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Anti-inflammatory Effect of Shea Butter Extracts in Canine Keratinocytes

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Abstract : Shea butter (*Vitellaria paradoxa*) is a fat extracted from shea tree nuts and contains relatively high levels of non-glycerides. Triterpenes, the main non-glyceride component, exhibit a variety of biological activities such as antitumor, antibacterial, and anti-inflammatory. Shea butter extract (SBE) has been used to treat various skin problems such as burns, eczema, and rash in human medicine, but little is known about the activity of SBE on canine skin. This study evaluated the cytotoxicity and anti-inflammatory effect of SBE in canine keratinocytes. Cytotoxicity of lipopolysaccharide (LPS, 5-50 ng/mL) and SBE (50-200 μg/mL) was evaluated using the CCK-8 assay. Non-cytotoxic concentrations of LPS and SBE were administered to canine cell cultures to evaluate anti-inflammatory effects. To evaluate the anti-inflammatory activity of SBE, the levels of IL-1β, IL-8, IL-12, and TNF-α were measured using ELISA kits. The concentration of each cytokine was quantified in control, LPS-treated, LPS + SBE-treated groups. Increased levels of IL-1β, IL-8, and IL-12 were found in LPS-treated groups relative to control groups. LPS + SBE-treated groups showed a lower level of IL-1β, IL-8, and IL-12 than LPS-treated groups. These results suggest that SBE may have application as a topical agent for canine inflammatory skin diseases. However, further *in vivo* study is needed to evaluate the safety and efficacy of SBE in dogs.

Key words **:** Shea butter extracts (SBE), anti-inflammatory, cytokine, keratinocyte, dog.

Introduction

Shea butter is a fat extracted from Shea trees (*Vitellaria paradoxa*) which grows predominantly in Africa (11,18). Shea butter contains higher levels of non-glyceride constituents compared to typical vegetable oils (6,12). The primary non-glyceride constituents are triterpenes such as lupeol, butyrospermol, α-amyrin, and β-amyrin (22). Triterpenes are bioactive compounds, and the composition of the trierpenes in shea butter have no striking regional differences (2). Triterpenes are reported to exhibit a variety of biological activities such as antitumor, antibacterial, and anti-inflammatory activities (3,8,9,30).

Canine Skin is the largest organ of the body and is exposed to numerous biological or environmental stresses; the body responds to changes caused by stress factors to restore tissue and maintain tissue hemostasis (6,27). The epidermis is the outermost layer of the skin and acts as a physical barrier against various agents that are physically, chemically, or biochemically harmful (7,15,27). Exogenous stimuli induce the biosynthesis of inflammatory mediators such as leukotrienes and prostaglandins, causing an immune response (7,14,17,28).

Keratinocytes are the predominant cell type of the epidermis and synthesize a variety of pro-inflammatory and anti-inflammatory cytokines, including TNF-α, IL-1, IL-6, IL-8, and I-10 (14,17). In several skin diseases, abnormal production of cytokines occurs, and this production is belived to contribute to the pathogenesis of inflammatory dermatosis (20,24).

In veterinary dermatology, the use of topical therapy to manage of skin diseases has been increasing (21,23). Topical agents are applied directly to the skin and have been used to treat numerous skin diseases, including allergic and infectious disorders (23). In allergic skin diseases, topical therapy reduce inflammation and pruritus by desensitizing or improving the barrier function of skin (21). Numerous commercial topical agents have been developed and used in veterinary dermatology (10).

Several studies have shown the anti-inflammatory effect of shea butter extract (SBE), indicating potential for reducing skin stress induced by environmental factors (6,13,15,25). Shea butter has been used to treat various skin problems such as eczema, dry skin, burns, rashes, and wrinkles in humans (1).

Although shea butter is reported to have beneficial effect on skin, little is known about the activity of SBE on canine skin. In this study, we evaluated the cytotoxicity and anti-inflammatory effects of SBE in lipopolysaccharide (LPS)-induced inflammation in canine keratinocytes, using the CCK-8 assay and ELISA.

Materials and Methods

Shea butter extracts (SBEs)

Refined shea butter extracts (ULAB SWITZERLAND, Co; Korea) were dissolved in dimethyl sulfoxide (Sigma-Aldrich; Korea) to make a stock solution (100 mg/mL) and stored at −80°C until required (30). Lipopolysaccharide (LPS) was dissolved in sterile phosphate-buffered saline (PBS) and prepared as a 1 μg/mL stock solution.

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