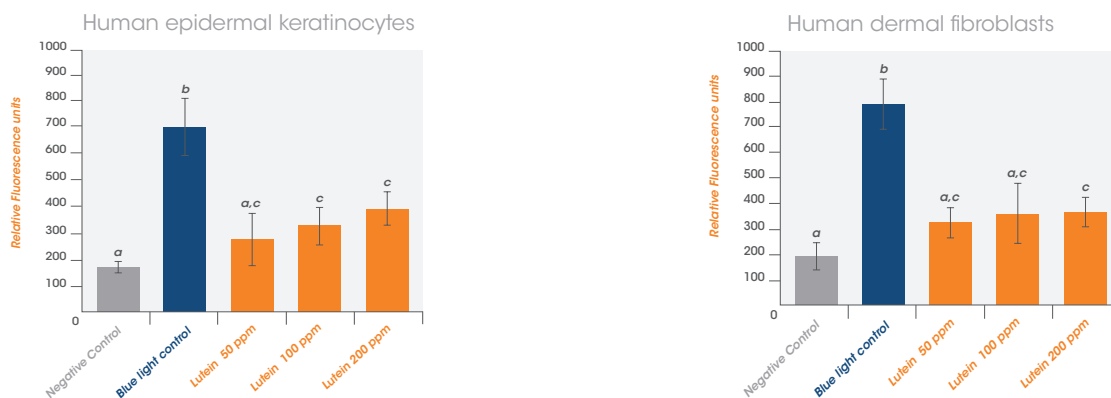
### Treatments:

- Negative control: cells not exposed to blue light at room temperature
- Blue light control: cells exposed to blue light without treatment of lutein
- FloraGLO Lutein solutions: 50, 100 and 200 ppm.

**Test:** Cells seeded in microplates were grown to confluence and treated with DCFH-DA (2', 7'-dichlorodihydrofluorescein diacetate) solution and incubated for 1 hour at 37 °C. Later, cells were washed 2-3 times with D-PBS (Dulbecco's Phosphate-Buffered Saline) and fresh medium was added and exposed to blue LED lights (476 nm) for 9 hours. At the end of treatment period, media was replaced with 100 µL of fresh medium and 100 µL of cell lysis buffer was added. After 5 minutes incubation, 150 µL of this medium was transferred to a black wall 96-well plate and fluorescence was measured.

### ROS assay for different treatments in response to exposure of blue light

data points represented are the average of  $n = 6 \pm S.D$



Means with same letters are not statistically significant from each other

**When HEK and HDF cells were exposed to blue light, the results showed a lower fluorescence signal with lutein treatments indicating that the presence of FloraGLO Lutein solutions at different concentrations (50, 100 and 200 ppm) can reduce the generation of ROS, by 44-60 % in HEK and 53-58 % in HDF, compared to the control (100%). The test confirms that lutein absorbs blue light and can protect the cells from oxidative stress generated by blue light exposure.**

## CONCLUSIONS

New studies show stunning properties of FloraGLO Lutein Topical. FloraGLO Lutein Topical is a powerful antioxidant that is able to:

- absorb harmful blue light rays
- reduce the exposure of blue light to keratinocytes and fibroblast skin cells and decrease cytotoxicity
- reduce Reactive Oxygen Species release that causes oxidative stress at cellular level.

FloraGLO Lutein Topical is an ideal plant-based active to reduce premature signs of aging induced by damaging visible light that surrounds us everyday.