MICROWAVE ASSISTED EXTRACTION OF TOTAL PHENOLIC COMPOUND FROM PINEAPPLE PEEL AND CORE

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Abstract— **A microwave assisted extraction system was used to extract total phenolic compound (TPC) from pineapple peel and core. The effect of microwave power (180,300,450W) and extraction time (2 and 3min) were investigated. Pineapple peel and core were extracted with ethanol solvent. The highest extraction time for peel and core were 3min(180W) and 2min (300W). Meanwhile, the optimum microwave power obtain for both peel and core was 300W. The analysis was determined by using UV spectrophotometer at 760nm and the result was compared with standard curve of Gallic acid equivalent (mg/L). The results demonstrated that MAE could be a fast and reliable method for quantitative analysis of phenolic compounds in pineapple peel and core.**

Keywords— **Microwave-assisted extraction, phenolic compound, antioxidant activity, pineapple peel and core, ananas comosus**

I. INTRODUCTION

Ananas comosus is the scientific name for pineapple that normally grown in several tropical and subtropical countries include Philippines, Thailand, Indonesia, Malaysia and China (Pavan *et al.*, 2012). Pineapple one of the most popular agricultural fruit in tropical and subtropical because of its appealing flavor and stimulating sugar-acid balance(Bartolome *et al.*, 1995). For worldwide production, after banana and mangoes, pineapple production is currently the third most important tropical fruit (Moyle *et al.*,2004). However, based on data of 2014, Malaysian has ranked to 15th position as a major producer of pineapple fruit in the world. Yet, it remains as a major fruit crop in Malaysia, amongst others such as mango, watermelon, papaya, banana and pomelo. The main force for planning and encouraging pineapple industry planting and its downstream industry is MPIB (Amar *et al.,*2015).

Through the processing of pineapple, the crown and stem are cut off before peeling. Then, for advance processing the core being removed and all this waste of peel, core, stem and leaves mostly present for 50% (w/w) of total pineapple weight. Hence, this give out of pineapple waste proportionally as increasing of pineapple production (Ketnawa *et al.*,2012). Only 40% being used as juice and the other 60% will go as waste disposed for composting or dumped in landfill of open site. It is usually in wet condition that useless and not suitable to dry and worthless to dry in order to use the fibrous residues as boiler fuel even inapplicable as cattle feed, because can cause diarrhea. Thus, this solid waste part can cause problem and impact the environment when being disposed to environment. In order to reduce the solid waste, value addition for these solid waste done by extracting the phenolic compound.

It is expected that throw away fruit and waste material can be useful for futher processing in industry like fermentation, bioactive compound extraction and etc. There have been various industry on application of waste gained from the fruit and vegetable, dairy and meat industry. Since the pineapple waste are beneficial source for sugars, vitamins, and factors of growth, it have been used as substrate for bromelain, organis acids, ethanol and etc (Upadhyay *et al.*, 2010). The solid waste of peel, core, leaves and crown of this pineapple can be useful in medical treatment if the bioactive compound of the phenolic compound being extracted.

Phenolic compound are the secondary metabolites which has been broadly spread throughout the plant kingdom(Pinela *et al.*, 2016). Generally, phenolic of antioxidant activity is split into two category: primary and secondary antioxidants. Primary antioxidants serve to stop chain reaction by donation of electron or hydrogen to free radical to form a stable and steady compound. Meanwhile, metal chelation can prevent the beginning of free radical chain reaction for secondary antioxidants (Yuris & Siow, 2014). It has been found that the antioxidant activity can slowing down the ageing process and lower the risks of many chronic disease, cancer and coronary heart disease(Li *et al.*, 2012). This has been supported by Bhuyan et al., (2015) stated that flavonoids and phenolics potency agent for therapeutic values and antimutagenic properties, cardioprotective and anticarcinogenic. Besides that, the phenolic compound establish to inhibit human immunodeficiency viral replication (HIV), human simplex virus (HSV), glucosyl transferases of Streptococcus mutans (dental carries), ascorbate auto-oxidation (green tea), cytotoxic effects, tumor promotion and xanthine and monoamine oxidases (Nayak *et al.*, 2015).

Therefore, inoder to fight any disease is by improve our body's antioxidant defenses. People can lower the incidence likes degenerative disease by high consumption of fruits and vegetables. Fruit likes pineapple also can improve our health for example relieve the sore throat and seasickness as primary due to the antioxidant activity in fruit (Hossain & Rahman, 2011). This antioxidant of phenolic compound plays an important role in fight the disease of human pathogen, thus it can be apply in nutraceutical, pharmaceutical and cosmetiucal industries (Pinela *et al.*, 2016).

