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Research Article

Photoprotective and antioxidant effects of *Citrus limon* and *Citrus sinensis* Peels: Comparative investigation of the efficiency of five extraction solvents

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ABSTRACT

Photoaging and photo-carcinogenesis are primarily caused by repeated exposure to ultraviolet (UV) radiation. The use of phytochemical with an antioxidant capacity as photoprotector has increased recently. Citrus fruit's waste such as peels are considered as one of the resources of antioxidant. The aim of this study was to investigate the effect of five Soxhlet extraction solvents i.e., hexane, chloroform, acetone, ethyl acetate and ethanol on the photoprotective activity of *Citrus limon* and *Citrus sinensis* peels. The antioxidant activity of the extract was evaluated using 2,2'-diphenyl-1-pierylhydrazyl (DPPH) assay while photoprotective properties was investigated based on sun protection factor (SPF) and UV absorption ability. The acetone extract of *C. limon* (IC_{50}:15.30±2.13mg/mL) and *C. sinensis* (IC_{50}: 26.05±5.19mg/mL) peels exhibited the highest antioxidant activity while ethanolic, ethyl acetate and chloroform extracts of *C. sinensis* peel exhibited similar scavenging activity with IC_{50} values ranging between 31-33mg/mL (p>0.05). Hexane extract of both fruit peels showed the lowest antioxidant activity (IC_{50}>50mg/mL). Interestingly, the opposite was observed in photoprotective activity for *C. limon* peel extracts with chloroform extract which showed the highest photoprotective activity (SPF:9.06±1.96) followed by hexane (SPF:6.96±0.22), ethyl acetate (SPF:5.11±1.63), ethanol (SPF:4.95±2.38) and lastly acetone (SPF:1.39±0.40). Similarly, acetone extract of *C. sinensis* peels demonstrated the least photoprotective activity (SPF:1.96±0.28) followed by ethanolic extract (SPF:2.70±0.51). Three extracts i.e., hexane (SPF:6.75±0.33), ethyl acetate (SPF:7.34±0.05) and chloroform (SPF:7.90±0.12) of *C. sinensis* peel revealed similar potential in photoprotection. The correlation between both DPPH IC_{50} and SPF values of *C. limon* and *C. sinensis* peel extracts are not significant (p>0.05). In terms of UV absorption, all extracts of *C. limon* peel demonstrated high UV absorption at UVB region (280-320nm) except acetone extract with high UV absorption at UVA region (320-400nm). Meanwhile, all extracts of *C. sinensis* peel showed broad absorption at UVA and UVC regions with the highest absorption detected at 310-350nm. This finding suggests that ethyl acetate extract of *C. sinensis* could be used as a natural sunscreen in pharmaceutics due to its valuable antioxidant and photoprotective activities.

Keywords: Antioxidant, *Citrus species* photoprotective, photo-carcinogenesis, sunscreen

INTRODUCTION

The skin is the largest organ acts as the main barrier protecting the body from environmental attack including solar radiation (Kanitakis, 2002). Solar radiation, an electromagnetic radiation emitted by the sun consists mainly infrared (IR), visible and ultraviolet (UV) light (Duthie *et al*., 1999). Repetitive exposure of the skin to

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sunlight lead to extrinsic aging or photoaging including wrinkling, scaling, dryness, hypo- and hyperpigmentation. The extreme consequence of photodamage is skin cancer (Pinnell, 2003). These harmful effects are mainly associated with the UV radiation which is divided into three different bands: UV-A (320-400 nm), UV-B (290-320 nm), and UV-C (200-290 nm). Different bands of UV radiation results in distinctly effects on living tissue (Tuchinda *et al*., 2006).

The UV-A radiation which can be subdivided into UV-A1 (340-400 nm) and UV-A2 (315-340 nm) is the major UV radiation (over 90%) reaching the earth's surface and penetrate deep into epidermis and dermis of skin. The skin cells including keratinocytes, melanocytes, fibroblasts, and endothelial cells in blood vessels are affected by 80% of UV-A radiation (Svobodová and Vostálová, 2010). Keratinocytes contain high levels of endogenous antioxidant such as glutathione, superoxide dismutase and catalases that are involved in antioxidant defense (Liochev and Fridovich, 1994). However, endogenous photosensitizer or chromophores interact with UV-A photons produce high levels of reactive oxygen and nitrogen species which can oxidize cellular proteins, lipids, and saccharides that overwhelm the body's natural defense to oxidative damage. The reactive oxidative species also induce various types of oxidative DNA lesions including single-strand breaks, and DNA bases alteration (Svobodová and Vostálová, 2010). Oxidative damage to biomolecules increases the expression of signalling molecules including inflammatory cytokines, transcription factors, matrix metalloproteinases (MMP), mitogen-activated protein kinases (MAPK), or pro- and anti-apoptotic genes resulting in the development of pathological changes in skin tissue (Skarupova *et al*., 2020).

