The Effect Of Various Processing Processes On The Characteristics Of Avocado Seed Flour (*Persea an Americana mill*)

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Abstract:
This study was conducted to determine the various processing, of avocado the characteristic seed flour, and well to find out the influence processing of avocado the characteristic seed flour levels. This research was conducted using a Completely Randomized Design (RAL) with five treatment techniques namely without treatmant, soaking in the water for 30 minutes, soaking in the water for 12 hours, boiling in the water at 80 °C, steam blanching 100 °C for 10 minutes. Each treatment was repeated 4 times, so that 20 experimental units were obtained. The data obtained were analyzed using the analysis of variance (ANOVA) at 5% and 1% levels. Parameter with significantly affect was further analyzed using duncan's new multiple range test (DnMRT) at the 5% level. Processing techniques have a very significant effect on tannin content, activiti antioxidants and the color of the flour produced. Soaking in 30 minutes of water, soaking in water for 12 hours, boiling blanching at 80 °C, and steam blanching for 10 minutes produce flour with tannin content that is not statistically different according to the DnMRT test, but soaking in water for 30 minutes and immersion in water for 12 hours produces flour with higher antioxidant activity than boiling blanching at 80 °C and steam blanching for 10 minutes at 100 °C. Thus the 30 minute soaking treatment was chosen as the best treatment that produced avocado seed flour with characteristics: tannin content 62.23 μg ATE / g, antioxidant activity value 62.82%, color test value L * 56, + a * 13 , 40, + b * 27.60, 7.25% moisture content.

Keywords: Processing Processes, Characteristic, Avocado Seed Flour

1. Introduction

The avocado plant (*Persea americana Mill*) is a type of fruit that is popular with the public because apart from its delicious taste, it is sweet, thick and also has a high antioxidant content and also nutrients such as fat, namely 9.8 g/100 g of fruit flesh (Afrianti, 2010). There are two types of avocado, namely round green avocado (*Persea americana Mill*) and long green avocado (*Persea gratissima Gaertn*). *Persea amaricana Mill* avocado has a fruit weight of around 0.3 kg with a round shape. Unripe avocados have dark green avocado skin, while ripe avocados have light green avocado skin, slightly rough and dull. Unripe fruit has thick, greenish or yellow flesh like butter, so it is better known as butter avocado. *Persea gratissima Gaertn* fruit weighs around 0.38 kg with a long fruit neck, the skin of the fruit is green and smooth, the flesh is thick with a savory taste (Widyastuti and Paimin, 1993). Avocados consist of 65% flesh, 20% seeds and 15% skin (Risyad et al., 2016). Most people only use avocados for the fruit while the other parts are thrown away and become waste. One of the wastes of avocados is the seeds, which are known to contain many benefits.

According to Zuhrotun (2007), avocado seeds contain rich benefits. The results of phytochemical screening of avocado seed extract show that avocado seeds contain polyphenols, flavonoids, triterpenoids, quinones, saponins, monoterpenoids, sesquiterpenoids and tannins. With the presence of these compounds, avocado seeds are functional and are believed to be able to treat toothache, chronic stomach ulcers, hypertension and diabetes mellitus (Monica, 2006).
Avocado production in Indonesia is quite high, this can be proven by data on avocado production in Indonesia in 2014 from the Central Statistics Agency (BPS), which reached 307,326 tons per year. From BPS data, avocado production in Indonesia continues to increase every year (BPS, 2013), along with increasing avocado production, the avocado seed waste produced also increases. Therefore, it is necessary to handle avocado seed waste by conducting research on the processing of avocado seed flour.

Food processing in this case focuses on processing avocado seed waste. So it is necessary to identify the chemical and physical properties of the material, namely avocado seeds which will first be made into flour. Flour is an alternative form of semi-finished product that is recommended because it is more resistant to storage or can extend shelf life, is easy to mix (make composites), is formed and is cooked faster according to the practical demands of modern life (Damarjati et al., 2000). One of the problems faced in making avocado seed flour is the presence of tannin compounds, which give the flour a thick taste, so it is necessary to remove the tannins using various processing processes. Tannin compounds are compounds that dissolve easily in water and give color to the water. The color of the tannin solution varies depending on the type of plant produced, from light to dark red or brown and the solubility increases when dissolved in hot water, but on heating to temperature 100°C will decompose into pyrogallol, pyrocatechol and phloroglucinol (Muryati., et al, 2015). This shows that the blanching temperature has an effect on reducing tannin levels. Where the higher the blanching temperature the tannin content decreases.

