



Identification of Five Acylated Anthocyanins and Determination of Antioxidant Contents of Total Extracts of a Purple-Fleshed *Ipomoea batatas* L. Variety Grown in Burkina Faso

**Dominique Saga Kaboré¹, Adama Héma^{1*}, Elie Kabré²,
Raoul Bazié², Abdoul Karim Sakira³, Moumouni Koala^{1,4},
Paul-André Koussao Somé⁵, Eloi Palé¹, Touridomon Issa Somé³, Pierre Duez⁶
and Mouhoussine Nacro¹**

¹Laboratoire de Chimie Organique et de Physique Appliquées, Université Joseph KI-ZERBO, 03 BP 7021 Ouagadougou 03, Burkina Faso.

²Laboratoire National de Santé Publique 09 BP 24 Ouagadougou 09, Burkina Faso.

³Laboratoire de Chimie Analytique et de Toxicologie, UFR/ Sciences de la Santé, Université Joseph Ki-Zerbo, Burkina Faso.

⁴Institut de Recherche en Sciences de la Santé (IRSS/CNRST) 03 BP 7192 Ouagadougou 03, Burkina Faso.

⁵CNRST/INERA, Département Productions Végétales, 03 BP 7047 Ouagadougou 03 Burkina Faso.

⁶Service de Chimie Thérapeutique et de Pharmacognosie, Université de Mons, 25 Chemin du Champ de Mars, 7000 Mons, Belgique.

Authors' contributions

This work was carried out in collaboration among all authors. Authors EK, RB and PD did HPLC-MS/MS analysis. Author AKS responsible for the cuttings of the varieties. Authors EP and MN manage laboratory analysis. Author TIS collaborate the project. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IRJPAC/2021/v22i730420

Editor(s):

(1) Dr. Farzaneh Mohamadpour, University of Sistan and Baluchestan, Iran.

Reviewers:

(1) Sinha Ashutosh Kumar, Bharat Pharmaceutical Technology, India.

(2) Ramazan Erenler, Tokat Gaziosmanpasa University, Turkey.

Complete Peer review History: <https://www.sdiarticle4.com/review-history/73800>

Original Research Article

**Received 05 July 2021
Accepted 15 September 2021
Published 25 September 2021**

ABSTRACT

Anthocyanins are bioactive compounds, which thanks to their anti-free radical properties, can protect the human body against oxidative stress. The latter can cause many diseases, such as cancer, aging. The extract of the green cap purple variety of the sweet potato (*Ipomoea batatas* L.) is active against the radical ABTS. Its antioxidant content is estimated at 0.183 mg E TEAC/ g fresh material. The characterization of anthocyanins was performed by high performance liquid chromatography-mass spectrometry-UV (HPLC-MS-UV) and high performance liquid chromatography-mass spectrometry (HPLC-MS/MS) analyses. These analyses allowed the identification of five anthocyanic compounds. These are: cyanidin 3-(6''-caffeoyl- 6'''-*p*-hydroxybenzoyl sophoroside) -5-glucoside; cyanidin 3-(6''-feruloyl 6'''-caffeoyl sophoroside) -5-glucoside; peonidin 3-(6'''-caffeoyl sophoroside)-5-glucoside; peonidin 3-(6''-caffeoyl-6'''-*p*-hydroxybenzoyl sophoroside) -5-glucoside; and peonidin 3-(6''-feruloyl -6'''-caffeoyl sophoroside)-5-glucoside.

Keywords: Anthocyanins; high performance liquid chromatography ; mass spectrometry.

1. INTRODUCTION

Anthocyanins are positively charged flavonoids with a C6-C3-C6 structure and are water-soluble pigments most commonly encountered in nature. The structures of anthocyanins vary according to the nature, number and position of the sugars attached to the aglycone; as well as the number of hydroxyl groups, aliphatic or phenolic acids that are attached to these different sugars [1]. This particular chemical structure makes them powerful natural antioxidants and therefore very reactive towards free radicals [2]. The six most common aglycones in plants are cyanidin, peonidin, delphinidin, pelargonidin, petunidin and malvidin [3]. The basic structures of purple sweet potato anthocyanins are cyanidin, peonidine and pelargonidin. Among them, the pelargonidin content is relatively low [1]. But the most abundant anthocyanins in reported PFSP were peonidin derivatives [4,5]. In these varieties, glycosylated and acylated anthocyanins account for over 93% [4,6].

The purple-fleshed sweet potato has a high nutritional value due to its abundance of phytochemicals beneficial to human health; it is therefore considered a functional food [7]. C3 and C5 were combined separately with sophorosis and glucose to form a glycosidic bond. C6 sophorosis was combined with *p*-hydroxybenzoic acid, ferulic acid, coumaric acid or conic acid to form one or two monoacylated bonds [8]. Acylation with various phenolic acids makes PSP anthocyanins unique and also offers some benefits in pH and heat resistance, light sensitivity, and overall stability. From a nutritional point of view, acylated anthocyanins have been reported to possess high antioxidant and anti-

mutagenicity activities [4]. When it comes to color food with water-soluble natural red pigments, the large group of anthocyanins is the first choice for coloring beverages, fruit preparations and confectionery [9].

Anthocyanins, especially acylated ones, play a very important role in human health. Indeed, a study on adult men with borderline hepatitis indicated that consumption of purple-fleshed sweet potato beverages (400 mg anthocyanins/day) may have a potential protective capacity for the liver against oxidative stress [10]. Acylated anthocyanins are known to have better stability and biological activities [11].

In order to contribute to the study of anthocyanins in purple sweet potato, the variety TUSKEGEE POURPRE CAP VERT, introduced and produced in the agroecological conditions of Burkina Faso, was studied. Indeed, the present work aims to determine the antioxidant contents and to identify anthocyanin structures from this variety by coupling High-Performance Liquid chromatography and Mass spectrometry methods (HPLC-ESI-MS/MS).

2. MATERIALS AND METHODS

2.1 Plant Material

The plant material studied consists of tubers of a local variety of sweet potato with purple flesh. It is the variety TUSKEGEE POURPRE CAP VERT produced in an experimental garden of Laboratory of pure and applied organic chemistry and physics in Joseph Ki-Zerbo University. The cuttings of this variety were provided by INERA

(Institut de l'Environnement et de Recherches Agricoles).

2.2 Methods

2.2.1 Extraction

Acidified methanol 1 % (hydrochloric acid) was used to extract anthocyanins of the tubers of PFSP. Crude extracts obtained were evaporated almost dry and then filtered and taken back with a minimum of acidified water. The filtrate was deposited on the Amberlite XAD-7 in a column of about 24 cm [12]. The extract was then washed with a large amount of acidified water to get rid of impurities and free sugars contained in the crude extract. Total anthocyanin extract was removed from the column using a minimum of acidified methanol. This total extract was evaporated to dry and dissolved in a minimum of pure acidified methanol. A few drops of pure ethyl acetate in the previous extract precipitate anthocyanins whose drying with Speed dryer makes it possible to obtain an amorphous powder of purple color. For HPLC-ESI-MS/MS tests, a 1 mg/mL concentration solution was prepared using the 95:5 (v/v) water-acid mixture as a solvent [13].

For the determination of antioxidant contents, acidified ethanol 1% (hydrochloric acid) was used for extraction. Indeed, 5 g (gram) of fresh plant material was extracted with 10 mL of the solvent in an ultrasonic for 1h. This operation was repeated twice.

2.2.2 Evaluation of antioxidant contents

The antioxidant content (TAO) was determined by the radical-cation reduction method of 2,2'-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS). Indeed, the Trolox Equivalent Antioxidant Capacity (TEAC) assay reported by Miller and Rice-Evans [14] is based on the ability of antioxidant compounds in an extract to scavenge the radical-cation. The radical-cation was chemically generated by mixing 10 mL of potassium persulfate solution $K_2S_2O_8$ to and 50 mL of 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) solution at 7.01 mM. The resulting mixture was stored in the refrigerator for 16 hours before use. The resulting greenish-blue solution was diluted to have an absorbance of approximately 0.700 ± 0.05 at 734 nm [15]. In addition, 50 μ L of the extract was added to 200 μ L of ABTS solution and the absorbance was measured after 10 min at 734

nm using MP96 microplate spectrophotometer, SAFAS.

2.2.3 Analysis using HPLC-DAD-MS-UV visible

PFSP extract was first analyzed using high-performance liquid chromatography coupled with mass spectroscopy equipped with a positive electrospray ionization source (ESI) and UV-visible spectroscopy. An agilent HPLC system was used. It has a column type "Column Oven (G7130A)" type. This was a 250 mm long C18 column. The elution was achieved with the mobile phase C (acetonitrile 5 % formic acid /H₂O 1:1 v: v) and the mobile phase D (H₂O 5 % formic acid v: v). The gradient of elution was 0 to 2 min, 20 % C; 2 to 15 min, 40 % C; 15 to 17 min, 50 % C; 17 to 27 min, 50 % C; 27 to 40 min, 95 % C and 40 to 50 min, 95 % C. The flow has been maintained at 1 mL/min and the injection volume was 10 μ L [16].

2.2.4 HPLC-ESI-MS/MS Analysis

HPLC coupled with tandem mass spectrometry by electrospray ionization in positive mode (HPLC-ESI-MS/MS) was used in this study for the identification of anthocyanins. Indeed, a HPLC system of Agilent technology infinitely better 1290 was used for chromatographic separation. It had a C18 reverse-phase column with a length of 250 mm and a diameter of 4.6 mm, the size of the particles was 5 μ m. The elution was achieved with the mobile phase A (water : 5% formic acid v: v) and the mobile phase B (acetonitrile : 5% formic acid, v: v). The gradient of elution according to the time expressed as a percentage of the volume of **A** and the mobile phase **B** was programmed as follows: 0 to 5 min, 5% B; 5 to 15 min, 10% B; 15 to 25 min, 10% B; 25 to 35 min, 12% B; 35 to 50 min, 15% B; 50 to 60 min, 18% B; 60 to 80 min, 25% B and 80 to 90 min, 30% B. The flow was maintained at 0.6 mL/min and the temperature of the column at 25°C [1].

