## In vitro Biological Activities of the Anti-aging Potential of Dimocarpus longan Leaf Extracts

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## ABSTRACT

The longan (Dimocarpus longan Lour.) leaves, which are the underutilized part of trimming longan trees to increase their fruit productivity, were of interest in this study. They have been reported to contain phytochemical components which might be used for anti-aging purposes. In this study, longan leaves were extracted using two different solvents including 95% ethanol and 50% ethanol, named ethanolic extract (ET) and hydroethanolic extract (HE), respectively. The extracts were investigated for antioxidation properties, inhibition of hyaluronidase, collagenase, MMP-2 and MMP-9 together with the determination of their total phenolic and flavonoid content. Additionally, HPLC-fingerprinting of the extract was performed. The results revealed that HE had a higher yield with the remarkable property of superior in vitro biological activity compared with ET. HE showed radical scavenging activity on DPPH and hydrogen peroxide with  $IC_{50}$  of  $30.03 \pm 7.64$  and  $71.40 \pm 15.30$ µg/ml, respectively. Moreover, it showed inhibition of lipid peroxidation with  $IC_{50}$  of 537.01 ± 42.32 µg/ml. For inhibition against hyaluronidase and collagenase, HE was detected with  $IC_{50}$  of 234.80  $\pm$  21.52 and 314.44  $\pm$  62.14 µg/ml, respectively. The extract also demonstrated MMP-2 and MMP-9 inhibition, which is more potent than gallic acid as determined by zymography at 1.0 mg/ml. In conclusion, hydroethanolic extract (HE) of longan leaves presented high potential as in vitro antioxidant and inhibitor of enzymatic

## activities. It might be a promising approach to the further development of anti-aging products.

**Keywords:** Longan leaves, Antioxidant activity, Anti-hyaluronidase activity, Anti-collagenase activity, Inhibition of MMP-2 and MMP-9 assay

## **INTRODUCTION**

Aging is associated with a progressive deterioration of cells that is a major risk factor for developing many degenerative diseases such as cardiovascular disease, neurodegenerative disease and even skin aging (Denicola and Lopez-Alarcon, 2013). Skin aging involves the degradation of extracellular matrix (ECM) in both epidermal and dermal layers. The results lead to skin dryness, thinning and sagging, while the loss of structure and integrity diminishes the skin's ability to protect the body and detect changes in temperature and pressure. Aging skin tends to be sensitive to infection, trauma and pressure ulcers (Davies, 2008).

Aging skin is caused by intrinsic and extrinsic factors. Exposure to sun rays and air pollution increases Reactive Oxygen Species (ROS) generation in the skin and induces oxidative stress. Oxidative damage may lead to lipid peroxide formation, mitochondrial and DNA damage and protein and gene modification, which alter protein structure and function. ROS also increases matrix metalloproteinase enzymes (MMPs) in the skin cells (Mukherjee et al., 2011). The combined actions of collagenase (MMP-1), 92 kDa gelatinase (MMP-2) and 72 kDa gelatinase (MMP-9) can fully degrade skin collagen and components of the elastic network. Collagenase is capable of cleaving in the triple helical domain of collagen. Furthermore, the denatured collagen can be further broken down by gelatinase (Jenkins, 2002). Hyaluronic acid is a predominant voluminous molecule in ECM. It plays a significant role in the maintenance of extracellular space, facilitate the transportation of nutrients and preserve tissue hydration (Manuskiatti and Maibach, 1996). Reduction of oxidative stress, as well as inhibition of enzymatic activities of collagenase, hyaluronidase, MMP-2, and MMP-9, can be used to evaluate anti-aging activity.

While considerable attention has been paid to plant components to replace synthetic additives, phytochemical components such as phenolics, flavonoids and terpenoids have been widely investigated and have shown anti-aging properties (Kim et al., 2008; Mukherjee et al., 2011).

Longan (*Dimocarpus longan* Lour.) is a member of the Sapindaceae family, widely distributed in China and Southern Asia, including Thailand, Vietnam and the Philippines (Yuge, 2012). Longan leaves have been reported to contain phenolics, flavonoids, terpenoids and sterols (Yuge, 2012; Xue et al., 2015). The longan leaf extract has shown high antioxidant activity, anti-hepatitis