

Phytochemicals and antioxidant activity in seed and pulp of wild grape (*Ampelocissus martinii* Planch.) extracts

Ansaya Thonpho, Chuleerat Wongnarat, Wilaiwan Simcheur
and Prasong Srihanam*

Innovative Chemistry and Innovation Research Unit, Center of Excellence for Innovation in Chemistry, Department of Chemistry, and Faculty of Science, Mahasarakham University, Kantharawichai, Maha Sarakham 44150, Thailand

*Corresponding author. E-mail: prasong.s@msu.ac.th

ABSTRACT

In this work, the phytochemical content and antioxidant activity of methanolic extract of the wild grape seed and pulp were examined at three different stages, namely, immature, mature and ripe. The results showed that the seed extracts produce higher phytochemical content and antioxidant activity than the pulp extracts. For both seed and pulp extracts, polyphenols decrease at the ripening period. The immature wild grape seed extract (GWGS) showed the highest number of phytochemicals and potent antioxidant activity. The content of phytochemical results was found to have a significant correlation with the antioxidant activities ($p < 0.01$). In conclusion, GWGS from the extract of wild grape seed is a source of phenolic compounds that might be potentially used as good supplement for health benefits.

Keywords: *Antioxidant activity, Extract, Phenolic compounds, Wild grape*

INTRODUCTION

For many years, traditional medicinal plants have been shown to be an alternative source of antioxidants potentially for treating microbial diseases in tropical and developing countries (Devika, 2012; Kuete et al., 2011; Saeed et al., 2014) and they also have many applications in food industry (Joshi, Su and D'Souza, 2015; Pozharitskaya et al., 2015; Yang et al., 2016). A variety of phytochemicals from the plant extracts, including terpenoids, phenolic acids, lignans, tannins flavonoids, quinones, coumarins or alkaloids, has been reported for antioxidant activity (Bakkali et al., 2008; Huang, Ou and Prior, 2005; Tagne et al., 2014). Currently, antioxidants can be obtained synthetically and naturally but, owing to the toxic issue raising on the synthetic antioxidants (Barros, Carvalho and Ferreira, 2010), antioxidants from plant extracts would thus become more environmentally friendly and human health (Jiménez et al., 2015; Sanchez-Bel et al., 2015; Tauchen et al., 2015). On the other hands, oxidant or free radicals produced in living organisms, via aerobic metabolism, could be a dangerous source of various degenerative diseases (Jothy, 2011; Sarikurkcu et al., 2008), owing to their high reactivity against biological macromolecules (Xia et al., 2011).

Article history:

Received 17 January 2019; Received in revised form 2 February 2019;

Accepted 7 March 2019; Available online 24 June 2019

Wild grape (*Ampelocissus martinii* Planch.) is a tropical plant that can generally be found in Thailand, Vietnam, Cambodia, Laos, Peninsular Malaysia, Borneo and Philippines (Wen, Lu and Boggan, 2013). It also has long been used as folk medicine in Thailand. At present, little information is known about antioxidant components in the wild grape. Here, we aim to investigate the phytochemical profile and antioxidant activities at different stages of the wild grape extracts. We have identified several potential antioxidants from wild grape seed and pulp extracts, which would be used as good supplement source for future medical treatment and health benefits.

MATERIALS AND METHOD

Chemicals and plant material

All chemicals used in the present study were of analytical grade and purchased from Sigma-Aldrich (St. Louis, Mo., USA).

Wild grape (*Ampelocissus martinii* Planch.) were categorized into three groups namely immature (green), mature (red) and ripe (black). All grape fruits were separated into seed and pulp, and then dried, ground into a fine powder. The powder of each sample was extracted with methanol (1:10 w/v) and stirred at room temperature for 3 h. The solvent was removed at 40 °C using rotary vacuum evaporator. The obtained residue was dissolved in 20 mL of methanol (99.8%) and stored at -4 °C until analysis.