Mass spectrometry scanning was performed in positive mode with a scanning interval of 200-1200 m/z. Nebulization was performed at 200°C with a simultaneous flow of N₂ to 15 psi. The hair strains were set to 3.5 kV. Data were analyzed using the LC/MS Data Acquisition software for 6400 series triple quadrupole version B.06.00 Bulld 6.0.6025.0.

3. RESULTS AND DISCUSSION

3.1 Antioxidant Contents

For the evaluation of antioxidant levels by the Trolox Equivalent Antioxidant Capacity (TEAC) method, a standard curve was previously established using Trolox as the reference antioxidant ($y = -24.371x + 0.5953$) (Fig.1) and the result is then expressed in mg Trolox Equivalents (TE) per gram of fresh material. The antioxidant content of the anthocyanin extract of TUSKEGEE POURPRE CAP VERT variety was $0,183 \pm 0.005$ mg d'E TEAC/ g; lower than those found by Yeong Ran Im et al which were 6 mg/ g [17].

3.2 HPLC-MS-UV Profiles of the Anthocyanin Extract

The chromatographic profile of the extract revealed thirteen peaks noted from 1 to 13 corresponding to 13 compounds of anthocyanin (Fig. 2). Among these thirteen peaks, only peaks 5, 10, 12 and 13 were major.

In ultraviolet-visible (UV) spectroscopy, all the anthocyanins in the extract showed the two characteristic absorption peaks of anthocyanins, notably at about 280 nm and 530 nm. In addition, most of these compounds showed an absorption peak around 330 nm characteristic of the presence of acid substituents. Through the different UV-visible spectra recorded we can note that all the anthocyanins of extract were glycosylated in position 3 and 5 (Fig. 3). Indeed, the position and number of these acid

substituents as well as the position of the different glycosyl moieties could be deduced from the A_{440}/A_{vis} and A_{acyl}/A_{vis} ratios (Table 1). Indeed, a ratio of A_{440}/A_{vis} between 29% and 35% or 15% and 24 % indicates respectively a monoside or a bioside [18,19]. Three different cinnamic acids were found acylating the anthocyanins of the studied extract. These were caffeic, *p*-hydroxybenzoic and ferulic acids.

3.3 HPLC-MS/MS Profiles

High performance liquid chromatography (HPLC) coupled to electrospray ionization tandem mass spectrometry (ESI-MS/MS) on a triple quadrupole instrument was used to study anthocyanins of the extract. The LC-ESI-MS in SIM mode "select ion monitoring" was used to obtain chromatograms of all molecular ions already identified in the previous study (LC-MS-UV). After obtaining these chromatograms, the LC-ESI-MS/MS in "ion product" mode was used to obtain individual chromatograms for the purpose of fragmentation of molecular ions.

Using the chromatographic conditions described above, and from the study previously carried out, we were interested in the five compounds corresponding to peaks 3, 5, 6, 7 and 9 at the respective retention times RT= 47.09; 51.552; 53.57; 55.14 and 58.65 min (Fig. 4). The UV-visible spectra corresponding to these compounds showed characteristic absorption bands of anthocyanins (cyanidin and peonidin) with wavelengths ranging from 518 nm to 528 nm [20].

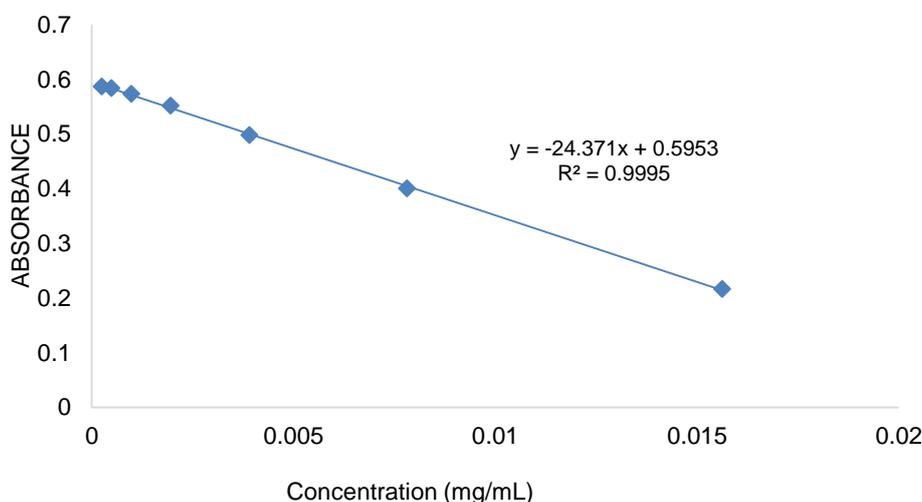


Fig. 1. Trolox standard curve by method (ABTS)

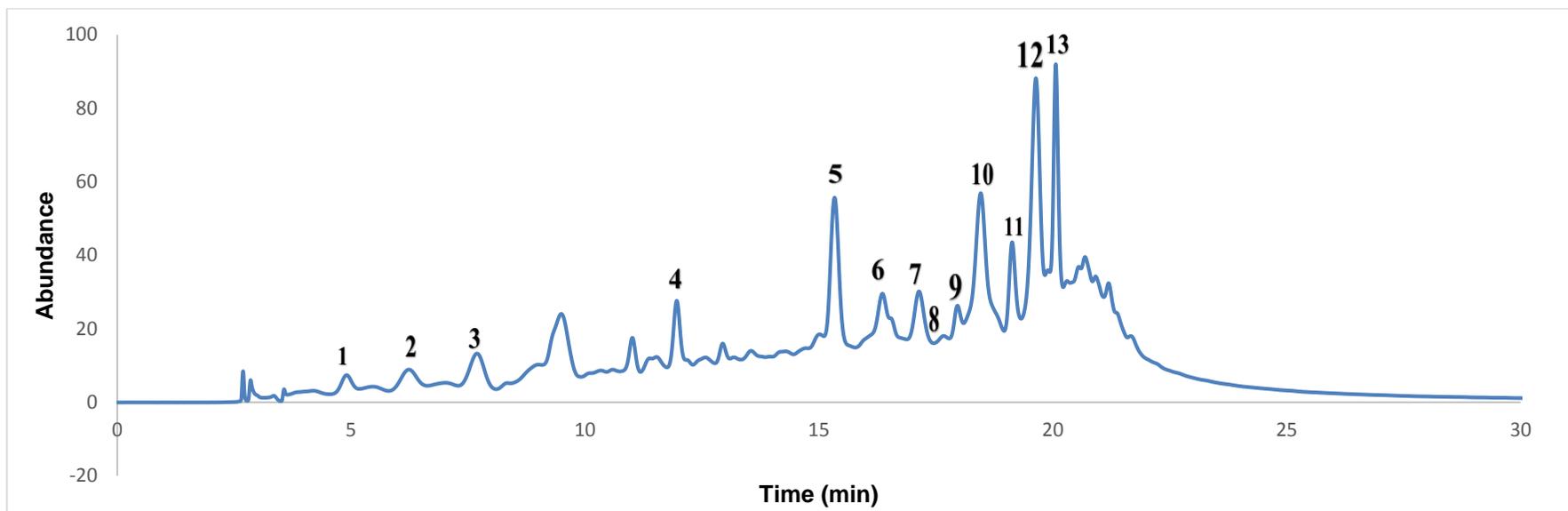
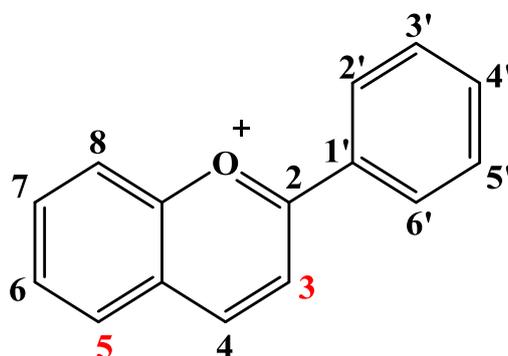


Fig. 2. HPLC-UV-MS chromatogram of the crude extract of TUSKEGEE POURPRE CAP VERT

Table 1. Data obtained from HPLC-UV-MS chromatogram and UV-visible spectra of anthocyanins

Peak number	Retention time	% Area	λ_{vis}	λ_{acy}	A_{440}/A_{maxvis} %	A_{acy}/A_{vis} %	M+H
1	5.06	1.19	514		18		773
2	6.5	2.09	514		18		611
3	7.92	3.59	514		18		787
4	12.32	3.67	520	330	17	30	907
5	15.49	10.18	520	328	14	25	963
6	16.55	5.98	520	328	18	62	935
7	17.17	3.9	518	328	18	51	1055
8	17.73	1.90	514		19		773
9	18.24	1.56	528	330	19	105	1111
10	18.5	13.13	522	330	18	60	949
11	19.15	3.78	526	330	15	49	1069
12	19.65	14.72	520		14		787
13	20.07	8.11	520	328	22	66	1125

λ_{vis} : visible UV wavelength; λ_{acy} : acylated wavelength; A_{440} : absorbance at wavelength 440; A_{maxvis} : maximum visible uv absorbance; A_{acy} : acylated absorbance; % : percentage

**Fig. 3. Structure of anthocyanidin**

3.3.1 Identification of compound 3

The electronic spectrometry spectrum of compound **3** showed an absorption band around 328 nm which could correspond to the presence of an acyl group confirmed by the $A_{acy}/A_{max.vis}$ ratio of about 51% (Fig. 5). Anthocyanins are more stable when position 3 of the aglycone is occupied as a priority.

