ANTIOXIDANT ACTIVITY OF FRUIT EXTRACTS USED IN COSMETIC PRODUCTS

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Abstract: The aim of this work was to evaluate the total phenolic content and the antioxidant activity of five commercial fruit extracts available on Polish cosmetics market: *Citrus grandis* (pomelo), *Cucumis melo* (muskmelon), *Prunus armeniaca* (apricot), *Prunus persica* (peach) and *Vitis vinifera* (grape). Two tests: DPPH radical scavenging activity assay and Trolox Equivalent Antioxidant Capacity (TEAC) assay were chosen to evaluate the antioxidant activity of extracts. The polyphenol content in extracts ranged from 53.9 mg/L in pomelo extract to 510.5 mg/L in apricot extract. DPPH radical scavenging activity, calculated as EC_{50} value, ranged from 0.82% for apricot extract to 1.8% for pomelo extract. Antioxidant activity, expressed as TEAC values, ranged from 0.45 mmol/L for pomelo extract to 2.72 mmol/L for apricot extract. High correlations between polyphenol content and antioxidant activity expressed as EC_{50} values ($r = 0.944$), and as TEAC values ($r = 0.992$) in fruit extracts were observed. These correlations indicate that the scavenging radical activity and antioxidant activity of fruit extracts significantly depend on their polyphenol content.

Key words: fruit extracts, antioxidant activity, cosmetic products

1. INTRODUCTION

The medicinal plants are useful for healing of human diseases because of their phytochemical constituents. Herbal medicines have a strong traditional base and the potential to be useful as drugs in terms of safety and effectiveness, leads for treating different diseases [Prasad 2014].

Extracts produced from different parts of plants are also valuable ingredients of cosmetics, mainly because of the fact that they contain various phytochemical molecules such as: sugars, proteins, fats, glycosides, flavonoids, alkaloids, essential oils, tannins, mucilages, pectins, minerals and vitamins. Due to presence of these compounds, plant extracts exhibit many properties, such as antioxidant, anti-inflammatory, antimicrobial, anti-phlogistic, antiallergic and UV protective activities [Xiao Xian 1999; Xiao Xian 2000; Majeed and Prakash 2002; F'guyer, Afaq and Mukhtar 2003; Jassim and Naji 2003; Prakash, Satyan and Majeed 2003; Thornfeldt 2005]. Many current studies are now focused towards antioxidant activity of plant extracts (from roots, seeds, barks, fruits, flowers, leaves) rich in polyphenols that can be applied for prolonging the stability of cosmetic and pharmaceutical products. Natural antioxidant phytochemicals are available in all parts of higher plants. They have been found to protect against a variety of disorders, particularly cardiovascular diseases [Acheson and Williams 1985; Verlangieri *et al.* 1985] and some types of cancer [Collins 2005].

In the past years there has been a renewed interest in studying and quantifying the phenolic metabolites of fruits and vegetables due to their health-promoting properties. Fruit polyphenols include a wide range of compounds with antioxidant activity, that is, hydroxycinnamates, flavan-3-ols (condensed tannins), gallic acid derivatives (hydrolyzable tannins), flavonols, and anthocyanins [Gil *et al.* 2002].

This study is a continuation of the previous research [Malinowska and Zieliński 2011; Malinowska 2013; Malinowska, Gliszczyńska-Świgło and Szymusiak 2014] of antioxidant activities of natural extracts used in the cosmetics industry. The study is aimed at a preliminary screening of antioxidant activity of extracts isolated from fruits: *Citrus grandis* (pomelo), *Cucumis melo* (muskmelon), *Prunus armeniaca* (apricot), *Prunus persica*

(peach) and *Vitis vinifera* (grape), which are offered by their producers to application in cosmetics.

