Food Research 6 (4) : 107 - 112 (August 2022)
Journal homepage: https://www.myfoodresearch.com

FOOD RESEARCH

FULL PAPER

Total phenolic compound and its antioxidant activity of by-product from pineapple

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Article history:
Received: 25 June 2021
Received in revised form: 30 July 2021
Accepted: 15 November 2021
Available Online: 14 July 2022

Keywords:
Pineapple,
By-product,
Radical scavenging activity,
Phenolic,
Fuzzy analytical method

DOI:
https://doi.org/10.26656/fr.2017.6(4).453

Abstract

Pineapple is a popular tropical fruit. This work aimed to investigate the total phenolic and total flavonoid content in pineapple by-products including peel, core and pomace. In addition, the antioxidant activities of their extract were determined. The results showed that pineapple peel gave the highest extract yield followed by core and pomace, while the peel and core extract contain a high amount of phenolic and flavonoid. In addition, the highest DPPH radical and ABTS+ radical scavenging activity was pineapple peel extract. The Fuzzy Analytical Method has been performed to evaluate the best value material by using 3 criteria which are composed of extract yield, amount of bioactive compound and its antioxidant activities. The results showed that the peel was the best value material followed by core. This can be pointed out that the pineapple peel could be a choice for food ingredients.

1. Introduction

Nowadays, food waste continuously increases. Viuda -Martos *et al*. (2019) reported that the by-product of fruit and vegetable as well as cereal was one of the main food wastes. Therefore, the reduction of food waste has been increasingly demanded. Many types of research have been reported that the by-products of fruits and vegetables composed of a large number of phenolic compounds contained in waste higher than edible parts (Zhu *et al*., 2019; Tlais *et al*., 2020). Consequently, the bioactive compounds and fibres from food waste can be interestingly extracted.

Pineapple is one of the popular tropical fruits. It has been widely cultivated in Thailand which was the 4th largest cultivated worldwide (Rodsamran and Sothornvit,2019). It has a high nutritional value such as vitamins A, B and C and several minerals and also bioactive compounds (Gardner *et al*., 2000). Due to its nutritional values, it can be produced as value-added compounds for example antioxidants, organic acids, bromelain, and phenolic compounds (Lobo and Yahia, 2016; Mohd Ali *et al*., 2020).

The pineapple can be consumed as fruit and also processed into many products for example canned, juice, jam, jelly and dried product. The high consumption of pineapple results in high pineapple waste such as peel, core, stem and leaves (Ketnawa *et al*., 2012; Rattanapoltee and Kaewkannetra, 2014; Rodsamran and Sothornvit, 2019). The pineapple waste can be used for fibre extraction (Martínez *et al*., 2012), pectin extraction (Rodsamran and Sothornvit, 2019) and bromelain extraction (Campos *et al*., 2020). Rodsamran and Sothornvit (2019) extracted pectin from pineapple peel and reported that the pectin yield was 1.02–2.12%. Martínez *et al*. (2012) reported that the dietary fibre content in pineapple peel from industries was in the range of 69.1 and 81.5 g/100 g dry matter. Campos *et al*. (2020) reported that the enzymatic fractions in stems and peels were observed between 4.8% (w/w) and 17.3% (w/w).

Besides that, many studies reported that the by-products of fruits composted phenolic compounds which can be used as a valuable material for bioactive compound extract. The structure of polyphenols is composed of a number of hydroxyl groups which showed the capability to donate the hydrogen atom and support the unpaired electron, while the glycosylation of a flavonoid would decrease (Cai *et al*., 2004; Zhu *et al*., 2019). Lobo and Yahia (2016) reported that pineapple juice contained mainly phenolic compounds and flavonoids. Campos *et al*. (2020) reported that the phenolic compounds in pineapple peel and stem after juice extraction was in the range of 157.8 – 1270.1 mg GAE/100 g.

