Optimization of Pectin Extraction from Pedada Fruit
(Sonneratia caseolaris)

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Abstract. Pedada has a high pectin content that can be used for produce pectin. The study aimed to determine the optimum conditions of extraction time and temperature on the quality of pectin. This study consists of 2 stages, the first stage determines the best treatment for temperature and extraction time. The second stage was to determine the optimization of pectin extraction using RSM (Response Surface Methodology). The best treatment for the first extraction was obtained at 80°C and 120 minutes, and the code and experimental level values were obtained +1.414 (94.142°C:162.426 minutes) ; +1 (90°C:150 minutes) ; 0 (80°C:120 minutes); -1 (70°C:90 minutes) ; -1.414 (65.857°C:77.573 minutes). The result of extraction optimization was 89.59°C and 128.55 minutes with a yield of 7.66%, equivalent weight 850.42mg, methoxyl content 3.71%, galacturonic content 85.17% and esterification degree 24.99% with desirability 0.75.

Keywords : Extraction, HCl, Pectin, Pedada Fruit, RSM

1. Introduction
Pedada fruit is one of mangrove fruit that contain enough high pectin which was reported by Jariyah [1] of 9%, so that pedada can be used as an alternative source of novel pectin. This pedada fruit is very abundant when harvested and has not been used optimally. Pectin is a polymer of D-galacturonic acid which is connected by glycosidic β -1.4 bonds [2]. Pectin is widely used in food fields such as jelly, jam, marmalade [3-4].

Pectin extraction process is influenced by several factors, two of which are very influential factors, namely the temperature and time of extraction. According to Chan [5] states that if low temperatures are not enough to hydrolyze the protopectin so yield of pectin is low. The extraction time also determine the amount of hydrogen ions that have succeeded in substituting ion Ca and Mg from protopectin so it determines the amount of pectin extracted [6]. These two factors affect the quality of pectin, so this study aims to extract pectin at the right temperature and time to produce yield, methoxyl content and optimal degree of esterification.

2. Methods
The ingredients used was pedada (Sonneratia caseolaris) obtained from Sawahan Sidoarjo, HCl, 96% alcohol, 0.1 N NaOH, and 0.25 N, 0.25 N HCl, phenol red indicator. The tools are magnetic stirers, digital scales, ovens, cabinet dryers, blenders, furnaces and other glassware.
This study consisted of 2 stages, the first stage was looking for the best treatment temperature (60°C, 70°C and 80°C) and time (60, 90 and 120 minutes). The second stage is using RSM (Response Surface Methodology) to obtain optimization of pectin extraction. The results of the first phase of research were obtained the best treatment at 80°C and 120 minutes, to determine the code and the level of the experiment (Table 1), so that the research design presented in Table 2.

| Code  | Temperature | Time | X<sub>1</sub> | X<sub>2</sub> |
|-------|-------------|------|---------------|---------------|
| +1.414| 94.1421     | 90   | -1            | -1            |
| +1    | 80          | 70   | 1             | -1            |
| 0     | 120         | 120  | 0             | 0             |
| -1    | 150         | 0    | 0             | 0             |
| -1.414| 77.5736     | 0    | -1.414        | 0             |
| -1.414| 120         | -1.414| 0             | 0             |

| Code  | Temperature | Time | X<sub>1</sub> | X<sub>2</sub> |
|-------|-------------|------|---------------|---------------|
| 80    | 70          | 120  | 0             | 0             |
| 80    | 90          | 120  | 0             | 0             |
| 80    | 120         | 77.5736 | 0 | -1.414        |
| 80    | 120         | 162.426 | 0 | 1.414        |
| 65.8579 | 120     | -1.414| 0             | 0             |
| 94.1421 | 120   | 1.414 | 0             | 0             |

3. Result and Discussion

3.1 Yield of Pectin

Yield of pectin showed using 3D Surface Plot which presented that the increase in yield corresponds to the increase in temperature and extraction time. The highest yield at extraction temperature was 85-90°C while extraction time was 105-120 minutes, this indicated that the longer extraction time affect the increase of kinetic energy in solution so the diffusion of solvent into tissue cells become higher, causing the weight of the pectin to increase. Tuhuloula [7], reported that contact between particles requires time, the longer time would made the contact process become greater and the chance extraction of the solvent become higher which increases the weight of the pectin. The higher temperature causes the hydrogen ions to substitute calcium and magnesium from the more protopectin, so hydrolyzed protopectin produces increased pectin, and continues to increase until the maximum state is reached [8-10].