Scientific literature contains data indicating that these fruit extracts exhibit antioxidant activity. Literature provides data that pomelo fruit (*Citrus grandis*) is an excellent source of antioxidants including vitamin C, phenolics, carotenoids and exhibits scavenging ability for different free radicals including DPPH, superoxide anion, and hydrogen peroxide free radicals, similar to the abilities exhibited by BHA and vitamin C [Tsai, Chang S.K and Chang S.J 2007]. Moreover, independent studies confirmed, that pomelo showed reducing power, antioxidant capacity [Lim *et al.* 2006; Cho, Yang and Kim 2009; Toh, Khoo and Azrina 2013], antidiabetic [Kim, Shin and Jang 2009] and anticancer effects [Lim *et al.* 2009]. Scientific literature contains also data [Vouldoukis *et al.* 2004; Menon and Ramana Rao 2012] indicating that muskmelon fruit (*Cucumis melo*) shows the antioxidant activity. Independent studies confirmed the lipophilic and hydrophilic antioxidant capacity of fresh and dried apricot fruit (*Prunus armeniaca*) cultivars [Davarynejad *et al.* 2010; Milošević *et al.* 2012]. Moreover, the results of other studies confirmed that the apricot fruit extracts decrease lipid peroxidation level and have scavenging effect on the DPPH radical [Ozsahin and Yilmaz 2010] and ABTS radical [Sharma, Satpathy and Gupta 2014]. There are also scientific reports indicating that *Prunus persica* (peach) fruit extracts show free radical scavenging capacity and iron-reducing capacity [Mariäa *et al.* 2002]. Moreover, literature provides data concerning antioxidant capacity [Yang, Martinson and Liu 2009] and radical scavenging activity of *Vitis vinifera* (grape fruit) [Dani *et al.* 2009].

The aim of this work was to determine and compare the content of phenolics and the antioxidant capacity of 5 above fruit extracts, which are used in commercially available cosmetic products. In order

to evaluate the total antioxidant activity of these extracts two tests were chosen: DPPH radical scavenging activity assay and Trolox Equivalent Antioxidant Capacity (TEAC) assay.

2. EXPERIMENTAL

2.1. Chemicals

Methanol were purchased from Chempur (Piekary Śląskie, Poland), 1,1-Diphenyl-2-picryl-hydrazyl radical (DPPH$^{\bullet}$) and Trolox® (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) were from Sigma Chemicals Co (St. Louis, MO, USA). Folin–Ciocalteu phenol reagent was purchased from Merck (Darmstadt, Germany), gallic acid was from Acros Organics (New Jersey, USA). ABTS (Diammonium 2,2'-azino-bis(3-ethylbenz thiazoline-6-sulphonate) was from Roche Diagnostics, potassium persulphate was from POCH S.A. (Poland). All chemicals were of analytical grade.

2.2. Fruit extracts

Five cosmetic fruit extracts were obtained from few cosmetic companies:

- *Prunus armeniaca* (apricot) hydroglycolic fruit extract;
- *Vitis vinifera* (grape) glycolic fruit extract;
- *Prunus persica* (peach) glycolic fruit extract;
- *Cucumis melo* (muskmelon) glycolic fruit extract;
- *Citrus grandis* (pomelo) hydroglycolic fruit extract.

Fruit extracts were preserved by parabens' mixture. They were stored at room temperature in the dark.

2.3. Determination of total phenolic content

The amount of total phenolics in fruit extracts was determined by colorimetric method according to the Folin-Ciocalteu procedure using gallic acid as the standard [Singleton and Rossi 1965]. The total phenolic content was expressed in mg of gallic acid equivalents (GAE) per liter of extract [mg GAE/L of extract]. Data are presented as mean ± SD of triplicate measurements.

2.4. DPPH radical scavenging assay

The free radical scavenging activity of fruit extracts using DPPH radical was measured according to the procedure described by Sanchez-Moreno, Laurrauri and Saura-Calixto [1998].

The $DPPH^{\bullet}$ concentration in the reaction mixture was calculated from its calibration curve. The percentage of remaining $DPPH^{\bullet}$ was calculated according following formula:

$$\%DPPH^{\bullet}_{REM} = \frac{[DPPH^{\bullet}]_{t=10}}{[DPPH^{\bullet}]_{t=0}} \times 100\%$$

where: $[DPPH^{\bullet}]_{t=0}$ is the remaining concentration of the stable radical without the extract,
$[DPPH^{\bullet}]_{t=10}$ is the remaining concentration of the stable radical after 10 min.

