Malaysian Journal of Analytical Sciences (MJAS)
Published by Malaysian Analytical Sciences Society

CORRELATION BETWEEN QUERCETIN CONTENT AND PHOTOPROTECTIVE ACTIVITY OF PLANT EXTRACTS

(Hubungan Antara Kandungan Quercetin dan Keupayaan Perlindungan Cahaya oleh Ekstrak Tumbuhan)

Amalia Sabirah Mohd Razali[1], Hannis Fadzillah Mohsin[1], Kathleen Jalani[2], and Nurhuda Manshoor[1,2*]

[1]*Faculty of Pharmacy, Universiti Teknologi MARA Selangor, Kampus Puncak Alam, 42300 Selangor, Malaysia*
[2]*Atta-ur-Rahman Institute for Natural Products Discovery, Universiti Teknologi MARA Selangor, Kampus Puncak Alam, 42300 Selangor, Malaysia*

[*]*Corresponding author: nurhuda15@uitm.edu.my*

Received: 18 March 2024; Accepted: 29 August 2024; Published: 29 December 2024

Abstract

Ultraviolet radiation (UVR) poses significant deleterious effects on human well-being and increases the risks of getting skin cancers. Sunscreen is used topically as a defence against UVR. The photoprotection value of a sunscreen is represented as a sun protection factor (SPF). Co-formulation of sunscreen with natural phytoconstituents is a key to solving the problems related to the cumulative dissemination of synthetic UV filters into the environment with the worst being ecosystem contamination and coral bleaching. Quercetin, a compound derived from natural flavonoids, has a prominent occurrence in plants as well as sun protection activities due to its UV-absorbing properties. Six edible plants and fruits high in quercetin content were tested for their photoprotective activity. Mint (*Mentha piperita*) leaves, tomato (*Solanum lycopersicum*) skin, apple (*Malus domestica*) skin, asparagus (*Asparagus officinalis*), banana (*Musa acuminata*) peels, and basil (*Ocimum basilicum*) leaves were extracted using 99.8% methanol. The quercetin content of the extracts was determined and quantified using analytical high performance liquid chromatography (HPLC) with UV detection. The photoprotective activity for each plant sample was calculated based on their ultraviolet absorption capacity. The relationship between quercetin content and photoprotective effect was computed using correlation and regression analysis. *Ocimum basilicum* leaf extract possesses the highest quercetin content of 28.4 ppm and an SPF value of 35. The correlation coefficient value obtained between quercetin content and its photoprotection activities is 0.6273 indicating a moderate positive relationship.

Keywords: sun protection factor, edible plant, quercetin, high performance liquid chromatography, photoprotective activity

Abstrak

Sinaran ultraungu (SUU) menimbulkan kesan buruk yang ketara terhadap kesejahteraan manusia dan berisiko menyebabkan kanser kulit. Pelindung matahari digunakan secara luaran sebagai pertahanan terhadap SUU. Nilai perlindungan pelindung matahari diwakili sebagai faktor perlindungan matahari (FPM). Formulasi campuran pelindung matahari dengan bahan fitokimia adalah kunci untuk menyelesaikan masalah yang ditimbulkan oleh pelindung matahari sedia ada. Pelindung UV sintetik boleh menyebabkan pencemaran ekosistem dan pelunturan karang. Quercetin, sebatian yang berasal daripada flavonoid semulajadi, adalah kandungan fitokimia yang penting dalam tumbuhan sebagai pelindung matahari kerana sifatnya yang menyerap UV. Enam

tumbuhan dan buah-buahan tempatan yang boleh dimakan dan tinggi kandungan quercetin telah diuji untuk aktiviti fotoprotektif mereka. Daun pudina (*Mentha piperita*), kulit tomato (*Solanum lycopersicum*), kulit epal (*Malus domestica*), asparagus (*Asparagus officinalis*), kulit pisang *(Musa acuminata*), dan daun selasih (*Ocimum basilicum*) telah diekstrak menggunakan 99.8% metanol. Kandungan quercetin di dalam ekstrak ditentukan dan dihitung menggunakan kromatografi cecair prestasi tinggi analitikal (KCPT) dengan cerapan ultra ungu. Aktiviti fotoprotektif untuk setiap sampel tumbuhan dihitung berdasarkan kapasiti penyerapan ultraviolet. Hubungan antara kandungan quercetin dan kesan fotoprotektif dihitung menggunakan analisis korelasi dan regresi. Ekstrak daun *Ocimum basilicum* mempunyai kandungan quercetin tertinggi iaitu 28.4 ppm dan nilai SPF 35. Nilai pekali korelasi yang diperolehi antara kandungan quercetin dan aktiviti perlindungan fotonya ialah 0.6273 menunjukkan hubungan positif yang sederhana.

Kata kunci: faktor perlindungan matahari, tumbuhan makanan, quercetin, kromatografi cecair prestasi tinggi, aktiviti fotoprotektif

Introduction

Sun ultraviolet radiation (UVR) is a well-known factor in the emerging prevalence of the most common type of skin cancer, basal cell carcinoma (BCC) which accounts for around 75% of all skin cancer diagnoses [1]. It is irrefutable that a trivial amount of UVR is needed for skin in terms of vitamin D production, however in the incidence of excessive quantity, it will cause multiple adverse effects such as sunburn, phototoxic, DNA damage, and immunosuppression which are all risks factors for cancer [2]. These adverse effects are associated as a result of an increased formation of reactive oxygen species (ROS) due to the interaction of skin chromophores with molecular oxygen and subsequent oxidative stress which then leads to lipid peroxidation, enzyme inactivation, DNA damage, and pathologic changes in the skin [3-5]. Active ingredients present in sunscreen were formulated to absorb solar radiation within the 290-400 nm range to prevent the occurrences of the associated adverse effects.

