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Effect of temperature and extraction time on quality of pectin from arabica coffee pulp

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ABSTRACT

Coffee is one of the largest commodities in the world. With coffee production more industrialized, waste generated a huge environmental problem. Coffee pulp is the main by-product of coffee processing. One of the components of coffee pulp is pectin. Pectin production from coffee pulp and its characteristics should be studied since extraction conditions used in production vary depending on raw material, desired type of pectin, and process economics. This research aimed to study the characteristics of pectin from coffee pulp extracted using oxalic acid under different times (80 and 90 minutes) and temperatures (100°C and 120°C). The parameters analyzed were yield, equivalent weight, water content, ash content, methoxyl, galacturonic acid, and degree of esterification. The result showed that extraction time and temperature affect the characteristics of pectin. The highest yield was obtained by extraction at 90°C for 90 minutes. The coffee pectin has colored light brown and is categorized as low methoxyl pectin. Overall pectin coffee pulp produced in this study has met the quality requirements of dried pectin by the International Pectin Producer Association (IPPA) standard.

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INTRODUCTION

Coffee is a very popular beverage that is consumed worldwide. As the demand for coffee increases, coffee productivity in Indonesia also increases. In 2021, BPS (2020) reported that Indonesia produced 774,60 thousand tonnes of coffee, an increase of circa 1.62% from the previous year. A coffee bean is produced by separating the bean from the coffee pulp (Harahap et al. 2022).

Pulping is the main stage in green bean processing, especially in the wet or semi-wet process method. To obtain the seeds (beans), the dried cherry coffee's exocarp, mesocarp, and endocarp must be removed by hulling, originating the so-called husk. In the wet processing procedure, the fruits are pulped, fermented, washed, and sun or artificially - dried; the pulping procedure removes the exocarp and most of the mesocarp, resulting in the so-called coffee pulp. Therefore, husk and pulp are the main by-products of coffee processing waste, and by-products of the coffee amount in the millions of tons in the world in terms of production (Mazzafera 2002; Bouafou 2011; Muzaifa et al. 2021). Then Ernawati et al. (2018) also added that during pulping process, 100 kg of coffee cherries produce 43,2 kg of the coffee pulp as the by-product.

As the most waste product in coffee processing, an effort is needed to utilize coffee pulp. With its high carbohydrate content, the coffee pulp can potentially be used as a raw material for pectin production. Pectin is a plant hydrocolloid found in the primary cell wall and middle lamella in all dicotyledonous plants, more commonly in the outer shell or skin than in the inner matrix of plants (Chandel et al. 2022). Pectin can be converted into hydrogels and forms a network of flexible polymer chains. It has a complex structural formed by homogalacturonan, rhamnogalacturonan, xylogalacturonan (Ma et al. 2020; Rodsamran et al. 2019).

Pectin is one of the structural carbohydrates present in plants. It is probably one of the most complex macromolecules found in nature. Pectin is commonly used as gelling, thickening, and emulsifying agent in various applications, from food to pharmaceutical products (Tsoga et al. 2004; Liu et al. 2013; Kaya et al. 2014). But nowadays, the large number of functional groups in the pectin structure can trigger different functions, allowing pectin to have various applications. Currently, the study of pectin mainly addresses its applications in food, agriculture, medicine, and biomedicine, with trends in the production of edible food coatings, bio-based antimicrobial films, and nanoparticles for studies in cancer treatment; in wound healing; and sanitary napkins by the pharmaceutical industry (Freitas et al. 2021).

Commercial pectin is usually extracted from apples and citrus fruits. However, recent research is starting to extract pectin from various industrial by-products, presenting itself as a green option for valorizing agro-industrial residues, in line with the circular economy concept. The coffee pulp should be considered as another source of commercial pectin. Coffee pulp represents 45% of the total weight of the coffee cherry (Muzaifa et al. 2021).