According to research by Rohaman, (1993), the tannin content of bentul taro in the boiling treatment can reduce the tannin content by 49.9%, while in the steaming treatment the tannin content can decrease by 50.2%. Based on research by Suprato (1997), he explained that the tannin content in sorghum seeds can be reduced by soaking in water, with the tannin content lost by this method being around 281.92 ppm. According to Narshih et al in Subagio (2014), stated that, to reduce tannin from sorghum, it is necessary to soak red sorghum seeds so that the tannin can be reduced, because the tannin found in sorghum will dissolve in water. Reducing tannin levels by soaking can be done by soaking the ingredients in clean water for several hours, then stirring until the water becomes white foam. Next, throw away the water and replace it with new clean water. According to research by Malangi, (2012) avocado seeds have tannin levels of 41 mg/kg. Avocado seeds contain quite high levels of tannin so that processing them into avocado seed flour produces flour with tannin levels which can cause a bitter taste in the derivative products.

Based on the description above, the author conducted research with the title "The Effect of Various Processing Processes on the Characteristics of Avocado Seed Flour (Persea americana Mill)"

2. Research Methods

Materials and Tools
The materials used are ripe avocado seeds, obtained from waste, one of which is fruit soup or fruit juice sellers in Jambi city, tissue, label paper, filter paper, masks, gloves, distilled water, aluminum foil, DPPH (1,1-diphenyl-2-picrylhydrazil), tannic acid, Na2CO3 20%, Folin Ciocalteu. Meanwhile, the tools used are analytical scales, digital scales, boiling pans, stoves, ovens, basins, knives, baking pans, blenders, 60 mesh sieves, cuvettes, spectrophotometers, glass funnels, micro pipettes, dropper pipettes, stirring rods, Erlenmeyer, test tube, test tube rack, desiccator, vortex, centrifuge, 100 ml volumetric flask, 10 ml measuring cup, beaker, stopwatch.

Research Design and Statistical Analysis
The research used a Completely Randomized Design (CRD) with treatments without soaking, soaking in plain water for 30 minutes, soaking in plain water for 12 hours, boiled blanching (hot water at 80 °C), steam blanching at 100 °C for 10 minutes. Each treatment was repeated 4 times to obtain 20 experimental units. The data obtained were analyzed using analysis of variance at the 1% and 5% levels and if they were significantly different then continued with Duncan's New Range Multiple Test (DnRMT) at the 5% level.

Research Procedures
The avocado seeds used are ripe avocado seeds, a round green avocado (Porsea americana Mill). The avocado seeds are washed with clean water so that no dirt sticks to them, after that the avocado seeds are weighed with a weight of ± 250 grams for each treatment with 1 L of water, then cut into small and thin
pieces, then followed by five treatments, then dried. Each treatment uses an oven for approximately 8 hours at a temperature of 60°C. Once dry, the avocado seeds are then ground using a blender, then once smooth, sifted using a 60 mesh sieve. Avocado seed flour is ready to be analyzed. Store flour at room temperature where it is not directly exposed to sunlight.

Water content (Sudarmadji, et al, 1997)

Determination of water content is carried out by weighing 2 grams of the sample and placing it in a cup which has first been dried in an oven at 105 °C for 3 hours and cooled in a desiccator, then the cup containing the sample is placed in an oven at 105 °C for 1 hour, then cooled again in a desiccator, and weighed. This work is carried out repeatedly until a constant weight is obtained (difference in successive weighings x ± 0.2 mg). Water content is obtained using the following formula:

\[
\text{Water Content} = \frac{\text{Initial Weight} - \text{Final Weight}}{\text{Initial Weight}} \times 100
\]

Tannin Content (Samosir, 2018)