Plants synthesize secondary metabolites, mainly flavonoids, to act as their sunscreen and increase tolerance in response to high penetrating sunlight [6]. Flavonoids absorb UVR to prevent DNA damage and reactive oxygen species (ROS) synthesis [7]. Quercetin (figure 1), a compound derived from natural flavonoids, has a perpetual occurrence in plants such as *Nasturtium officinale* (Brassicaceae), *Apium graveolens* (Apiaceae), *Asparagus officinalis* (Aspargaceae), *Malus domestica* (Rosaceae), *Capparis spinosa* (Capparaceae), *Vitis vinifera* (Vitaceae) and *Ginkgo biloba* (Ginkgoaceae) [8]. The potential of quercetin as a natural sunscreen due to its photoprotective enhancing potential is currently being studied [9-17]. The integration of quercetin in plant extracts is believed to enhance the photoprotective synergy of UV filters in sunscreen formulations. As much as plants benefit from quercetin biosynthesis in photoprotection activities, humans may also utilize this naturally derived compound to protect themselves from the molecular damage induced by UV irradiation.

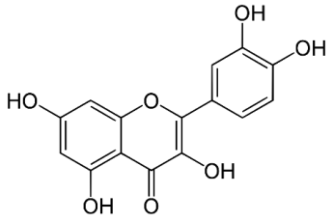

Figure 1. Structure of quercetin

The foundation of the studies originated from the observably significant elevation of the biosynthesis of quercetin compound in the plants being continuously projected to UVB radiation [18,19]. Recent studies circulate the supplementation of quercetin in the conventionally used UV filters in sunscreen formulations to observe the photoprotection activities outcomes [20]. Enhancement of photoprotection activities of the UV filters was reported to be achieved when supplemented with natural extract from plants as compared to using the UV filters alone [16,20-23]. The excess excitation energy quenching effects of quercetin are believed to play a vital role in stimulating the synergism of the photoprotection effect [21]. This experiment is to examine the correlation between the increased amount of quercetin biosynthesis and the following photoprotection activities. Six (6) local plants reported rich in quercetin were selected plant for this experiment. The hypothesis for this experiment is that the photoprotection activities are correlated to the

quercetin content in plants.

Materials and Methods

Chemicals and materials

Standard Quercetin (solid 2-(3,4-Dihydroxyphenyl)-3,5,7-trihydroxy-4H-1-benzopyran-4-one,3,3',4',5,6-Pentahydroxyflavone) was purchased from Sigma-Aldrich, Germany. Plant samples mint leaves (*Mentha piperita*), tomato (*Solanum lycopersicum*) skin, apples (*Malus domestica*) skin, asparagus (*Asparagus officinalis*), banana (*Musa acuminata*) peels, and basil (*Ocimum basilicum*) leaves were purchased from a local supermarket. Solvents for extraction are of analytical grade. Chromatographic solvents are of HPLC grades and ultra-pure water with a resistivity of 18 MΩ at 25ºC.

Preparation of plant samples

The plant samples were washed and cut into smaller pieces. The plant materials were frozen at -40°C before being lyophilized using a Labconco freeze dryer. The dried plant materials were then ground using mortar and pestle into fine powders. The dried plant powders were kept in airtight bottles and stored in the refrigerator at 4°C before use.

Preparation of reference standard solution

50 mg of quercetin reference standard were weighed and dissolved in 25 mL of 99.8% methanol. The stock solution with a concentration of 2000 µg/mL was sonicated for 30 minutes. The stock solution was further diluted to obtain a 100 µg/mL standard reference solution. The reference solution was diluted serially by a two-fold dilution factor to obtain a set of reference standards at concentrations of 100. 50, 25, 12.5, and 6.25 µg/mL.

Preparation of Sample Solution

The dried plant samples (10.0 ± 0.5 g) were weighed and sonicated with 99.8% methanol for 1 hour to allow extraction of the quercetin compound. The solution was filtered using Whatman filter paper to produce a clear solution. Then, the solvent was evaporated using the rotary evaporator at 60 rpm to further concentrate the extract. The remaining sample extracts were weighed together with the round-bottomed flask and then the weight of the empty flask was subtracted to obtain the individual weight of the plant extracts. The extracts were then dissolved in 10 mL of 99.8% methanol.

Determination of Quercetin

HPLC Parameters: A mobile phase consisting a gradient of ACN:H2O (30:70 to 60:40 for 14 minutes) delivered by a double reciprocating pump; flow/rate of 1 ml/min; Separation made using pH resistant Inertsil, C18, 150 × 4.6 mm, 5 µm column; UV detection at a wavelength of 370 nm, output process, and record by means of compatible integrator; temperature maintained at 35°C; The injection volume is 20 µL; Chromatographic run of 14 min; Injection procedure is blank (diluent), reference solution (three times), test solution, and blank solution to rinse.

Standard calibration curve: The quercetin reference standards were injected directly into the HPLC system in triplicate runs. A linear regression was carried out on known quercetin concentration values against the corresponding mean peak area. The regression coefficient (r) slope and intercept of the resulting calibration curve were determined. The concentration range for the standard solution was 5-100 µg/mL. The concentrations of quercetin in the extracts were then determined using the calibration curve method.

Sun protection factor

The dried plant samples (2-5 g) were weighed and sonicated in 40 ml of 75% methanol for 1 hour. The solutions were filtered with Whatman No. 1 filter paper to obtain a clear solution. The filtrates were diluted to obtain a 1 mg/ml solution. Each solution was measured for its absorbance within the range of UVB wavelength (290-320 nm) with 5-nm increments using a BMG Labtech SPECTROstar® Nano spectrophotometer and 99.8% methanol as blank. The SPF values were calculated using the following equation [24]:

$$SPF = CF \sum_{290}^{320} EE(\lambda).I(\lambda).Abs(\lambda) \quad (1)$$

Where, CF = Correction factor which is 10 (constant), EE = erythemogenic effect of radiation at each wavelength, I = intensity of the sun, and Abs = absorbance of the sample. The constant EE and I are shown as listed in Table 1.

