Research on the Antioxidant and Anti-Inflammatory Effects of Hawthorn Polyphenols

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Abstract: *Hawthorn (Crataegus pinnatifida) polyphenols are important natural plant chemicals with significant antioxidant and anti-inflammatory effects. This study aims to systematically explore the chemical components of hawthorn polyphenols and their impact on oxidative stress and inflammatory responses. Hawthorn polyphenols mainly include hawthorn acid, flavonoids, and tannins, which exhibit notable antioxidant properties, effectively scavenge free radicals, and reduce oxidative damage. Additionally, hawthorn polyphenols show significant inhibitory effects on inflammation by modulating relevant signaling pathways and inflammatory mediators, thereby alleviating inflammatory symptoms. This paper summarizes the extraction and separation techniques of hawthorn polyphenols, analyzes their biochemical mechanisms of antioxidant and anti-inflammatory effects, and validates their bioactivity through in vitro and in vivo experiments. The findings provide scientific evidence for the application of hawthorn polyphenols in health fields and offer directions for their future functional development and clinical applications.*

Keywords: *Hawthorn polyphenols; Antioxidant; Anti-inflammatory; Chemical components; Biochemical mechanisms; In vitro experiments; In vivo effects*

Introduction

With changes in modern lifestyles and increasing environmental pollution, oxidative stress and inflammatory responses have become significant factors in the development of various diseases. Hawthorn, as a traditional medicinal herb and functional food, contains polyphenolic compounds that are believed to have strong antioxidant and anti-inflammatory properties. Hawthorn polyphenols not only exhibit notable pharmacological activities but are also widely used in the development of foods, health supplements, and pharmaceuticals. However, despite existing research indicating the beneficial biological activities of hawthorn polyphenols, further systematic investigation into their specific chemical components, mechanisms of action, and effects in different biological systems is needed. This study aims to provide theoretical basis for the application of hawthorn polyphenols in health fields by comprehensively analyzing their chemical components and biological effects. In-depth exploration of the mechanisms underlying the antioxidant and anti-inflammatory actions of hawthorn polyphenols will help reveal their potential in preventing and treating related diseases and provide scientific evidence for the development of functional products.

1. Chemical Components and Properties of Hawthorn Polyphenols

1.1 Main Chemical Components of Hawthorn Polyphenols

Hawthorn (Crataegus pinnatifida) polyphenols primarily include several key compounds: flavonoids, tannins, and hawthorn acid. Flavonoids, such as quercetin, rutin, and isorhamnetin, are widely present in hawthorn. These flavonoids exhibit excellent antioxidant properties, effectively neutralizing free radicals and reducing oxidative damage.

Tannins, such as hawthorn tannins, are another significant polyphenolic component in hawthorn. These compounds not only possess antioxidant activity but also form stable complexes by binding with proteins, thus inhibiting harmful enzyme activities and further demonstrating their anti-inflammatory effects.

Hawthorn acid is a bioactive triterpenoid compound that can regulate various physiological processes,

including antioxidant and anti-inflammatory responses. Additionally, hawthorn acid has certain antibacterial and antiviral activities, which may offer potential protective effects on health. [1]

1.2 Physicochemical Properties of Hawthorn Polyphenols

The physicochemical properties of hawthorn polyphenols include solubility, stability, and spectral characteristics. Flavonoids typically have good solubility in both water and organic solvents, while tannins mainly dissolve in water. Hawthorn acid is relatively stable under neutral and mildly acidic conditions but tends to degrade in strongly acidic or alkaline environments.

The spectral characteristics of hawthorn polyphenols can be characterized using ultraviolet-visible (UV-Vis) spectroscopy and Fourier-transform infrared (FTIR) spectroscopy. Flavonoids generally exhibit distinct absorption peaks in the UV-Vis spectrum, which are related to the phenolic hydroxyl groups and benzene rings in their structure. FTIR spectroscopy reveals the presence of functional groups such as phenolic hydroxyl groups and carbonyl groups.

Furthermore, the antioxidant capacity of hawthorn polyphenols is usually evaluated through methods such as oxidation-reduction potential (ORP) measurement, DPPH radical scavenging assays, and ABTS radical scavenging assays. In-depth studies of these physicochemical properties help in understanding the biological activities of hawthorn polyphenols and provide a theoretical basis for their applications.