The LC-MS/MS analysis of the mass spectrometry of compound **3** revealed a molecular ion $[M+H]^+$ at m/z 1055 at retention time $RT= 47.09$ min (Fig. 6) corresponding to the mass calculated from molecular formula $C_{49}H_{51}O_{26}$ and three fragment ions.

The fragment ion at m/z 449 $[M+H-606]^+$ would correspond to the loss of 2 glycosyl + *p*-

hydroxybenzoyl and caffeoyl moiety; all in position 3.

The $A_{440}/A_{max.vis}$ ratio about 18% obtained from its UV-visible spectrum showed that this compound was glycosylated in position 5 [19]. A fragment ion at m/z 893 with formula $C_{43}H_{41}O_{21}$.

This fragment ion corresponded to the loss of an hexose $[M+H-hexose]^+$. This fragment ion could correspond either from the loss of a glycosyl (glucosyl or galactosyl) in position 5 or from the loss of caffeoyl $[M+H-caffeoyl]^+$ bound to sophoroside in position 3 of aglycone.

In addition, the presence of the fragment ion at m/z 286.700 (≈ 287) revealed that **3** was cyanidin derivative (Fig. 7). Compound **3** could be identified as cyanidin 3-(6''-caffeoyl- 6'''-*p*-hydroxybenzoyl sophoroside)-5-glucoside (Fig. 8).

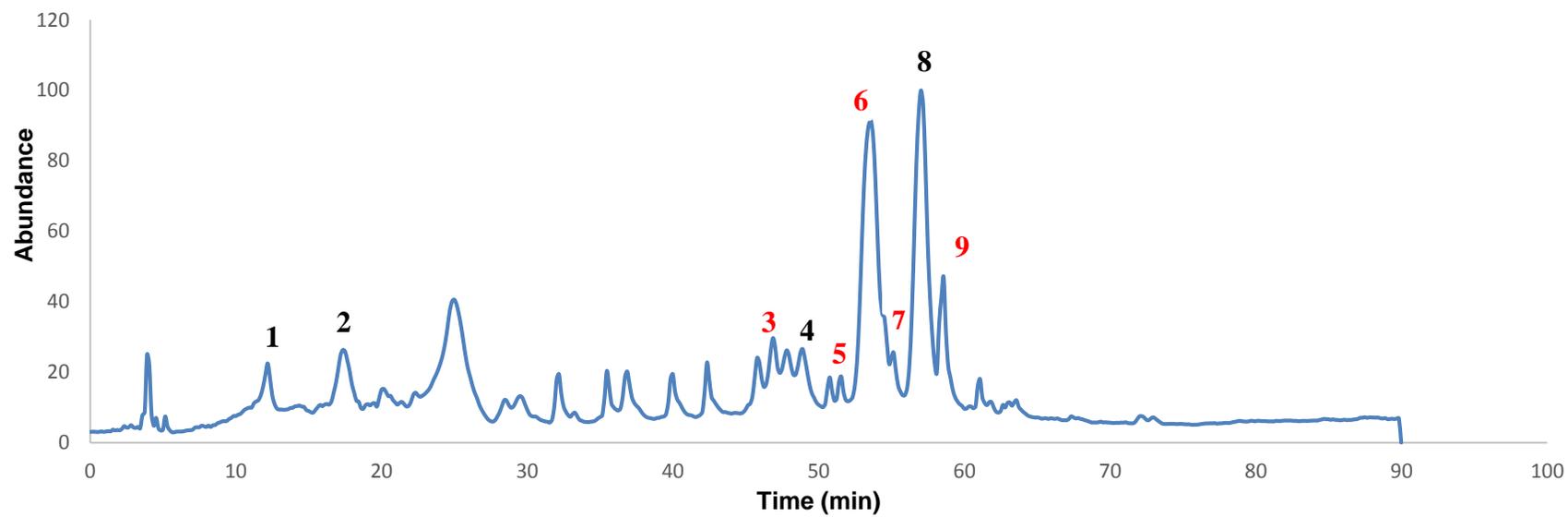


Fig. 4. HPLC/MS-MS chromatogram of the crude extract of TUSKEGEE POURPRE CAP VERT