Table 1. EE and I constants for the calculation of sun protective factor (SPF)

λ (nm)	EE(λ) × I(λ)
290	0.0150
295	0.0817
300	0.2874
305	0.3278
310	0.1864
315	0.0839
320	0.0180
Σ	1.0000

The correlation between the SPF values and the concentration of quercetin was computed using Pearson's correlation coefficient and regression analysis by Microsoft Excel.

Results and Discussion

Calibration curve of quercetin standards

Standard reference quercetin solutions with concentrations of 100. 50, 25, 12.5, and 6.25 µg/mL were injected into the HPLC column. The analyses were carried out in triplicates for each reference standard. The retention time is 6.531 min (Figure 2). The mean of the peak area of the standards was extracted and plotted against the concentration to obtain a calibration curve. The response data were plotted to form a calibration curve (Figure 3). The obtained linear equation, y = 6.0191x - 2.0417, $R^2 = 0.9999$ was used to determine the concentration of quercetin in the plant extract samples by comparing the peak area under the curve obtained from HPLC.

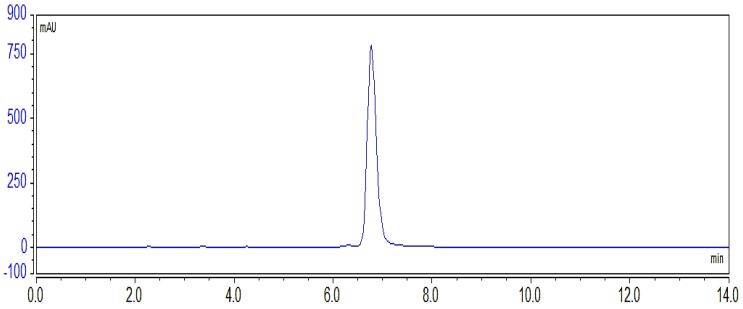

Figure 2. Chromatogram of quercetin standard

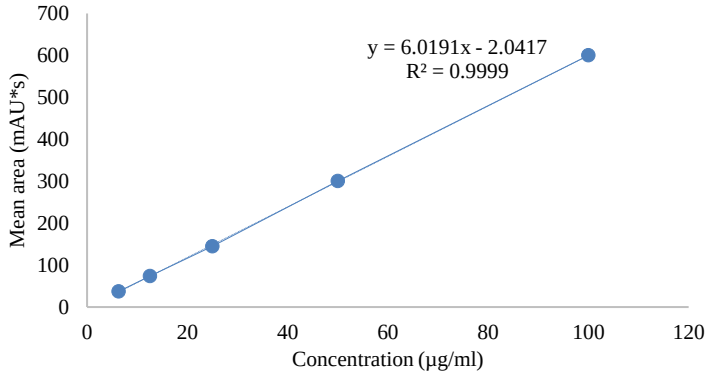

Figure 3. Calibration curve of quercetin standard

Quantification of quercetin in sample extracts

The plant crude extracts were chromatographed using the same chromatographic conditions as the standard quercetin. The identification of quercetin in the extracts was determined by comparing the retention time of quercetin in the standard (Figure 4). The concentration of quercetin for each sample was determined by plotting its peak area against the calibration curve.

The concentration of quercetin was standardized to the mass of the dried plant materials and expressed in ppm. The highest quercetin content was observed in basil (*Ocimum basilicum*) extract at 28.40 ppm, followed by tomato skin (*Solanum lycopersicum*) extract, mint leaves (*Mentha piperita*), asparagus (*Asparagus officinalis*), banana peels (*Musa acuminata*), and the lowest apple skin (*Malus domestica*) extract with quercetin concentration of 4.30 ppm. The chromatographic data and quercetin concentration for each sample are summarized in Table 2.