Pectin production from coffee pulp and its characteristics should be studied since extraction conditions used in production vary depending on raw material, desired type of pectin, and process economics. Pectin is usually extracted from raw materials by aqueous extraction, and common methods include direct boiling, microwave heating, ultrasonic, autoclaving, and electromagnetic induction (Chandel et al. 2022). Freitas et al. (2021) emphasized that on an industrial scale, acid extraction and alcohol precipitation are commonly used to extract pectin commercially. Pectin acid extraction is based on the fact that protopectin hydrolysis occurs at higher temperatures. The pectin extracted with organic acid has highly methoxylated based on the analysis of the degree of methyl esterification. Based on the degree of methyl esterification, pectin is classified as high methoxyl pectin or high ester (if the degree of esterification is greater than 50%) and as low ester or low methoxyl pectin (if the degree of esterification is less than 50%).

Studies on the extraction of pectin from coffee pulp and its quality are still rare. Chandel et al. (2022) stated that the pectin quality varies due to the influence of extraction conditions such as time, extraction temperature, pH, and raw materials.Extraction time is very important to extract pectin; if the extraction process is extended for a long time, high temperatures can destroy pectin content (Vo and Luong 2010). An extraction condition is critical in obtaining high pectin yields without compromising the desired quality. This research aimed to study the characteristics of pectin from the coffee pulp at different times and temperatures. Therefore this study aims to study the characteristics of pectin from the coffee pulp at different times and temperatures.

MATERIALS METHODS

Sample Preparation

Fresh coffee pulp obtained from a coffee industry in Gayo Highland Takengon, Aceh, Indonesia. The fresh pulp was washed and immersed in 0,1% sodium metabisulphite to prevent a browning reaction. The coffee pulp was sun-dried for 8 h, and the dried pulp was ground using a grinder into powder (60 mesh). The powdered coffee pulp was stored in an air-tight vacuum container before pectin production.

Statistical Methods

This study used one-way ANOVA with extraction condition as an independent factor with four levels of treatments. T1= extraction temperature 80^{o}C with 90 minutes of extraction time; T2 = extraction temperature 80^{o}C with 120 minutes of extraction time; T3 = extraction temperature 90^{o}C with 90 minutes of extraction time and T4 = extraction temperature 80^{o}C with 120 minutes of extraction time. All treatments were replicated 3 times, as in total, we had 12 samples. The dependent factors were pectin yield (%), moisture contents, ash (%), pectin equivalency weight (mg), methoxyl contents (%), galacturonate acids contents (%), and degree of esterification (%). Suppose the measured dependent factors showed a significant result of Anova, the Least Significance Difference (LSD) performed for further posthoc test. The statistical analysis was performed by IBM-SPSS 23.

Pectin Extraction Methods

The procedure followed Ramli and Asmawati (2011) method with slight modifications. The extraction conditions were applied using ammonium oxalate with a rotary vacuum evaporator. Coffee pulp (50 g sample) was placed onto a 1000 ml beaker glass and stirred with 500 ml of ammonium oxalate solution with 2.5% concentration until homogenized. Then 50 g of ammonium oxalate with 10% concentration was added separately and stirred until the pH reached 3.2. This comparison was applied for each treatment. The extraction occurred by heating the beaker glass in the waterbath at a specific temperature (80 and 90 ^{O}C) and designed time (90 and 120 minutes). After extraction, the extract was separated from the solid residue by filtration through a muslin cloth and squeezed until the liquid came out as much as possible. The residue was re-extracted again with a similar procedure to obtain residual pectin.

Then the filtrates of both extraction processes were mixed. Pectin was precipitated by adding ethanol 96% and left for 18 hours at room temperature. To separate the precipitate of pectin, the mixed sludge of pectin and ethanol was filtered with Whitman filter paper. Pectin was purified and washed eight times with 1000 ml ethanol 96%. Pectin purification validity was done by dripping 10% of AgNO3. The acid purification is successful if any white coagulation is present in the pectin solution. Then, the purified coffee pulp pectin solution was dried in an oven at 45 ^{O}C for 5 h. Before further analysis, the coffee pulp pectin was stored in an air-tight vacuum container.