About 50% of UV-induced photodamage is caused by the generation of reactive oxidative species *via* cellular photosensitization (Bernstein *et al*., 2004). It has been established that most of the endogenous chromophores absorb in UV-B range although UV-A exposure results in more oxidation than UV-B (Trautinger, 2001). UV-B is directly absorbed by DNA in epidermal cells causing the DNA mutations which activate skin melanogenesis or formation of "sunburn" cells in the epidermis *via* apoptosis of keratinocytes induced by p53 proteins. Prolonged irradiation can suppress the p53 proteins resulting in the accumulation of damaged cells and the initiation of mutagenesis and photocarcinogenesis (Gilchrest *et al*., 1996).

Photoprotection of human skin using sunscreen is focused on the prevention of acute sunburn and chronic skin damage including photoaging and skin cancer that may result from exposure of UV-A and UV-B radiation (Mc Daniel *et al*., 2018). Many research studies are being conducted on evaluation and development of sunscreen containing plant-derived antioxidant compounds, especially carotenoids and flavonoids because they protect against sun damage by filtering the UV and neutralized the UV-induced free radicals. Phenolic compounds such as flavonoids can act as sun filters due to the presence of aromatic rings that can absorb UV-A and UV-B at a wavelength range of 200-400 nm (De Cooman *et al*., 1998; Wang *et al*., 2011).

Carotenoids, pigments that responsible for the citrus fruit peel's color are synthesized using light as essential source during fruit development. The intense coloration of citrus fruit was primarily cause by the high concentration of total carotenoids (Yokoyama and Vandercook, 1967). Major carotenoids in citrus fruits such as α-carotene, β-carotene, lutein, zeaxanthin, and β-cryptoxanthin have two health care effects including anti-oxidative activity and as vitamin precursor to slow down aging process and promote immune functions (Shan, 2016). Previous study done by DellaPenna and Pogson (2006) revealed that carotenoids were the best natural component served as UV protector. Meanwhile, the presence of antioxidants such as vitamins C and E, and polyphenols of *Citrus limon* peel contribute to endogenous photo-protection (Fernández-García, 2014). Therefore, citrus fruit peels have gained considerable attention for their potential use as effective agent for preventing or reducing UV-induced oxidative damage, photoaging and skin cancer. There are wide range of carotenoids components in the nature with wide or varied properties. Comparative study of different extraction method for carotenoids is important to enhance its true properties. In this study, fruit peels of lemon (*Citrus limon*) and orange (*Citrus sinensis*) were selected to access the influence of the extraction solvents (hexane, chloroform, acetone, ethyl acetate and hexane) on the photoprotective activity of their crude extracts.

MATERIALS AND METHODS

Sample collection

About 30 *C. limon* and *C. sinensis* fruits were purchased at fruit market in Giant, Shah Alam, Selangor. The fruits were carefully selected without any blemish and defects. The fruits were washed with tap water while the peels were removed and cut into smaller pieces before freeze-dried. The dried peels were grinded into uniform powder by using electrical blender.

Extraction

The dried peels powders were extracted using Soxhlet extractor based on modified method adapted from Dumbravã *et al*. (2010). The organic solvents i.e., hexane, chloroform, acetone, ethyl acetate and ethanol were used, and extraction were done with 4 cycles. The obtained extracts were concentrated under vacuum at 45°C.

2,2-Diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging activity

Antioxidant activity of extract was measured by using DPPH free radical scavenging activity according to (Shirazi *et al*., 2014), with some modifications. About 3 mL of DPPH solution (4 mg in 200 mL of methanol) was mixed with 50 µL of extracts from various concentrations (10-500 µg/mL) and incubated in darkness for 15 minutes. The absorbance was measured at 515 nm. Dimethyl sulfoxide (DMSO) was used as negative control while ascorbic acid was used as positive control. An antioxidant reduces DPPH solution from deep violet to yellow color. The percentage of inhibition of DPPH by extracts was calculated by using the following formula:

$$\text{DPPH radical scavenging activity (\%)} = [(Abs_{negative\ control} - Abs_{sample})] / (Abs_{negative\ control})] \times 100$$

Where, $Abs_{negative\ control}$ is the absorbance of DPPH radical + DMSO

Abs_{sample} is the absorbance of DPPH radical + sample extract/ standard

The IC_{50} values of extracts (the concentration at which 50% of DPPH solution is scavenged) were calculated based on the linear regression graph. Ascorbic acid with different concentrations (20, 40, 60, 80 and 100 µg/mL) was used as a positive control for comparison. All experiments were performed in triplicate.