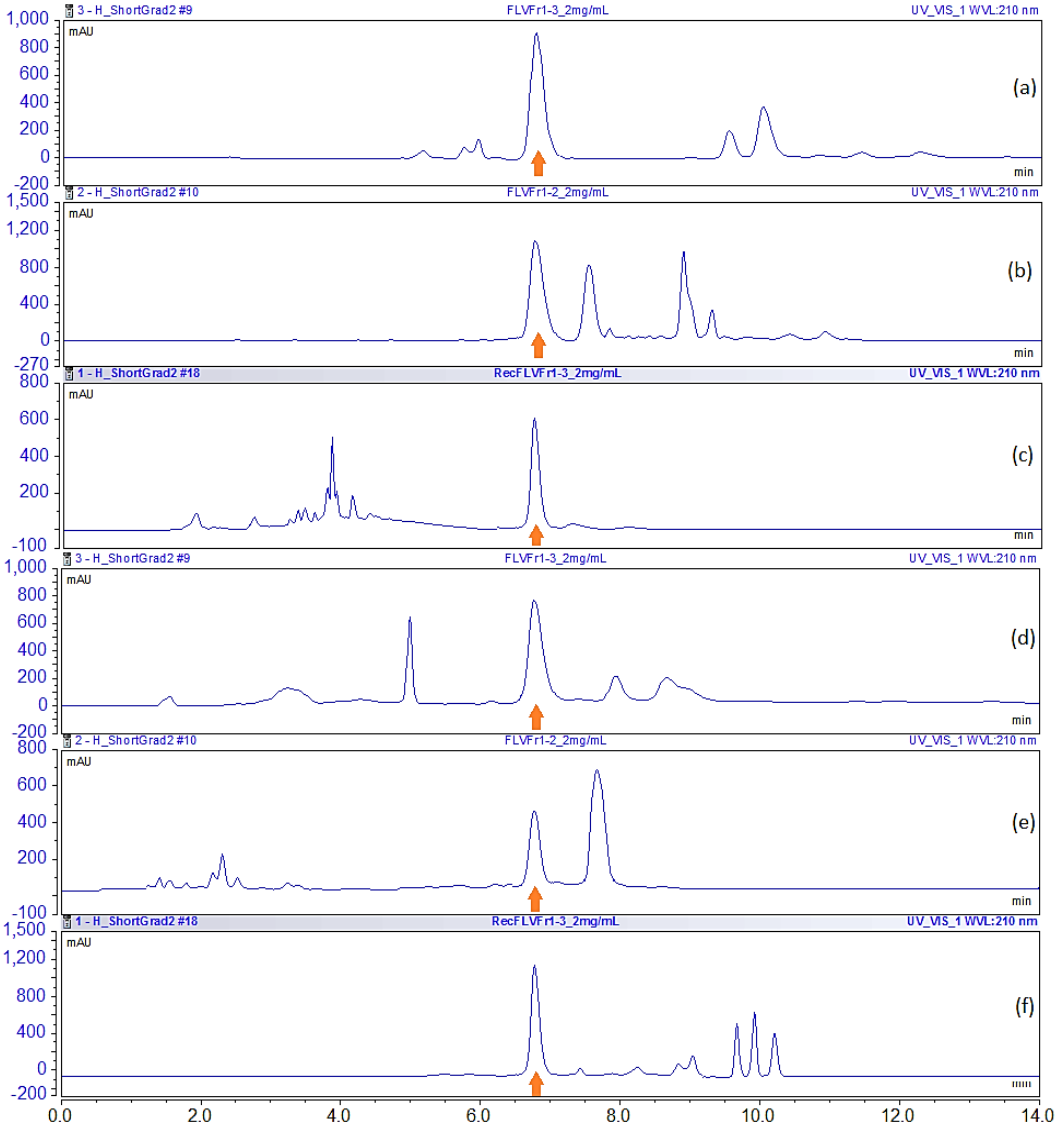

Figure 4. Chromatograms of *Mentha piperita* (a), *Solanum lycopersicum* (b), *Malus domestica* (c), *Asparagus officinalis* (d), *Musa acuminata* (e), and *Ocimum basilicum* (a); all samples are prepared at 10mg/mL. The peaks corresponding to quercetin are marked with arrows.

The HPLC analyses of all extracts show every sample comprises of similar peak at the retention time (Rt) of 6.511 – 6.532 min. The UV spectra of the peaks were like flavonol quercetin. Thus, the quercetin was identified from the retention time comparison using standard. The quantification of quercetin in sample extracts was done by comparing the peak area with the standard. The linearity was confirmed by preparing standard solutions of quercetin solutions in methanol at five concentrations. A calibration curve was plotted and determined using the standard data, which were linear for the specified concentration ranges. The detection limit was 0.145 µg/mL and the quantification limit was 0.480 µg/mL for quercetin. The quantification results show that the amounts of quercetin present in the extracts were high above the detection and quantification limits, further emphasizing the reliability of the method. The precision and accuracy of the measurements were within allowable values; the accuracy did not allow values that exceeded 2.09%.

Sun protection factor (SPF) values

To provide effective protection against UV-induced skin damage, the plant samples should have a wide range of absorbance in the UVB region, which is between 290 nm to 320 nm [25]. Therefore, in vitro sun protection factor (SPF) determination was conducted for each sample at different concentrations. The SPF values ranged from 35.00, the highest value, to 6.82, the lowest. Mint leaves (*Mentha piperita*) and basil (*Ocimum basilicum*) share the highest SPF value, followed by apple skin (*Malus domestica*), tomato skin (*Solanum lycopersicum*), asparagus (*Asparagus officinalis*) and the lowest is banana peels (*Musa acuminata*).

Table 2. Quercetin concentration was measured from chromatogram peak areas and SPF values were calculated form the samples' UV absorption

Sample	Retention Time (min)	Area (mAu*s)	Quercetin Concentration (µg/mL)	SPF (mean ± SD)
Mentha piperita	6.524	133.5084	22.52	35.00 ± 0.43
Solanum lycopersicum	6.531	167.7571	28.21	16.08 ± 0.22
Malus domestica	6.527	25.0443	4.50	17.45 ± 0.15
Asparagus officinalis	6.532	74.8222	12.77	6.92 ± 0.20
Musa acuminata	6.511	56.9455	9.80	6.83 ± 0.13
Ocimum basilicum	6.519	169.8036	28.55	35.00 ± 0.55

The extracts of all plant samples had the sunscreen activity evaluated with the method developed by Mansur et al. [24]. According to the USFDA, only SPF greater than or equal to 6 is suitable for use in cosmetic products with photoprotective activity. All tested extracts had satisfactory sunscreen activity (6.92 to 35.00), higher than the minimum required by the regulatory agency. The SPF values of the extracts tested were not proportional to the concentration of quercetin in the sample. This observation can be attributed to the other flavonoid compounds present in the samples also contributed to the photoprotective activity. A previous report showed that plant extracts rich in flavonoids are efficient in absorbing ultraviolet light, and usually show two maximum peaks of ultraviolet absorption, one between 240-280 nm and another 300-550 nm [26].

Statistical analysis

The correlation coefficient between the SPF values and quercetin content was computed and tabulated alongside the summary of the data obtained from the experiment. The computed correlation coefficient was 0.6273 which indicates a moderate positive relationship. A clustered bar chart was generated to better visualize the data obtained and represented by Figure 5.

