

Aqueous and hydro-alcoholic extracts of *Echinacea purpurea* (L) Moench as traditional herbal remedies with immunotropic activity

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Echinacea purpurea preparations are derived from the aerial parts of purple coneflower, *Echinacea purpurea* (L) Moench Asteraceae=Compositae, which grows throughout North America and Europe. Information about use of *Echinacea* first came from American Indians. They used leaves and roots/rhizomes of *Echinacea purpurea*, *Echinacea angustifolia* DC and *Echinacea pallida* Nutt, internally and externally, as remedies for many ailments.

Plants were used fresh or, most often, dried, as infusion, succus or decoction [1-6].

The most ancient form of medical herbs use, aside from consuming the fresh or dried plant, is aqueous extract which may be dried to form powder. According to *Materia Medica* [7] contemporary "*Echinacea purpurea* preparations include liquid(aqueous or hydro-alcoholic)extracts of fresh or dried whole plant or aerial parts; fresh or dried preparations of

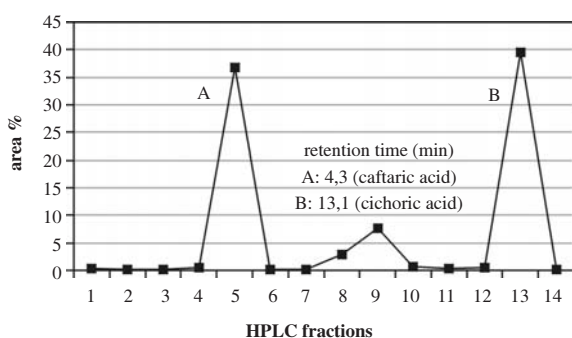


Fig. 1. High performance liquid chromatography (HPLC) of phenolic compounds from aqueous extract of dried *Echinacea purpurea* herb (Klęka)

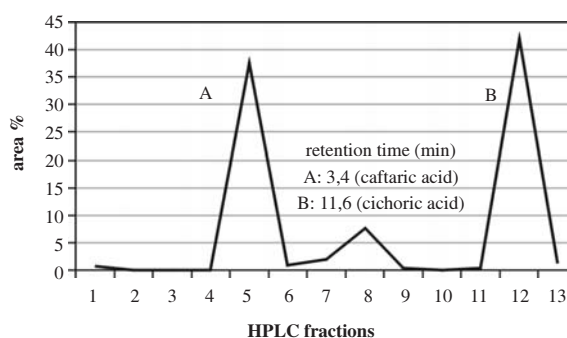


Fig. 2. High performance liquid chromatography (HPLC) of phenolic compounds from hydro-alcoholic extract of fresh *Echinacea purpurea* herb (succus *Echinaceae* Klęka)

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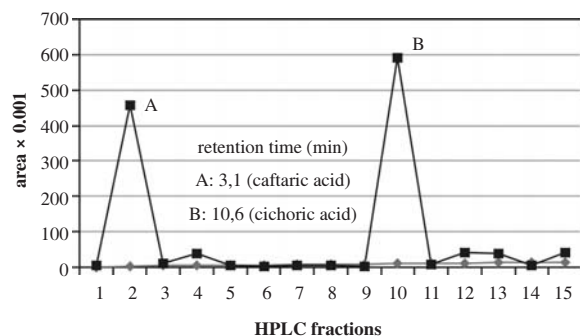


Fig. 1a. High performance liquid chromatography (HPLC) of phenolic compounds from ECHINERBA tablets

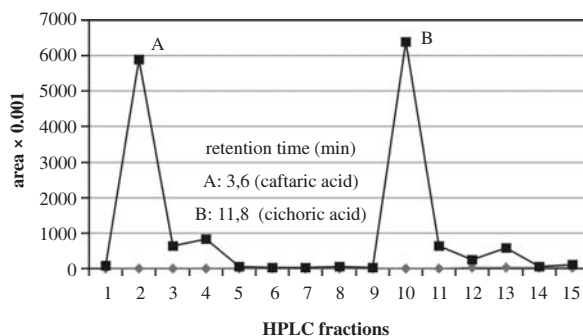


Fig. 2a. High performance liquid chromatography (HPLC) of phenolic compounds from ECHINAPUR tablets