3.2 Equivalent weight

Equivalent weight of pectin showed in Figure 2 below, indicated that the higher temperature and the longer extraction time result in decrease of pectin’s the equivalent weight and vice versa. This due to pectin turn into pectic acid by the longer extraction time which lead the decrease of the equivalent weight. Based on this study, the highest equivalent weight occurs at a temperature range of 57-91°C
and extraction time of 57-138 minutes. Rizqi [11] reported that the longer extraction time result in transform pectin into pectic acid so that the galacturonic acid group which is unesterified becomes more numerous. In addition, deesterification also form the pectic acid that increase the number of free acid groups and induce the decrease of the equivalent weight [7].

3.3 Methoxyl content
Pectin’s methoxyl content showed in Figure 3 by 3D surface plot which give result that the highest methoxyl content of 4.5% was at 76.66°C and the time range 96.66-103.33 minutes. The extracted methoxyl pectin level ranged from 3.14-4.73. Referring to Sundar [12] methoxyl content of the pectin result in this study included in the low category, due to the deesterification process of pectin which caused a decrease in methoxyl content [13]. According to Sayah [14] and Khan [15], low pH of HCl lead to increase the de-esterification process of pectin which had been extracted into galacturonate followed by a decrease in methoxyl content.

3.4 Galacturonic acid content
Galacturonic acid content represented in Figure 4 using 3D surface plot. Based on Figure 4, the highest level of galacturonic acid occurs at the temperature range of 69-86°C and 100-140 minutes while the lower level occurs at 80-87°C and 90-140 minutes. This caused by decomposing of galacturonic compounds which decrease the galacturonic levels. Wignyanto [16] reported that the reduction of galacturonic acid by the strong acid HCl increases the decarboxylation process of pectin and lead to the decomposition and reduction of galacturonic acid.

3.5 Esterification degree
Esterification degree represented in Figure 5 by 3D surface plot. Figure 5 showed the increase of esterification degree when the temperature and extraction time are lower and vice versa. The greatest degree of esterification occurs in the temperature range 70-74°C and 60-75 minutes. The longer the time and the higher extraction temperature cause the decrease of esterification degree. This due to the glycosidic bonds of the methyl ester group from pectin tend to be hydrolyzed into galacturonic acid [17-19].
Roikah [20] also reported that the longer extraction time induce the degradation of pectin’s methyl ester group into carboxylic acid. The presence of acid in the extraction process made the degree of esterification to decrease, if the extraction time was longer, pectin could turn into moderate pectic acid. The methyl ester group changes to free galacturonic acid. The number of methyl ester groups indicates the number of unesterified carboxylic groups or degrees of esterification.
3.6 Optimization

The results of the analysis of the optimization criteria for the relationship between temperature and time of extraction of pedada pectin optimization are presented in Table 3. Optimum conditions reached at 89.59°C and extraction time of 128.55 minutes with the highest desirability value of 0.75. The optimum condition has a response value ($y^*$) presented in Table 4.

| Criteria          | Goal     | Limit          | Weight | Importance |
|-------------------|----------|----------------|--------|------------|
| Temperature       | in range | 60 90          | 1 1    | +++        |
| Time              | in range | 60 150         | 1 1    | +++        |
| Methoxyl          | maximize | 3.14 4.73      | 1 1    | +++        |
| Equivalent Weight | in range | 600 1600       | 1 1    | +++        |
| Galacturonic Acid | maximize | 35 100         | 1 1    | ++++       |
| Esterification Degree | in range | 24.92 25.53 | 1 1 | ++++       |
Table 4. Response Value (Y^) in Optimum Condition.

| No | Response                          | Value (Y^) | Real value (Lab Test) |
|----|----------------------------------|------------|-----------------------|
| 1  | Yield                            | 6.79       | 7.66                  |
| 2  | Equivalent Weight                | 971.13     | 850.42                |
| 3  | Methoxyl Content                 | 4.13       | 3.71                  |
| 4  | Galacturonic Acid Content         | 94.44      | 85.17                 |

4. Conclusion
The optimum conditions in extracting pectin were achieved at a temperature of 89.59°C and extraction time of 128.55 minutes. The optimum value included in the optimum criteria is yield 7.66%, equivalent weight 850.42 mg, methoxyl content 3.71%, galacturonic acid content 85.17% and the degree of esterification 24.99%.

5. References
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