Sun protection factor (SPF) measurement

The absorbance of the prepared extracts (100ìg/mL) was determined between 290 and 320 nm by using UV-Visible spectrophotometer (Kaur and Saraf, 2010). Dimethyl sulfoxide (DMSO) was used as a blank and measurements were made in triplicates. The obtained absorbance (λ) was multiplied with the respective EE (I) values and their summation was taken and multiplied with the correction factor (10). The formula as stated below:

$$\text{SPF spectrophotometric} = CF \times \sum_{290}^{320} EE(\lambda) \times I(\lambda) \times Abs(\lambda)$$

Where, EE (λ) is the erythmogenic effect of radiation with wavelength λ

I (λ) is the solar intensity spectrum

EE (λ) x I (λ) are constants

Abs (λ) is the absorbance values at wavelength λ of test sample

CF is the correction factor (= 10)

UV absorption spectrum measurement

The UV absorption spectrum for each test sample (100 µg/ mL) was measured over a wavelength range of 200 to 400 nm by using UV-Visible spectrophotometer. The absorption spectrum of the extracts was compared to that of epigallocatechin gallate (EGCG) standard (100 µg/mL) (Patil *et al*., 2009).

Statistical analysis

All results presented as mean ± standard deviation. Data were analyzed statistically using ANOVA with significance level set at $p<0.05$ and post-hoc Tukey procedure was carried out with SPSS 21 for Windows. Correlation between antioxidant activities and sunscreen protection activities was performed using the Pearson's method (Fidrianny *et al*., 2018).

RESULTS AND DISCUSSION

The peel's extracts of *C. limon* and *C. sinensis* produced from Soxhlet method using various solvents such as ethanol, ethyl acetate, acetone, chloroform, and hexane showed different yield in this study (Table 1). Extraction yield ranged from 4.2% to 15.0% and 3.0% to 23.1% for *C. limon* and *C. sinensis*, respectively. It was noticed that extraction yield increases with increasing polarity of the solvent used in extraction for *C. limon* peels: hexane < chloroform < acetone < ethyl acetate < ethanol. Generally, variation in the extraction yield can be explained *via* different types of bioactive compounds and their different solubilities in various types of solvents. Lipophilic carotenoids can be easily extracted *via* organic solvents, whereas hydrophilic bioactive compounds such as ascorbic acid, pectin, and sugars can be extracted *via* aqueous or aqueous alcoholic solvents (Papoutsis *et al*., 2016).

Table 1: Extraction yield of *C. limon* and *C. sinensis* peels produced from Soxhlet extraction

Solvent	*C. limon* peels	*C. sinensis* peels
Ethanol	15.0%	23.1%
Ethyl acetate	9.9%	6.1%
Acetone	6.6%	22.8%
Chloroform	5.9%	11.2%
Hexane	4.2%	3.0%

In terms of extraction yield, solvent such as alcohol were found to be the best options. According to Gotmare and Gade (2018), methanolic extract of orange peels recorded the highest extraction yield of 55.6% compared to distilled water (3.2%), acetone (2.2%) and hexane (1.8%). Similar findings were also reported by Arora & Kaur (2013) on extraction yield of orange peels where methanolic extract gave the highest yield of 60.6% compared to distilled water (12.7%), acetone (1.5%) and hexane (1.2%). Extraction yield for acetone extract of orange peels obtained in this study was higher compared to the aforementioned studies. This differences in extraction yield could be due to slow hours of shed drying (92 hours) and air drying (48 hours) methods. In this study, freeze drying (24 hours) method was used. Besides processing methods, the variation of extraction yield could also be explained by the intrinsic and extrinsic factors like age of plant, geographical climate, nature of the soil, and season. Muraina and coworkers (2008) reported variation of percentage yield in *Anoigeissus leiocarpus* from two different regions of Nigeria: Zaria and Jos (cooler due to lower ambient temperature). Similarly, Iloki-Assanga *et al*. (2015) reported significant differences in extraction yield of phenolic compounds from *Phoradendron californicum* of oak and mesquite, revealing that environment in which the plants lived could be one of the contributing factors.

