

GREAF

Medicago sativa Extract: Resist the Blue Light (HEVL) Damage in Cell and Skin

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Introduction:

Blue light, also known as high energy visible light (HEVL), which wavelength is among $400 \sim 500$ nm in visible light. Studies have reported that HEVL can penetrates the epidermis of the skin and reaches the dermis, causing oxidative stress and inflammation in cells, which leads to apoptosis^[1,2]. It also causes melanocytes to produce more melanin and reduce the extracellular matrix, leading to photoaging. In this paper, the function of *Medicago sativa* Extract (MS) against HEVL damage was identified from molecular and cellular terms to the human skin. Data suggest that *Medicago sativa* Extract could resist the HEVL damage through ROS reduction.

Materials & Methods:

1. Determination of ABTS free radical scavenging in vitro

Under treatment of oxidizing agent, ABTS can be oxidized into ABTS·+ with green color. Dilute MS and positive control VC (PC) into 7-8 concentrations. Added 20 μl samples and 180 μl ABTS·+ reagent in a 96-well plate, reacting for 10 min, and then detected the OD value in 405 nm.

2. Measurement of ROS production in vitro

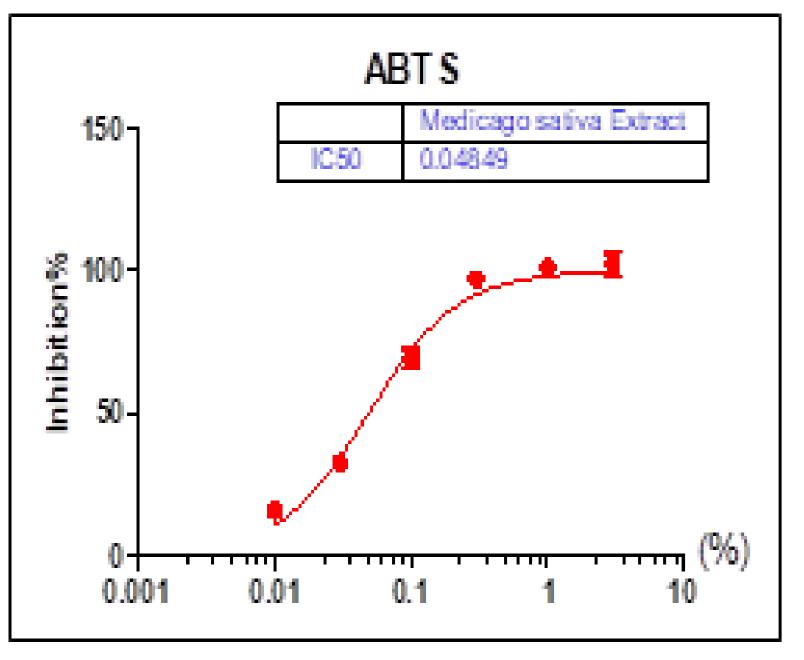
HaCaT cells were inoculated into a slide at 1×10⁵ cells per well overnight, and then exposed to 36 J/cm² HEVL. After treatment, cells were incubated with culture medium contained MS again for 18 h. Immunofluorescence probes were added to detect the ROS expression under a fluorescence microscope.

3. Clinical trials of HEVL irradiation

13 Volunteers wore customized opaque cuff containing several 1cm×1cm exposure areas, applied with or without 0.5%~5.0% MS samples in the exposed area. 10 min later, the forearm was irradiated with HEVL for 30 J/cm² or 60 J/cm², quantitatively detected the skin ITA°, Melanin and Erythem value by CK probe, meantime took pictures by VISIA to analyze the red area.