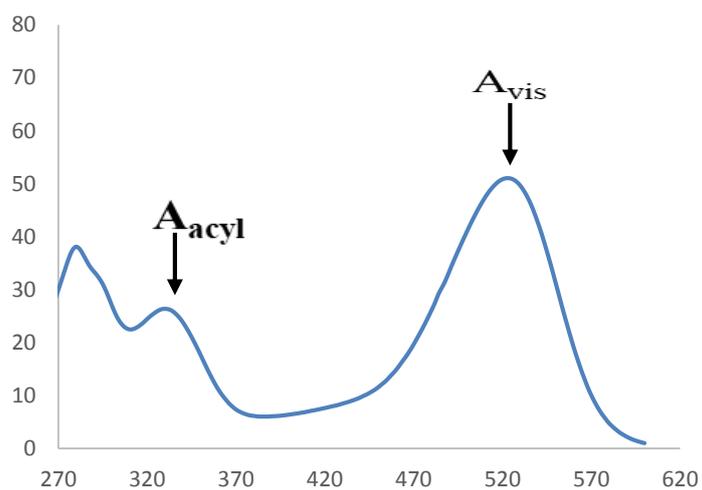


Fig. 5. UV -Visible spectrum of compound 3

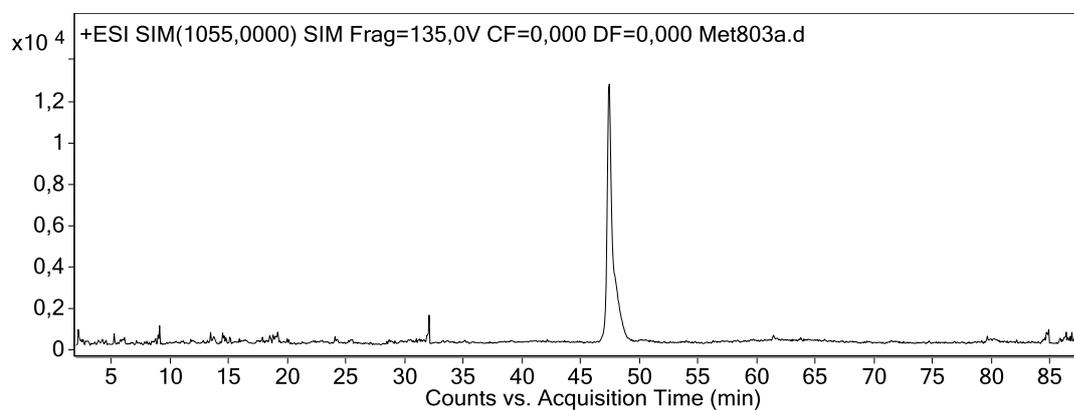


Fig. 6. Chromatogram of compound 3

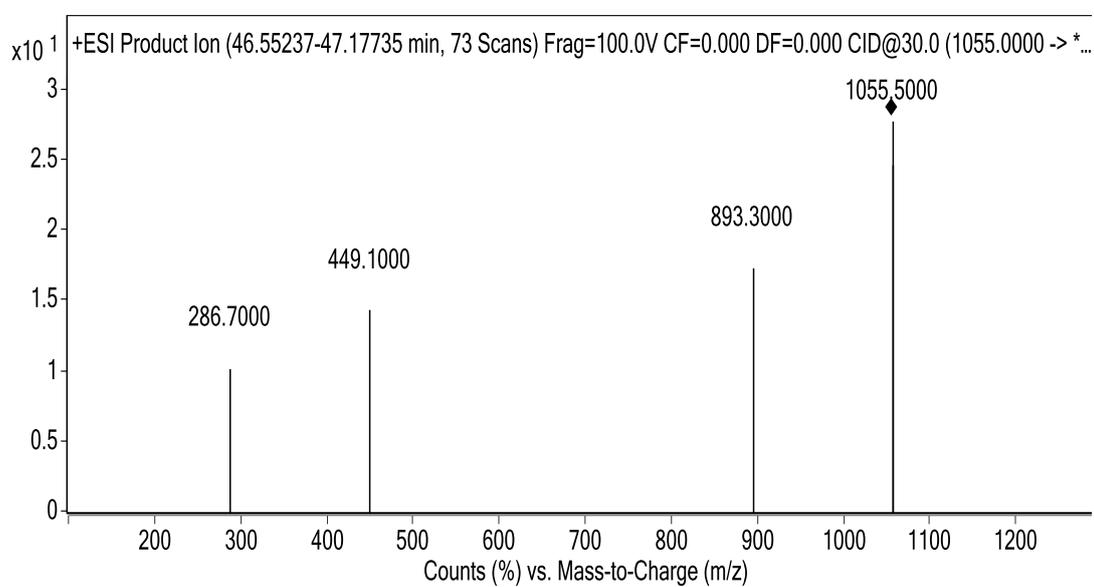


Fig. 7. MS/MS spectra of compound 3

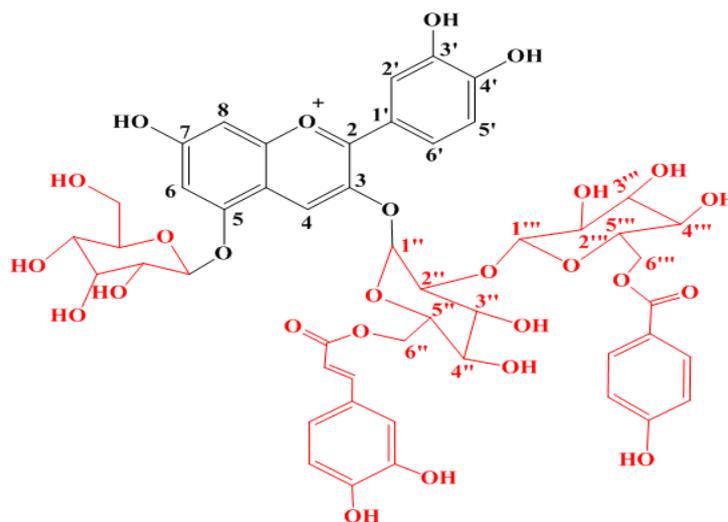


Fig. 8. Structure of compound 3

3.3.2 Identification of compound 5

In addition to the absorption band at 528 nm characteristic of the 3-glucosylated cyanidin, the electronic spectrum of compound 5 (Fig. 9) showed an absorption band around 330 nm characteristic of the presence of an acyl group. This result was confirmed by the $A_{acyl}/A_{max.vis}$ ratio of about 105%. The UVvisible spectrum gave a ratio $A_{440}/A_{max.vis}$ of about 19 %. This result showed that compound 5 was glycosylated at position 5. The chromatographic analysis revealed two peaks at retention times 51.72 min and 54.23 min (Fig. 10). The molecular ion corresponding to these peaks had a mass of m/z 1111. It corresponded to the mass calculated from the formula $C_{52}H_{55}O_{27}$. By comparing the intensity of the peaks, the compound with retention time at 51.72 min was major in the studied extract. To determine the structure of this compound 5 two techniques were used. The MRM (multiple reaction monitoring) and ion product techniques.

The MRM technique was used specifically in this case due to some troubles with the separation. It is a very specific and sensitive mass spectrometry technique that allowed the selective quantification of compounds in an extract. It uses a triple MS quadrupole that first targets the molecular ion corresponding to the compound with a later fragmentation of that compound to produce a range of fragment ions. Only the specific fragment ions from the targeted molecular ion are isolated in the mass spectrometry [21]. In this case, the mass spectrum of the compounds gave three masses

corresponding to the fragment ions at m/z 949; 463 and 449 (Fig.11). This result revealed that the studied extract contained two molecular ions at m/z 1111. A molecular ion that gave the fragment ions at m/z 949 and 463 corresponding to a peonidin derivative; the other which gave fragment ions at m/z 949 and 449 corresponding to a cyanidin derivative. This result confirmed that of high-performance liquid chromatography where the chromatogram (Fig.10) showed two peaks at different retention times (51.72 and 54.23 min). The cyanidin derivative being more polar had the lowest retention time compared to that of peonidin which had a methyl group on the B ring of aglycone.

LC-MS/MS mass spectrum analysis by the ion product technique showed one molecular ion at m/z 1111 and two fragment ions at m/z 949 and 287 (Fig. 12). The fragment ion at m/z 949 would correspond to the loss of a hexose $[M+H-glycosyl]^+$. This peak corresponded to the loss of a glycosyl which could be at position 5 as reported by Monalisa et al. [16]; the position 3 being occupied by a caffeoyl-feruloyl sophoroside group. The fragment ion at m/z 287 corresponds to the aglycone of cyanidine. It corresponds to the loss of caffeoyl-feruloyl sophoroside $[M+H-caffeoyl-feruloyl\ sophoroside]^+$. This compound can therefore be identified as cyanidin 3-(6''-feruloyl-6'''-caffeoyl sophoroside)-5-glucoside (Fig. 13). As the molecular ion corresponding to the peonidin derivative was not abundant (Fig. 10), fragmentation could not be performed for the confirmation of the structure of the peonidin derivative.

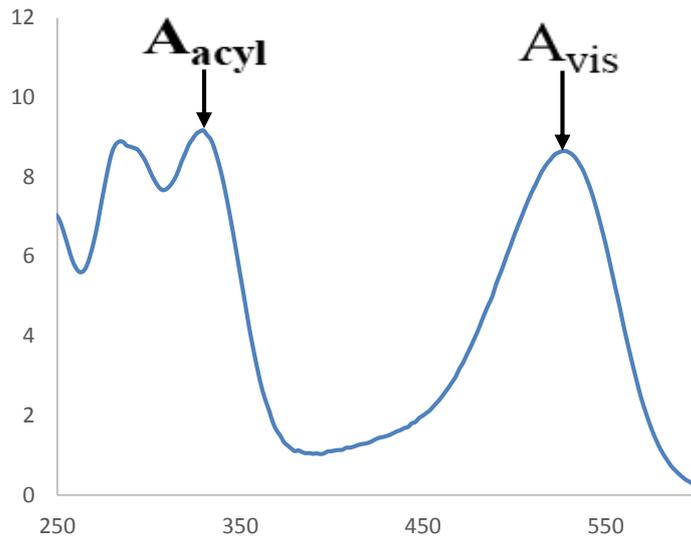


Fig. 9. Visible UV spectrum of compound 5

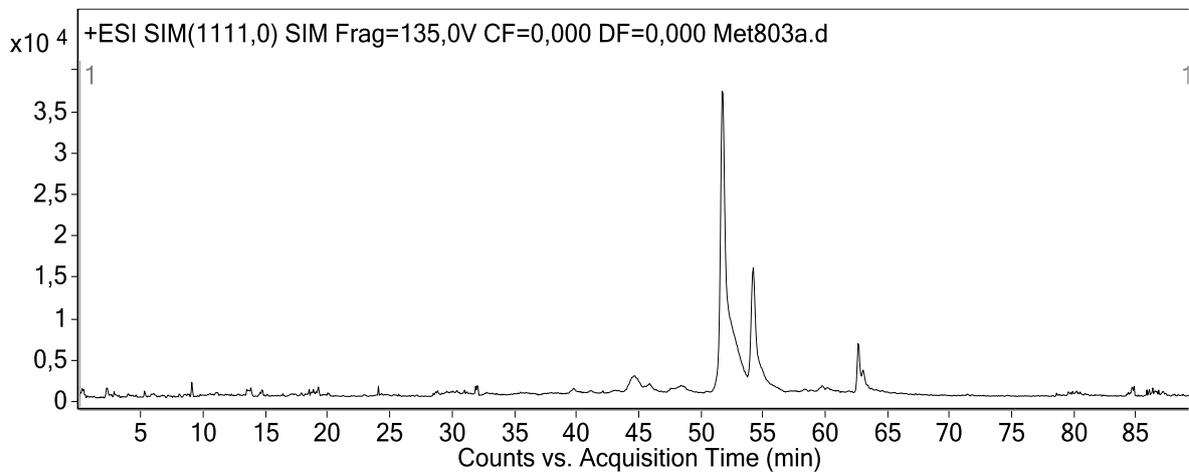
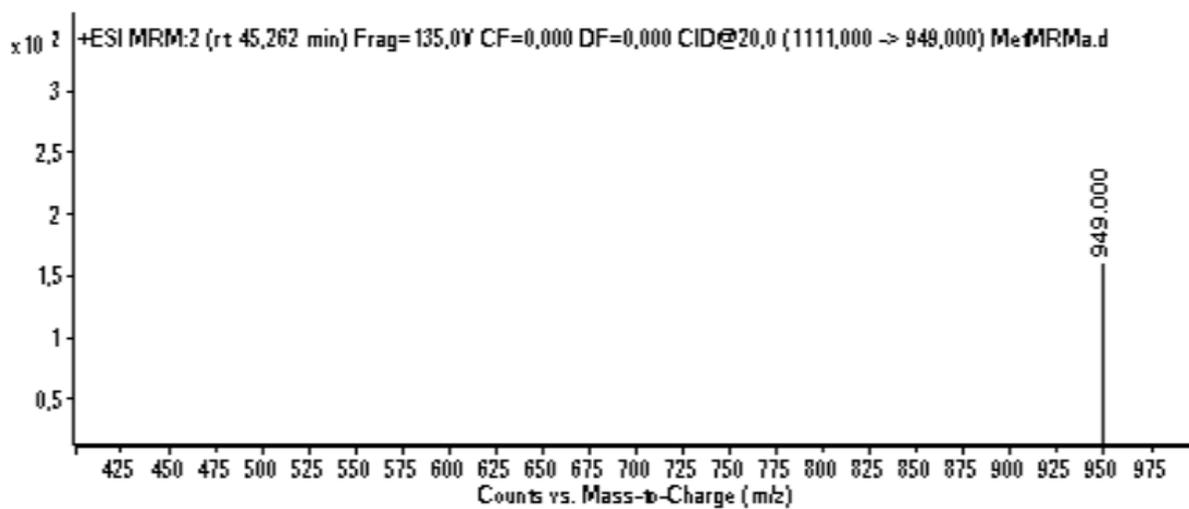