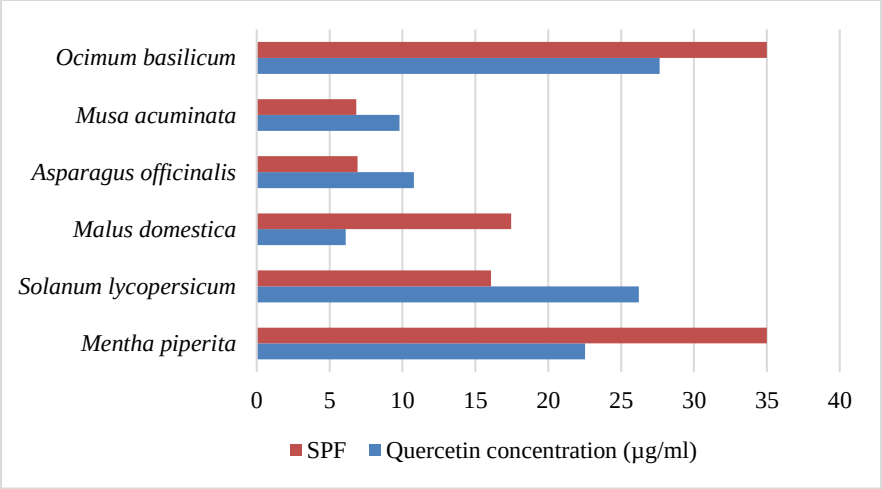

Figure 5. Clustered bar chart representing a comparison of SPF values and quercetin content in all plant samples

The SPF evaluation shows satisfactory results for all extracts, which are observed at SPF ≥ 6 at the lowest value. For mint and basil leaves, the values were close to the SFP standard of benzophenone 5%, in the fixed protective factor of 30-50, which indicates that the formulations containing the plants may be suitable for very sensitive skin sunburn, because of their high sun protection, as demonstrated in the in vitro methodology [27].

Quercetin is naturally present in an abundance of plants. It has an SPF value of 12.34, and when used in association with titanium dioxide, the SPF is around 30 [22]. The only difference is the expression of the phytochemical in the plants itself. Some may need more quercetin to protect themselves from the intense molecular damage resulting from radiation and ROS production [8]. Some may require less quercetin and produce different important phytochemicals with respective functions instead. Besides different plants synthesizing a different quantity of quercetin, different organs of the same plant also synthesize different amounts of quercetin as orchestrated by the plant-environment communication [28]. In this experiment, three extracts were obtained from the leaves while the other three were sampled from the outer skin of the fruit. Green leafy plants are known to be significantly rich in quercetin [29,30]. Basil extract displays the highest content of quercetin followed by tomato skin extract, mint leaves extract, asparagus extract, banana peel extract, and apple skin extract. Taking environmental factors into consideration, the enhanced nutrient uptake by the plants may leverage the production of quercetin in *Ocimum basilicum* [31]. In apple skins, most of the quercetin is present in moiety forms specifically rutinoside, galactoside, glucoside, and rhamnoside [29]. The standard used comparatively in this experiment was pure quercetin which presented no moieties and hence the low concentration. Quercetin may also be synthesized in de novo biosynthesis by engineered yeast *Saccharomyces cerevisiae* which may potentially be utilized as a promising candidate for sustainable bulk production in the future to replace the need to conduct a massive extraction procedure from native plants [32].

There is a vast method to detect the presence of quercetin in plants as it is a frequently studied compound due to its myriad benefits to humans [33-36]. The method used in this experiment employs reverse-phased high-performance liquid chromatography (RP-HPLC) to accurately determine the concentration of quercetin in the studied plants. This method lined closely with the previously reported method [37] which allows detection at 370 nm with isocratic elution of acetonitrile and 2% v/v acetic acid which is replaced by 2% v/v formic acid in this experiment, (40:60 v/v). The method was linear in the range of 5-100 µg/mL with r^2 value of 0.9999. Primarily, various combinations of acetonitrile with water in both isocratic and gradient elution were tried but the peak resolution was not achieved in both the

standard and samples. Later, it was decided to change the solvent water with acid as it has a vital role in producing an apparent peak and preventing tailing [38]. The resulting peaks resolution were achieved, and visually apparent peaks were observed in the standard and the samples.

However, the separation of the peaks in the samples was not achieved with some peaks being overlapped with each other such that can be observed in the *Solanum lycopersicum* chromatographic profile. The retention time of quercetin in the sample obtained was not the same value as the retention time of the standard, which renders it inaccurate. There are only two samples, *Malus domestica* and *Asparagus officinalis* that achieved the peak at retention time within the acceptable range of standard quercetin. To overcome this obstacle and improve the analysis, the ratio of acetonitrile should be decreased to allow slower movement of the compound from the column. The aglycone form of quercetin which lacks carbohydrate moiety is well dissolved in acetonitrile [39]. Adjusting the solvent composition in the mobile phase changes the polarity of the chromatographic condition, resulting in an alteration of the distribution coefficient. The relatively polar quercetin being retained longer in the column thus improves the resolution. This way, a peak attributable to the quercetin compound in the samples can be positively identified. It is also notable to mention that most quercetin present in nature is attached to the sugar moiety. The glycosidic form renders them to become more lipophobic.

The photoprotective activities of the plant extracts were studied by assessing their SPF values. SPF is defined by the ratio of the minimum UV radiation needed to induce sunburn on the skin layered with the sunscreen relative to the amount to cause the same degree of sunburn in the skin not protected by sunscreen [11]. Therefore, a higher SPF value signifies higher protection against sunlight. According to the FDA, an SPF value below 15 is not suitable for the preparation of pharmaceutical and cosmeceutical applications [40]. All extracts have SPF values greater than 15 with the exceptions being *Asparagus officinalis* and *Musa acuminata* extracts which obtained 6.9160 and 6.8294 respectively. This indicates the suitability of the extracts to be integrated or developed as one of the active photo protectants in a sunscreen formulation. The active ingredient in sunscreen is an organic compound that has UVR absorbing properties and can dissipate the energy absorbed into heat or light. To date, the chemical ingredients commonly used are sulisobenzone, oxybenzone, homosalate, octinoxate, and avobenzone. Studies suggested that sunscreen formulated along with topical antioxidants will reduce the production of reactive oxygen species and cytokines besides helping to downregulate the metalloproteinases expression [41]. The studied compound, quercetin fulfils the criteria to become the active ingredient as it presents the chromophores which are responsible for UV assimilation and antioxidant properties [22,33,42].