1.3 Extraction and Separation Techniques for Hawthorn Polyphenols

Extraction and separation techniques are crucial for obtaining and purifying the effective components of hawthorn polyphenols. Common extraction methods include water extraction, alcohol extraction, and ultrasonic-assisted extraction. Water extraction is simple and easy but may be less effective for extracting certain polyphenols. Alcohol extraction significantly improves the extraction rate of hawthorn polyphenols, with 70% ethanol showing the best results. Ultrasonic-assisted extraction enhances the extraction efficiency through high-frequency ultrasound and reduces extraction time.

For separation techniques, high-performance liquid chromatography (HPLC) and thin-layer chromatography (TLC) are commonly used analytical methods. HPLC efficiently separates the various components of hawthorn polyphenols and, when combined with mass spectrometry (MS), provides qualitative and quantitative analysis. TLC is used for preliminary screening and separation of polyphenol components.

In recent years, solid-phase extraction (SPE) technology and high-performance reversed-phase liquid chromatography (RP-HPLC) have also been applied to the separation and purification of hawthorn polyphenols. These techniques improve the separation purity and extraction efficiency of hawthorn polyphenols by optimizing separation conditions and refining separation media. Through these advanced extraction and separation techniques, high-purity hawthorn polyphenols can be effectively obtained, providing a reliable foundation for their applications in antioxidant and anti-inflammatory research. [2]

2. Mechanisms of Antioxidant Action of Hawthorn Polyphenols

2.1 Biochemical Mechanisms of Antioxidant Action

The antioxidant action of hawthorn polyphenols is primarily achieved through the following biochemical mechanisms:

2.1.1 Free Radical Scavenging

Hawthorn polyphenols exhibit significant free radical scavenging capabilities. Their phenolic hydroxyl groups can react with free radicals to form stable products, effectively neutralizing free radicals. This action is particularly notable against hydroxyl radicals (·OH) and hydrogen peroxide radicals (H2O2), helping to alleviate oxidative stress-induced cellular damage.

2.1.2 Redox Reactions

Hawthorn polyphenols can inhibit the activity of redox enzymes through redox reactions. By inhibiting peroxidase (P450), hawthorn polyphenols reduce the generation of oxidants, thereby mitigating oxidative damage.

2.1.3 Metal Ion Chelation

Hawthorn polyphenols can form chelates with transition metal ions (such as iron and copper), reducing the oxidative reactions catalyzed by these metal ions. This mechanism effectively reduces Fenton reactions in oxidative stress, protecting cells from oxidative damage.

2.1.4 Regulation of Antioxidant Enzyme Systems

Hawthorn polyphenols can upregulate the expression of endogenous antioxidant enzymes, such as superoxide dismutase (SOD), catalase (CAT), and glutathione reductase (GR). These enzymes enhance cellular antioxidant capacity and remove oxidative products from the body.

Through these mechanisms, hawthorn polyphenols exert broad antioxidant effects in vivo, effectively preventing oxidative stress-related damage to tissues and cells. [3]

2.2 In Vitro Experimental Studies on the Antioxidant Activity of Hawthorn Polyphenols

In vitro experiments typically evaluate the antioxidant activity of hawthorn polyphenols using the following methods:

2.2.1 DPPH Radical Scavenging Assay

This assay measures the ability of hawthorn polyphenols to scavenge 1,1-diphenyl-2-picrylhydrazyl (DPPH) radicals. The degree of discoloration of DPPH upon reaction with hawthorn polyphenols is proportional to the antioxidant activity. Studies have shown that hawthorn polyphenols can significantly reduce the absorbance of DPPH, indicating strong antioxidant effects.

2.2.2 ABTS Radical Scavenging Assay

The ABTS radical scavenging assay assesses the inhibitory capacity of hawthorn polyphenols against 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) radicals. Hawthorn polyphenols perform excellently in this assay, significantly reducing the absorbance of ABTS radicals, demonstrating their potent antioxidant ability.

2.2.3 Oxidation-Reduction Potential (ORP) Measurement

By measuring the oxidation-reduction potential of hawthorn polyphenol solutions, their in vitro antioxidant activity can be evaluated. Hawthorn polyphenol solutions show lower ORP values, indicating strong reducing capabilities.

2.2.4 Lipid Peroxidation Measurement

The inhibitory effect of hawthorn polyphenols on lipid peroxidation is measured to further verify their antioxidant activity. Experimental results indicate that hawthorn polyphenols can effectively reduce the generation of lipid peroxidation products (such as malondialdehyde, MDA), confirming their antioxidant capacity.

These in vitro experimental results demonstrate that hawthorn polyphenols have significant antioxidant activity, providing a solid foundation for their applications in the biomedical field.

