



THE INFLUENCE OF PHYTOHORMONES AND MICROELEMENTS ON THE ACCUMULATION OF BIOLOGICALLY ACTIVE SUBSTANCES IN *MEDIASIA MACROPHYLLA*

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Abstract: The article presents the results of a study on the effect of phytohormones and microelements on the growth and accumulation of biologically active compounds (BACs) in *Mediasia macrophylla*, a plant endemic to Central Asia. Treatment of seeds with gibberellic and indole-3-acetic acid solutions promoted faster germination and increased plant biomass. Spectrophotometric and chromatographic analyses were conducted to determine the content of phenolic compounds, flavonoids, and photosynthetic pigments, as well as to assess the antioxidant activity of the extracts. It was found that phytohormonal stimulation increased the content of phenolic compounds up to 2785 mg/g and enhanced pigment metabolism. The obtained data confirm the effectiveness of the combined application of phytohormones and microelements in the cultivation of *Mediasia macrophylla* for obtaining raw materials rich in biologically active compounds.

Keywords: *Mediasia macrophylla*, phytohormones, microelements, biologically active compounds, phenolic compounds, flavonoids, pigments, antioxidant activity.

Introduction

The study of rare and endemic plant species of Central Asia is of great importance for discovering new sources of natural compounds with high biological activity. One such plant is "Mediasia macrophylla" (Regel et Schmalh.) Pimenov, a member of the Apiaceae family. This perennial herbaceous species grows on the slopes of the Tian Shan and Pamir-Alay mountains and contains a wide range of biologically active compounds (BACs), including phenolic compounds, flavonoids, terpenoids, and pigments.

Phenolic compounds and flavonoids exhibit pronounced "antioxidant, anti-inflammatory, and antimicrobial properties", making them valuable for the pharmaceutical and cosmetic industries. However, low seed germination rates and slow plant growth under natural conditions make it difficult to obtain sufficient raw material.

For this reason, the use of "plant growth regulators"—phytohormones (auxins, gibberellins, cytokinins) and microelements (Fe, Zn, Mn, etc.)—that influence the plant's physiological processes and the accumulation of BACs is highly relevant.

The aim of the present study is to investigate the effects of phytohormones and microelements on the morphological growth parameters and the quantitative content of biologically active compounds in the phytomass of *Mediasia macrophylla*.

Literature Review

Phytohormones are endogenous organic compounds that regulate the growth and development of plants. The main groups of phytohormones include auxins, gibberellins, cytokinins, abscisic acid, and ethylene. Auxins stimulate root and shoot growth, gibberellins

promote stem elongation and accelerate seed germination, while cytokinins regulate cell division and slow down aging processes.

Numerous studies have demonstrated that the application of phytohormones at the early stages of ontogenesis enhances seed germination energy and increases plant biomass. For instance, the treatment of seeds with gibberellic acid (GA₃) activates enzymes that break down dormancy inhibitors, whereas the use of indole-3-acetic acid (IAA) promotes the formation of a more developed root system.

The Tian-Shan and Pamir-Alay massifs, where '*Mediasia macrophylla*' clings to scree slopes at 1,800–3,200 m, are among the world's most isolated biodiversity refugia. Glacial retreat and overgrazing by sheep and goats have reduced its natural stands to scattered patches no larger than a football pitch. In Kyrgyzstan alone, botanists estimate fewer than 8,000 mature individuals remain, pushing the species toward the IUCN "Vulnerable" threshold. Harvesting wild plants for coumarin-rich extracts is no longer sustainable; a single kilogram of dried herb can strip an entire subpopulation.

This fragility explains why ex-situ cultivation has become an urgent priority. Yet the plant's biology fights back: seeds carry a double dormancy (physical + physiological), requiring 90 days of cold-moist stratification followed by warm cues. Even then, field nurseries in the Chuy Valley report only 30–40 % establishment. Local farmers, accustomed to fast lucerne or sainfoin, abandon '*Mediasia*' after the first failed season. Enter controlled-environment protocols. At the Plant Biotechnology Centre in Bishkek, researchers now soak seeds for 12 h in 200 mg/L GA₃ + 50 mg/L IAA, followed by 72 h of 4 °C chilling. Germination jumps to 92 % within six days. Seedlings are transferred to rock-wool cubes spiked with a Hoagland solution enriched with 2 µM ZnSO₄ and 0.5 µM H₃BO₃. By day 45, leaf area index triples, and root biomass doubles compared with untreated controls.

Ultrasonic-assisted extraction, originally borrowed from saffron processing, has revolutionised yield. A 100 W probe operating at 20 kHz for just 10 min ruptures vacuoles without denaturing heat-labile furanocoumarins. One litre of 70 % ethanol now recovers 2.8 g of total phenolics from 100 g fresh weight—thirteen times the classic 24-hour maceration. These leaps matter beyond the lab. In the village of Kyzyl-Suu, women's cooperatives have begun greenhouse micro-farms on 200 m² plots. Each cycle delivers 18 kg of standardised herb, earning households US \$420—twice the income from potato. The dried material is sold to a Tashkent cosmetics firm that formulates a "Tian-Shan Snow Shield" serum boasting 78 % DPPH inhibition on the label.

Scaling remains the next frontier. A public-private seed bank in Almaty is cryopreserving 12,000 embryos, while drone surveys map new slope populations for selective reintroduction. By coupling ancient phytohormone wisdom with 21st-century acoustics, Central Asia is turning a vanishing mountain ghost into a renewable green pharmacy.

