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ANTI-INFLAMMATORY ACTIVITIES OF *VICIA CANESCENCE* AND *VICIA HIRSUTA* AERIAL PARTS

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Abstract

Many pharmacological activities have been reported in Papilionaceae family. The aim of present study was to investigate anti-inflammatory activities of *Vicia canescence* and *Vicia hirsuta* aerial parts. Inhibition of carrageenan induced edema was used to evaluate anti-inflammatory activity. The effect was dose-dependent. At all tested doses, extract produced statistically significant inhibition of inflammation when compared to the control group. V. canescence extract was more potent than V. hirsute extract (88.1 vs. 86.8% inhibition at 800 mg kg-1 i.p.) but no statistically difference was observed between them ($p>0.05$). V. canescence extract at 200 mg kg-1 (62.8%) and V. hirsute extract at 400 mg kg-1 (67.7%) were equipotent with diclofenac ($p>0.05$). Extracts were safe and did not exhibit any toxicity up to 3 g kg-1. This study indicates the potential therapeutic uses of *V. canescence* and *V. hirsuta* as potent antiinflammatory agents.

Key words: *Vicia canescence*; *Vicia hirsuta*; Carrageenan;

Introduction

Inflammation is a part of the complex process, which is frequently associated with pain and involves occurrences such as the increase of vascular permeability, increase of protein denaturation and membrane alteration. When cells in the body are damaged by any microbials, physical or chemical agents, the injury is in the form stress. Inflammation of tissue is due to response to stress. It is a defensive response that is characterized by redness, pain, heat, and swelling and loss of function in the injured area [1]. When tissue cells become injured they release kinins, prostroglandins and histamine. These cause increased vasodilatation and permeability of the capillaries. This leads to increased blood flow to the injured site [2]. Non-steroidal anti-inflammatory drugs (NSAIDS) and corticosteroids are the most widely used medications in the world, due to their efficacy in reducing pain and inflammation [3]. Although the traditional drugs are effective in relieving pain and inflammation, they are associated with adverse effects including alterations in blood pressure, hepatic injury, platelet inhibition and the significant risk of serious gastrointestinal and cardiovascular events in chronic use [4,5]. Nowadays, the development of drugs from natural sources is recommended to overcome the side effects of many of the synthetic drugs. Recent research on medicinal plants has generated a great deal of information about the biologically active chemical components that are responsible for the claimed medicinal effects [6]. Vicia genus (Papilionaceae) has 45 species in Iran [7]. V. Sativum has protective role in cancer invasion and metastasis, insecticidal activity and also shows hepatoprotective activity [8]. Antioxidant activities of V. sativum [8], V. faba [9], V. cracca and V. sativa [10] and V. canescence [11] have been reported previously. Also anti-inflammatory and antinociceptive activity of V. sativa [12] and antimicrobial and cytotoxic activity of V. faba [13] have been reported. We have recently reported good antioxidant, antidepressant and antihemolytic activities of V. sojakii [14,15].

To the best of our knowledge, anti-inflammatory activities of *V. canescence* and *V. hirsuta* have not been reported to date and nothing was found about these activities. The aim of the present work was to determine the anti-inflammatory activities of V. canescence and *V. hirsuta* aerial parts in mice in carrageenan induced edema in order to understand the usefulness of this plant in medicine.

Methods

Plants materials and preparation of extract

V. canescence and V. hirsuta aerial parts were collected, in July 2014 from Sari, Iran. The sample was authenticated by Dr. B. Eslami and the voucher specimen was deposited (No. 545 and 546) have been deposited in the Sari School of Pharmacy herbarium. Plant materials were dried under dark conditions at r. t. for 2 weeks. The dry materials were milled, obtaining 2-3 mm particles and then extracted by methanol for 24 h at room temperature. The extracts were then separated from the sample residues by filtration through Whatman No.1 filter paper and repeated three times. The resulting extracts were concentrated over a rotary vacuum at 35-40 °C until a crude solid extracts were obtained which then were freeze dried (MPS-55 Freeze-drier, Cperon, Korea) for complete solvents removal.

Animals

All experiments were performed on male Wistar rats (180-200 g) obtained from Institute Pasteur. Animals were randomly housed in polypropylene cages at an ambient temperature, 25 ± 1°C and 45-55% relative humidity, with a 12 h light: 12 h dark cycle (lights on at 7 a.m.). The animals had free access to standard pellet and water and libitum. Experiments were conducted between 9:00 and 14:00 h. The experiments were conducted according to the norms of Committee for the Purpose of Control and Supervision of Experiments in Animal. Each animal was used once only. Seven mice were used in each experiment.

Antiinflammatory activity (Carrageenan-induced paw edema test)

Carageenan (50 µL of 1% suspension, Sigma Chemicals Co. USA) was injected into the sub planar tissue of the right hind paw of each rat. Extract (200-800 mg kg-1) or diclofenac sodium (50 mg kg-1) was administered i.p. to rats 1 hour before carageenan injection. The volume of edema was measured prior and 3 h after carageenan injection. The degree of swelling was the ratio of the volume of hind paw before to after carageenan treatment [16-18].

Non-fatal dose

Three mg kg-1 doses of extracts were injected to separated groups of seven. After 48h, any mortality was considered as the maximum non-fatal dose.

Statistical Analysis

Results are expressed as means ± SD. One-way

analysis of variance (ANOVA) followed by Newman-Keuls multiple comparisons tests were used. Differences with $p<0.05$ were considered significant.