Pectin Analysis and Characterization

Pectin from the coffee pulp was analyzed for moisture and ash content by oven method (AOAC 2005), yields, equivalent weight, methoxyl content, galacturonic acid (Owens and Albany 1952), anddegree of esterification was calculated from the observed value of methoxyl content and anhydrogalacturonic acid as described by Schultz (1965).

RESULTS AND DISCUSSION

The Yield of Coffee Pulp Pectin

The yield of pectin from coffee pulp varied from 12.03% to 37.28%, with an average yield is 23.80%. The value of each treatment is presented in Table 1. One-way ANOVA shows that extraction condition (temperature and time) significantly influences pectin's obtained yield, as illustrated in Figure 1. Figure 1 shows a post hoc test with LSD for four treatments of extraction condition.

The highest yield is obtained from the third extraction condition, and pectin is extracted at 90^{o}C for 90 minutes. This third treatment is significantly different from the other three treatments. The first treatment, where the coffee pulp is extracted at 80^{o}C for 90 minutes, produces the lowest yield, significantly different from other treatments. As for prolonged extraction time of up to 120 minutes, the yields were not significantly different, whether for lower or higher extraction temperatures.

Table 1 Mean and standard deviation of pectin characterization parameters (n=3)

Measured Parameters and Treatments		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Min.	Max.
						Lower	Upper		
Yield	80oC, 90 min	3	14.9867	3.98616	2.30141	5.0845	24.8888	12.03	19.52
	80oC, 120 min	3	24.9767	5.65035	3.26223	10.9404	39.0129	18.76	29.80
	90oC, 90 min	3	32.6833	5.12955	2.96154	19.9408	45.4258	27.15	37.28
	90oC, 120 min	3	22.6333	3.57799	2.06576	13.7451	31.5216	18.51	24.92
	Total	12	23.8200	7.69804	2.22223	18.9289	28.7111	12.03	37.28
Moisture	80oC, 90 min	3	5.9700	.81166	.46861	3.9537	7.9863	5.13	6.75
	80oC, 120 min	3	6.9000	.64815	.37421	5.2899	8.5101	6.19	7.46
	90oC, 90 min	3	7.8233	.17616	.10171	7.3857	8.2609	7.62	7.93
	90oC, 120 min	3	6.2733	2.05656	1.18735	1.1646	11.3821	3.90	7.53
	Total	12	6.7417	1.23245	.35578	5.9586	7.5247	3.90	7.93
Ash	80oC, 90 min	3	.9333	.00577	.00333	.9190	.9477	.93	.94
	80oC, 120 min	3	.9367	.00577	.00333	.9223	.9510	.93	.94
	90oC, 90 min	3	.9267	.02309	.01333	.8693	.9840	.90	.94
	90oC, 120 min	3	.9400	.01000	.00577	.9152	.9648	.93	.95
	Total	12	.9342	.01240	.00358	.9263	.9420	.90	.95
Metoxyl_ Contents	80oC, 90 min	3	4.1467	.43616	.25182	3.0632	5.2301	3.88	4.65
	80oC, 120 min	3	6.4267	.09713	.05608	6.1854	6.6679	6.32	6.51
	90oC, 90 min	3	7.5233	.18771	.10837	7.0570	7.9896	7.32	7.69
	90oC, 120 min	3	6.4600	.32970	.19035	5.6410	7.2790	6.08	6.67
	Total	12	6.1392	1.31086	.37841	5.3063	6.9720	3.88	7.69
Galacturonate_Acid	80oC, 90 min	3	44.9400	2.04051	1.17809	39.8711	50.0089	42.77	46.82
	80oC, 120 min	3	65.7667	.71220	.41119	63.9975	67.5359	65.12	66.53
	90oC, 90 min	3	69.7533	1.63087	.94158	65.7020	73.8046	67.94	71.10
	90oC, 120 min	3	60.7767	1.59544	.92113	56.8134	64.7400	58.96	61.95
	Total	12	60.3092	9.93603	2.86828	53.9961	66.6222	42.77	71.10
Esterification_ Degree	80oC, 90 min	3	52.3233	3.72390	2.14999	43.0727	61.5740	49.08	56.39
	80oC, 120 min	3	55.4767	.34588	.19969	54.6175	56.3359	55.10	55.78
	90oC, 90 min	3	61.2400	.90703	.52367	58.9868	63.4932	60.37	62.18
	90oC, 120 min	3	60.3967	4.02731	2.32517	50.3923	70.4011	56.20	64.23
	Total	12	57.3592	4.48867	1.29577	54.5072	60.2111	49.08	64.23