There are numerous of technique that can be utilized for phenolic compound extraction and this technique separated into traditional and modern. Therefore, previous study shown that the conventional method that usually apply to extract phenolic content include soxhlet extraction, maceration and maceration assisted with stirring (Ćujić *et al.*, 2016). However, this conventional method have limitations for example low yield of extraction, need high temperature, use of large volume of solvents, not efficient, mass transfer resistance and risk to health (Safdar *et al.*, 2016). As the procedure, it need certain period of time with the high temperature for extraction by strring the solvent consist of sample (Albuquerque *et al.*, 2016).

Microwave assisted extraction (MAE) recently is one of the most valuable extraction method for bioactive compound. In this era, technologies focus on discovery the technique that can produce high yield, low consumption of solvent and short extraction time. As MAE have many advantages, it has been used in many industry like pharmaceutical and food industry. Study by Simic *et al*. (2016) stated that MAE only need shorter extraction time, low cost, and according to environmental standards it can be considered as "green" technologies.

The objectives of the current study were to investigate the effects of MAE on the extraction efficiency and recovery of phenolic compound of antioxidant from pineapple peeal and core and to optimize the extraction process. Optimisation parameter included microwave power and extraction time.

II. METHODOLOGY

A. *Chemicals and standards*

Ethanol used for extraction solvent, Folin-Ciocalteu reagent and sodium carbonate solution were used for absorbance measurement via UV-visible specthrophotometer.

Sodium hypochloride used for washing material, hydrochloric acid aqueous solution for pH adjustment, and Gallic acid standard solution for standard curve.

B. *Plant Materials*

The pineapple fruit that obtained from local markets in Shah Alam is washed with sodium hypochloride for 5 min. It is to remove the debris and dust. Then, it is cut into quarters and then the stem is removed. The peels and core are taken and finely cut with a sharp knife. The cut segment is placed on stainless strays and dried in hot oven 50◦C to constant weight obtained. The final product is in the form of powder where the dried peel need to ground and sieve it by using 40-mesh sieve size. After that, it can be stored in dark bags and ensure it was in a dry environment prior to experimental analysis.

C. *Microwave assisted extraction (MAE)*

After preparation of raw material, the desired amount of ground fruit, 1.5 g is soaked in a solvent ethanol for 2 hr in a beaker. The pH is adjusted to 2.5 by addition of hydrochloric acid aqueous solution and then transferred and subjected to a microwave heating. The solutions are extracted with three powers of 180, 300, 450W at different extraction time of 2min and 3min. The mixture of solution is allowed to cool down at room temperature (25◦C), filtered using filter paper, and then it being centrifuged (10 000 x g for 15 min). After centrifugation, the supernatant is carefully collected for futher analysis.

D. *UV-Spectrohotometer analysis*

By using UV Spectrophotometer, the analysis to determine the total phenolic compound in the pineapple peel and core are carried out. Firstly, 0.2mL of extract supernatant transferred into 10mL volumetric flask. Then, 0.5mL of Folin- Ciocalteu reagent and 1.0mL of saturated Na_2CO_3 solution are added into the flask. The Na_2CO_3 with 35g is dissolve in 100mL of water. The total mixture is increase up to 10mL using the distilled water. The final mixture is maintained in the dark place for 30min before take for absorbance measurement via UV – visible spectrophotometer. It is calculated based on gallic acid equivalents (TAE) per gram of sample.

E. *Gallic Acid Standard Curve*

Gallic acid standard solutions are prepared at 0.020, 0.035, 0.050, 0.065, 0.110, 0.125 mg/L . The procedure of analysis was repeated inorder to obtain gallic acid standard. .Then, the comparison is make to determined the concentration of total phenolic compound where the absorbance obtain for extract sample is compared with gallic acid standard solutions. Total phenolic content (TPC) was expressed as mg gallic acid equivalents (GAE) per g dry sample (Ballard *et al*., 2010)