Determination of phytochemicals

The total phenolic content (TPC) was determined using Folin-Ciocalteu reagent as followed by Pastrana-Bonilla et al. (2003), using gallic acid as a standard. Total flavonoid content (TFC) was analyzed using a spectrophotometric method described by Abu Bakar et al. (2009) using catechin as a standard. Total proanthocyanidin content (TPAC) was determined following the procedure described by Li et al. (2006) using catechin as a standard. Total saponin content (TSC) was determined as described by Hiai, Oura and Nakajima method (1976) using aescin as standard. In addition, total monomeric anthocyanin content (TMAC) was performed using the method described by Giusti and Wrolstad (2001) and following their direction for buffer preparation. Total anthocyanins were calculated using the following equation:

$$\text{TMAC (mg/g)} = (A \times Mw \times DF \times 1000) / (\epsilon \times l)$$

Where A is the absorbance of $(A_{510} - A_{700})_{\text{pH } 1.0} - (A_{510} - A_{700})_{\text{pH } 4.5}$; Mw is the molecular weight of cyanidin-3-O-glucoside ($449.38 \text{ g mol}^{-1}$); DF is the dilution factor; l is the path length in cm; ϵ is the 26900 M extinction coefficient in L/mol/cm for cyanidin-3-O-glucoside; and 1000 is the conversion from g to mg.

Antioxidant activity assay

Free radical-scavenging activity of the extracts was determined by using of a stable 2,2-diphenyl-2-picrylhydrazyl radical (DPPH \cdot) according to the previous method (von Gadow, Joubert and Hansmann, 1997) with some modifications. Cation radical (ABTS $^{+\cdot}$) scavenging activity was also measured according to the method of Re et al. (1999). The DPPH and ABTS $^{+\cdot}$ radicals scavenging activity was represented

in the term 50% of inhibition (IC_{50}) value. The ferric reducing activity was measured by FRAP assay according to the method of Benzie and Strain (1996) with some modifications. The cupric reducing antioxidant was also carried out with the method of Apak et al. (2004) with some modifications using trolox as standard.

Statistical analysis

All experiments were performed in triplicate and expressed the results as mean \pm standard deviation (SD). Analysis of variance (ANOVA) was carried out to determine the significant differences of measurements by SPSS statistical software, considering the confidence level of 95%. Linear correlations between different assays were calculated using the correlation coefficient statistical option in Pearson test.

RESULTS AND DISCUSSION

Extraction yield and phytochemical content

The extraction yield and phytochemical content in wild grape pulp and seed extracts are presented in Table 1. As expected, different amounts of the TPC, TFC, TMAC, TPAC and TSC among selected sample. The yield of all extracts varied from 6.17 to 39.85% (w/w) with a significant difference ($p < 0.05$). BWGP exhibited the highest yield, followed by GWGP, RWGP, BWGS, GWGS and RWGS, respectively.