The concentration of fruit extract necessary to decrease the initial $DPPH^{\bullet}$ concentration by 50% (EC_{50} [v/v]) was obtained from $DPPH^{\bullet}$ calibration curve. Radical scavenging activity of fruit extracts (AA_{DPPH}) was calculated as $1/EC_{50}$ according to method described by Maisuthisakul, Suttajit

and Pongsawatmanit [2007]. The lower EC_{50} value indicates the higher DPPH radical scavenging activity. AA_{DPPH} value is opposite one, the higher value means the higher antiradical activity.

2.5. TEAC (Trolox Equivalent Antioxidant Capacity) assay

Trolox equivalent antioxidant capacity (TEAC) of fruit extracts was measured according to the procedure described by Re *et al.* [1999]. The resulting TEAC values were expressed in mmol Trolox per liter of extract [mmol Trolox/L of extract]. Data are presented as mean ± SD of three replicates.

2.6. Statistical analysis

All analyses were performed in triplicate. The recorded results were subjected to statistical analysis using SPSS Statistics 14.0. The results were interpreted at the significance level $p = 0.05$.

3. RESULTS AND DISCUSSION

The polyphenol content varied widely in extracts tested and ranged from 53.9 mg/L in pomelo extract to 510.5 mg/L in apricot extract (Fig. 1). No significant difference was observed for grape and peach extracts. The polyphenol content in apricot extract was 2-fold higher than in grape and peach extracts, 3-fold higher than in muskmelon extract, and 9.5-fold higher than in pomelo extract.

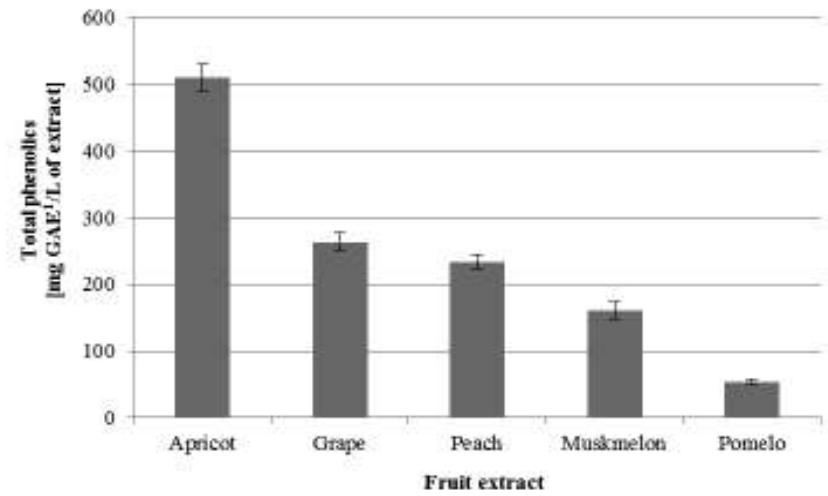

Figure 1. Total phenolics content in fruit extracts. [1]GAE – gallic acid equivalents. Source: Author's own work

DPPH radical scavenging activity, calculated as EC_{50} value, ranged from 1.8% for pomelo extract to 0.82% for apricot extract (Fig. 2), and calculated as AA_{DPPH}, ranged from 0.56 for pomelo extract to 1.22 for apricot extract (Fig. 3).

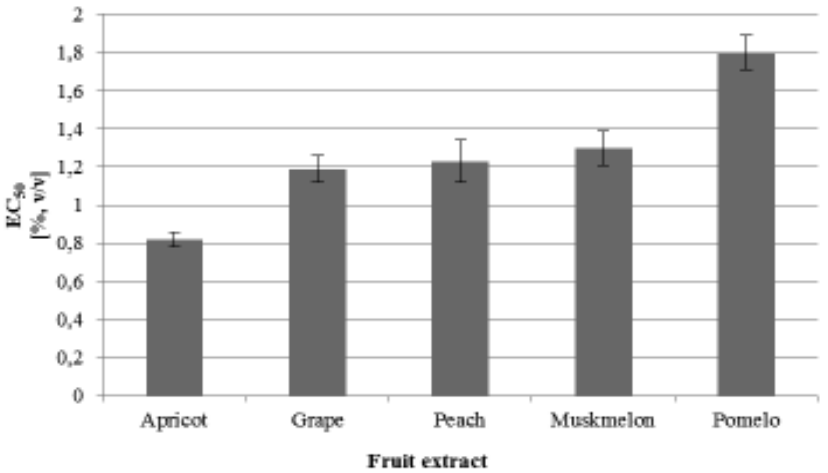

Figure 2. DPPH radical scavenging activity of fruits extracts, expressed as EC_{50}. Source: Author's own work