The DPPH radical scavenging activity of *C. limon* and *C. sinensis* peels extracts varied from 12.00-90.35% and 6.13-71.87%, respectively at concentrations ranging from 8-49 mg/mL (Figure 1 and 2). Among the tested extracts, greatest antioxidant activity was observed in acetone extract for *C. limon* (IC_{50}:15.30 ± 2.13 mg/mL) and *C. sinensis* (IC_{50}: 26.05±5.19 mg/mL) (Table 2). With the same solvent (acetone), the highest extraction yield was observed for *C. sinensis* peels in this study. Comparative studies of antioxidant activity using different solvent extracts of orange peels done by Park *et al.* (2014) also suggesting acetone as the best solvent for extracting antioxidant compounds i.e., phenolics with good radical scavenging action when compared to other solvents like ethanol and methanol. Phenolics and other phytochemicals such as flavonoids, alkaloids, and terpenoids possess powerful antioxidant activity which can protect the human body against oxidative damage through scavenging various types of reactive oxygen species (Truong *et al*., 2019). Meanwhile, hexane extract for both citrus fruit peels were poor in free radical scavenging activity with IC_{50} value of more than 50 mg/mL. All *C. limon* extracts (except for hexane extract) exhibited good radical scavenging activity, which is comparable to the previous findings reported by Chatha *et al*. (2011) for methanolic *C. limon* peels i.e., 20-40 mg/mL. The statistical analysis showed that scavenging activity of *C. limon* peels was found to be varied significantly (p=0.000) between all extracts (ethanol, ethyl acetate and acetone) and chloroform extract. The antioxidant results obtained in this study is also similar to that reported by Park *et al*. (2014). The authors had shown that the DPPH radical scavenging activity of orange peel extract obtained by using acetone (IC_{50}: 0.78 mg/mL) as extracting solvent was higher than that achieved by the using of solvents such as ethanol (IC_{50}: 1.14 mg/mL) and methanol (IC_{50}: 1.40 mg/mL). Variations in the antioxidant potential can be explained by the extraction methods used.

In contrary, the photoprotective activity was opposite to the findings in DPPH activity. Chloroform extract showed

Figure 1: DPPH radical scavenging activity of different Soxhlet extracts from peels of *C. limon* by different solvents at various concentrations. Each value represents a mean ± SD (n = 3)

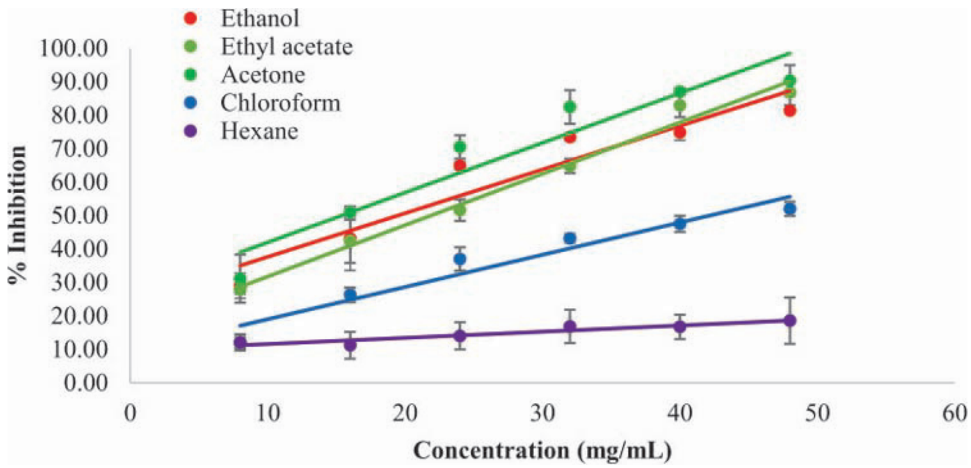

Figure 2: DPPH radical scavenging activity of different Soxhlet extracts from peels of *C. sinensis* by different solvents at various concentrations. Each value represents a mean ± SD (n = 3)

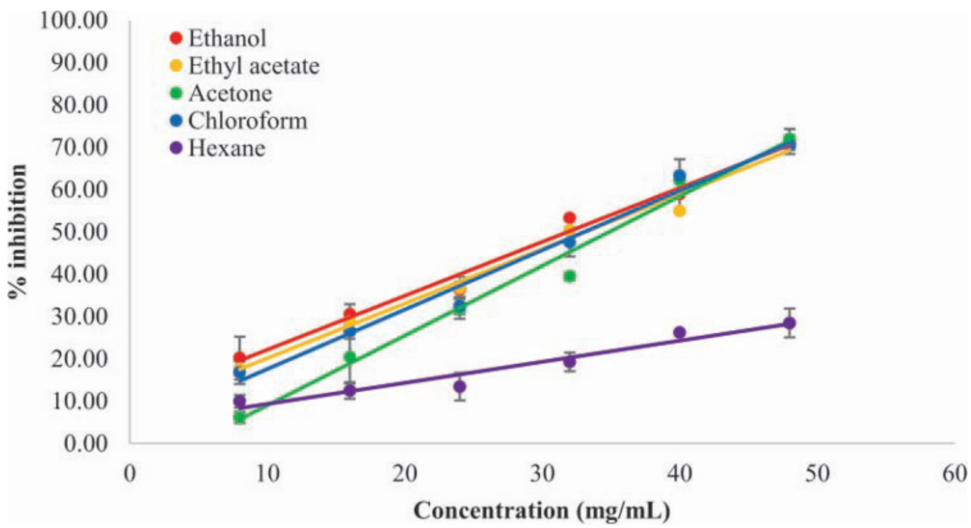

Table 2: IC_{50} value of DPPH scavenging activity of *C. limon* and *C. sinensis* peel extracts. Each value represents a mean ± SD (n = 3).