2.3 Evaluation of In Vivo Antioxidant Effects of Hawthorn Polyphenols

The in vivo antioxidant effects of hawthorn polyphenols are mainly evaluated through animal models and clinical trials:

2.3.1 Animal Experiments

In animal studies, hawthorn polyphenols are typically administered orally or by injection to assess their inhibition of oxidative stress in vivo. Research findings indicate that hawthorn polyphenols significantly lower oxidative biomarkers in animals, such as malondialdehyde (MDA) and hydrogen peroxide levels, while enhancing the activity of antioxidant enzymes like superoxide dismutase (SOD) and catalase (CAT).

2.3.2 Biomarker Measurement

The antioxidant effects of hawthorn polyphenols are assessed by measuring oxidative stress-related biomarkers in vivo, such as 8-hydroxydeoxyguanosine (8-OHdG) and glutathione (GSH). Studies show that hawthorn polyphenols significantly reduce the levels of these biomarkers, further proving their

antioxidant effects. [4]

2.3.3 Clinical Trials

In clinical trials, the antioxidant effects of hawthorn polyphenols are evaluated by measuring oxidative stress status and antioxidant enzyme levels in serum. Trial results indicate that hawthorn polyphenols significantly improve patients' oxidative status, enhance antioxidant capacity, and alleviate symptoms of oxidative stress-related diseases.

2.3.4 Tissue Damage Assessment

Tissue slices and pathological analyses are used to evaluate the protective effects of hawthorn polyphenols against tissue damage. Research finds that hawthorn polyphenols effectively reduce tissue damage and improve tissue structure, supporting their clinical application potential in antioxidant effects.

Overall, these in vivo studies show that hawthorn polyphenols exhibit significant antioxidant effects, indicating a promising application in the prevention and treatment of oxidative stress-related diseases.

3. Mechanisms of Anti-inflammatory Action of Hawthorn Polyphenols

3.1 Molecular Mechanisms of Anti-inflammatory Action

The anti-inflammatory effects of hawthorn polyphenols are primarily achieved through the following molecular mechanisms:

3.1.1 Inhibition of Inflammatory Mediators' Release

Hawthorn polyphenols mitigate inflammation by inhibiting the synthesis and release of inflammatory mediators such as prostaglandin E2 (PGE2), tumor necrosis factor-alpha (TNF-α), and interleukin-6 (IL-6). They achieve this by suppressing the activity of cyclooxygenase (COX) and lipoxygenase (LOX), thereby reducing the production of PGE2 and alleviating inflammation.

3.1.2 Regulation of Nuclear Factor κB (NF-κB) Signaling Pathway

Hawthorn polyphenols inhibit the activation of the NF-κB signaling pathway, a crucial transcription factor involved in inflammatory responses. They reduce the degradation of IκB proteins and inhibit the nuclear translocation of NF-κB, leading to decreased expression of inflammation-related genes.

3.1.3 Inhibition of Reactive Oxygen Species (ROS) Generation

Hawthorn polyphenols reduce oxidative damage and inflammation by inhibiting the generation of reactive oxygen species (ROS). They enhance the activity of antioxidant enzymes, lowering ROS levels within cells and thus mitigating the inflammatory response.

3.1.4 Regulation of Macrophage Function

Hawthorn polyphenols influence macrophage activity, reducing its excessive activation during inflammation. They achieve this by inhibiting the release of chemotactic factors and inflammatory mediators from macrophages, thereby decreasing their contribution to inflammation.

These molecular mechanisms indicate that hawthorn polyphenols regulate inflammation through multiple pathways, thereby exhibiting significant anti-inflammatory effects.

3.2 Experimental Studies on the Anti-inflammatory Activity of Hawthorn Polyphenols In Vitro

The anti-inflammatory activity of hawthorn polyphenols in vitro is typically assessed using the following methods:

3.2.1 Cell Model Experiments

Cellular models, such as lipopolysaccharide (LPS)-induced macrophage models or other inflammation cell models, are used to study the inhibitory effects of hawthorn polyphenols on the release of inflammatory factors. Studies have shown that hawthorn polyphenols significantly reduce the secretion of TNF-α, IL-6, and PGE2 induced by LPS, demonstrating effective anti-inflammatory properties.

3.2.2 Cell Signaling Pathway Analysis

Western blotting or immunofluorescence techniques are used to examine the impact of hawthorn polyphenols on the NF-κB signaling pathway. Results indicate that hawthorn polyphenols significantly reduce NF-κB activation and nuclear translocation, suppressing the expression of associated inflammatory genes.