The TPC in wild grape was, on average, 53-fold more concentrated in the seed than in the pulp. The highest value of TPC was found in GWGS (417.35 mg GAE/g DW). Moreover, in all samples the TFC is higher for seed extracts than the pulp and, the highest value was found in GWGS (390.78 mg CE/g DW). The pulp extracts showed lower TFC than that of seed, ranging from 6.03 to 6.42 mg CE/g DW. It was previously reported that grape seeds are rich in phenolic compound than grape pulp. This was due to the fact that the seeds act mainly as a reservoir during the development of the sprouts (Pająk et al., 2014). In addition, the wild grape seed extracts contain the amount of TPAC higher than the pulp extracts. The TPAC is highest for GWGS (304.39 mg CE/g DW). In previous work, we studied phytochemical contents from fruit (all parts) of wild grape but the stages are the same. In this work, we need to know the content in each part, especially seed and pulp of wild grape fruit. The results indicated that almost phytochemical content, except TSC in young (green) stage of wild grape pulps have lower than all parts. In mature (red) stage, the TPC has higher than fruit but TFC and TMAC have lower. In ripe (black) stage, the TPC and TFC have slightly higher content than fruit but TMAC has lower. Among them, the TSC of pulp in all stages have higher contents than fruit. Interestingly, the phytochemical contents of all stages have dramatical high than the fruit extract (Wongnarat and Srihanam, 2016). As suggested by Hernández-Jiménez et al. (2009), proanthocyanidins are the major phenolic compounds found in grape seed, and are higher than those found in other part of grapes (*Vitis vinifera* L.). Saponins are important secondary metabolites derived from various plants and have been used extensively in drug-related industry due to their pharmaceutical properties. The saponin distribution in variable content depending on an individual part of plants (Cheok, Salman and Sulaiman, 2014). The extract of GWGS had the highest saponins content (687.33 mg Aes/g DW). The results indicated that, the saponin content both in seed and pulp extracts decrease during the development stage of fruit, as can be seen from the color change at immature (green) to ripe (black). These findings were in agreement with previous studies that found the largely decrease in the polyphenols of grape during ripening stage (Kennedy, Matthews and Waterhouse, 2000). From TMAC analysis, anthocyanins were found only in pulp extracts,

with the BWGP extract having the highest anthocyanins content (19.17 mg C3GE/g DW). This is because anthocyanins are pigments and mainly exist in grape skins and pulps (Xia et al., 2010).

Antioxidant activities of wild grape extract

There is no universal assay that accurately reflects all the antioxidants in a complex system. Therefore, it is necessary to use different methods to evaluate the antioxidant capacity to ensure obtained results (Jin et al., 2012). The antioxidant activities of wild grape seed and pulp extracts were determined using DPPH, ABTS, FRAP and CUPRAC assay and the results are shown in Table 2. DPPH and ABTS assay were expressed in IC₅₀ value and compared to trolox and ascorbic acid as a positive control. The DPPH[•] scavenging activity of wild grape seed extracts (IC₅₀ = 39.37 to 51 µg/mL) showed a higher scavenging activity for the pulp extracts (IC₅₀ = 2665.68 to 4196.85 µg/mL). The order of DPPH[•] scavenging activity of GWGS, BWGS and RWGS was the same activity of ascorbic acid and trolox ($p < 0.05$), respectively. A similar trend to the DPPH[•] scavenging results was found in the ABTS⁺ scavenging activity. The seed extracts showed IC₅₀ values in the range of 6.34 to 12.21 µg/mL which is more active on ABTS⁺⁺ than the pulp extracts (IC₅₀ ranging from 750.34 to 1639.68 µg/mL). The highest activity on ABTS⁺⁺ scavenging was found on GWGS, BWGS and RWGS. FRAP and CUPRAC assay have been used mainly to detect the reducing ability of the antioxidant, i.e., the ability to reduce iron and copper ions. In the ferric reducing antioxidant assay, the seed extracts showed FRAP value (1203.96 to 1702.05 mmol Fe²⁺/g DW), which was 68-fold (on average) higher than those of the pulp extracts (14.79 to 29.63 mmol Fe²⁺/g DW). The relative activity was in the highest in GWGS. The cupric reducing potency of the wild grape extracts was expressed as mg TE per g dry weight. The extract of GWGS possessed the strongest (111.56 mg TE /g DW) reducing activity. It is well known that there are correlation between the antioxidant activities and the polyphenol compositions. In particular, many studies have confirmed that the phytochemicals from plants is affected by various factors: cultivars, maturity, colors, part of fruits as well as genetic, climate, geographic origin, and cultivation act main roles on the plant phytochemicals (Boonsod, Sangdee and Srihanam, 2014; Bruno and Sparapano, 2007). Higher antioxidant activity was observed in GWGS as expected from high contents of phenolic compounds. This result agrees with both DPPH[•] and ABTS⁺⁺ scavenging activity, and ferric and cupric reducing potency activities, in which GWGS is the highest efficient antioxidant. Moreover, GWGS was equally effective against trolox and ascorbic acid by DPPH and ABTS assay. Thus, GWGS appears to be a good potential source of natural antioxidants which would be applied in reduction of oxidative damage in the human body and for providing health protection.