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eISSN: 2550-2166
Published by Rynnye Lyan Resources

Fuzzy Analytical Method (FAM) is a mathematical technique that generated criteria into a fuzzy scale [0 to 10] and then integrated the fuzzy grade matrix with their relative weights into the overall index. This overall index can be used for comparison and determination or decision. FAM has been applied in food production for quality control (Perrot *et al*., 2006), for sensory evaluation (Singh *et al*., 2012; Routray and Mishra, 2012; Lasunon and Sengkhamarn, 2016), and for deciding on the best condition for extraction (Tongkham *et al*., 2017; Lasunon *et al*., 2021). Tongkham *et al*. (2017) studied the effect of the pectin extraction condition from the dragon fruit peel and used the value of pectin yield and pectin viscosity as a criterion for the decision of the best condition. In addition, Lasunon *et al*. (2021) studied the effect of bioactive compound extraction condition from tomato waste and used the value using 2 quality criteria which were the values for DPPH radical scavenging activity of hydrophobic and hydrophilic fractions for choosing the best condition.

This study aimed to determine the bioactive compound and its antioxidant activity of the pineapple waste including peel, core and pomace after laboratory juice extraction. The best valuable material for bioactive compound extraction will be evaluated by FAM using 3 criteria.

2. Materials and methods

2.1 Fruit material

The pineapples, Smooth Cayenne, were collected from the same local farm in Nongkhai Province, Thailand. The pineapples were washed, peeled and then cut into the small piece and the core was discarded. The pineapple pomace of its juice extract was collected. The by-product from pineapple juice in which pineapple peel, core and pomace were then soaked in water for 6-8 hrs to remove the small sugars. Then the pineapple peel, core and pomace were dried at 60°C for 24 hrs then ground and kept in a plastic bag for further experiment.

2.2 Bioactive compound extraction

The bioactive compounds of dried pineapple by-product were extracted according to Zhu *et al*. (2019) with some modifications. In brief, the dried sample (10 g) was microwave extracted with 100 mL of absolute ethanol in which 450 W of microwave power for 2 mins. The extraction process was performed twice. After filtration, the extract was then rotary evaporated at 60oC until dry. The dry extracts were kept at 4oC for further determination.

2.3 Total phenolic contents

The total phenolic content of the extract was determined using the Folin-Ciocalteu method as described by Lou *et al*. (2014) with some modifications. Briefly, the extract was dissolved in absolute ethanol at a concentration of 1 mg/mL. An aliquot (1 mL) of extract solution was mixed with 1 mL of Folin-Ciocalteu reagent and left in the mixed solution for 3 mins. The 5 mL of distilled water and 3 mL of Na_2CO_3 (10 g/100 mL) were added to the solution. After 30 mins at room temperature, the absorbance of the mixture at 764 nm was measured. The total phenolic content was expressed as gallic acid equivalent/100 g of extract.

2.4 Total flavonoid contents

The total flavonoid content of the extract was performed by the aluminium chloride colourimetric technique. The 0.5 mL of extract (1 mg/mL in ethanol) was mixed with 0.1 mL of Aluminum chloride (10%), 0.1 mL of Potassium acetate and 4.3 mL of distilled water. After incubation at room temperature for 40 mins, the absorbance at 415 nm was measured. The total flavonoid content was expressed as Quercetin equivalent/100 g of extract.

2.5 Antioxidant activity

2.5.1 DPPH radical scavenging activity

The DPPH radical scavenging activity of the extracts was performed according to Chen *et al*. (2008). In brief, the 1 mL of extract (1 mg/mL in ethanol) was mixed with 2 mL of 95% Ethanol and 2 mL of DPPH in ethanolic. After 30 mins at room temperature in the dark, the absorbance was measured at 517 nm. The free radical scavenging activity was calculated as follows:

$$\% \text{ Scavenging activity} = [(AB-AA)/AB] \times 100$$

When AA was the absorbance of the extract and AB was the absorbance of blank (95% ethanol)

2.5.2 ABTS+ radical scavenging activity

The ABTS+ radical scavenging activity was performed according to An *et al*. (2016) with some modifications. The ABTS+ working solution was prepared by dissolving in 2.45 mM potassium persulfate and allowed to stand in the dark for 12–16 hrs. This solution was further diluted with phosphate buffer (pH 7.4) to reach the absorbance of 0.70±0.02 at 734 nm. The 10 µL of extract (1 mg/mL in ethanol) was mixed with 1 mL of ABTS+ working solution. After 6 mins at room temperature in the dark, the absorbance was measured at 734 nm. Results of the ABTS+ radical scavenging activity were expressed as mg Trolox/g dry extract.