Radical scavenging activity of grape extract was statistically the same as observed for peach and muskmelon extracts. Apricot extract showed about 1.5-fold DPPH radical scavenging activity higher than grape, peach and muskmelon extracts, and 2,2-fold higher than pomelo extract. The order of DPPH radical scavenging activity of fruit extracts was as follows: apricot > grape ≥ peach ≥ muskmelon > pomelo.

Figure 3. DPPH radical scavenging activity of fruits extracts expressed as AA_{DPPH}. Source: Author's own work

Antioxidant activity, expressed as TEAC values, ranged from 0.45 mmol/L for pomelo extract to 2.72 mmol/L for apricot extract (Fig. 4). No significant difference was observed for peach and muskmelon extracts. The highest TEAC value was observed for apricot extract, which contained also the highest amount of polyphenols. It showed about 1.8-fold higher antioxidant activity than grape extract, 2.5-fold higher than peach extract, 2.7-fold higher than muskmelon extract, and 6.8-fold higher than pomelo extract. The order of antioxidant activity of fruit extracts (expressed as TEAC value) was as follows: apricot > grape > peach ≥ muskmelon > pomelo.

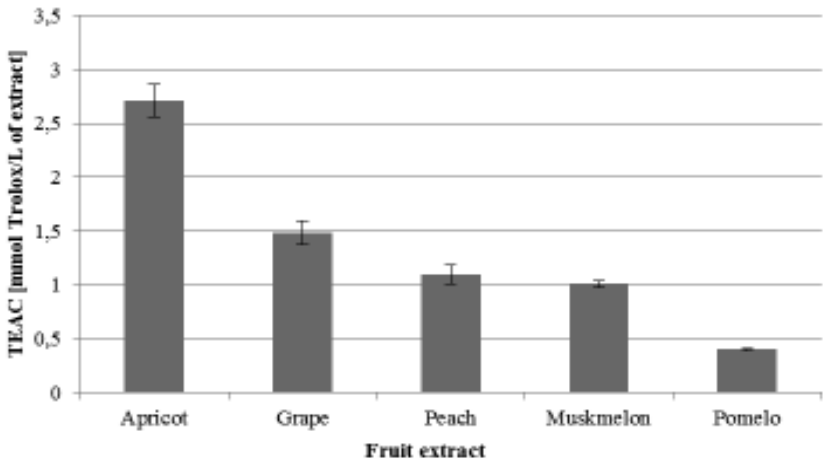

Figure 4. Antioxidant activity of fruits extracts expressed as TEAC values. Source: Author's own work

High correlations between polyphenol content and antioxidant activity expressed as EC_{50} values (r = 0.944), and as TEAC values (r = 0.992) in fruit extracts were observed. These correlations indicate that the scavenging radical activity and antioxidant activity of fruit extracts significantly depend on their polyphenol content.

Table 1. Cosmetic properties and application of tested fruit extracts

Plant extract	Cosmetic properties	Application in cosmetics
Apricot *(Prunus armeniaca)*	- softening - moisturizing - antioxidant - free radical scavenging - sebum reducing - trans-epidermal water loss (TEWL) regulatory - cell regeneration stimulating - exfoliating	- creams, lotions, moisturizers, masks, and products formulated specifically for dry and mature skin - anti-ageing cosmetics - light cream-gels for oily skin - photo-protective products - after sun products - body and hand preparations - bath products, and soaps - peelings