Figure 1 showed a tendency that a higher temperature (90^{O}C) caused a higher percentage of yield than a lower one since the higher temperature can destroy membrane cells, making solvent easier to extract the pectin contents. Previous results also mentioned similar findings, as Sulihono et al. (2012), who produced pectin with orange skin. Moreover, the high temperature must be matched with the optimum time. The combination of high temperature and length of extraction time (90^{O}C, 120 minutes) caused pectin hydrolysis and further produced pectate acid (Utami 2014), which impacted lower results (22.63%). Yujaroen et al. (2008) also stated that the longer the extraction time, the higher the percentage of pectin derived.

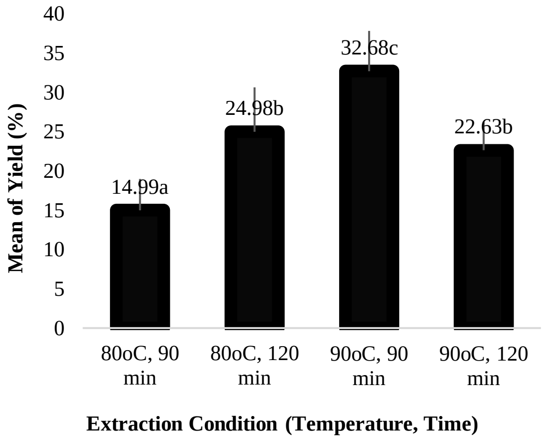

Figure 1 Effect of extraction condition of pectin yield (n = 3), LSD = 0,030.

These findings emphasize that extraction at 90°C is more effective and produces optimum results with prolonged extraction time. On the other hand, the use of ammonium oxalate as a solvent in this research showed a higher yield than any previous studies of pectin extraction from other materials such as ambarella, mangoes, and oranges (Koubala et al. 2008a; Koubala et al. 2008b) or dragonfruit (Ismail et al. 2012).

Moisture Contents

The moisture contents of pectin from arabica pulp coffee range from 3,90% to 7,93%, with average moisture content of 6,74%, as presented in Table 1. Furthermore, one-way ANOVA shows that extraction condition has any significant influence ($P>0,05$) on the moisture contents of pectin from arabica pulp coffee.

Even though there is no statistical evidence, Table 1 shows that extraction condition with lower temperature (80°C) and shorter extraction time tends to have higher moisture contents. Meanwhile, as temperature increases to 90°C with a prolonged extraction time of up to 120 minutes, the produced pectin tends to have lower moisture content. This value was similar to soy hull pectin, with a moisture content of 6 to 7% that were extracted based on different temperature and times (Kalaptahy and Proctor, 2001). These findings might occur since all treatments went through similar drying conditions. Based on this finding, all pectin arabica pulp coffee samples have moisture contents that have fulfilled the standard of the International Pectin Producers Association (IPPA 2001) and The Council of The European Communities (1998), which mentioned that moisture contents must not exceed 12%.

Ash Contents

Pectin from arabica coffee pulp has ash contents varied from 0.90-0.95% with the average score is 0.93%±0.12% as can be seen in Table 1. in range within 0.90-0.95% and has fulfilled the standard of International Pectin Producers (2001) as well as The Council of The European Communities (1998) which mentioned that moisture contents must not exceed 10%. Lower ash contents are related to lower mineral residues, for instance, calcium and magnesium, which co-hydrolysate with protopectines. Moreover, ash contents also showed pectin pureness; the higher the as contents, the lower the pectin pureness. Meanwhile, the use of ammonium oxalate as a solvent in this extraction seems to positively impact the ash contents, and pectin pureness since the temperature and length of extraction showed any differences. Furthermore, one-way ANOVA shows that extraction condition has any significant influence ($P>0,05$) on the ash contents of pectin from arabica pulp coffee.