2.5.3 β-carotene bleaching test

The β-carotene bleaching test of the extract was determined according to Derakhshan *et al*. (2018) with some modifications. The β-carotene solution was prepared by dissolving 0.5 mg of β-carotene in 100 µL chloroforms, and then the solution was added with 25 µL linoleic acid, 200 mg Tween-40 and 100 mL of oxygenated distilled water. The 350 µL of extract in ethanolic solution was mixed with 2.5 mL of β-carotene solution and the absorbance of the sample at 490 nm and BHT solution were measured at zero time and after incubation for 48 hrs. The β-carotene bleaching activity of the extracts was calculated as follows:

$$\% \text{ β-carotene bleaching activity} = 100\ [1 - (A_0 - A_t) / (B_0 - B_t)]$$

When A was the absorbance of the extract and B was the absorbance of BHT solution.

2.6 Statistical analysis

The experimental design was done in triplicate and compared using Duncan's New Multiple Range Test at a significance level of $p<0.05$.

2.7 Fuzzy Assessment Method

The most suitable parts of pineapple by-products have been determined using the overall performance index which was calculated with Fuzzy Analytical Method according to Lasunon (2016). The overall performance index was calculated using 3 criteria including the extract yield, bioactive compound content and antioxidant activity of the extract as a weight of 40, 30 and 30, respectively. The results of each criterion were converted to performance scores from 0 to 10 by using the lowest and highest from all values of each criterion. After that, the fuzzy performance grade matrix and the overall performance index were calculated (Tongkham *et al*., 2017).

3. Results and discussion

3.1 Extraction yield

Pineapple is a tropical fruit that contains high nutritional values such as vitamin C, polyphenols and dietary fibre. Due to the high consumption of both fruit and fruit product, hence the by-product from the manufacturing is continuously increased. The by-product from pineapple juice was microwave extracted and the extracted yield was shown in Table 1. The results pointed out that the extracts from Peel were the highest while the core and pomace extract was similar. However, the bioactive compounds in each extract were determined and would be discussed in the next part.

3.2 Total phenolic and total flavonoid contents

Lopes *et al*. (2021) reported that by-products of fruits can be used as a material source for phenolic compounds. Therefore, the total phenolic contents of each extract were determined, and the results have been shown in Table 1.

Table 1. The extract yield, total phenolic content and total flavonoid content in each extract

By product	% Yield	TPC (mg GAE/g dry extract)	TFC (mg Quercetin /g dry extract)
Peel	24.06±1.29[a]	5803.21±1304.60[a]	9067.09±0.43[a]
Core	11.11±0.57[b]	1543.51±50.21[b]	8725.21±195.84[a]
Pomace	11.01±0.40[b]	598.39±21.25[b]	3267.95±46.23[b]

Values are presented as mean±SD. Values with different superscripts within the same column are significantly different ($p<0.05$).

The results (Table 1) showed that pineapple peel extract contained the highest amount (5803.21 mg GAE/ g dry extract) of phenolic compound followed by core extract and pomace extract which was 1543.51 and 598.39 mg GAE/g dry extract, respectively. Furthermore, the level of phenolic content in peel exhibited approximately 3.76-folds higher than that found in the core. Considering the flavonoid content, the results exhibited that the highest flavonoid content was found in pineapple peel extract. Campos *et al*. (2020) reported that the pineapple peel extracts contained total phenolic content higher than pineapple stem extracts. Derakhshan *et al*. (2018) compared the total phenolic content from pomegranate peel, seed and juice and they found that pomegranate peel extract showed the highest levels of total phenolic content. Moreover, Tlais *et al*. (2020) reported that bioactive compound such as polyphenols in fruit and vegetable has been found higher in the waste than in their edible parts.

3.3 Antioxidant activity

The phenolic compound and flavonoids have been reported to be effective in inhibiting the occurrence of oxidation. Therefore, the antioxidant activities of extracts were determined using DPPH, ABTS+ and β-carotene bleaching test and the results are shown in Table 2.