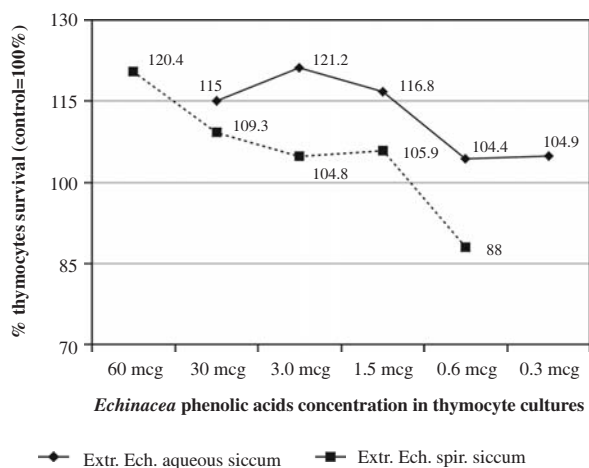


Fig. 3. Thymomimetic activity of *Echinacea* extracts

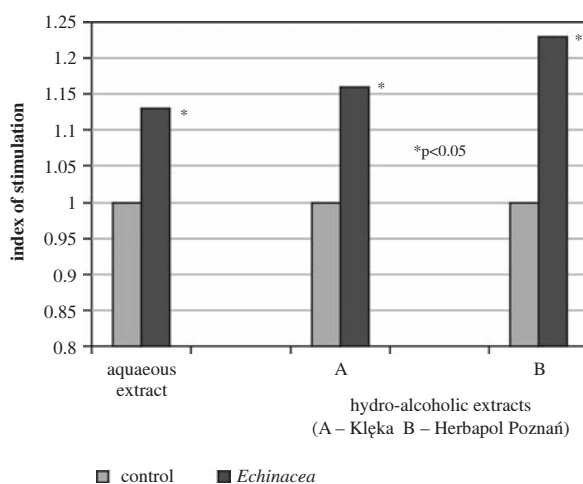


Fig. 4. The *in vitro* effect of *Echinacea purpurea* extracts on chemokinetic activity of human blood mononuclear cells in cell culture

root and rhizome; stabilized juice of the flowering tops, mixtures of any of the above preparations and tablets and capsules based on any of the above plant parts or corresponding extracts". According to WHO Monograph [8] *Herba Echinaceae purpureae* consists of the fresh or dried aerial parts of *Echinacea purpurea* (L) Moench harvested in full bloom, and dosage forms are: powdered aerial parts, pressed juice and galenic preparations for internal and external use, as supportive therapy for colds, infections of respiratory and urinary tract, wound healing and various inflammatory skin diseases.

This is a general agreement, that therapeutic proprieties of *Echinacea purpurea* herb are connected with its immunostimulatory effects [9-30]. Various compounds are known to contribute to this activity – the most important are caffeic acid derivatives CADs (among them caftaric and cichoric acid), alkalimides and polysaccharides [31-34]. It

was found, that the persistence of enzymatic activity in fresh plant products, catalyzed the breakdown of cichoric acid. This reaction may be stopped by drying of plant material or by stabilization with alcohol. Since this observation a lot of commercial products of *Echinacea purpurea* are stabilized with alcohol – there are on the market preparations of fresh pressed juice stabilized later with alcohol (Immunal drops LEK, and Echinacin Madaus since 1938), or preparations of succus from fresh plants macerated with alcohol (Succus Echinaceae Phytopharm in liquid form produced for almost 18 years) and Echinapur Herbapol Poznań tablets, present on the market more than 12 years; and Echinaforce (Vogel) produced since half of the 20th century. There are also products made from aqueous extracts of dried herb (Echinerba Labofarm, Alchinal Gemi, Echin Elder Traditional Medicinals) and products where water extracts are mixed with various alcoholic ones to obtain the best

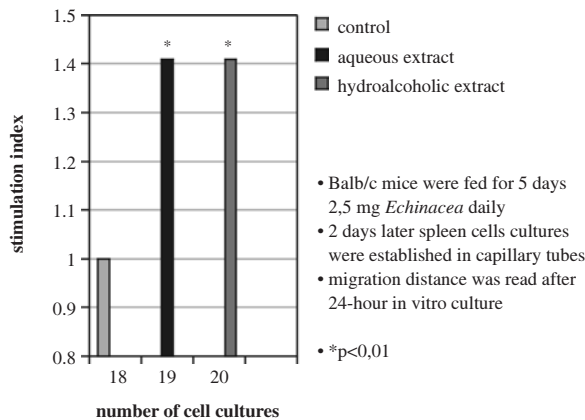


Fig. 5. The *in vivo* effect of *Echinacea purpurea* extracts on the chemokinetic activity of murine splenic lymphocytes

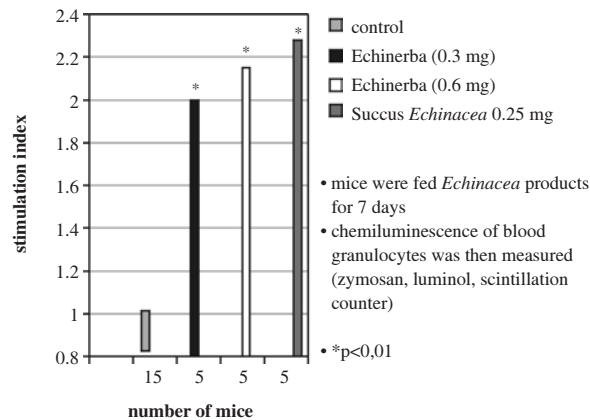


Fig. 6. The effect of *Echinacea purpurea* products on metabolic activity of blood granulocytes

composition of active compounds (Echinilin, Factors R&D Technologies) [35-44].