Grape *(Vitis vinifera)*	- antioxidant - free radical scavenging - protecting skin stem cells against UV stress - preventing the photo-aging - anti-inflammatory - reducing the skin irritations - vasoprotective - reducing the damage of hair during styling - hair growth stimulating	- skin creams, lotions, toners, moisturizers for sensitive skin and for skin with dilated capillaries - anti-ageing products - sun and after sun products - hair care products, especially conditioners
Peach *(Prunus persica)*	- emolient - refreshing - antioxidant - free radical scavenging - moisturizing - trans-epidermal water loss (TEWL) regulatory - cellular regeneration stimulating - soothing - stimulating the hair growth	- creams, moisturizers, lotions, milks, serums, gels and masks for oily and sensitive skin - anti-ageing products - photoprotective cosmetics - after sun products - bath products, and soaps - body and hand preparations - hair care products, especially conditioners
Muskmelon *(Cucumis melo)*	- moisturizing - softening - antioxidant - free radical scavenging - nourishing - softening - emollient - exfoliating - revitalizing	- moisturizing creams, masks, gels for dry and mature skin - anti-aging skin care products - sun and after sun products - body care products, such as lotions, balms, milks - peelings - hair care products, such as shampoos, conditioners - bath care products
Pomelo *(Citrus grandis)*	- antiseptic - astringent - antioxidant - free radical scavenging - protective properties on capillary vessels - normalizing the permeability of capillary vessels - revitalizing - hair growth stimulating	- creams, lotions, toners, moisturizers for oily and acne-prone skin care - skin creams, lotions, toners, moisturizers for skin with dilated capillaries - make-up products - hair care products, especially shampoos for oily hair and conditioners - cleansing product: soaps, bath capsules, oils, tablets, and salts

Source: Technical data sheets of fruit extracts

Tested fruit extracts, apart from phenolics, contain different constituents, which can influence their activities. Activities and application of studied fruit extracts in cosmetics are presented in table 1. Tested fruit extracts, especially apricot and grape extracts, can be proposed as a source of natural antioxidants to cosmetic topical formulations. Apart from the antioxidant activity, these fruit extracts have many other properties, therefore their application allows to formulate comprehensive cosmetics.

4. CONCLUSIONS

The number of fruit extracts used in skin and hair care products is very large and it has increased lately as a result of the additional benefits being proven by contemporary cosmetic researchers. Fruit extracts were chosen for antioxidant tests, as they are used as active ingredients in cosmetic products with many caring properties. Present results confirmed that some fruit extracts can be also a potential source of natural antioxidants for prolonging the oxidative stability of skin care products as well as the active ingredients in anti-ageing products. As a result of this study it was found that antioxidant and free radical scavenging activities of fruit extracts are significantly affected by their polyphenol content. Commercial cosmetic fruit extracts are available and used in cosmetics industry, however, their polyphenol content and the antioxidant as well as free radical scavenging activity vary among different extracts significantly.

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STRESZCZENIE

Celem pracy było zbadanie zawartości polifenoli oraz aktywności przeciwutleniającej pięciu handlowych kosmetycznych ekstraktów z owoców dostępnych na polskim rynku kosmetycznym: pomelo (*Citrus grandis*), melona (*Cucumis melo*), moreli (*Prunus armeniaca*), brzoskwini (*Prunus persica*) i winogron (*Vitis vinifera*). Do oceny właściwości przeciwutleniających zastosowano test zdolności wygaszania rodnika DPPH oraz metodę TEAC.

Zawartość związków polifenolowych różniła się znacznie między badanymi ekstraktami i mieściła się w granicach od 53.9 mg/L w ekstrakcie z pomelo do 510.5 mg/L w ekstrakcie z moreli. Zdolność do wygaszania rodnika DPPH, wyrażona jako parametr EC_{50}, mieściła się w granicach od 0.82% dla ekstraktu z moreli do 1.8% dla ekstraktu z pomelo. Potencjał przeciwutleniający TEAC mieścił

się w granicach od 0.45 mmol/L dla ekstraktu z pomelo do 2.72 mmol/L dla ekstraktu z moreli. Zaobserwowano istnienie wysokich korelacji między zawartością związków polifenolowych w badanych ekstraktach a ich właściwościami przeciwutleniającymi wyrażonymi jako EC_{50} (r = 0.944), oraz TEAC (r = 0.992). Otrzymane korelacje wskazują na znaczący wpływ zawartości polifenoli w badanych ekstraktach na ich zdolność do wygaszania rodnika DPPH oraz ich potencjał przeciwutleniający TEAC.

Słowa kluczowe: ekstrakty z owoców, aktywność przeciwutleniająca, produkty kosmetyczne