Solvent	**IC_{50} value (mg/mL)**	
	***C. limon* peels**	***C. sinensis* peels**
Ethanol	19.21 ± 2.75	31.84 ± 1.62
Ethyl acetate	21.82 ± 1.33	33.14 ± 0.72
Acetone	15.30 ± 2.13	26.05 ± 5.19
Chloroform	42.16 ± 1.49	33.08 ± 0.75
Hexane	> 50	> 50

the highest photoprotective activity with SPF value of 9.06 ± 1.96 for *C. limon* and 7.90 ± 0.12 for *C. sinensis* (Table 3 & 4). All reported SPF results of *C. limon* peel extracts were found not significantly different, except for chloroform and acetone extract (p=0.001); and acetone and hexane extract (p=0.015). Nevertheless, all reported SPF results of *C. sinensis* were found significantly different (p=0.000) except for chloroform and acetone extracts (p=0.131). All peel extracts of *C. limon* except for acetone extract showed comparatively higher SPF values than reported study done by Kaur and Saraf (2010) for hydroalcoholic dilutions of lemon oil i.e., 2.810. Similarly, the SPF value of *C. sinensis* peel extracts (chloroform, ethyl acetate and hexane) in this study was higher as compared to the study done by Khelker *et al*. (2017) for 200 µg/mL of petroleum ether extract of orange peels produced from Soxhlet extraction method, i.e., 3.086±0.14. Thus, it can be concluded that chloroform solvent is efficient for enhancing the photoprotective activity of citrus fruit peels. This is probably due to the

Table 3: *In vitro* Sunscreen protection factor (SPF) of *C. limon* peel extracts. Each value represents a mean ± SD (n = 3)

nm	Ethanol	Ethyl acetate	Acetone	Chloroform	Hexane
290	0.06 ± 2.21	0.04 ± 1.53	0.03 ± 1.14	0.15 ± 2.26	0.06 ± 0.16
295	0.38 ± 2.10	0.34 ± 1.58	0.11 ± 0.27	0.97 ± 0.02	0.46 ± 0.15
300	1.39 ± 2.25	1.38 ± 1.61	0.35 ± 0.21	3.18 ± 0.06	1.89 ± 0.17
305	1.62 ± 2.38	1.68 ± 1.63	0.46 ± 0.40	3.34 ± 0.02	2.28 ± 0.22
310	0.95 ± 2.51	0.99 ± 1.64	0.36 ± 0.08	1.74 ± 0.03	1.29 ± 0.23
315	0.44 ± 2.63	0.45 ± 1.65	0.17 ± 0.37	0.65 ± 1.65	0.53 ± 0.21
320	0.09 ± 1.99	0.09 ± 1.61	0.05 ± 1.16	0.13 ± 1.54	0.10 ± 0.20
SPF	4.95 ± 2.38	5.11 ± 1.63	1.39 ± 0.40	9.06 ± 1.96	6.96 ± 0.22

Table 4: *In vitro* Sunscreen protection factor (SPF) of *C. sinensis* peel extracts. Each value represents a mean ± SD (n = 3)

nm	Ethanol	Ethyl acetate	Acetone	Chloroform	Hexane
290	0.02 ± 1.31	0.10 ± 1.62	0.02 ± 0.30	0.06 ± 1.70	0.10 ± 1.34
295	0.16 ± 1.06	0.56 ± 1.03	0.13 ± 0.55	0.41 ± 1.12	0.54 ± 0.91
300	0.65 ± 0.75	1.99 ± 0.63	0.28 ± 0.85	1.62 ± 0.74	1.88 ± 0.51
305	0.82 ± 0.55	2.33 ± 0.44	0.64 ± 0.28	2.05 ± 0.50	2.17 ± 0.33
310	0.51 ± 0.38	1.38 ± 0.31	0.27 ± 0.21	1.29 ± 0.31	1.24 ± 0.23
315	0.24 ± 0.27	0.64 ± 0.25	0.16 ± 0.10	0.63 ± 0.20	0.55 ± 0.19
320	0.05 ± 0.19	0.14 ± 0.21	0.02 ± 0.42	0.14 ± 0.12	0.11 ± 0.18
SPF	2.70 ± 0.51	7.34 ± 0.05	1.96 ± 0.28	7.90 ± 0.12	6.75 ± 0.33