Correlation analysis

The correlation coefficients (r) between the phytochemical contents and antioxidant activities obtained from each assay were analyzed and the results are presented in Table 3. The strong positive correlation coefficients were observed between TPC, TFC, TPAC, TSC and antioxidant activities (DPPH[•], ABTS⁺⁺, FRAP and CUPRAC assay), the r -values is in range of 0.825 to 0.998 ($p < 0.01$). The results in this study suggest that the content and composition of some phenolic compounds in

the wild grape extracts could play an important role in antioxidant activity. These findings are consistent with previous studies that demonstrated on a positive correlation between phenolic compounds and antioxidant activity (Kim et al., 2006; Meng et al., 2012).

CONCLUSION

Our results indicate that the wild grape extracts from seed gave the higher phytochemical content than pulp extracts. The polyphenols in seed and pulp extracts decrease during ripening period. Both the content and composition of phenolics, flavonoids, proanthocyanidins and saponins of the wild grape extracts are strongly correlated with the antioxidant activity. The highest antioxidant activity was observed for GWGS, the extract that has the highest total phenolic compound content and is equally effective against trolox and ascorbic acid by DPPH and ABTS assay. Thus, the immature wild grape seed (GWGS) would be a potential source of natural antioxidants that can be used as food supplement for health benefits.

ACKNOWLEDGEMENT

The authors gratefully acknowledge the higher education research promotion (HERP), the Center of Excellence for Innovation in Chemistry (PERCH-CIC), Commission on Higher Education, Ministry of Education, Thailand and Mahasarakham University for financial support.

Extracts	Yield (%)	TPC (mg GAE/g DW)	TFC (mg CE/g DW)	TPAC (mg CE/g DW)	TMAC (mg C3GE/g DW)	TSC (mg Aes/g DW)
GWGP	36.73 ± 0.86 ^b	4.22 ± 0.17 ^d	6.42 ± 0.17 ^d	1.49 ± 0.05 ^d	0.04 ± 0.02 ^b	17.74 ± 0.62 ^e
RWGP	24.67 ± 1.10 ^c	6.13 ± 0.37 ^d	6.07 ± 0.21 ^d	1.18 ± 0.03 ^d	0.08 ± 0.02 ^b	69.74 ± 4.21 ^d
BWGP	39.85 ± 1.16 ^a	8.06 ± 0.80 ^d	6.03 ± 0.17 ^d	6.36 ± 0.26 ^d	19.17 ± 0.71 ^a	96.92 ± 2.24 ^d
GWGS	8.10 ± 0.10 ^e	417.35 ± 22.54 ^a	390.78 ± 7.10 ^a	304.39 ± 34.73 ^a	ND	687.33 ± 17.30 ^a
RWGS	6.17 ± 0.19 ^f	239.78 ± 7.67 ^c	232.41 ± 6.29 ^c	94.17 ± 0.85 ^c	ND	318.65 ± 33.38 ^c
BWGS	10.35 ± 0.05 ^d	316.66 ± 8.08 ^b	296.20 ± 16.98 ^b	158.55 ± 10.63 ^b	ND	378.97 ± 20.97 ^b

Results are expressed as mean ± SD of triplicate measurements. Means with different letters in the same column represent significant differences at $p < 0.05$. GWGP, green wild grape pulp; RWGP, red wild grape pulp; BWGP, black wild grape pulp; GWGS, green wild grape seed; RWGS, red wild grape seed; BWGS, black wild grape seed; WGP, white grape pulp; RGP, red grape pulp; WGS, white grape seed; RGS, red grape seed. ND, not detected. Means with different letters (a-e) in the same column represent significant differences at $p < 0.05$. For example, %yield of GWGP significant differences from RWGP at $p < 0.05$.