A total of 1 gram of avocado seed flour was added with 10 ml of distilled water, 0.1 ml of the extract was taken, added with 0.1 ml of Folin Ciocalteu reagent and vortexed for 15 minutes, added with 2 ml of 20% Na₂CO₃ and vortexed again for 15 minutes. Then leave it for 30 minutes in a dark place and cover with aluminum foil. After that, it was put into a cuvette and the absorbance was read at a wavelength of 760 nm. The results obtained were plotted against a tannic acid standard curve prepared in the same way. Tannin content is expressed in µg ATE/g flour. The tannic acid concentrations used were 500, 450, 400, 350, 300, 250, 200 and 150 µg/ml. So we get a linear regression line from the curve with a value of R²= 0.9869.

Antioxidant Activity (Bahriul et al., 2014)

A total of 1 gram of avocado seed flour with 10 ml of distilled water added, 1 ml of the sample was pipetted using a micro pipette and put into a test tube, then 5 ml of DPPH solution was added. The solution mixture was homogenized with a vortex and kept for 30 minutes in a dark room and covered with aluminum foil. Absorption was measured with a UV-Vis spectrophotometer at a wavelength of 517 nm. For the positive control, vitamin C or ascorbic acid is used as a comparison to determine the absorbance obtained. Calculations are carried out using the following formula:

\[
\% \text{ Inhibition} = \left( \frac{\text{Control Absorbance} - \text{Sample Absorbance}}{\text{Control Absorbance}} \right) \times 100\%
\]

Color (Andarwulan et al., 2011)

Color analysis is carried out using a Color reader tool. Measurement by placing the sample in uniform white plastic (transparent plastic). Next, the L*, a*, and b* values were measured on the sample. The Hunter color notation system is characterized by 3 color parameters, namely, color intensity with L* notation for brightness, a* notation and b* notation for color intensity. Each value is L*, a*, and b* with a value range of 0-100. The L* value represents the brightness parameter which has a value from 0 (black) to 100 (white). The a* value represents a mixed chromatic color of red – green with +a (positive) from 0-100 for red and –a (negative) value from 0-(−80) for green. +b (positive) value from 0-70 for yellow and –b (negative) value from 0-(−70) for blue.

Color descriptions based on L*, a*, and b* values are:

L* = brightness black (0) white (100)

a* = degrees red (+80) green (-50)

b* = degree yellow (+70) blue (-70)

3. Results and Discussion

Water Content

Water content is the amount of water contained in a material and is usually expressed in percent. The water content in food also determines the freshness and durability of the food. The lower the water content,
the slower the growth of microorganisms so that the food can last longer. On the other hand, the higher the water content, the faster the microorganisms will reproduce, so that the decay process will take place more quickly (Winarno, 2004). The following is Table 1. Average value of water content of avocado seed flour based on avocado seed processing techniques:

Table 1. Average value of water content of avocado seed flour with various avocado seed processing processes.

<table>
<thead>
<tr>
<th>Avocado Seed Processing Process</th>
<th>Water Content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Without Soaking</td>
<td>6,81</td>
</tr>
<tr>
<td>Soaking in water for 30 minutes</td>
<td>7,25</td>
</tr>
<tr>
<td>Soaking in water for 12 hours</td>
<td>7,69</td>
</tr>
<tr>
<td>Boiled blanching (hot water temperature 80°C)</td>
<td>7,96</td>
</tr>
<tr>
<td>Steam blanching at 100 °C for 10 minutes.</td>
<td>8,14</td>
</tr>
</tbody>
</table>

Based on the analysis of variance, various avocado seed processing techniques did not have a significant effect on the water content of avocado seed flour. According to Causgrove, (2004) water content is important in processed flour products, because water content of more than 14.5% is a good medium for the growth of fungi, bacteria and insects which can damage flour during the storage process. According to Hendra, (2007) avocado seeds have a water content of 12.56%. The aim of reducing the water content is to extend the shelf life, to reduce the volume and weight of the material so that it makes packaging easier and more economical (Winarno, 2008). According to SNI 01-3751-2009, the maximum water content in flour is 14%, so it can be seen that the water content in avocado seed flour in this study still meets the Indonesian National Standard, namely that avocado seed flour has a water content below 14%. Based on Table 1 above, it can be seen that the water content of avocado seed flour meets the Indonesian National Standard.