Methoxyl Content

Methoxyl pectin contents from arabica coffee pulp varied from 3,88 to 7,69 with an average score of 6,14%. Metoxil contents measure hydrolyzed methyl groups of protopectin which changed to pectin at extraction (Mohnen 2008). Anova shows that the extraction condition has significantly influenced ($P<0,05$) the methoxyl content of obtained pectin, as can be seen in Figure 2.

Based on the average score of methoxyl contents, the obtained pectin is classified as low methoxyl contents (below 7%), except the pectin from 90°C and 90 minutes extraction time which its methoxyl content exceeds 7,00% and is considered as high methoxyl contents.

The value of methoxyl in pectin is responsible for the ability to perform gel, the preparation time, and its consistency. To gelation formation, high methoxyl contents require a high amount of sugar and are sensitive to acids, and vice versa for low methoxyl groups. Therefore low methoxyl pectin is preferred by the food industry for low-sugar food applications since it is not required the addition of high sugar to form gel (Pagarra et al. 2018).

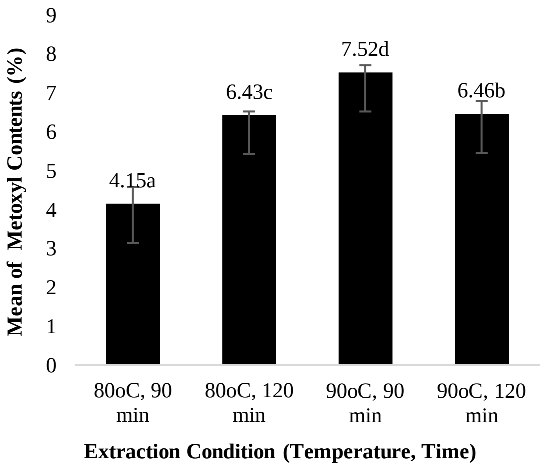

Figure 2 Effect of extraction condition on methoxyl content of resultant pectin of arabica coffee pulp (n = 3), LSD = 0,002.

According to Koubala et al. (2008), pectin extraction using ammonium oxalate produced pectin with high methoxyl contents, which significantly impacts the gel characterization and pectin solubility. Susanti (2021) also stated that the higher the methoxyl content, the greater its ability to thicken the medium. IPPA also recommends the maximum permissible level of methoxyl in pectin is 7,12%.

Galacturonate Acids Content

Galacturonate acids content of resultant pectin varied from 42,10-71,10%, as seen in Table 1. Anova shows that extraction condition (time and temperature) significantly influences the galacturonate acids (GA) contents of resultant pectin. The LSD of each treatment is presented in Figure 3.

$LSD_{0,05}$ shows that each treatment level is significantly differentiated. A prolonged extraction time of up to 120 min for 80°C temperatures produced pectin with the highest GA content. On the other hand, when the set temperature is higher, specifically at 90°C, prolonged extraction time to 120 min produced pectin with the lowest GA contents, of 60,78%. Ramli et al. (2014) mentioned that The GA content was higher by increasing the extraction time, which the hydrolysis reaction of protopectin might cause became D-anhydrogalacturonic or galacturonic acid that a basic component of pectin. However, when the higher temperature is set and combined with a longer extraction time, the GA is low due to the depolymerization is occurred.

Galacturonate acids content refers to the purity of resultant pectin (Passikalio et al., 2017). As GA content increases, the inorganic content in pectin tends to decrease and lead to an increasing level of pectin purity (Susanti et al. 2021). The arabica coffee pulp pectin has GA content above the minimum level of IPPA, which is 35%. Therefore arabica coffee pulp is a potential source of pectin production.