Fig. 10. chromatogram of compound 5



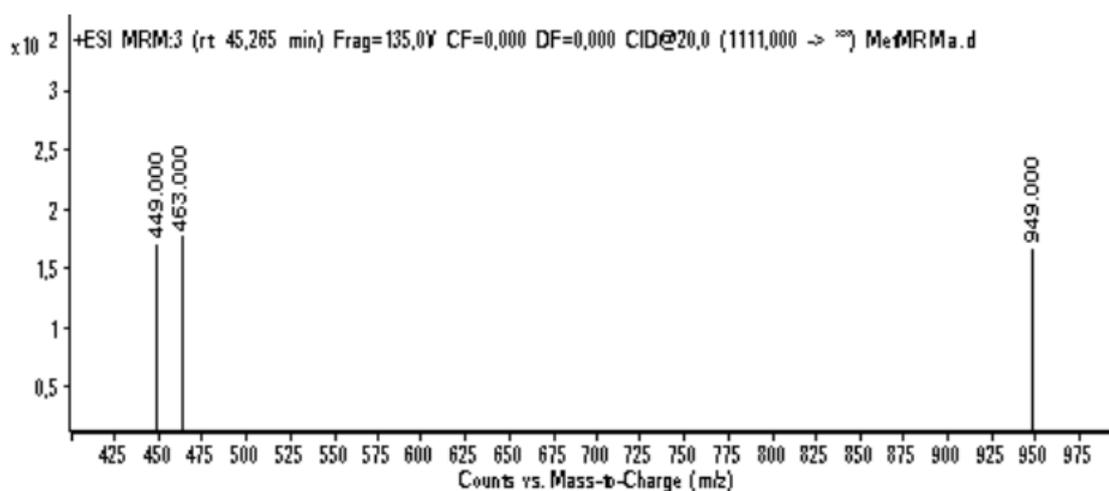


Fig. 11. MS/MS spectrum in MRM mode

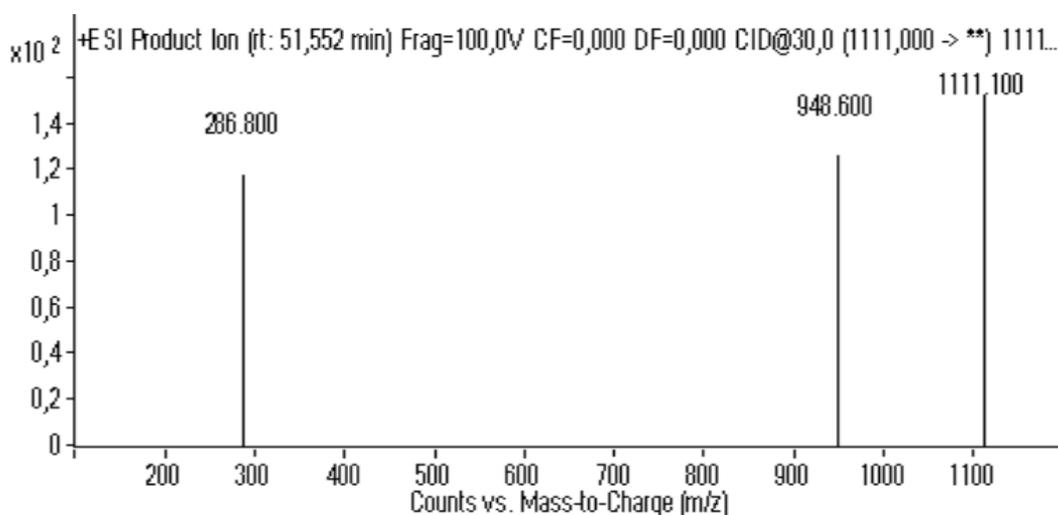


Fig. 12. Structure of compound 5

3.3.3 Identification of compound 6

The electronic spectrum of compound **6** showed an absorption band around 522 nm which could correspond to a glucosylated peonidin derivative in position 3 (Fig. 14). From the UV-visible spectrum of this compound, $A_{440}/A_{\max \text{ vis}}$ and $A_{\text{acy}}/A_{\max \text{ vis}}$ ratios obtained which were 18% and 60% respectively. These values revealed that this compound was not only glucosylated in position 5 but it had in its structure an acylated derivative. This result was in agreement with the results of the LC-MS/MS analysis. Indeed, the MS/MS (Fig. 15) spectrum showed a molecular ion at m/z 949 corresponding to the mass calculated from the molecular formula $C_{43}H_{49}O_{24}$. Like the previous cases this molecular ion gave

three fragment ions (Fig. 16). A fragment ion at m/z 787 corresponding to $[M+H-160]^+$. This peak could correspond to the loss of either the caffeoyl bound to the sophoroside or the glycosyl in position 5 of the aglycone. The fragment ion at m/z 463 corresponded to the loss of 2 glycosyl + caffeoyl fragments all in position 3. The fragment ion at about m/z 301 corresponds to the aglycone of peonidin. The absence of a fragment ion at m/z 449 showed that it was a peonidin derivative. The MS/MS spectrum showed that the peak corresponding to $[M+H-\text{acyl-sophoroside}]^+$ was weaker than the peak corresponding to $[M+H]^+$. So, the caffeoyl group could be in position 6'''. Compound **6** could then be identified as Peonidin 3-(6'''-caffeoyl sophoroside)-5-glucoside (Fig.17).