Tannin Level

Tannin is a very complex organic substance and consists of phenolic compounds which have high molecular weight. Tannin has the property of being soluble in water or alcohol because it contains many phenols which have OH groups (Sajarutad, 2013). The tannin content of avocado seed flour extract using various avocado seed processing techniques can be seen in Table 2.

Table 2. Tannin content, antioxidant activity and color of avocado seed flour using various avocado seed processing techniques.

<table>
<thead>
<tr>
<th>Processing Techniques</th>
<th>Tannin Content (μg ATE/g)</th>
<th>Decrease in Tannin Content (%)</th>
<th>Antioxidant Activity (%)</th>
<th>Color (L* a* b*)</th>
<th>Color Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Without soaking</td>
<td>93,32b</td>
<td>-</td>
<td>67,76c</td>
<td>55,93a +12,88 +27,55</td>
<td>Dark moderate orange</td>
</tr>
<tr>
<td>Soaking in water for 30'</td>
<td>62,23a</td>
<td>33,32</td>
<td>62,82bc</td>
<td>56a +13,40 +27,60</td>
<td>Dark moderate orange</td>
</tr>
<tr>
<td>Soaking in water for 12 hours</td>
<td>59,57a</td>
<td>36,16</td>
<td>60,72ab</td>
<td>56,33a +13,36 +27,65</td>
<td>Dark moderate orange</td>
</tr>
<tr>
<td>Boiled blanching (water 80°C)</td>
<td>59,07a</td>
<td>36,70</td>
<td>57,28ab</td>
<td>56,75a +13,48 +27,65</td>
<td>Dark moderate orange</td>
</tr>
<tr>
<td>Steam blanching 100°C for 10’</td>
<td>54,07a</td>
<td>42,06</td>
<td>56,29a</td>
<td>58,54b +13,70 +28,18</td>
<td>Dark moderate orange</td>
</tr>
</tbody>
</table>

Same superscript in the same column means no difference statistically using DnMRT (p>0.05)
Avocado seed processing techniques have a significant effect on the tannin content of avocado seed flour. Based on Table 3, the value of tannin content in the treatment without soaking was 93.32 μgATE/g and in the steam blanching treatment at 100 °C 10 minutes it was 54.07 μgATE/g. Tannin levels in processing without soaking had higher tannin levels than previous research results. According to Malangi, (2012) avocado seeds have tannin levels of 41 mg/kg. The high tannin content in the treatment without soaking is due to tannin compounds being sensitive to heating, so the concentration of tannin without treatment is higher than in the blanching processing technique, because with the heating process the tannin content bound to the protein of the material will be released and the tannin content will be lost by heating to high temperatures.

The processing technique without soaking was significantly different in the treatment of soaking in water for 30 minutes and in the treatment of soaking in water for 12 hours, because the soaking time had an influence on the tannin content. The longer the soaking time, the lower the tannin content in avocado seed flour. Decline tannin levels are due to tannins soluble in soaking water, where tannins are one of the compounds that dissolve in water. According to Suryaningrum, (2007), one of the factors that influences the solubility of a substance is the soaking time, where the longer the contact time between the material and the solvent (water), the more compounds from the material will be dissolved.

In the treatment, the processing technique was boiled blanching at a temperature of 80°C with a tannin content of 59.07 μgATE/g, while in the steam blanching treatment at 100 °C 10 minutes the tannin content was 54.07 μgATE/g. The effect of temperature during boiling and steaming causes proteins to denature resulting in changes to the tannin protein complex. Apart from that, the decrease in tannin levels was also caused by the influence of water in both treatments, where tannins are soluble in water (Turner, 1956).

Reducing tannin levels using the boiled blanching and steam blanching methods was able to reduce tannin levels by 36.70% and 42.06% and this was lower than previous research, namely according to Rohaman's research, (1993) the tannin levels of bentul taro in the boiling treatment could reduce the tannin levels. amounted to 49.9%, while the steaming treatment could reduce the agricultural content by 50.2%. The processing process with boiled blanching and steam blanching can have the same effect on reducing tannin levels.

