

Preventive effects of water extract of *Chlorella* on MMP1 production induced by PMA and IL-1 β in human skin fibroblast

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Purpose: Solar UV radiation damages human skin, affecting skin tone and resiliency and leading to premature ageing (photoaging). Oxidative stress caused by UV radiation, ozone, hydrogen peroxide & free radicals are known to increase PKC activity. Skin damage by oxidants may lead to activation of PKC, thus increasing Matrix metalloproteinases (MMPs) expression and collagen degradation. UV radiation activates cell surface growth factor and cytokine receptors, and therefore mimics the actions of receptor ligands. Administration of *Chlorella* has been shown to play some biochemical functions, such as promoting the growth rate of animals, ameliorating blood glucose and lipids in animals, boosting immune function, preventing stress-induced ulcer, and influencing oxidative stress in ethionine treated rats. In many cosmetic products also claim to contain the components of extract of *Chlorella*. In this study, the effects of extract of *Chlorella* on alleviating skin ageing were studied by inducing MMP production through activation of PMA and IL-1 β .

Method: MMP1 production was induced by PMA (100nM, a PKC activator) or IL-1 β (2ng) + PDGF-BB (10ng) treatment in Human skin fibroblast (966SK) cells, which were planted into 96-well plate (1×10^4) 24h prior to use. Water extract of *Chlorella* [WEC257] (2 or 1 mg/ml), vitamin C (125 μ M) or MMP inhibitor GM6001 (0.4nM) were then added and incubated for 5 hours prior to MMP1 concentration assay (ELISA kits). Concentrations of tissue inhibitor of metalloproteinase (TIMPs) after the same treatment, is the endogenous inhibitor of MMP, were also measured.

Results: Vitamin C, GM6001 and both doses of WEC257 all prevented IL-1 β + PDGF-BB-induced MMP1 production ($p < 0.05$, t-test). However, PMA-induced MMP1 production was only significantly prevented by both doses of WEC257. High dose of WEC257 also statistically significant increased production of TIMP1 ($p < 0.05$, t-test).

Conclusion: This study shows that application of WEC257 prevent MMP1 production possibly through a PKC activation pathway in skin fibroblast cells.

Protective effects of water extract of *Chlorella* on short UV wave-induced cell damage in human skin fibroblast

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Purpose: Short wave of UV light is known to possess higher energy than long wave to penetrate materials and cause damage to skin. Cell damage caused by UV radiation can lead to cell death and it is also believed that this damage is due to oxidative damage. Administration of *Chlorella* has been shown to play some biochemical functions, such as promoting the growth rate of animals, ameliorating blood glucose and lipids in animals, boosting immune function, preventing stress-induced ulcer, and influencing oxidative stress in ethionine treated rats. In many cosmetic products also claim to contain the components of extract of *Chlorella*. However, the real effects of extract of *Chlorella* on skin protection have not been published. Therefore, we treated human skin fibroblast cells with and without water extract of *Chlorella* (WEC257) during the short UV wave exposure to study whether the extract possessed any skin protection from UV light damage.

Methods: Human skin fibroblast (966SK) cells were planted into 96-well plate (1×10^4) 24h prior to application of WEC257 (2 or 1mg/ml), Vit C (150 μ M), or Vit E (25 μ M). The cells were then exposed to 2 consecutive of day short wave (254nm) UV for 30min 24h later. During the UV treatment, cell mediums were either replaced with PBS or remained with the same culture condition. After the second UV exposure, the cells were then cultured with normal culture medium for further 24, 48, or 72h prior to cell proliferation assay (XTT).

Results: Cell proliferation showed that short UV wave treatment for 2 consecutive days caused cell death as agreed with other investigators. When PBS was replaced by culture medium during the exposure of UV light, none of the drug pre-treatments effectively protected the cells. However, when the cells cultured in the drug-contained culture medium during the UV exposure, the cells were well protected from the damage caused by short UV wave in WEC257-treated groups 24, 48, or 72 h after the exposure. In the same experiments, the treatment of Vit C and Vit E could not prevent the UV exposure-caused cell death.

Conclusion: This study shows that treatment of WEC257 has cell-protection from UV radio-hazard.