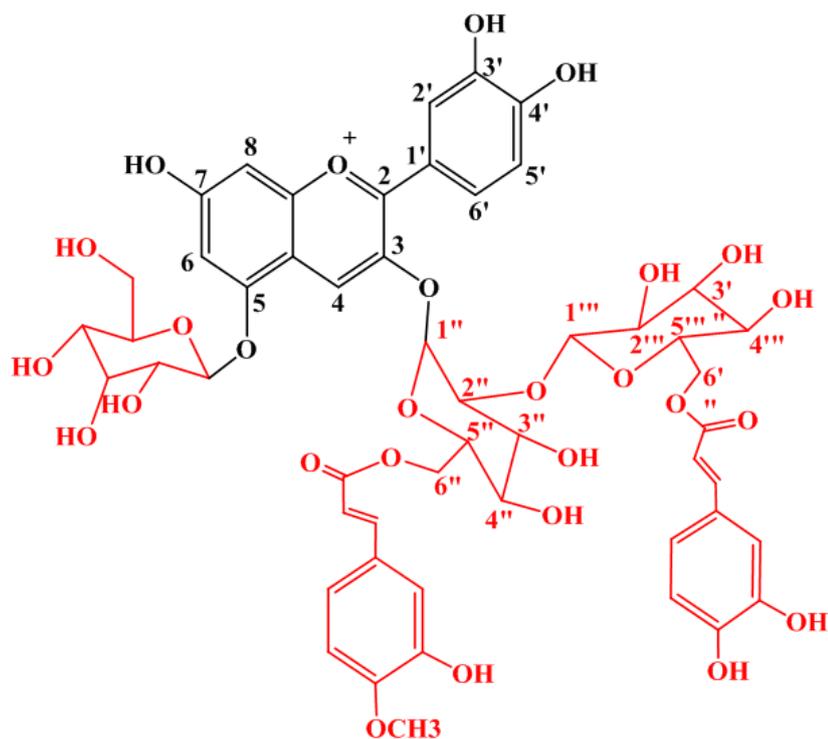


Fig. 13. MS/MS spectrum of compound 5 in "ion product" mode

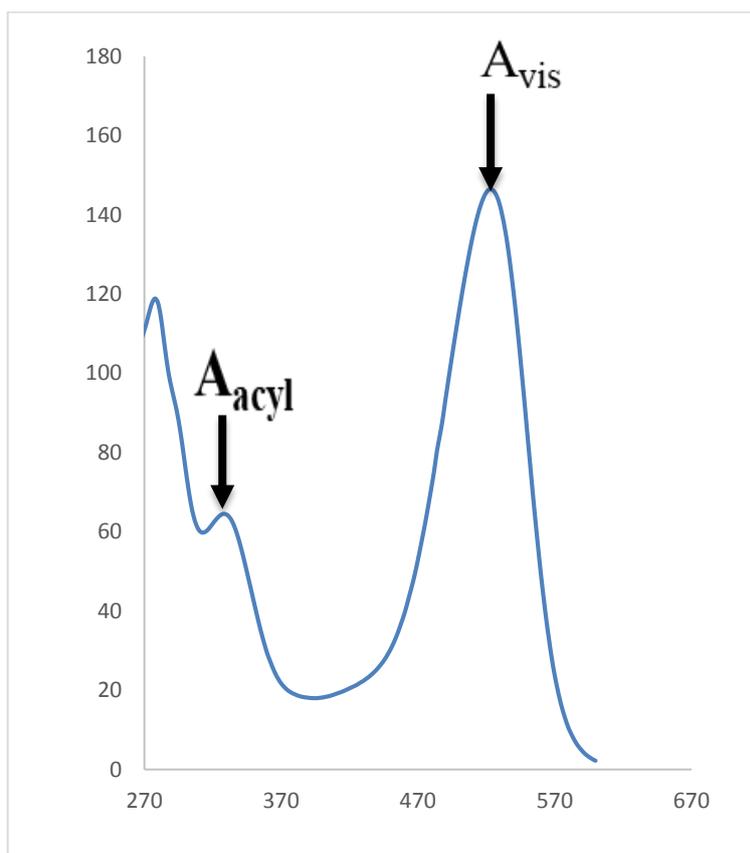


Fig. 14. Visible UV spectrum of compound 6

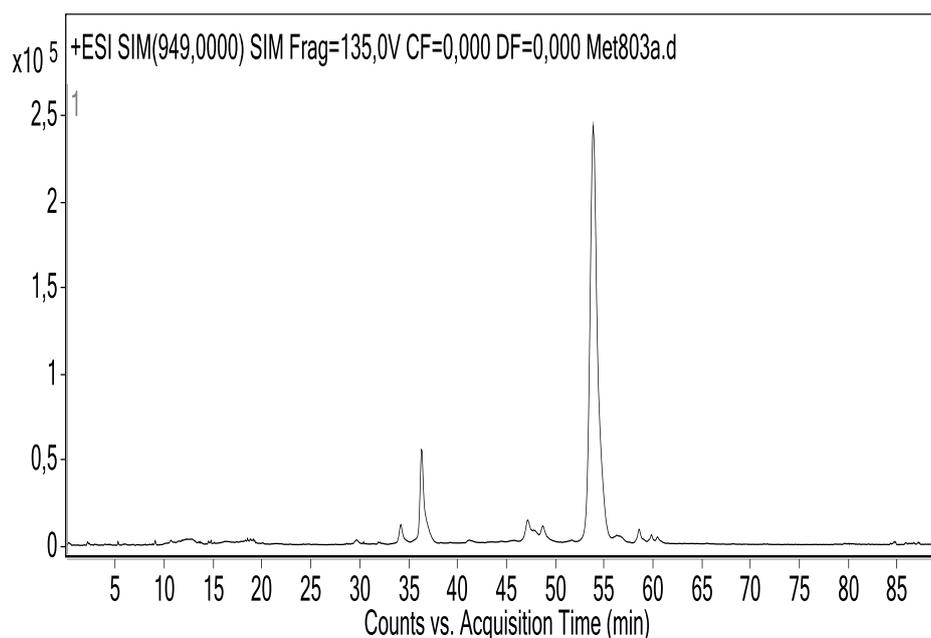


Fig. 15. Chromatogram of compound 6

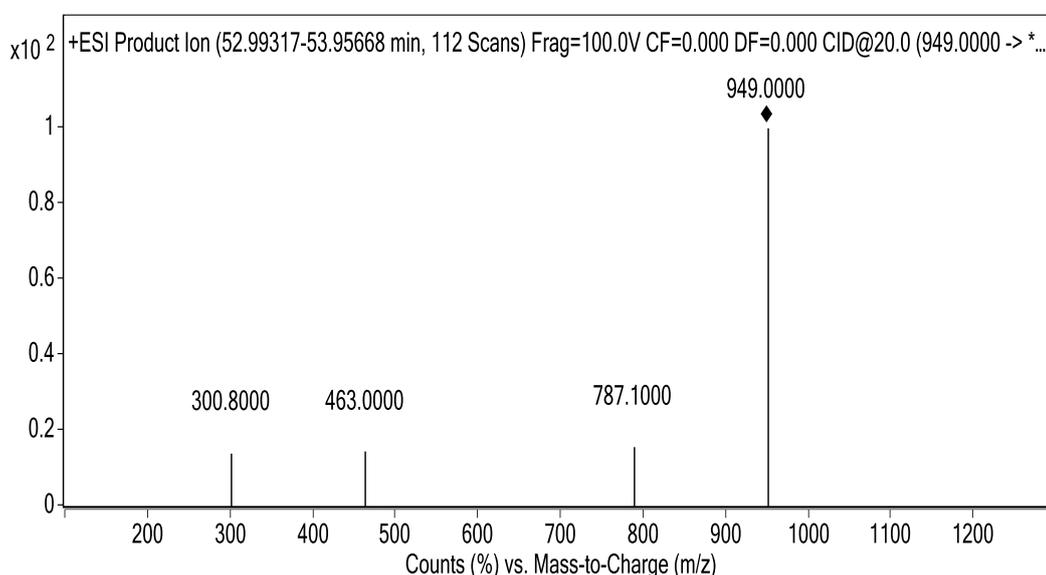


Fig. 16. MS/MS spectrum of compound 6

3.3.4 Identification of compound 7

In addition to the absorption band at 526 nm characteristic of 3-glucosylated peonidin, the electronic spectrum of compound 7 showed an absorption band around 330 nm characteristic of the presence of an acyl moiety (Fig. 19). This was confirmed by the $A_{\text{acyl}}/A_{\text{max.vis}}$ ratio of about 49%. From UV-visible spectrum a $A_{440}/A_{\text{max.vis}}$ ratio 15 % was calculated. This result showed that compound 7 was glycosylated at position 5. The LC/MS chromatogram showed that

compound 3 is more polar than compound 7. This could be explained by the presence of a methyl group in compound 7. The chromatogram of compound 7 showed a peak with a retention time of 55.14 min (Fig. 19). LC-MS/MS analysis of mass spectrometry spectrum of compound 7 showed a molecular ion $[M+H]^+$ at about m/z 1069 corresponding to the mass calculated from molecular formula $C_{50}H_{53}O_{26}$ and three fragment ions (Fig. 20). A fragment ion at m/z 907 corresponding to the formula $C_{41}H_{47}O_{23}$ would correspond to either the loss of a hexose

(glycosyl) bound to the aglycone in position 5 or to the loss of the caffeoyl bound to the sophoroside. The fragment ion at m/z 463 would correspond to the loss of 2 glycosyl + caffeoyl moieties and the *p*-hydroxybenzoyl all in position 3 of the aglycone. The fragment ion at m/z 301 corresponded to the aglycone of peonidin. When acylation occurs at the 6''' position of the carbon belonging to the sophoroside (Fig. 21), then the intensity of the $[M+H-acyl-sophoroside]^+$ peak is lower than the $[M+H]^+$ one [16]; this was the case. Then, the *p*-hydroxybenzoyl moiety was in

the 6''' position. Compound 7 can be identified as peonidin 3-(6''-caffeoyl-6'''-*p*-hydroxybenzoyl sophoroside)-5-glucoside (Fig. 21).

3.3.5 Identification of compound 9

The observation of the electronic spectrum of compound 9 showed three absorption bands characteristic of anthocyanins. One band in the visible at 520 nm, two others in the ultraviolet at 282 nm and at 328 nm (Fig. 22). The last one in uv is characteristic of the presence of an acyl