Results and Discussion

Inflammation is a part of the complex biological response of vascular tissues to harmful stimuli, such as pathogens, damaged cells or irritants. It is characterized by redness, swollen joint that is warm to touch, joint pain, its stiffness and loss of joint function. Inflammation is a defence reaction caused by tissue damage or injury. The primary objective of inflammation is to localize and eradicate the irritant and repair the surrounding tissue. There are basic two types of inflammation Acute and chronic inflammation. Acute inflammation is of short duration and represents the early body reactions. Acute inflammation may be an initial response of the body to harmful stimuli. An increased movement of plasma and leukocytes, especially granulocytes from the blood into the injured tissues is observed. A cascade of biochemical events propagates and matures the inflammatory response, involving the local vascular system, the immune system and various cells within the injured tissue. Mast cells in the tissues, the key players of inflammation, are loaded with mediators of inflammatory response[19,20]. Phagocytes produce reactive oxygen species (ROS). Macrophages and activated platelets release interleukin-1 which causes fever by stimulating the release of prostaglandins (PGs). Chemical mediators such as histamine and bradykinin induce the production of PGs and leukotrienes with a role to potentiate the plasma exudation. These potent mediators of inflammation are derivatives of arachidonic acid [19,20]. Carrageenin-induced edema is a non-specific inflammation resulting from a complex of diverse mediators. Since edema of this type is highly sensitive to NSAIDs, carrageenin has been accepted as a useful agent for studying new anti-inflammatory drugs [21]. Extracts produced statistically significant inhibition of edema induced by carrageenan at all doses when compared to the control groups ($p<0.001$, Figure 1). The effect was dose-dependent. V. canescence extract was more potent than V. hirsute extract (88.1 vs. 86.8% inhibition at 800 mg kg-1 i.p.) but no statistically difference was observed between them ($p>0.05$). This activity was better than that of diclofenac at 50 mg kg-1 i.p. (74.3%) but no statistically significance were observed between them ($p>0.05$). V. canescence extract at 200 mg kg-1 (62.8%) and V. hirsute extract at 400 mg kg-1 (67.7%) were equipotent with diclofenac ($p>0.05$). Diclofenac, like most of the non-steroidal anti-inflammatory compounds, inhibits the biosynthesis of prostaglandins and this effect might explain its antiinflammatory activity in carrageenan-induced rat paw edema.

Thus, the anti-inflammatory action of these agents may be related to the inhibition of prostaglandins and leukotriene synthesis [22]. There are some reports concerning the anti-inflammatory effect of flavonoids [21,22]. Vicia spp. contains flavonoids [11,12], it is possible that these compounds are responsible compounds for anti-inflammatory activity of this plant. In conclusion, these results introduced Vicia canescence and Vicia hirsute as easily accessible sources of natural products. This study indicates the potential therapeutic use of this plant as a potent anti-inflammatory agent. Elucidation of exact mechanism of action and active components responsible for good activities requires further investigations.

Acknowledgments

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Conflict of Interest

The authors declare that there are no conflicts of interest.

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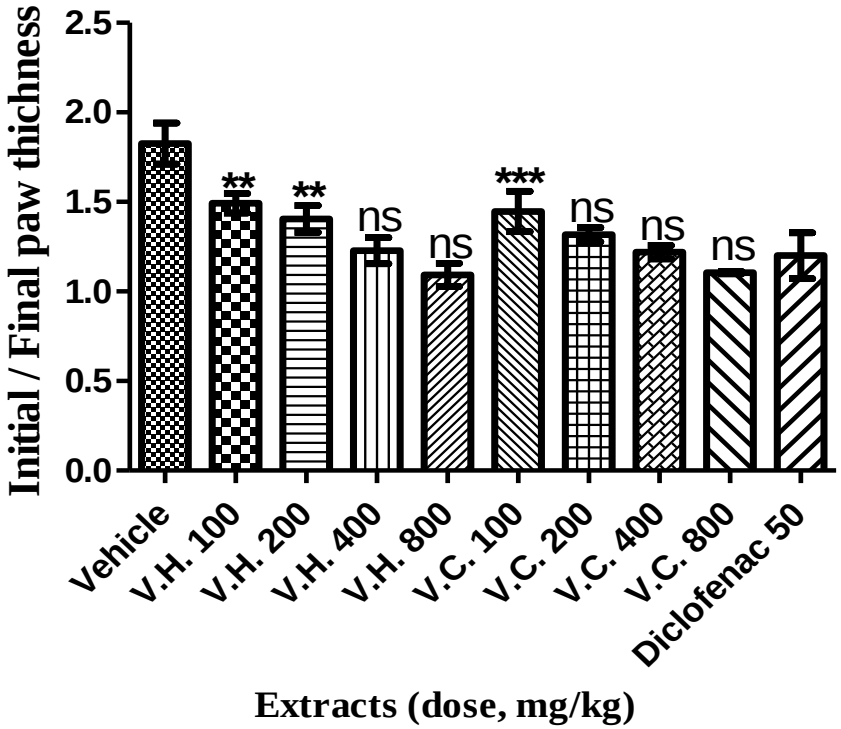

Figure 1. Antiinflammatory activities of *Vicia canescence* and *Vicia hirsuta* extracts in mice. Values are mean ± SD. (n = 7). All groups were different from control with ***p < 0.001.