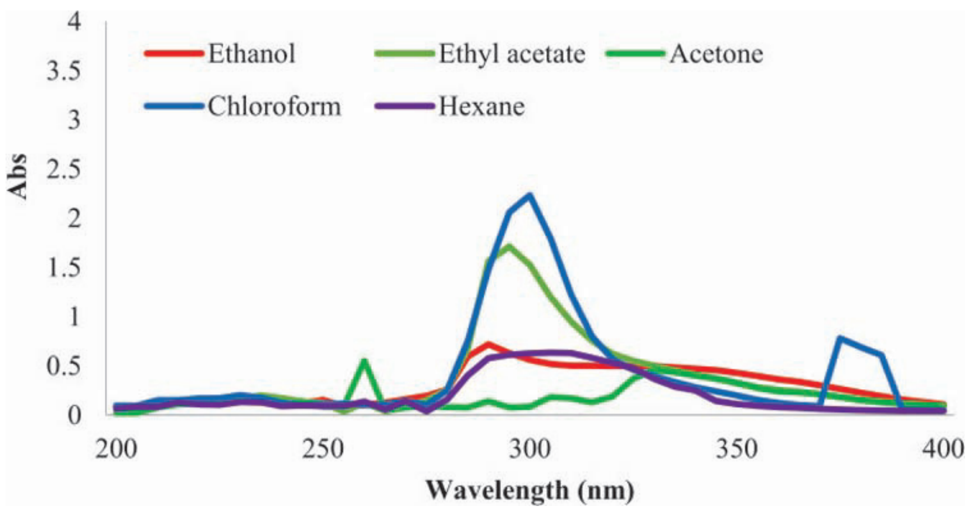

Figure 3: UVA/UVB absorption spectrum of different Soxhlet extracts from peels of *C. limon* by different solvents

presence of high concentration of apolar bioactive compounds like flavonoids (isoflavones, flavones and methylated flavones) and carotenoids since chloroform, an apolar solvent is commonly used to extract apolar nature compounds (Chávez-González *et al*., 2020; Saini & Keum, 2018).

In terms of UV absorption (Figure 3 & 4), all peel extracts of *C. limon* displayed a higher UVB absorption, except of acetone extract which showed higher UVA absorption; however, all peel extracts of *C. sinensis* showed broad absorption at UVA and UVC regions, while highest absorption was observed between 310-350nm (UVB and UVA regions). This result indicates the potential of *C. sinensis* peel extracts in preventing skin cancer as higher doses of UVB may cause sunburn which increase the likelihood of developing cancer. The potential of *Citrus*

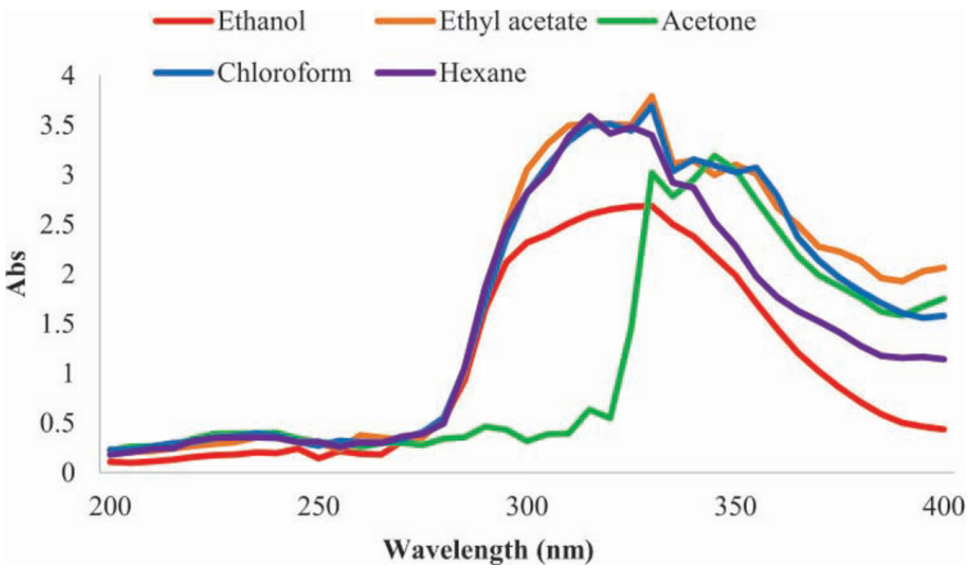

Figure 4: UVA/UVB absorption spectrum of different Soxhlet extracts from peels of *C. sinensis* by different solvents

Table 5: Correlations between the IC_{50} values of free radical scavenging activity and SPF value of different Soxhlet extracts from peels of *C. limon* and *C. sinensis* by different solvents. Each value in the table is represented as mean ± SD (n = 3). * Indicates significance at $p< 0.05$

IC_{50} of DPPH	Correlation R^2	
	SPF (*Citrus limon*)	SPF (*Citrus sinensis*)
Ethanol	0.825	-0.646
Ethyl acetate	0.603	-0.372
Acetone	-0.768	0.825
Chloroform	-0.981	0.975

sinensis as photoprotector against UV damage was supported by Yoshizaki *et al*. (2014). The authors had reported orange peel extract suppressed UVB-induced COX-2 expression and prostaglandin E_2 (PGE_2) production due to the presence of high levels of polymethoxy flavonoids (PMFs).