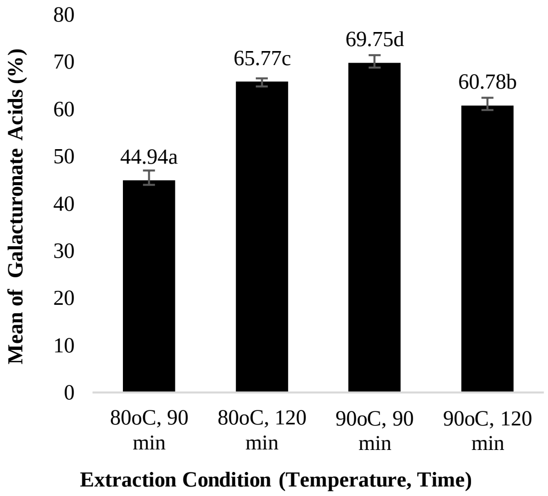

Figure 3 Effect of extraction condition on galacturonate acids content of resultant pectin of arabica coffee pulp (n = 3), LSD = 0,05.

Degree of Esterification

The degree of esterification (DE) calculates the percentage of D-galacturonic acid in which the carboxyl group passed through the ethanol esterification process. A higher esterification degree refers to the good capability of gel (Whistler and Daniel 1985; Ramli et al. 2014). Esterification degree is defined a comparison of methoxyl and galacturonate contents divided into two groups, high ester groups (min. 50%) and low ester group (max. 50%) (Sandarani, 2017). The high ester group, also known as high methoxyl pectin, has a degree of esterification above 50% and vice versa with low methoxyl ones (Susanti et al. 2021).

Pectin from this study has an esterification degree within 49,08-64,23% with an average score of 57,36%. These values classify pectin from arabica coffee pulp as high ester molecules or methoxyl pectin (IPPA 2001). When pectin is used as gelling agent, this class of pectin requires soluble solids and low pH conditions to perform a stable gel condition (Voragen et al. 1995; Tsoga et al. 2004).

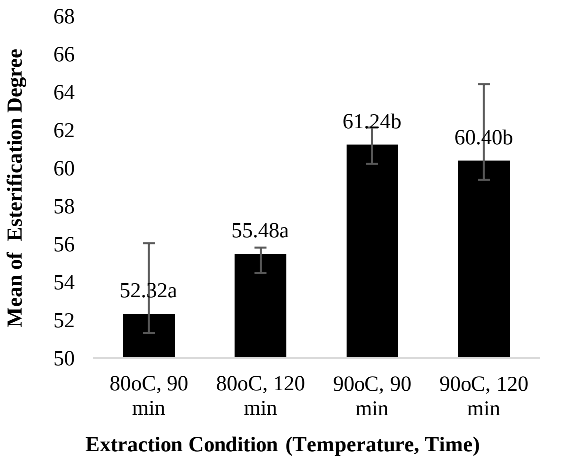

Figure 4 Effect of extraction condition of esterification degree of pectin (n = 3), LSD = 0,04

Anova shows that combined extraction time and temperature treatments significantly influenced ($P<0,05$) the esterification degree of pectin, as can be shown in Figure 4. From Figure 4, it can be seen that higher extraction temperatures up to 90°C produced high methoxyl pectin than lower temperatures. In lower extraction temperatures, pectin has a lower esterification degree. It also can be assumed that extraction of pectin at 90°C for 90 minutes is an optimum condition for obtaining the better quality pectin of arabica coffee pulp.

CONCLUSION

Coffee pulp is considered rich in pectin. Extraction condition (time and temperature) affects the characteristics of pectin. The highest yield was obtained by extraction at 90°C for 90 minutes. The coffee pectin has colored light brown and is categorized as low methoxyl pectin. Overall pectin coffee pulp produced in this study has met the quality requirements of dried pectin by the International Pectin Producer Association (IPPA) standard. According to methoxyl content, coffee pectin can use in the food industry, especially beverages and jam processing.

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