Table 1 Extraction yield and phytochemical content in the wild grape pulp and seed extracts.

Extracts	DPPH [*] IC ₅₀ ^A (µg/mL)	ABTS ⁺⁺ IC ₅₀ ^A (µg/mL)	FRAP assay (mmol Fe ²⁺ /g DW)	CUPRAC assay (mg TE/g DW)
GWGP	3479.21 ± 164.74 ^b	1433.23 ± 47.34 ^b	14.79 ± 0.47 ^d	2.90 ± 0.01 ^e
RWGP	4196.85 ± 223.40 ^a	1639.68 ± 16.94 ^a	18.05 ± 0.74 ^d	3.21 ± 0.03 ^d
BWGP	2665.68 ± 132.91 ^c	750.34 ± 36.55 ^c	29.63 ± 2.17 ^d	2.86 ± 0.01 ^e
GWGS	39.37 ± 0.39 ^d	6.34 ± 0.11 ^d	1702.05 ± 32.55 ^a	111.56 ± 0.22 ^a
RWGS	51.00 ± 1.89 ^d	12.21 ± 0.8 ^d	1203.96 ± 14.67 ^c	110.22 ± 0.00 ^c
BWGS	45.83 ± 1.03 ^d	9.56 ± 0.44 ^d	1370.81 ± 25.42 ^b	110.74 ± 0.17 ^b
Trolox ^B	12.12 ± 0.19 ^d	12.78±0.25 ^d	-	-
Ascorbic acid ^B	7.89 ± 0.10 ^d	8.911±0.42 ^d	-	-

Table 2 Antioxidant activities of the wild grape seed and pulp extracts.

	TPC	TFC	TPAC	TAC	TSC	DPPH [•]	ABTS ^{•+}	FRAP	CUPRAC
TPC	1	.998*	.956*	-.423	.965*	.985*	.995*	.993*	.953*
TFC	-	1	.947*	-.431	.962*	.988*	.995*	.995*	.957*
TPAC	-	-	1	-.356	.981*	.893*	.968*	.918*	.825*
TAC	-	-	-	1	-.320	-.442	-.415	-.435	-.451
TSC	-	-	-	-	1	.923*	.981*	.943*	.866*
DPPH [•]	-	-	-	-	-	1	.972*	.998*	.990*
ABTS ^{•+}	-	-	-	-	-	-	1	.984*	.932*
FRAP	-	-	-	-	-	-	-	1	.980*
CUPRAC	-	-	-	-	-	-	-	-	1

* Correlation is significant at the 0.01 level.

^A Concentration of the plant extract that scavenges 50% of free radical. Lower IC₅₀ values indicate higher radical scavenging activity.

^B Standard synthetic antioxidants were used as a reference for radical scavenging activity. Results are expressed as mean ± SD of triplicate measurements. Means with different letters (a-e) in the same column represent significant differences at $p < 0.05$.

Table 3 Correlation coefficients (r) between phytochemical contents and different antioxidant assays.