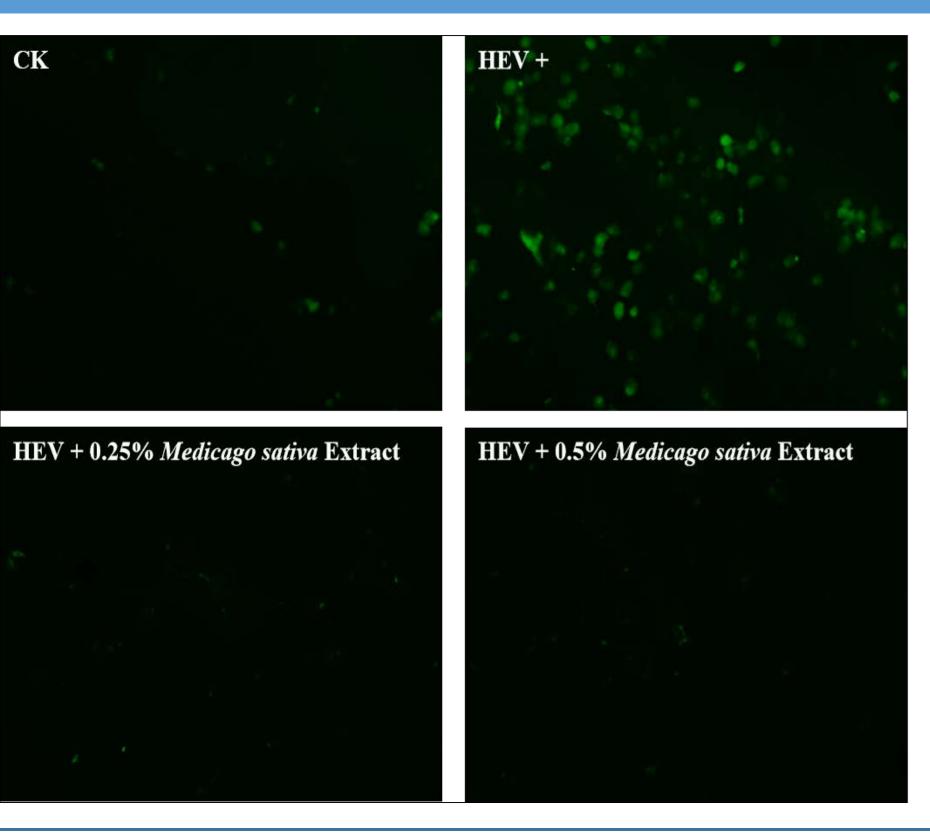
Results & Discussion:

Medicago sativa Extract scavenged ABTS free radicals



The IC50 of *Medicago sativa*Extract was 0.048%, indicating it can scavenge ABTS free radicals.
The inhibition rate reached to 97.5% at 0.3% concentration.

Medicago sativa Extract reduced HEVL-induced ROS free radicals



The cellular ROS level was increased after 36 J/cm² HEVL irradiation. While treated with 0.25% and 0.5% MS, ROS amount was reduced to normal group.

Medicago sativa Extract reduced HEVL damage in human skin ITA° change Melanin content Erythema content -90% **-80%** HEV 30J/cm² HEV 30J/cm² HEV 30J/cm² **HEV + 1%MS** HEV + **HEV + 5% MS** HEV + **HEV + 2% MS** HEV + Vehicle

The ITA°, Melanin and Erythem value were reduced in a dose-dependent manual when applied with 0.5%~5.0% MS after HEVL, indicating that *Medicago sativa* Extract could resist the damage of blue light on human skin.

Conclusions:

Medicago sativa Extract has obvious ability to clear ABTS free radicals, reduce cellular ROS production, and also decreased the skin ITA°, melanin and erythem caused by HEVL irradiation. It will be applicated in photo protection and skin care.

References:

- 1. Yuya Nakashima, et al. Blue light-induced oxidative stress in live skin. Free Radical Biology & Medicine 108 (2017) 300-310.
- 2. Alexa C. Wall, et al. Oxidative stress and endoreduplication induced by blue light exposure to CHO cells. Mutat Res Gen Tox En 841 (2019) 31-35.