III. RESULTS AND DISCUSSION

A. *The Extraction Time of Peel on Total Phenolic Compound*

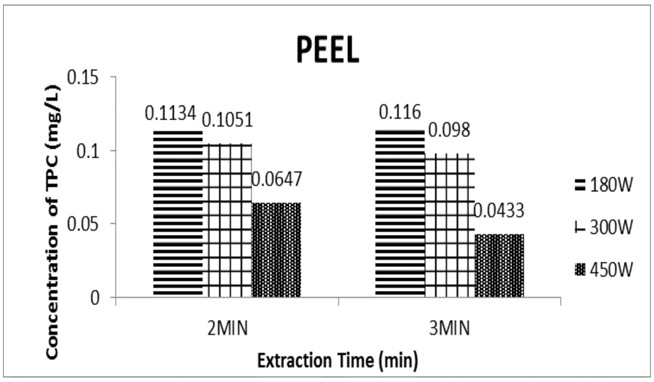

Fig. 1: The comparison of extraction TPC in peel at 2min and 3 min

Table 1: Comparison of peel between time of 2min and 3 min

Peel	2 min	3 min
180 W	0.1134	0.1160
300 W	0.1051	0.0980
450 W	0.0647	0.0433

From Figure 1, it shown that total phenolic compound concentration obtain from peel has the highest value at 3 min with microwave power of 180W. The concentration gained was 0.116mg/L. The trend of graph showed the similarity for 2min and 3min as the increase in microwave power will decrease the total phenolic compound extraction. If being compared for 2 min and 3 min, the extraction of TPC at 2min has higher TPC extraction for 300W (0.1051mg/L) and 450W (0.0647mg/L). This can be explained based on research by Ahmad & Langrish (2012) stated that with an increase in extraction time, the extent of antioxidant degradation increase. Prolonged time degrades the already extracted antioxidant instead of doing more extraction.

Besides that, the prolonged extraction will contribute to decrease in total phenolic compounds by lengthen the exposure to environment factors like light and oxygen. When using the conventional extraction process that carried out at 30min, 60min, 90min and 120min, the optimum condition gained at 30 min to extract the TPC. After 30min, the increase in extraction time will not affect or does not develop the extraction yield. This has been explained from Fick's second law diffusion which expressed that after a certain time, it will be a final equilibrium between extraction solvent and the plant sample where in this case the peel of pineapple (Benmeziane et.al, 2014)

Dahmoune et al (2014) also reported that efficiency of extraction will increase as the time of extraction was increase from 30 to 60/90s and then significantly decrease after 120s. This happen because probably without temperature control as exposed to longer irradiation induced thermal degradation of phenolic compounds. Besides that, 30-90s range is favourable compared to 120s as it reduces the energy cost. This explained why the highest extraction time obtained was 3min but at lower microwave power (180W). This is because when extraction happens at lower microwave power, the plant matrix exposed at low temperature gradient and need more time to extract the TPC.

B. *The Extraction Time of Core on Total Phenolic Compound*

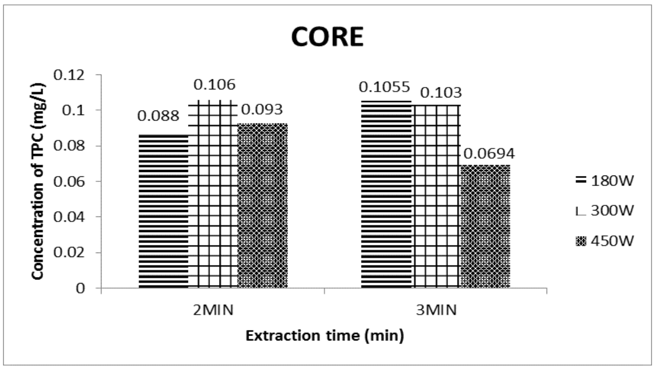

Fig 2 :The comparison of extraction TPC in core at 2min and 3 min

Table 2 :Comparison of Core between time 2 min and 3 min

Core	2 min	3 min
180 W	0.088	0.1055
300 W	0.106	0.103
450 W	0.093	0.0694

From Figure 2, the highest TPC extraction was gained at 2 min (300W) with concentration of 0.1060 mg/L. However, at 3min (180W) the result shown at highest extraction of 0.1055mg/L with 99.5% similar to result of 2min (180W). This related with the level of microwave power where lower in microwave power, higher extraction time is needed. This occurrence happen because the mass transfer at low rate with low temperature, therefore it needed more time for this phenolic compound to be dissolve in raw meterials into solution(Li et al., 2012). The research stated that at high level of temperature, it does not affected by the changes in time of extraction as the phenolic compound dissolution can achieve the equilibrium in a shorter time. As tomato use as the raw material that being extracts, it shown that higher microwave power and short extraction time are more effective in extracting the phenolic compound. It is related to the research study as the core of pineapple shown the highest extraction of 2min at with higher microwave power, 300W.