REFERENCES

- Abu Bakar, M.F., Mohamed, M., Rahmat, A., and Fry, J. (2009). Phytochemicals and antioxidant activity of different parts of bambangan (*Mangifera pajang*) and tarap (*Artocarpus odoratissimus*). *Food Chemistry*, 113, 479-483.
- Bakkali, F., Averbeck, S., Averbeck, D. and Idaomar, M. (2008). Biological effects of essential oils -A review. *Food and Chemical Toxicology*, 46, 446-475.
- Barros, L., Carvalho, A.M. and Ferreira, I.C.F.R. (2010). Leaves, flowers, immature fruits and leafy flowered stems of *Malva sylvestris*: A comparative study of the nutraceutical potential and composition. *Food and Chemical Toxicology*, 48, 1466-1472.
- Boonsod, Y., Sangdee, A. and Srihanam, P. (2014). Phytochemical and Biological Activities of the Wild Grape Fruit Extracts Using Different Solvents. *British Journal of Pharmaceutical Research*, 4, 23-36.
- Bruno, G. and Sparapano, L. (2007). Effects of three esca-associated fungi on *Vitis vinifera* L.: V. Changes in the chemical and biological profile of xylem sap from diseased cv. Sangiovese vines. *Physiological and Molecular Plant Pathology*, 71, 210-229.
- Cheok, C.Y., Salman, H.A.K. and Sulaiman, R. (2014). Extraction and quantification of saponins: A review. *Food Research International*, 59, 16-40.
- Koilpillai, Y.J. (2012). Phytochemical and in vitro micropropagation studies of *Clerodendrum phlomidis* L. *Journal of Pharmacy Research*, 5, 4396-4398.
- Giusti, M.M. and Wrolstad, R.E. (2001). Characterization and measurement of anthocyanins by UV-Visible spectroscopy. *Current Protocols in Food Analytical Chemistry*. John Wiley & Sons, Inc.
- Hernández-Jiménez, A., Gómez-Plaza, E., Martínez-Cutillas, A. and Kennedy, J.A. (2009). Grape skin and seed proanthocyanidins from Monastrell × Syrah grapes. *Journal of Agricultural and Food Chemistry*, 57, 10798-10803.
- Hiai, S., Oura, H. and Nakajima, T. (1976). Color reaction of some saponinins and saponins with vanillin and sulfuric acid. *Planta Medica*, 29, 116-122.
- Huang, D., Ou, B. and Prior, R.L. (2005). The Chemistry behind antioxidant capacity assays. *Journal of Agricultural and Food Chemistry*, 53(6), 1841-1856.
- Jiménez, N., Carrillo-Hormaza, L., Pujol, A., Álzate, F., Osorio, E. and Lara-Guzman, O. (2015). Antioxidant capacity and phenolic content of commonly used anti-inflammatory medicinal plants in Colombia. *Industrial Crops and Products*, 70, 272-279.
- Jin, L., Zhang, Y., Yan, L., Guo, Y. and Niu, L. (2012). Phenolic compounds and antioxidant activity of bulb extracts of six *Lilium* species native to China. *Molecules*, 17, 9361-9378.
- Joshi, S.S., Su, X. and D'Souza, D.H. (2015). Antiviral effects of grape seed extract against feline calicivirus, murine norovirus, and hepatitis A virus in model food systems and under gastric conditions. *Food Microbiology*, 52, 1-10.
- Jothy, S. L. Z., Z., and Sasidharan, S. (2011). Phytochemicals screening, DPPH free radical scavenging and xanthine oxidase inhibitory activities of *Cassia fistula* seeds extract. *Journal of Medicinal Plants Research*, 5, 1941-1947.
- Kennedy, J. A., Matthews, M.A. and Waterhouse, A.L. (2000). Changes in grape seed polyphenols during fruit ripening. *Phytochemistry*, 55, 77-85.

