INTRODUCTION

Phenolic acids possess valuable properties for therapy, e.g. antioxidant, anticancer and immunostimulating activities. Interesting large amounts may be produced in plant cell cultures. The studies are focused on phenolic acids produced by tissue cultures of *Aronia melanocarpa* (Michx.) Elliott, *Anethum graveolens* L. and *Schisandra chinensis* (Turcz.) Balli.

MATERIAL AND METHODS

I. In vitro cultures

- **medium:** acc. to Murashige and Skoog (MS) and acc. to Linsmaier and Skoog (LS)
- **plant growth regulators:** BAP (6-benzyladenine) from 0.1 to 3.0 mg/l and NAA (naphthaleneacetic acid) from 0.1 to 2.0 mg/l
- **temperature:** 25 ± 2°C
- **growth cycles:** 4 weeks, 3 series
- **type of cultures:** agar cultures
- **light conditions:** constant artificial light (4 W/m², LF-40W lamp, daylight, Piła)

II. Plant material

- **fruits** and/or **leaves** of native plants growing in the Rogów Arboretum, Warsaw University of Life Sciences, Forest Experimental Station in Rogów (Poland)

III. Hydrolysis and extraction

- **methylene extracts of biomass from in vitro cultures** collected after 4 weeks growth cycles and from plant material were analyzed after acid hydrolysis (2 M HCl, 2h)

IV. HPLC conditions

- **HPLC apparatus:** Merck - Hitachi
- **column:** Kinex™ 18C (150 x 4.6 mm, 2.6 µm)
- **solvent system:** 0.1% trifluoroacetic acid (A) and acetonitrile (B) (gradient program) [1]
- **injection volume:** 5µl
- **flow rate:** 1ml/min
- **detector UV-λ = 254 nm
- **standards:** caffeic, cinnamic, chlorogenic, protocatechuic, rosmarinic, salicylic, syringic, vanillic acids (Sigma); p-coumaric, ferulic, p-hydroxybenzoic, vanillic acids (Fluka)

RESULTS

1. The maximum total contents of phenolic acids in cell cultures on LS and MS medium were as follows:
   - *A. melanocarpa* shoot cultures - 165.40 mg/100 g d.w. and 230.25 mg/100 g d.w.
   - *A. melanocarpa* callus cultures - 163.34 and 230.25 mg/100 g d.w., respectively [4,5] (Tab. 3)
   - *A. graveolens* shoot-differentiating callus cultures - 115.64 mg/100 g d.w. and 230.25 mg/100 g d.w. (Tab. 4).

2. The cultures mainly produced p-hydroxybenzoic, salicylic, p-coumaric and chlorogenic acids (Tab. 1-4).

CONCLUSIONS

The basic components and plant growth regulators (BAP and NAA) concentrations in culture media as well as the degree of differentiation in tissue cultures are of great influence on the accumulation of phenolic acids. Medicinal plant tissue cultures can become good source of some therapeutically important phenolic acids.

REFERENCES


ACKNOWLEDGMENTS

The authors wish to express their sincere gratitude to Mr. Piotr Banaszak and Mr. Jarosław Słupik from the Rogów Arboretum - Warsaw University of Life Sciences, Forest Experimental Station in Rogów (Poland) for plant material.

![Fig. 1. Aronia melanocarpa shoot culture cultivated on the MS medium containing 2 mg/l BAP and 1 mg/l NAA](image)

![Fig. 2. Aronia melanocarpa callus culture cultivated on the MS medium containing 2 mg/l BAP and 1 mg/l NAA](image)

![Fig. 3. Aronia graveolens shoot-differentiating callus culture (A) and callus cultures (B) cultivated on the MS medium containing 3 mg/l BAP and 1 mg/l NAA](image)

![Fig. 4. Schisandra chinensis shoot-differentiating callus culture (A) and callus cultures (B) cultivated on the MS medium containing 3 mg/l BAP and 1 mg/l NAA](image)

![Fig. 5. Shoot-differentiating callus cultures and callus cultures of *Schisandra chinensis* cultivated on different MS and LS medium variants and their contents in fruits of native plant](image)

![Fig. 6. Phenolic acids contents in biomass extracts from callus cultures of *Aronia melanocarpa* cultivated on different MS and LS medium variants and their contents in fruits of native plant](image)