The difference in raw material to extract the total phenolic compound must be considered. Compared to Figure 1, core of pineapple needed higher microwave power (300W) but at shorter time (2min) inorder to extract highest total phenolic compound. Throughout MAE process, microwave diffuse within extraction mixtures contain of solvent and plant particles, then with certain moisture content that contain in the vacuole. Inside the vacuole, it's become heated after the intracellular moisture of the plant particle absorb with a portion of incident microwave energy referable to the favorable dielectric properties and vaporized to generate the internal pressure within cell vacuoles. After that, the membrane surrounding the vacuole will stretch and expand, finally get to the cell wall. Ahead that, the cell wall will expands together and as long as the cell wall of plant remains intact, the membrane of the vacuole eventually may or may not break. The bioactive compound will be release into surrounding solvent when the internal pressure surpass the strength of cell wall cause plant cell rupture (Chan et.al, 2016).

At extraction time of 2min, the result indicated that from 180W to 300W, the extraction of phenolic compound were increase and then decrease back at 450W. At 3 min, the graph showed the decrease in trend as microwave power increases. During 180W to 300W at 2min, the release of intracellular product happen by broken down the cell wall due to distruption of hydrogen bonds that being expressed from microwave induced dipole rotation of molecule and migration of ions which has enhanced the penetration of the solvent into the matrix, lastly allowing the compound to be extract. However, the decrease of component yield at 2min (450W) must be due to the over-consumption of energy (Phongthai et al,. 2016). Meanwhile, as the level of microwave power increase with extending extraction time, 3min causes the thermal degradation of phenolic compounds (Ballard et al., 2010)

C. The Effect of Microwave Power on Peel and Core

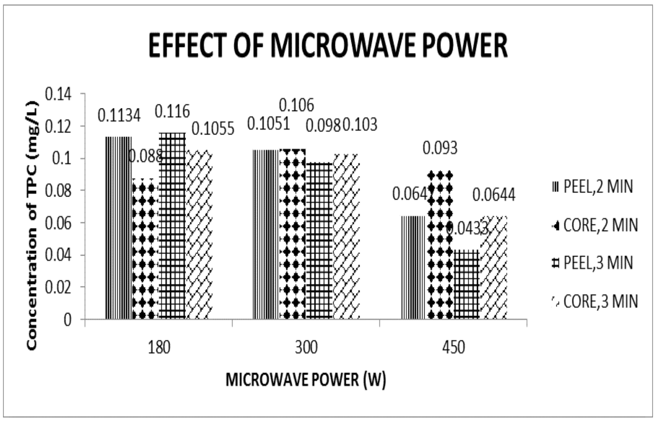

Figure 3: Effect of Microwave Power, 180W,300W , 450W

From Figure 3, the result shown that the highest extraction point for peel and core at 180W and the lowest extraction at 450W, meanwhile the optimum condition expressed at 300W. For microwave power 180W and 450W, the trend of graph was not consistent compared to 300W. Total phenolic compound extracted from apple pomace shown that decomposition of extracted components happened when higher absorbance of microwave power by polar molecule result in increase of temperature. Razaei et.al (2013) stated that inorder to extract this phenolic compound from apple pomace, the lower microwave power is more suitable and efficient as extreme heat will result in deterioration of part phenolic compound and cause less TPC obtained.

It is similar with our result as the optimum condition show at lower microwave power of 300W. Eventhough 180W of microwave power is the lowest power, the optimum condition shown at 300W can be explained from study by John Swamy & Muthukumarappan (2016) that increase microwave power level can soften the tissue of plant and cut down the protein/ carbohydrate interface and also the phenolic compound. Therefore, the solubility of the phenolic compounds become increase. This situation also has been approved by Pinela et al. (2016) that MAE at higher power will increase the temperature thus it improve efficiency of extraction by raise the desorption antioxidants from active site in the tomato matrix, and respectively recover sample wetting and penetration of matrix because the reduce in surface tension also solvent viscosity.

The microwave power powerfully dependent on time and extraction temperature. Experimental study by Elez Garofulić et al. (2013) for extraction of phenolic acid from sour cherry Marasca has obtain the optimum condition at 400W. The experiment has been conducted at 350W, 400W and 500W. The result obtain explained that as a general, the high temperature can improve extraction yield, but it also can reduce extraction selectivity as non-targeted compounds and matrix materials also be extracted. The thermal degradation can also be occurring at elevated temperature as well as longer extraction times.

IV. CONCLUSION

From the result obtained, it have demonstrated that total phenolic compound can be extracted from pineapple peel and core using microwave assisted extraction process. Besides that, this MAE process can produce high yield, low consumption of solvent and short extraction time compared to conventional method. From the results, the total phenolic compound for peel obtain highest at 3min with 180W and core was 2min at 300W. This shown that when increase in microwave power, the extraction time was decrease. Meanwhile, the optimum microwave power for both peel and core can be seen at 300W where all the parameter shown to be consistent at that point compared to others.

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