Due to undeniably increased risks for hypersensitivity reactions in humans as well as deteriorating effects on the environment by synthetic sunscreens, many studies have been conducted to make shifting from synthetic sunscreen to natural sunscreen more possible. Among the advantages of using a natural substitute for sunscreen is that natural sunscreens have higher compatibility with any type of skin and are less likely to induce irritating hypersensitivity reactions when compared to synthetic ones [2,43]. This is due to the formation and accumulation of damaging and toxic photoproducts on the skin after a continuous photochemical reaction of the synthetic chemical and sunlight [44]. It is also common in sunscreen formulation to use more than one synthetic UV filter intending to establish a larger and broader spectrum of UV light to be absorbed. The downside of this strategy is that it causes filter-filter interaction which either reduces the efficacy of the sunscreens or induces deleterious adverse effects to the skin it is being applied to [44,45]. These occurrences contradict the original idea of formulating a sunscreen to protect the skin from suffering the harmful effects of UV radiation. Therefore, this eventually put weight on the search for suitable natural UV filters to elucidate the active photo-protectant role in sunscreen.

However, despite being a strong candidate for active photo protectants in sunscreen, natural components such

as quercetin are not cut out to carry the UV filtering action alone. This is because in formulating an ideal sunscreen, the delivery of the active ingredients to the skin is one of the important factors in ensuring its intended action is achieved. Quercetin exhibits low hydrophilicity and poor percutaneous absorption which disrupt its potential development in sun protection products [46]. Even so, a novel nanoparticles-based topical drug delivery system approach introduces a way to encapsulate quercetin in a nanostructured particle to facilitate delivery and increase permeation to the skin [47]. The bigger problem is that quercetin or natural crude extracts which premiered as a potential candidate for sunscreen development generally have poor thermal and photo stability. This makes it harder to reach the ideal photoprotection threshold which is why most of the studies suggested quercetin or natural extracts to be used only as the supplementary ingredient to the existing chemical UV filters in commercial sunscreens [22,48,49]. It is proven that the addition of natural extracts to synthetic UV filters such as homosalate and oxybenzone will enhance the photoprotection action instead of using synthetic UV filters alone.

Conclusion

The results showed that *Mentha piperita* (mint leaves) and *Ocimum basilicum* (basil leaves) contain high concentrations of quercetin and a high sun protection factor. The result addresses the relationship between quercetin and photoprotective activity against UV radiation. Both plants are promising sources for future studies in the development of new skincare products associated with sunscreen competence. Studies from the analyzed extracts can be extended to the cosmetology and pharmacology fields since the extracts showed an SPF of 35, both of which presented a high sun protector factor with the potential to be used in sun protector formulations.

Most classical sunscreens are a combination of UVA and UVB chemical filters and other physical components to provide broad-spectrum protection. However, they are not based on renewable resources and may cause side effects i.e. under certain conditions can generate dangerous free radicals. Of all the plant samples studied, the leaves sample showed the best potential to be developed as natural sunscreen additives. The leaf part not only has great UV protection effects, but also it is easy to collect and has a high yield. Using the leaves as a source will not hurt or kill the plant. Therefore, the leaf part is the most promising renewable source. Thus, the plant leaves characterized by high SPF value must be taken into consideration. The finding from this study can supplement an explanation of the relationship between the photoprotection activities and the quercetin content in plant extracts. The resulting data may be beneficial for estimating the potential strategy or outcomes of using plant derivatives in the development of brand-new sunscreens. The outcomes from this study may project the potential resources for the development of innovative nature-based photoprotective cosmetics and medication.

Acknowledgment

The authors acknowledge Atta-ur-Rahman Institute for Natural Products Discovery (AuRIns) and the Faculty of Pharmacy, Universiti Teknologi MARA, Malaysia for technical support and research supplies. Our highest gratitude is dedicated to the laboratory technicians for their help and assistance throughout this study.

References

1. Newlands, C., Currie, R., Memon, A., Whitaker, S. and Woolford, T. (2016). Non-melanoma skin cancer: United Kingdom National Multidisciplinary Guidelines. *The Journal of Laryngology & Otology,* 130(2): 125-132.
2. Ghazi S. (2022). Do the polyphenolic compounds from natural products can protect the skin from ultraviolet rays? *Results in Chemistry*, 4: 100428.
3. Pavelkova, R., Matouskova, P., Hoova, J., Porizka, J. and Marova, I. (2020). Preparation and characterisation of organic UV filters based on combined PHB/liposomes with natural phenolic compounds. *Journal of Biotechnology*, 324: 100021.
4. Yu, Z. C., Zheng, X. T., Lin, W., He, W., Shao, L. and Peng, C. L. (2021). Photoprotection of Arabidopsis leaves under short-term high light treatment: The antioxidant capacity is more important than the anthocyanin shielding effect. *Plant Physiology and Biochemistry*, 166: 258-269.