3.2.3 Oxidative Stress Detection

The inhibition of oxidative stress by hawthorn polyphenols is evaluated by measuring intracellular ROS levels and antioxidant enzyme activity. Experiments reveal that hawthorn polyphenols significantly decrease ROS levels and increase antioxidant enzyme activity, thereby reducing cellular oxidative damage.

3.2.4 Cell Migration and Invasion Assays

The effects of hawthorn polyphenols on cell migration and invasion are assessed using wound healing assays and Transwell assays. Studies have found that hawthorn polyphenols significantly inhibit the migration and invasion of inflammatory cells, further confirming their anti-inflammatory activity.

These in vitro experimental results demonstrate that hawthorn polyphenols have significant anti-inflammatory effects, providing a basis for their application in inflammation-related diseases.[5]

3.3 Evaluation of the Anti-inflammatory Effects of Hawthorn Polyphenols In Vivo

The anti-inflammatory effects of hawthorn polyphenols in vivo are evaluated through animal models and clinical trials:

3.3.1 Animal Experiments

In animal models, the anti-inflammatory effects of hawthorn polyphenols are assessed through oral or injectable administration. Common models include acute and chronic inflammation models in mice or rats, such as the ear swelling model induced by xylene or models of experimental colitis. Studies show that hawthorn polyphenols significantly lower levels of inflammatory markers, including TNF-α, IL-6, and C-reactive protein (CRP) in serum. Additionally, hawthorn polyphenols effectively reduce leukocyte infiltration and tissue damage in inflamed tissues, further confirming their anti-inflammatory effects.

3.3.2 Histopathological Analysis

Histopathological analysis provides a visual assessment of the protective effects of hawthorn polyphenols on tissue inflammation. Techniques such as tissue sectioning and microscopy are used to observe inflammation in tissues. Results indicate that hawthorn polyphenols significantly alleviate tissue inflammation, improving tissue structure. For example, in colitis models, hawthorn polyphenols reduce mucosal damage, leukocyte infiltration, edema, and ulcer formation in the intestine. Pathological analysis also shows decreased apoptosis and tissue necrosis, demonstrating superior tissue protection.

3.3.3 Biomarker Measurement

Biomarker measurement provides quantitative evidence for evaluating the anti-inflammatory effects of hawthorn polyphenols. By detecting inflammation-related biomarkers such as interleukin-1β (IL-1β) and tumor necrosis factor-beta (TNF-β), a comprehensive understanding of hawthorn polyphenols' anti-inflammatory mechanisms is gained. Studies show that hawthorn polyphenols significantly lower the concentrations of these biomarkers, reducing inflammation. Additionally, assessments of antioxidant enzyme systems reveal a synergistic effect of hawthorn polyphenols in both antioxidant and anti-inflammatory activities.

3.3.4 Clinical Trials

In clinical trials, the anti-inflammatory effects of hawthorn polyphenols are validated by assessing patient inflammation indicators and symptom improvement. Clinical trials typically include randomized controlled trials and double-blind studies to ensure data reliability and scientific accuracy. Results show that hawthorn polyphenols significantly alleviate inflammation symptoms in patients, improving quality of life. Specific improvements include reductions in joint pain, swelling, and functional impairment. Clinical trials also evaluate the safety and tolerability of hawthorn polyphenols, with results indicating good tolerance and no significant adverse effects.

In summary, in vivo studies demonstrate that hawthorn polyphenols have significant anti-

inflammatory effects. Comprehensive evaluations through animal experiments, histopathological analysis, biomarker measurement, and clinical trials confirm their potential in treating inflammation-related diseases. These findings provide a solid scientific basis for the future clinical application of hawthorn polyphenols and lay the groundwork for further research in related fields.[6]

Conclusion

Hawthorn polyphenols exhibit significant antioxidant and anti-inflammatory effects, primarily due to their ability to scavenge free radicals and regulate inflammatory mediators. In vitro experiments have demonstrated that hawthorn polyphenols effectively reduce oxidative damage and show superior anti-inflammatory effects across various cell models. In vivo studies further confirm the alleviation of inflammation-related diseases by hawthorn polyphenols and indicate good biocompatibility and potential for practical applications. Future research should explore the combined effects of hawthorn polyphenols with other plant components or drugs, and larger-scale clinical trials will be beneficial in advancing the use of hawthorn polyphenols in functional foods and pharmaceuticals, thereby enhancing their health-promoting effects.

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