The correlation between DPPH IC_{50} and SPF values of *C. limon* and *C. sinensis* peel extracts which estimated using Pearson's method were found to be not significant (p> 0.05) in this study (Table 5). Positive correlations were observed in ethanol (*r*= 0.825) and ethyl acetate (*r*= 0.603) extracts of *C. limon*; in acetone (r= 0.825) and chloroform (r= 0.682) extracts of *C. sinensis*. Meanwhile, negative correlations were observed in acetone (*r*= -0.768) and chloroform (*r*= -0.981) extracts of *C. limon*; in ethanol (r= -0.646) and ethyl acetate (r= -0.372) extracts of *C. sinensis*. These findings were in agreement with previous study done by Ebrahimzadeh *et al*. (2014) that no correlation was found between SPF and antioxidant activity (*r*= 0.014, *p*= 0.560) in tested extracts i.e., *S. ebulus*, *Zea maize*, *F. sellowiana* and *C. pentagyna*.

CONCLUSION

Solvent chloroform is efficient in enhancing the photoprotective activity of citrus fruit peels due to its highest SPF value. High absorption was detected at both UVA and UVB regions from all tested extracts, suggesting its great potential as UV protector against harmful UV radiations. In general, the photoprotective activity of tested extracts did not has any negative impact on the antioxidant components. Further research should be conducted on identifying the photoprotective compounds in citrus extracts to increase its application and uses as cosmeceuticals for improving the quality of life.

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Conflict of interest

The authors declare no conflict of interest.

REFERENCES

Arora M and Kaur P (2013). Phytochemical screening of orange peel and pulp. *Intl. J. Res. Eng. & Technol.*, 2(12): 517-520.

Bernstein EF, Brown DB, Schwartz MD, Kaidbey K and Ksenzenko SM (2004). The polyhydroxy acid gluconolactone protects against ultraviolet radiation in an *in vitro* model of cutaneous photoaging. *Dermatologic Surg.*, 30(2): 189-196.

Chatha SAS, Hussain AI and Asi MR (2011). Evaluation of antioxidant potential of citrus peel extracts. *J. Chem. Soc. Pakistan*, 33(6): 863-868.

Chávez-González ML, Sepúlveda L, Verma DK, Luna-García HA, Rodríguez-Durán LV, Ilina A and Aguilar CN (2020). Conventional and emerging extraction processes of flavonoids. *Processes*, 8(4): 434.

De Cooman L, Everaert E and De Keukeleire D (1998). Quantitative analysis of hop acids, essential oils and flavonoids as a clue to the identification of hop varieties. *Phytochem. Analysis*, 9(3): 145-150.

DellaPenna D and Pogson BJ (2006). Vitamin synthesis in plants: tocopherols and carotenoids. *Ann. Rev. Pl. Biol.*, 57: 711-738.

Dumbrava DG, Hadaruga NG, Hadaruga DI, Moldovan C and Raba D (2010). Determination by RP-HPLC of β-carotene concentration from orange (*Citrus sinensis* L.) fruits peel extracts. *J. Agro alimentary Proc. & Technol.,* 16(2): 242-246.

Duthie MS, Kimber I and Norval M (1999). The effects of ultraviolet radiation on the human immune system. *The British J. Dermatol.*, 140(6): 995-1009.

Ebrahimzadeh MA, Enayatifard R, Khalili M, Ghaffarloo M, Saeedi M and Charati JY (2014). Correlation between sun protection factor and antioxidant activity, phenol and flavonoid contents of some medicinal plants. *Iranian J. Pharmaceu. Res.*, 13(3): 1041-1047.

Fernández-García E (2014). Skin protection against UV light by dietary antioxidants. *Food & Function*, 5(9): 1994-2003.

Fidrianny I, Anggraeni NAS and Insanu M (2018). Antioxidant properties of peels extracts from three varieties of banana (Musa sp.) grown in West Java-Indonesia. *Intl. Food Res. J.*, 25(1): 57-64.

Gilchrest BA, Park HY, Eller MS and Yaar M (1996). Mechanisms of ultraviolet light-induced pigmentation. *Photochem. and Photobiol.*, 63(1): 1-10.

Gotmare S and Gade J (2018). Orange peel: A potential source of phytochemical compounds. *Intl. J. Chem. Tech. Res.*, 11(2): 240-243.