5. Cestari, T. F., de Oliveira, F. B. and Boza, J. C. (2012). Considerations on Photoprotection and Skin Disorders. *Annales de dermatologie et de vénéréologie,* 139: 135-143.
6. Agati, G., Brunetti, C., Di Ferdinando, M., Ferrini, F., Pollastri, S. and Tattini, M. (2013). Functional roles of flavonoids in photoprotection: New evidence, lessons from the past. *Plant Physiology and Biochemistry*, 72: 35-45.
7. Hernández-Rodríguez, P., Baquero, L. P. and Larrota, H. R. (2019). Flavonoids: Potential therapeutic agents by their antioxidant capacity. in *Bioactive Compounds: Health Benefits and Potential Applications*, 1: 265- 288.
8. Singh, P., Arif, Y., Bajguz, A. and Hayat, S. (2021). The role of quercetin in plants. *Plant Physiology and Biochemistry*, 166: 10-19.
9. Yakoubi, R., Megateli, S., Sadok, H. T. and Gali, L. (2021). Photoprotective, antioxidant, anticholinesterase activities and phenolic contents of different Algerian *Mentha pulegium* extracts. *Biocatalysis and Agricultural Biotechnology*, 34: 102038.
10. Albuquerque Nerys, L. L., Jacob, Í. T., Silva, P. A., da Silva, A. R., de Oliveira, A. M., Rocha, W. R., Pereira, D. T., da Silva Abreu, A., da Silva, R. M., Da Cruz Filho, I. J. and de Lima, M. D. (2022). Photoprotective, biological activities and chemical composition of the non-toxic hydroalcoholic extract of *Clarisia racemosa* with cosmetic and pharmaceutical applications. *Industrial Crops and Products*, 180: 114762.
11. Saucedo, G., Vallejo, R. S., and Giménez, J. C. (2020). Effects of solar radiation and an update on photoprotection. *Anales De Pediatria*, 92 (6): 3771-3779.
12. Catelan, T. B. S., Gaiola, L., Duarte, B. F. and Cardoso, C. A. L. (2019). Evaluation of the in vitro photoprotective potential of ethanolic extracts of four species of the genus *Campomanesia Journal of Photochemistry and Photobiology B: Biology*, 197: 111500.
13. Lassoued, M. A, Ben Fatma, N. E. H, Haj Romdhane, M., Faidi, A., Majdoub, H. and Sfar, S. (2021). Photoprotective potential of a Tunisian halophyte plant *Carpobrotus edulis* L. *European Journal of Integrative Medicine*, 42: 101286.
14. da Silva, A. C. P., Paiva, J. P., Diniz, R. R., dos Anjos, V. M., Silva, A. B. S. M., Pinto, A. V.(2019). Photoprotection assessment of olive (*Olea europaea* L.) leaves extract standardized to oleuropein: In vitro and in silico approach for improved sunscreens. *Journal of Photochemistry and Photobiology B: Biology*, 193: 162-171.
15. Amparo, R. T., Silva, C. P. A., Seibert, B. J., da Silva, D. D., dos Santos, M. R. V., Vieira, M. A. P., Brandão, C. G., de Souza, B. B. G. and Santos, A. M. C. B. (2022). In vitro and in silico investigation of the photoprotective and antioxidant potential of *Protium spruceanum* leaves and its main flavonoids. *Journal of Photochemistry and Photobiology A: Chemistry*, 431: 114037.
16. Wróblewska, K. B., Baby, A. R., Grombone Guaratini, M. T. and Moreno, P. R. H. (2019). In vitro antioxidant and photoprotective activity of five native Brazilian bamboo species. *Industrial Crops and Products*, 130: 208-215.
17. Fardiyah, Q., Ersam, T., Suyanta, Slamet, A., Suprapto and Kurniawan, F. (2020). New potential and characterization of *Andrographis paniculata* L. ness plant extracts as photoprotective agent. *Arabian Journal of Chemistry*, 13(12): 8888-8897.
18. Shourie, A., Tomar, P., Srivastava, D. and Chauhan, R. (2014). Enhanced biosynthesis of quercetin occurs as a photoprotective measure in Lycopersicon esculentum Mill. under acute UV-B exposure. *Brazilian Archives of Biology and Technology*, 57 (3): 317-325.
19. Guidi, L., Brunetti, C., Fini, A., Agati, G., Ferrini, F., Gori, A. (2016). UV radiation promotes flavonoid biosynthesis, while negatively affecting the biosynthesis and the de-epoxidation of xanthophylls: Consequence for photoprotection? *Environmental and Experimental Botany*, 127: 14-25.
20. Mota, M. D., da Boa Morte, A. N., Silva, L. C. R. C. and Chinalia, F. A. (2020). Sunscreen protection factor enhancement through supplementation with rambutan (*Nephelium lappaceum* L) ethanolic extract. *Journal of Photochemistry and Photobiology B: Biology*, 205: 111837.
21. Almeida, W. A. da S., Antunes, A. dos S., Penido,

R. G., Correa, H. S. da G., Nascimento, A. M., Andrade, Â. L. (2019). Photoprotective activity and increase of SPF in sunscreen formulation using lyophilized red propolis extracts from Alagoas. *Revista Brasileira de Farmacognosia,* 29(3): 373-380.