- Kim, S.-Y., Jeong, S.-M., Park, W.-P., Nam, K.C., Ahn, D.U. and Lee, S.-C. (2006). Effect of heating conditions of grape seeds on the antioxidant activity of grape seed extracts. *Food Chemistry*, 97, 472-479.
- Kuete, V., Kanga, J., Sandjo, L., Ngameni, B., Poumale, H., Ambassa, P. and Ngadjui, B. (2011). Antimicrobial activities of the methanol extract, fractions and compounds from *Ficus polita* Vahl. (Moraceae). *BMC Complementary and Alternative Medicine*, 11, 1-6.
- Li, Y., Guo, C., Yang, J., Wei, J., Xu, J. and Cheng, S. (2006). Evaluation of antioxidant properties of pomegranate peel extract in comparison with pomegranate pulp extract. *Food Chemistry*, 96(2), 254-260.
- Meng, J.-F., Fang, Y.-L., Qin, M.-Y., Zhuang, X.-F. and Zhang, Z.-W. (2012). Varietal differences among the phenolic profiles and antioxidant properties of four cultivars of spine grape (*Vitis davidii* Foex) in Chongyi County (China). *Food Chemistry*, 134, 2049-2056.
- Pająk, P., Socha, R., Gałkowska, D., Rożnowski, J. and Fortuna, T. (2014). Phenolic profile and antioxidant activity in selected seeds and sprouts. *Food Chemistry*, 143, 300-306.
- Pastrana-Bonilla, E., Akoh, C.C., Sellappan, S. and Krewer, G. (2003). Phenolic content and antioxidant capacity of muscadine grapes. *Journal of Agricultural and Food Chemistry*, 51, 5497-5503.
- Pozharitskaya, O.N., Shikov, A.N., Kosman, V.M., Selezneva, A.I., Urakova, I.N., Makarova, M.N. and Makarov, V.G. (2015). Immunomodulatory and antioxidant properties of fixed combination of fish oil with plant extracts. *Synergy*, 2, 19-24.
- Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M. and Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine*, 26, 1231-1237.
- Saeed, A., Sultana, B., Anwar, F., Mushtaq, M., Alkharfy, K.M. and Gilani, A.-H. (2014). Antioxidant and antimutagenic potential of seeds and pods of green cardamom (*Elettaria cardamomum*). *International Journal of Pharmacology*, 10, 461-469.
- Sanchez-Bel, P., Romojaro, A., Egea, I. and Pretel, M.T. (2015). Wild edible plants as potential antioxidant or nutritional supplements for beverages minimally processed. *LWT - Food Science and Technology*, 62(1, Part 2), 830-837.
- Sarikurkcu, C., Tepe, B., Daferera, D., Polissiou, M. and Harmandar, M. (2008). Studies on the antioxidant activity of the essential oil and methanol extract of *Marrubium globosum* subsp. *globosum* (Lamiaceae) by three different chemical assays. *Bioresource Technology*, 99, 4239-4246.
- Tagne, R.S., Telefo, B.P., Nyemb, J.N., Yemele, D.M., Njina, S.N., Goka, S.M.C., et al. (2014). Anticancer and antioxidant activities of methanol extracts and fractions of some Cameroonian medicinal plants. *Asian Pacific Journal of Tropical Medicine*, 7, Supplement 1, S442-S447.
- Tauchen, J., Doskocil, I., Caffi, C., Lulekal, E., Marsik, P., Havlik, J., et al. (2015). In vitro antioxidant and anti-proliferative activity of Ethiopian medicinal plant extracts. *Industrial Crops and Products*, 74, 671-679.
- Wen, J., Lu, L. and Boggan, J.K. (2013). Diversity and Evolution of Vitaceae in the Philippines. *Philippine Journal of Science*, 142, 223-244.

- Wongnarat, C. and Srihanam, P. (2016). Phytochemical, antioxidant and antibacterial activities in different maturity stages of wild grape (*Ampelocissus martinii* Planch.) fruits. *International Journal of Applied Chemistry*, 12, 337-346.
- C. Wongnarat Srihanam PrasongXia, D.Z., Yu, X.F., Zhu, Z.Y. and Zou, Z.D. (2011). Antioxidant and antibacterial activity of six edible wild plants (*Sonchus* spp.) in China. *Natural Product Research*, 25, 1893-1901.
- Xia, E.-Q., Deng, G.-F., Guo, Y.-J. and Li, H.-B. (2010). Biological Activities of Polyphenols from Grapes. *International Journal of Molecular Sciences*, 11, 622-646.
- Yang, H.-J., Lee, J.-H., Won, M. and Song, K.B. (2016). Antioxidant activities of distiller dried grains with solubles as protein films containing tea extracts and their application in the packaging of pork meat. *Food Chemistry*, 196, 174-179.