The DPPH radical scavenging activity assay was a general tool for estimating the free radical-scavenging activity of antioxidants. This assay determined the radical scavenging of the extract on the DPPH radical which is a stable free radical. The results (Table 2) showed that the DPPH radical scavenging activity of the peel extract and the core extract was significantly similar and higher than pomace extract for approximately 3.44-folds. This was probably due to the bioactive compound

in their extract. The peel and core extract showed higher the total phenolic compound and total flavonoid content than in pomace extract. The phenolic molecule is composed of the number and position of hydroxyl groups particularly the position of hydroxyl groups. This phenolic structure showed the hydrogen atom denoting abilities and unpaired electron supporting capacities hence supporting the phenolic compound to exhibit its antioxidant capacity (Zhu *et al*., 2019).

Table 2. The antioxidant activity of the extracts

By product	DPPH (%)	ABTS (mg Trolox/g dry extract)	β-carotene blanching (%)
Peel	93.12±0.43[a]	3.19±0.02[a]	5.74±0.10[a]
Core	93.22±3.11[a]	3.07±0.01[b]	5.62±0.02[ab]
Pomace	27.03±1.18[b]	3.04±0.01[b]	5.51±0.03[b]

Values are presented as mean±SD. Values with different superscripts within the same column are significantly different ($p<0.05$).

The ABTS+ radical scavenging activity was performed in order to determine the ability of extract in the donation of unpaired electrons on ABTS+ radical. The results (Table 2) exhibited that the peel extract was the highest ABTS+ radical scavenging activity. While the ABTS+ radical scavenging activity of core and pomace extract was similar. This was probably due to the type of phenolic and flavonoid compounds. The high number of hydroxyl groups of polyphenols promoted the radical scavenging activity. However, the reaction was random and depended on structure and interaction between molecules (Dias *et al*., 2019; Rudke *et al*., 2021).

The β-carotene bleaching test has been performed to determine the inhibition of the oxidation activity of the extract in the food system. The mixture of the emulsion was prepared and the β-carotene bleaching of the emulsion was measured. The results showed that the peel extract was the highest percentage of β-carotene bleaching while the core and pomace were slightly different. However, the percentage of β-carotene bleaching in all extracts was quite low value.

3.4 Fuzzy Assessment Method

The fuzzy Analytical Method is a mathematical technique that calculated the overall performance index to evaluate the valuable material for bioactive compound extract. The overall performance was calculated using a triangular fuzzy grade which considered 3 criteria consisting of extract yield, bioactive compound content and antioxidant activity of the extract.

Each result was converted into a performance score from 0 to 10 by using the lowest and highest value as lower and upper bounds, respectively, for each of the results. The weighing value has been decided to be 40, 30 and 30 for extract yield, bioactive compound content and antioxidant activity of the extract, respectively. In the current study, two bioactive compounds, phenolic and flavonoid were weighed as equal which was 15 each. Moreover, the antioxidant activity of extract has been measured in three different assays comprising DPPH radical scavenging activity, the ABTS+ radical scavenging activity and β-carotene bleaching test and the weighting value was equal which was 15 for each. The overall performance index was calculated using a SciLab Software Package developed by Lasunon (2016) and the results were shown in Table 3.

Table 3. Overall Performance Index of the extracts

By product	Overall Performance Index
Peel	8.545±0.337
Core	5.071±1.056
Pomace	0.500±0.144

The results showed that the first valuable material for bioactive compound extract was pineapple peel and followed by core and pomace, respectively. The overall performance index of peel extract was higher than core extract for 1.7-folds and that of pomace extract was the lowest. This was in accordance with Tlais *et al*. (2020) who reported that the bioactive compound of fruit was found lower in the edible parts than in their waste.

4. Conclusion

To conclude, the more pineapples consumed, the more waste was produced. The bioactive compound in the by-product has been extracted using microwave-assisted extraction and it was found that the pineapple peel gave the highest extract yield in which it contained a high amount of phenolic and flavonoid content. Additionally, the antioxidant activities of its extract also pointed out that pineapple peel exhibited high antioxidation, particularly as the DPPH radical scavenging activity. A fuzzy analytical method has been performed to evaluate the best valuable by-product. The overall performance index proved that the pineapple peel was the best and higher in content than the pineapple core for 1.7-folds.

Conflict of interest

The authors declare no conflict of interest.

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