22. Choquenet, B., Couteau, C., Paparis, E. and Coiffard, L. J. M. (2008). Quercetin and rutin as potential sunscreen agents: Determination of efficacy by an in vitro method. *Journal of Natural Products*, 71(6): 1117-1118.
23. Solovchenko, A. and Schmitz-Eiberger, M. (2003). Significance of skin flavonoids for UV-B-protection in apple fruits. *Journal of Experimental Botany*, 54 (389): 1977-1984.
24. Mansur, J. S., Breder, M. N. R., Mansur, M. C. A. and Azulay, R. D. (1986). Determination of sun protection factor by spectrophotometry. *Anais Brasileiros de Dermatologia*, 61: 121-124.
25. Nigel, E. E. A., Nasir, N. S. M., Aspa, A. A. and Manshoor, N. (2023) Determination of sun protection factor (SPF) of Malaysian fruit and vegetable extracts using UV-Visible Spectroscopy. *Malaysian Journal of Chemistry*, 25(2): 87-97.
26. Bobin, M. F., Raymond, M. and Martini, M. C. (1995). UVA/UVB absorption properties of natural products. *Cosmet Toiletries,* 7: 44-50.
27. Costa, C. C., Detoni, C. B., Branco, R. C., Botura, M. B. and Branco, A. (2015). In vitro photoprotective effects of Marcetia taxifolia ethanolic extract and its potential for sunscreen formulations. *Revista Brasileira de Farmacognosia* 25: 413-418.
28. Becker, C., Klaering, H. P., Schreiner, M., Kroh, L. W. and Krumbein, A. (2014). Unlike quercetin glycosides, cyanidin glycoside in red leaf lettuce responds more sensitively to increasing low radiation intensity before than after head formation has started. *Journal of Agriculture and Food Chemistry*, 62(29): 6911 – 6917.
29. Dabeek, W. M. and Marra, M. V. (2019). Dietary quercetin and kaempferol: Bioavailability and potential cardiovascular-related bioactivity in humans. *Nutrients*, 11: 1-19.
30. Jan, R., Khan, M., Asaf, S., Lubna, Asif, S. and Kim, K. M. (2022). Bioactivity and therapeutic potential of kaempferol and quercetin: New insights for plant and human health. *Plants*, 11: 1-18.
31. Scagel, C. F. and Lee, J. (2012). Phenolic composition of basil plants is differentially altered by plant nutrient status and inoculation with Mycorrhizal fungi. *Hortscience*, 47 (5): 660-671.
32. Tartik, M., Liu, J., Mohedano, M. T., Mao, J. and Chen, Y. (2023). Optimizing yeast for high-level production of kaempferol and quercetin. *Microbial Cell Factories*, 2 (74): 1-11.
33. Yang, D., Wang, T., Long, M. and Li, P. (2020). Quercetin: Its main pharmacological activity and potential application in clinical medicine. *Oxidative Medicine and Cellular Longevity*, 2020: 1-13.
34. Zhang, L., Tu, Z. C., Xie, X., Wang, H., Wang, H., Wang, Z. X., Sha, X. M. and Lu, Y. (2017). Jackfruit (*Artocarpus heterophyllus* Lam.) peel: A better source of antioxidants and a-glucosidase inhibitors than pulp, flake and seed, and phytochemical profile by HPLC-QTOF-MS/MS. *Food Chemistry*, 234: 303-313.
35. Aguirre-Hernández, E., González-Trujano, M. E., Martínez, A. L., Moreno, J., Kite, G., Terrazas, T. (2010). HPLC/MS analysis and anxiolytic-like effect of quercetin and kaempferol flavonoids from Tilia americana var. mexicana. *Journal of Ethnopharmacology*, 127(1): 91-97.
36. Mondal, S. and Bandyopadhyay, A. (2023). Photoprotective, antioxidant potential and DNA damage protection assay of leaf methanolic extract of *Holoptelea integrifolia* (Roxb) plant and determination of some bioactive phenolic compounds by RP-HPLC. *Biocatalysis and Agricultural Biotechnology*, 50: 102728.
37. Ang, L. F., Yam, M. F., Fung, Y. T. T., Kiang, P. K. and Darwin, Y. (2014). HPLC method for simultaneous quantitative detection of quercetin and curcuminoids in traditional Chinese medicines. *Journal of Pharmacopuncture*, 17(4): 36-49.
38. Pawanpreet, K. and Baljeet, S. (2019). Analytical method development and validation of quercetin: A review. *International Journal of Pharmaceutical and Clinical Research*, 11 (2): 49-56.
39. Chebil, L., Humeau, C., Anthony, J., Dehez, F.,

Engasser, J. M. and Ghoul, M. (2007). Solubility of flavonoids in organic solvents. *Journal of Chemical Engineering Data*, 52(5): 1552-1556.

40. Dlugos, J. F. A (2022). spectroscopic in vitro method for the calculation of sunscreen SPF values. Case study; Customer Product, PerkinElmer, Inc. Shelton, CT USA.
41. Alvares, B. A., Miola, A. C., Schimitt, J. V., Miot, H. A. and Abbade, L. P. F. (2022). Efficacy of sunscreen with photolyase or regular sunscreen associated with topical antioxidants in treating advanced photodamage and cutaneous field cancerization: a randomized clinical trial. *Anais Brasileiros de Dermatologia*, 97(2): 157-165.
42. Hatahet, T., Morille, M., Hommoss, A., Devoisselle, J. M., Müller, R. H. and Bégu, S. (2016). Quercetin topical application, from conventional dosage forms to nanodosage forms. *European Journal of Pharmaceutics and Biopharmaceutics*, 108: 41-53.
43. He, H., Li, A., Li, S., Tang, J., Li, L. and Xiong, L. (2021). Natural components in sunscreens: Topical formulations with sun protection factor (SPF). *Biomedicine and Pharmacotherapy*, 134: 111161.
44. Gadgil, V. R., Darak, A., Patil, S. J., Chopada, A., Kulkarni, R. A., Patil, S. M. (2023). Recent developments in chemistry of sunscreens & their photostabilization. *Journal of the Indian Chemical Society*, 100: 100858.
45. Dutta, S., Kumar, S. P. J. and Banerjee, R. (2023). A comprehensive review on astaxanthin sources, structure, biochemistry and applications in the cosmetic industry. *Algal Research*, 74: 103168.
46. Michala, A. S. and Pritsa, A. (2022). Quercetin: A molecule of great biochemical and clinical value and its beneficial effect on diabetes and cancer. *Diseases*, 10(3): 37-52.
47. Wadhwa, K., Kadian, V., Puri, V., Bhardwaj, B. Y., Sharma, A., Pahwa, R. (2022). New insights into quercetin nano formulations for topical delivery. *Phytomedicine Plus*, 2(2): 100257.
48. Bastin, A., Teimouri, M., Faramarz, S., Shabani, M., Doustimotlagh, A. H. and Sadeghi, A. (2023). In vitro and molecular docking analysis of quercetin as an anti-inflammatory and antioxidant. *Current Pharmaceutical Design*, 29(11): 883-891.
49. Karrat, A., Palacios-Santander, J. M., Amine, A. and Cubillana-Aguilera, L. (2022). A novel magnetic molecularly imprinted polymer for selective extraction and determination of quercetin in plant samples. *Analytica Chimica Acta*, 1203: 339709.