Our scientific team have performed for many years research on the immunotropic activity of various *Echinacea purpurea* aerial part products, and we present below a brief minireview of our articles on this topic, illustrated with Figures prepared for this article purpose on the basis of published results [45-57]. In this minireview we compare the main phenolic compounds profiles and various immunotropic activities of hydro-alcoholic extracts (prepared from fresh aerial parts) and water extracts (prepared from dried aerial parts) of *Echinacea purpurea* plants of polish origin and derived from them products – Echinerbera tablets, Echinapur tablets and Succus Echinacea drops.

HPLC analysis of these products have revealed high similarity of their polyphenols profiles, with high peaks of caftaric and cichoric acids (Figures 1, 1a, 2, 2a).

Biological activity tests have revealed further similarities. In *in vitro* experiments, performed by Dr Teresa Sawicka and her team, both aqueous and hydro-alcoholic extracts exerted thymomimetic activity in cultures of murine thymocytes with hydrocortisone [47]. Activity of aqueous extract was more pronounced than water-alcoholic one (Figure 3).

In other *in vitro* test, where the effect of aqueous and hydro-alcoholic extracts were compared, their effect on migratory (chemokinetic) activity of human peripheral blood mononuclear cells in 24-hour cell culture was also similar (presented on the Figure 4).

High similarity was also observed when *ex vivo* chemokinetic activity of murine spleen lymphocytes was studied. In this experiment mice were fed Echinacea extracts for 5 days before they served as spleen cells donors (Figure 5).

Other studies have revealed further similarities. Feeding mice aqueous and hydro-alcoholic extracts of *Echinacea*

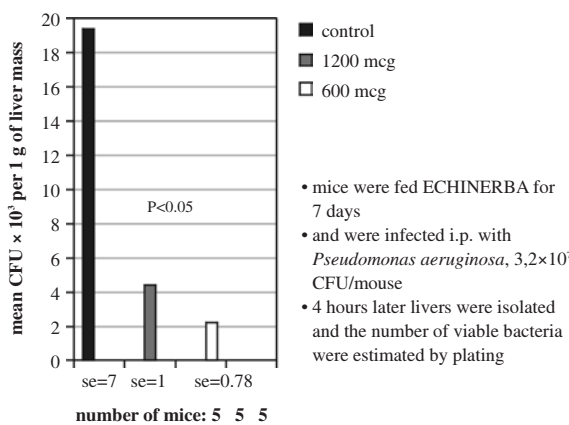


Fig. 7. The effect of feeding mice ECHINERBA on their anti-bacterial immunity

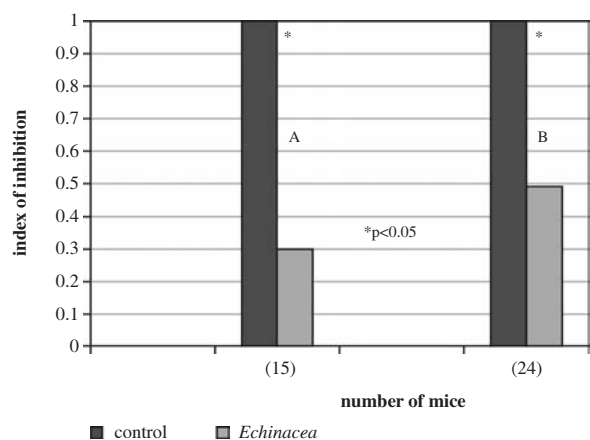


Fig. 8. Comparison of the effects of ECHINERBA (A) and hydro-alcoholic extract (B) on the development of *Pseudomonas aeruginosa* infection in mice

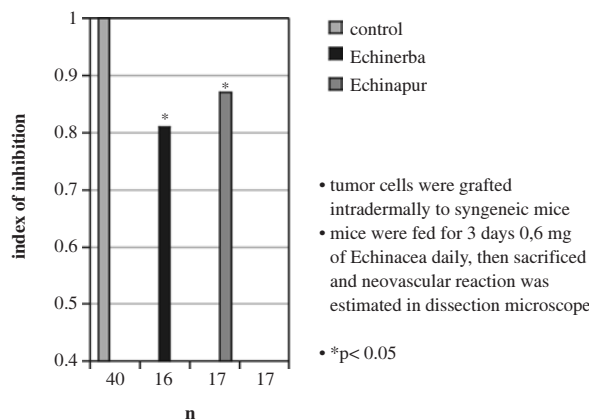


Fig. 9. The effect of various *Echinacea* extracts on neovascular reaction induced in mice skin by L-1 sarcoma tumor cells.

purpurea herb, or they products in tablets form, resulted in stimulation of blood granulocytes activity (Figure 6) and in increase of anti-bacterial activity of peritoneal phagocytes (Figures 7 and 8). Studies of anti-bacterial activity were performed by scientific team of prof. Janusz Bany from Military Institute of Hygiene and Epidemiology in Warsaw [45, 49, 52, 58].

Comparison of the effect of various Echinacea extracts on the angiogenic activity of murine L-1 sarcoma tumor cells, and on this tumor vascularization, revealed similar inhibitory influence of aqueous and hydro-alcoholic extracts on these parameters (Figure 9).

Histological analysis performed on H+E stained paraffin sections of 7-days tumors revealed less and smaller blood vessels at a tumor margin in material collected from Echinacea-fed animals than in the controls.

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