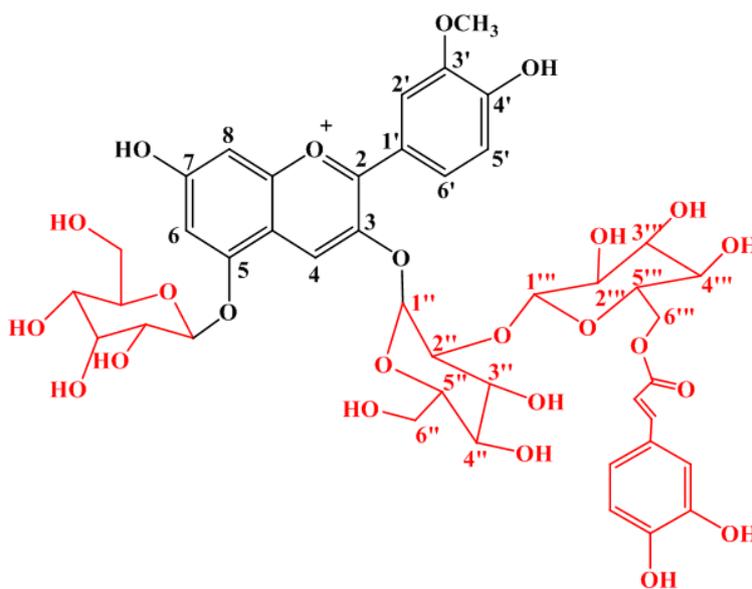


Fig. 17. Structure of compound 6

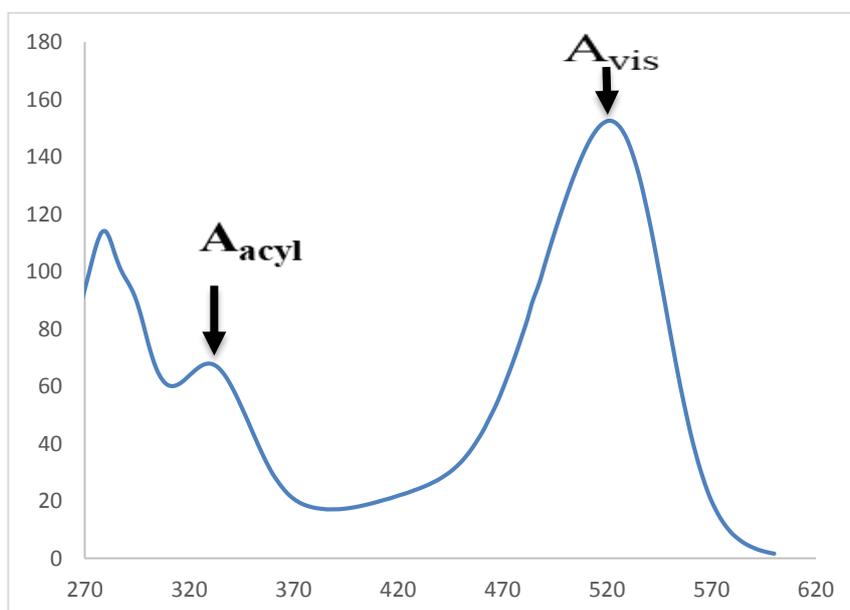


Fig. 18. Visible UV spectrum of compound 7

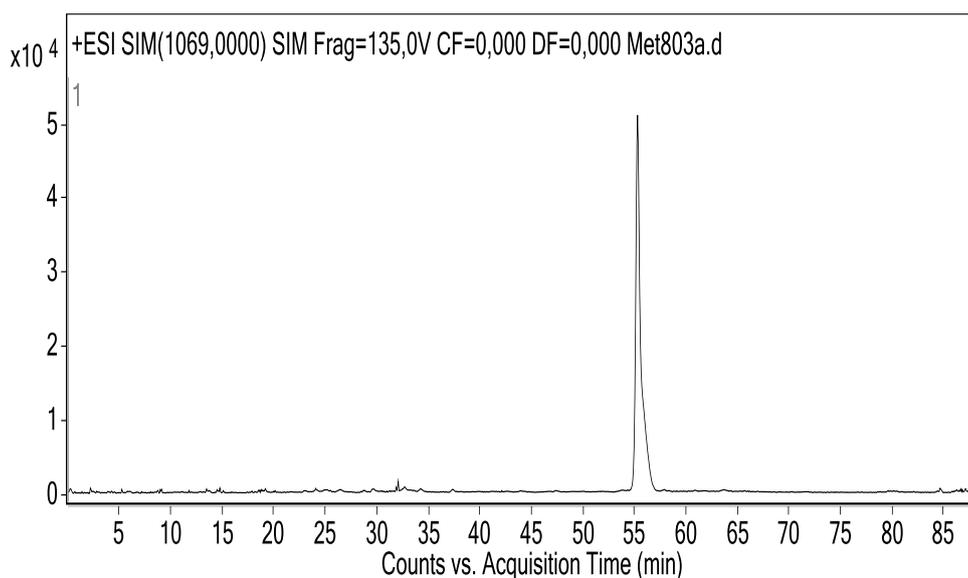


Fig. 19. Chromatogram of compound 7

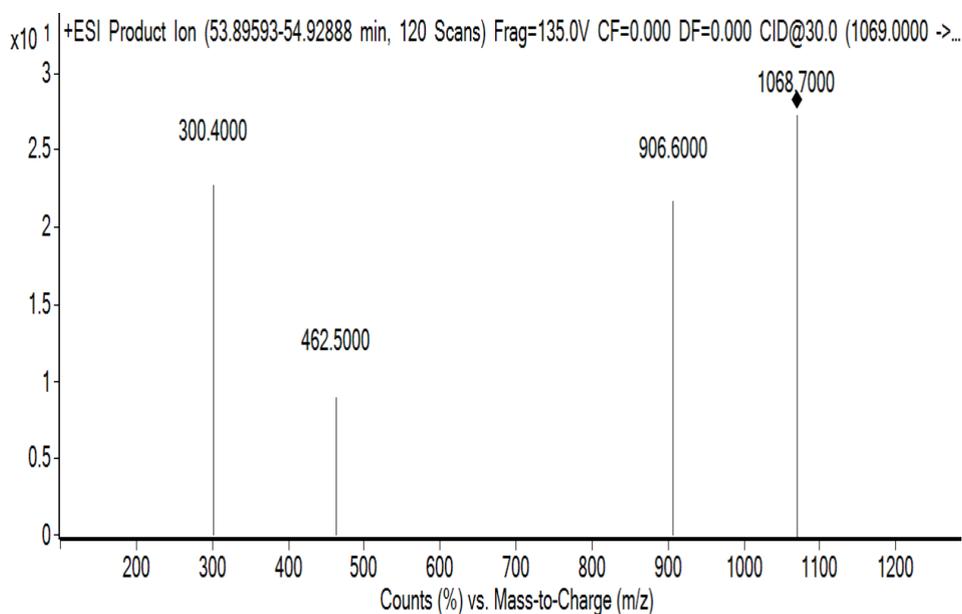


Fig. 20. MS/MS spectrum of compound 7

moiety. This compound could correspond to an acylated derivative of peonidin or cyanidin. This result was confirmed by the $A_{acyl}/A_{max.vis}$ ratio (66%) showing the presence of two acyl groups [22]. This spectrum gave an $A_{440}/A_{max.vis}$ ratio of 22 % which revealed that this compound was glycosylated in position 5.

The chromatogram of extract showed that compound 9 had a retention time $RT = 58.65$ min (Fig.23). ESI-MS/MS signal analysis of this compound showed a molecular ion $[M+H]^+$ at m/z 1125 corresponding to the mass calculated from

molecular formula $C_{53}H_{57}O_{27}$ and three fragment ions (Fig. 24). There was a fragment ion at m/z 963. According to the work reported by Jiu-Liang Zhang et al, [18] this fragment ion corresponded to $[M+H-caffeoyl]^+$. The fragment ion at m/z 463 would correspond to the loss of 2 glycosyl + caffeoyl fragments and the -feruloyl all in position 3 of the aglycone ; the fragment ion at m/z 301 corresponding to the aglycone of peonidin. Compound 9 can be identified as peonidin 3-(6''-feruloyl-6'''-caffeoyl sophoroside)-5-glucoside (Fig. 25).