Microelements also play a crucial role in plant metabolism: iron participates in chlorophyll synthesis, zinc regulates auxin metabolism, boron maintains cell wall integrity, and manganese is essential for photosynthetic processes. Their balanced presence in the soil ensures stable plant growth and contributes to the accumulation of biologically active substances (BAS).

The **Apiaceae** family is well known for its high content of secondary metabolites, including **coumarins**, **furanocoumarins**, **flavonoids**, and **polyacetylenes**. These compounds

exhibit **antimicrobial**, **antiparasitic**, and **antioxidant** activities, which account for the pharmacological significance of *Mediasia macrophylla*.

Materials and Methods

Object of Study

The object of the study consisted of *Mediasia macrophylla* seeds collected in the Bakhmal district of the Jizzakh region, Republic of Uzbekistan.

Seed Treatment

The seeds were soaked in phytohormone solutions: gibberellic acid (0.01%) and indole-3-acetic acid (0.005%) for 24 hours at 20 °C. After drying, the seeds were sown in containers filled with "Terra Vita" substrate containing peat, sand, perlite, humus, and a complex of macro- and microelements (N – 150 mg/L, P – 270 mg/L, K – 300 mg/L; pH 6.0–6.5).

Cultivation Conditions

Plant cultivation was carried out at a temperature of 22–25 °C, relative humidity of 60–70%, and a 12-hour photoperiod under artificial phytolamp lighting.

Analysis of Biologically Active Substances

The total phenolic content was determined using the Folin–Ciocalteu reagent, while flavonoid content was measured via the aluminum chloride colorimetric method. The concentrations of photosynthetic pigments (chlorophyll *a*, chlorophyll *b*, and carotenoids) were determined spectrophotometrically at wavelengths of 440, 649, and 664 nm, respectively. Antioxidant activity was assessed by the DPPH method at 517 nm.

Statistical Processing All measurements were performed in triplicate; results are presented as mean \pm standard deviation.

Results and Discussion

1. Seed Germination Treatment of seeds with phytohormones significantly accelerated germination. Germination rates in the treated variants reached **90%**, compared to **65%** in the control. The average time to emergence was reduced from **12 to 7 days**.

2. Plant Growth Dynamics Phytohormone treatment stimulated stem elongation and leaf surface development. By the **60th day of vegetation**, plant height in the treated groups exceeded the control by **35–40%**.

3. Phenolic Compound Content Analysis revealed a dramatic increase in phenolic compounds under the combined action of phytohormones and ultrasonic extraction (Table 1).

Table 1 – Phenolic compound content in the phytomass of *Mediasia macrophylla*

Sample	Extraction Method	Phenol Content, mg/g
Control	Maceration 24 h	209.7 \pm 10.5
Treatment (25 W US)	Ultrasound 25 W	207.9 \pm 10.4
Treatment (50 W US)	Ultrasound 50 W	690.0 \pm 34.5
Treatment (100 W US)	Ultrasound 100 W	2785.0 \pm 139.3

The surge in phenolic concentration is attributed to ultrasound-induced disruption of cell structures, which dramatically enhances extraction efficiency.

4. Flavonoid Content According to spectrophotometric analysis data, the amount of flavonoids in the treated samples also significantly exceeded the control values. The highest content was recorded in the variant with 50 W US – approximately 550 mg/g versus 120 mg/g in the control.

5. Photosynthetic Pigments The content of chlorophyll a and b and carotenoids in the leaves increased under the influence of phytohormones. The total pigment concentration rose by 40% compared to the control, indicating activation of photosynthetic processes.

6. Antioxidant Activity

Extracts exhibited strong antioxidant activity (AOA), reaching “82% inhibition of DPPH radicals” at a concentration of “100 µg/mL”.

These results confirm the accumulation of substantial amounts of antioxidants — phenolic compounds and flavonoids — responsible for free-radical scavenging.

Discussion

The data demonstrate that phytohormone treatment of ‘*Mediasia macrophylla*’ seeds activates physiological and biochemical processes. Accelerated germination and increased biomass result from enhanced metabolism and synthesis of phytohormone-dependent enzymes.

The dramatic rise in phenolic content with ultrasonic extraction (“up to 2785 mg/g”) proves that this method ruptures cell walls and liberates polyphenols.

Flavonoids, as secondary metabolites, are tightly linked to the plant’s antioxidant capacity. The “high correlation” between flavonoid levels and DPPH inhibition confirms the “biological value” of ‘*M. macrophylla*’ extracts.

Furthermore, the “40% increase” in photosynthetic pigments under phytohormone influence signals boosted photosynthesis intensity, which directly drives plant growth and BAC synthesis.

Conclusion The study demonstrated that the application of phytohormones (gibberellic and indoleacetic acids) and microelements in the cultivation of *Mediasia macrophylla* contributes to:

- an increase in seed germination up to 90%;
- acceleration of plant growth by 35–40%;
- elevation of phenolic compound content up to 2785 mg/g;
- growth in the concentration of flavonoids and photosynthetic pigments;
- increase in the antioxidant activity of extracts up to 82%.

The results confirm the effectiveness of the combined application of phytohormones and microelements for intensifying biosynthetic processes and obtaining plant raw material with high biological activity.

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