Iloki-Assanga SB, Lewis-Luján LM, Lara-Espinoza CL, Gil-Salido AA, Fernandez-Angulo D, Rubio-Pino JL and Haines DD (2015). Solvent effects on phytochemical constituent profiles and antioxidant activities, using four different extraction formulations for analysis of *Bucida buceras* L. and *Phoradendron californicum*. *BMC Res. Notes*, 8(1): 1-14.

Kanitakis J (2002). Anatomy, histology and immunohistochemistry of normal human skin. *Eur. J. Dermatol.*, 12(4): 390-401.

Kaur CD and Saraf S (2010). *In vitro* sun protection factor determination of herbal oils used in cosmetics. *Pharmacognosy Res.*, 2(1): 22-25.

Khelker T, Haque N and Agrawal A (2017). Ultraviolet Protection potential of *Curcuma longa* L. and *Citrus sinensis* (L.) Osbeck. *Res. J. Pharm. Technol.*, 10(12): 4282-4284.

Liochev SI and Fridovich I (1994). The role of O_2 in the production of HO.: *in vitro* and *in vivo*. *Free Radical Biol. Medicine*, 16(1): 29-33.

McDaniel D, Farris P and Valacchi G (2018). Atmospheric skin aging-contributors and inhibitors. *J. Cosmetic Dermatol.*, 17(2): 124-137.

Muraina IA, Adaudi AO, Mamman M, Kazeem HM and Eloff JN (2008). Effects of geographical location on the yield and bioactivity of *Anoigeissus leiocarpus*. *J. Pharm. Bioresources*, 5(2): 68-72.

Papoutsis K, Pristijono P, Golding JB, Stathopoulos CE, Scarlett CJ, Bowyer MC and Van Vuong Q (2016). Impact of different solvents on the recovery of bioactive compounds and antioxidant properties from lemon (*Citrus limon L.*) pomace waste. *Food Sci. and Biotechnol.*, 25(4): 971-977.

Park JH, Lee M and Park E (2014). Antioxidant activity of orange flesh and peel extracted with various solvents. *Preventive Nutr. Food Sci.*, 19(4): 291-298.

Patil PR, Rakesh SU, Dhabale PN and Burade KB (2009). Simultaneous estimation of ramipril and amlodipine by UV spectrophotometric method. *Res. J. Pharm. Technol.*, 2(2): 304-307.

Pinnell SR (2003). Cutaneous photodamage, oxidative stress, and topical antioxidant protection. *J. Am. Acad. Dermatol.*, 48(1): 1-22.

Saini RK and Keum YS (2018). Carotenoid extraction methods: A review of recent developments. *Food Chem.*, 240: 90-103.

Shan Y (2016). *Comprehensive utilization of citrus by-products*. Academic Press.

Shirazi OU, Khattak MMAK, Shukri NAM and Nasyriq MN (2014). Determination of total phenolic, flavonoid content and free radical scavenging activities of common herbs and spices. *J. Pharmacog. Phytochem.*, 3(3): 104-108.

Skarupova D, Vostalova J and Svobodova AR (2020). Ultraviolet A protective potential of plant extracts and phyto-chemicals. *Biomedical Papers of the Medical Faculty of Palacky University in Olomouc*, 164(1): 1-22.

Svobodová A and Vostálová J (2010). Solar radiation induced skin damage: review of protective and preventive options. *Intl. J. Radiation Biol.*, 86(12): 999-1030.

Trautinger F (2001). Mechanisms of photodamage of the skin and its functional consequences for skin ageing. *Clin. Exp. Dermatol.*, 26(7): 573-577.

Truong DH, Nguyen DH, Ta NTA, Bui AV, Do TH and Nguyen HC (2019). Evaluation of the use of different solvents for phytochemical constituents, antioxidants, and *in vitro* anti-inflammatory activities of *Severinia buxifolia*. *J. Food Quality*, 2019(1): 1-9.

Tuchinda C, Srivannaboon S and Lim HW (2006). Photoprotection by window glass, automobile glass, and sunglasses. *J. Am. Acad. Dermatol.*, 54(5): 845-854.

Wang SQ, Osterwalder U and Jung K (2011). *Ex vivo* evaluation of radical sun protection factor in popular sunscreens with antioxidants. *J. Am. Acad. Dermatol.*, 65(3): 525-530.

Yokoyama H and Vandercook CE (1967). Citrus carotenoids. I. comparison of carotenoids of mature green and yellow lemons. *J. Food Sci.*, 32(1): 42-48.

Yoshizaki N, Fujii T, Masaki H, Okubo T, Shimada K and Hashizume R (2014). Orange peel extract, containing high levels of polymethoxyflavonoid, suppressed UVB induced COX 2 expression and PGE 2 production in HaCaT cells through PPAR γ activation. *Exp. Dermatol.*, 23(1): 18-22.

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Dean

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