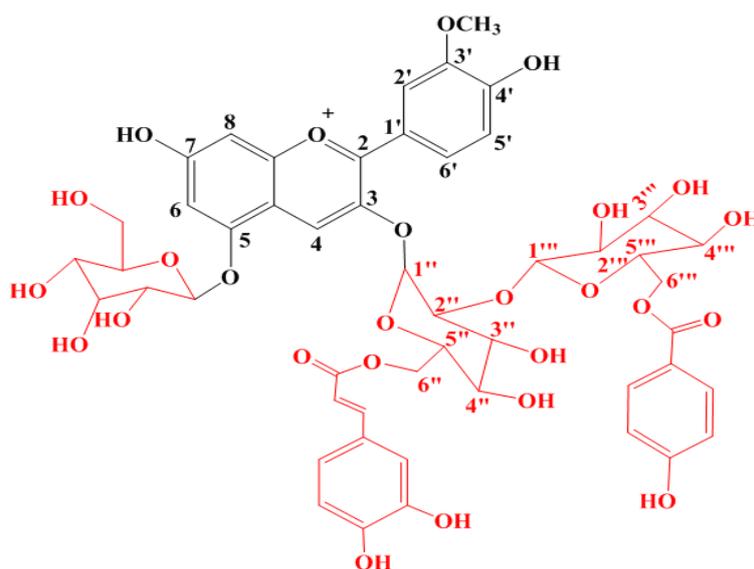


Fig. 21. Structure of compound 7

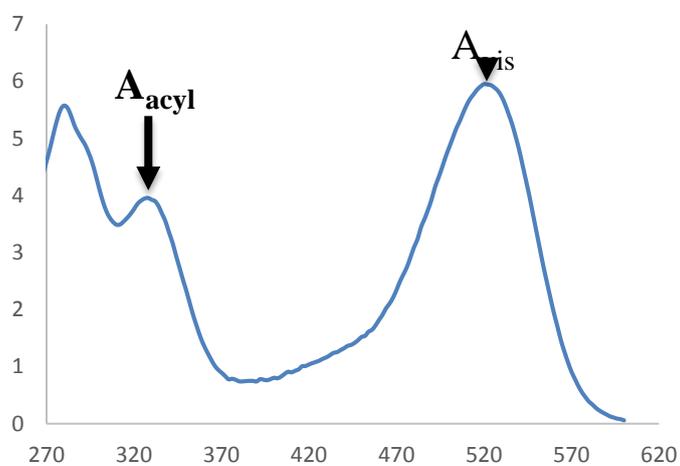


Fig. 22. Visible UV spectrum of compound 9

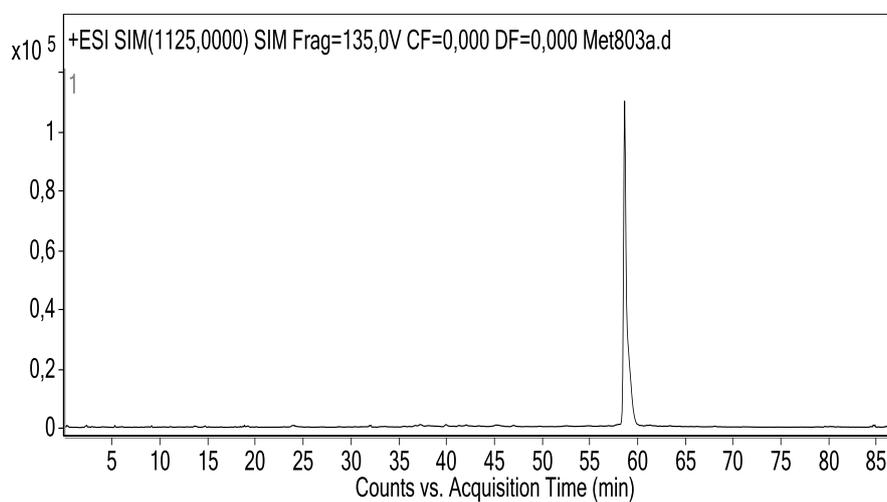


Fig. 23. Chromatogram of compound 9

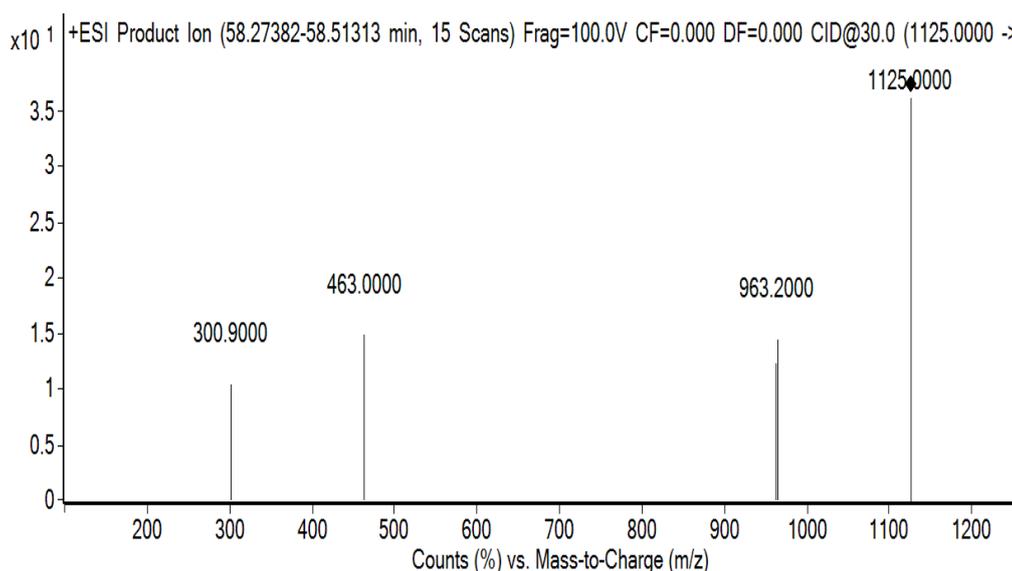


Fig. 24. MS/MS spectrum of compound 9

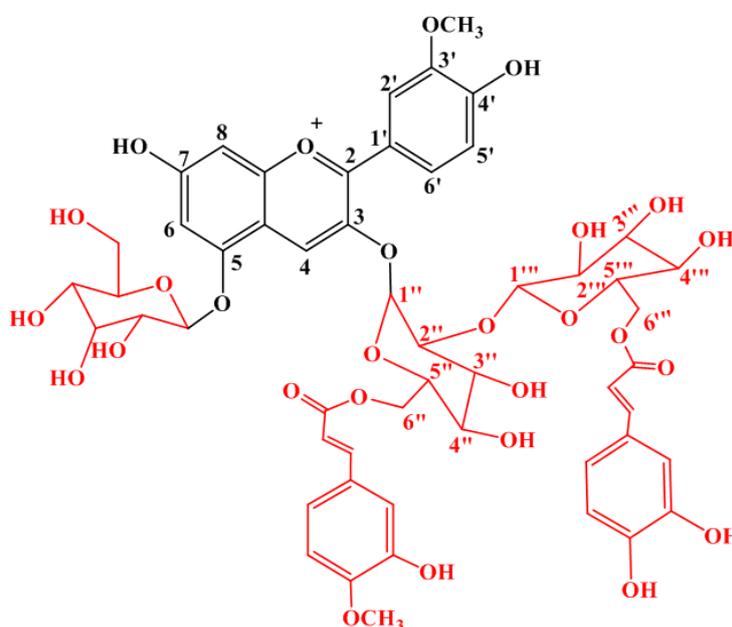


Fig. 25. Structure of compound 9

4. CONCLUSION

From TUSKEGEE POURPRE CAP VERT, a variety of sweet potato produced in a garden in Joseph Ki-Zerbo University, five acylated anthocyanins were identified using successively HPLC-UV-MS and HPLC-ESI-MS/MS. Thirteen anthocyanins (five non-acylated and eight acylated) were identified by HPLC-UV-MS. Among these thirteen anthocyanins; five were analyzed by HPLC-MS/MS. These were cyanidin 3-(6''-caffeoyl-6'''-p-hydroxybenzoyl

sophoroside)-5-glucoside; cyanidin 3-(6''-feruloyl-6'''-caffeoyl sophoroside)-5-glucoside ; peonidin 3-(6'''-caffeoyl sophoroside)-5-glucoside; peonidin 3-(6''-caffeoyl-6'''-p-hydroxybenzoyl sophoroside)-5-glucoside and peonidin 3-(6''-feruloyl-6'''-caffeoyl sophoroside)-5-glucoside. The antioxidant content assessed using TEAC method was estimated to be 0.183 mg TEAC/ g fresh material. This local sweet potato variety could be a potential source of natural antioxidants and beneficial for the health of consumers.

ACKNOWLEDGEMENT

The authors are grateful to the Academy of Research and Higher Education/Commission for Development Cooperation (ARES/CCD) for funding the ARES-CCD Mini PRD No. 3 in which this study was conducted. Also, the authors are grateful to Joseph Ki-ZERBO University, CNRST/INERA and LNSP (National Public Health Laboratory) for conducting this study.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

- Lee MJ, Park JS, Choi DS, Jung MY. Characterization and quantitation of anthocyanins in purple fleshed sweet potatoes cultivated in Korea by HPLC-DAD and HPLCpESI-QTOF-MS/MS. *J. Agric. Food Chem.* 2013;61:3148– 3158.
- Jin-Ge Z, Qian-Qian Y, Ren-Yu X, Jian Z, Yu-Qing Z. Isolation and identification of colourless caffeoyl compounds in purple sweet potato by HPLC-DAD–ESI/MS and their antioxidant activities. *Food Chem.* 2014; 161:22–26.
- Kim HW, Kim JB, Cho SM. Anthocyanin changes in the Korean purple-fleshed sweet potato, Shinzami, as affected by steaming and baking. *Food Chem.* 2012;130:966– 972.
- Jianteng X, Xiaoyu S, Soyoung L, Jason G, Edward C, Benjamin Katz, John Tomich, Scott JS, Weiqun W. Characterisation and stability of anthocyanins in purple-fleshed sweet potato. *Food Chem.* 2015;186:90–96.
- Zhang ZC, Su GH, Luo CL. Effects of anthocyanins from purple sweet potato (*Ipomoea batatas* L. cultivar Eshu No. 8) on the serum uric acid level and xanthine oxidase activity in hyperuricemic mice. *Food Funct.* 2015;6:3045–3055.
- Truong V, Deighton N, Thompson RT, Mcfeeters RF, Dean LO, Pecota KV. Characterization of anthocyanins and anthocyanidins in purple fleshed sweet potatoes by HPLC-DAD/ESIMS/MS. *J. Agric. Food Chem.* 2010;58:404– 410.
- Bovell-Benjamin A. Sweet potato: A review of its past, present, and future role in human nutrition. *Adv Food Nutr Res.* 2007;52:1 59.
- Galan-Vidal CA, Castaneda-Ovando A, Pacheco-Hernandez ML, Paez-Hernandez ME, Rodriguez JA. Chemical studies of anthocyanins. *a Rev. Food Chem.* 2009;113:859–71.
- Gras CC, Nemetz N, Carle R, Schweiggert RM. Anthocyanins from purple sweet potato (*Ipomoea batatas* (L.) Lam.) and their color modulation by the addition of phenolic acids and food-grade phenolic plant extracts. *Food Chem.* 2017;235:265– 27,.
- Suda I, Ishikawa F, Hatakeyama M, Miyawaki M, Kudo T, Hirano K, Ito A, Yamakawa O, Horiuchi S. Intake of purple sweetpotato beverage affects on serum hepatic biomarker levels of healthy adult men with borderline hepatitis. *Eur. J. Clin. Nutr.* 2008;62:60–67.
- Kamiloglu S, Pasli AA, Ozcelik B, Van CJ, Capanoglu E. Colour retention, anthocyanin stability and antioxidant capacity in black carrot (*Daucus carota*) jams and marmalades: effect of processing, storage conditions and *in vitro* gastrointestinal digestion. *J. Funct. Foods.* 2015;13:1– 10..
- Yang Z, Tang C, Zhang J, Zhou Q, Zhang Z. Stability and antioxidant activity of anthocyanins from purple sweet potato (*Ipomoea batatas* L. cultivar Eshu No. 8) subjected to simulated *in vitro* gastrointestinal digestion. *Int. J. Food Sci. Technol.* 2019;1–12.
- Monica GM, Maria PF, Huseyin A, David T, Ivan M. Characterization and Quantitation of Anthocyanins and Other Phenolics in Native Andean Potatoes. *J. Agric. Food Chem.* 2014;62(19): 4408–4416.
- Miller NTJ, Diplock AC, Rice-Evans J, Davies MV, Gopinathan, Milner A. A novel method for measuring antioxidant capacity and its application to monitoring the antioxidant status in premature neonates. *Clin. Sci.* 1993;84:407-412.
- Santiago S, Taihua M, Hongnan S, María C A. Antioxidant activity, nutritional, and phenolic composition of sweet potato leaves as affected by harvesting period. *Int. J. Food Prop.* 2020;178–188.
- Monalisa SCJ, Manuela CPAS, Sidney P, Ana CMGS, Luzimar dSMDn, Renata GB, José GMG, José AAE, Ronoel LOG. Acylated anthocyanins from organic purple-fleshed sweet potato (*Ipomoea batatas* (L.) Lam) produced in Brazil, *Food Sci. Technol.* 2020 ;78(4):1

17. Yeong IR, Inhwan K, Jihyun L. Phenolic Composition and Antioxidant Activity of Purple Sweet Potato (*Ipomoea batatas* (L.) Lam.): Varietal Comparisons and Physical Distribution. *Antioxidants*. 2021;10:462.
18. Zhang JL, Chun LL, Qing Z, Zhang ZC. Isolation and identification of two major acylated anthocyanins from purple sweet potato (*Ipomoea batatas* L. cultivar Eshu No. 8) by UPLC-QTOF-MS/MS and NMR. *International Journal of Food Science and Technology*. 2018;53(8):1932-1941.
19. Qingguo T, Izabela K, Schwartz J, Steven. Probing Anthocyanin Profiles in Purple Sweet Potato Cell Line (*Ipomoea batatas* L. Cv. Ayamurasaki) by High-Performance Liquid Chromatography and Electrospray Ionization Tandem Mass Spectrometry. *J. Agric. Food Chem.* 2005;53:6503–6509
20. Giusti MME, Wrolstad R. Acylated anthocyanins from edible sources and their applications in food systems. *Biochem. Eng. J.* 2003;14:217-225.
21. Pitt JJ. Principes et applications de la chromatographie en phase liquide-spectrométrie de masse en biochimie clinique. *Clin Biochem.* 2009;30(1) :19-34.
22. XIANLI W, RONALD PL. Identification and Characterization of Anthocyanins by High-Performance Liquid Chromatography–Electrospray Ionization–Tandem Mass Spectrometry in Common Foods in the United States: Vegetables, Nuts, and Grains. *J. Agric. Food Chem.* 2005;53:3101–3113.

© 2021 Kaboré et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:

*The peer review history for this paper can be accessed here:
<https://www.sdiarticle4.com/review-history/73800>*