**Antioxidant Activity**

Antioxidant activity is a compound that gives electrons. Antioxidants work by donating one electron to a compound that is an oxidant so that the activity of the oxidant compound can be inhibited. Antioxidant activity stabilizes free radicals by completing the lack of electrons that free radicals have, and inhibiting chain reactions from the formation of free radicals (Winarsi, 2007). Testing the free radical reducing activity of natural or synthetic compounds can be carried out using chemical reactions using DPPH as a stable free radical compound. The interaction of antioxidants with DPPH, either by transferring electrons or hydrogen radicals to DPPH, will neutralize the free radical character of DPPH and form reduced DPPH. If all the electrons in the DPPH free radical become paired, the color of the solution changes from dark purple to bright yellow and the absorbance at a wavelength of 517 nm will disappear (Rohman et al., 2010).

Based on the analysis of variance, various avocado seed processing techniques have a significant effect on the antioxidant activity value of avocado seed flour. Based on Table 2, the antioxidant activity value in the treatment without soaking was 67.76% and in the steam blanching treatment at 100 °C for 10 minutes it was 59.29%. The processing of avocado seeds has a significant effect on antioxidant activity because antioxidant properties are susceptible to heat. This is in line with research (Alfia, 2018) which states that the decrease in antioxidant activity is caused by changes in compounds due to the heating process, which affects phenolic compounds and other antioxidant compounds which undergo oxidation.

Degradation of antioxidant compounds can be caused by oxidation reactions, resulting in the breaking of covalent bonds or increasing the rate of oxidation reactions by heat. Antioxidant compounds that have been oxidized will be damaged and reduce their ability to reduce and ward off free radicals (Patras et al., 2010 in Huzaimah, 2018). According to Kawiji et al., (2011), the difference in antioxidant activity values is due to, among other things, the nature of antioxidants which are susceptible to temperature, oxygen, pH, peroxide and light. Antioxidant activity is not always correlated with phenol or tannin, this can also be due to several factors such as differences in active components in plants, synergistic or antagonistic effects between...
the active components contained, research conditions, and processing techniques used can influence antioxidant activity in plants. (Gengaihi et al., 2014)

In the treatment without soaking it was 68.76% significantly different from the treatment soaking in water for 30 minutes which was 62.82%, this is because antioxidant compounds can dissolve in water. This may be due to the influence of the phenolase enzyme contained in the extract fluid. The decrease in phenolic compound content can reduce the antioxidant activity contained in the extract. Avocado seeds contain 67.65% antioxidants (Liberty, 2012). In table 4, the standard value of ascorbic acid was obtained at 83%. It can be seen that the various processing processes for avocado seed flour did not meet the requirements, namely the treatment without soaking, soaking in plain water for 30 minutes, soaking in plain water for 12 hours, boiled blanching (hot water temperature 80 °C) and steam blanching 100 °C 10 minutes with respective values of 67.76%, 62.82%, 60.72%, 57, 28 and 56.29%.

Color

Avocado seed flour has an orange color with a hint of brownish. The results of color measurements produce data on the values of L, a, and b. The value data is the color notation used in the hunter notation system. According to Soekarto (1990), the hunter notation system has three parameters to describe colors, namely L, a and b.

The L parameter has values ranging from 0 (black) to 100 (white). The L value represents reflected light that produces white, gray and black achromatic colors. The notation a represents a mixed achromatic color of red – green with a range of +a values ranging from 0 to +100 for red and –a values ranging from 0 to -80 for green. The notation b states the chromatic color of a mixture of blue and yellow, with a +b value ranging from 0 to +70 for blue and a –b value ranging from 0 to -70 for yellow (Soekarto, 1990). The L, a, and b values obtained from colourreader readings can be described through color hexsa which shows the color that is visible visually, the color of avocado seed flour obtained from color hexsa shows a dark moderate orange color or a slightly dark orange color. The results of color measurements using various avocado seed flour processing techniques can be seen in Table 2.

The results of the L* values in Table 2 show a real effect on the color brightness level from 0-100, namely black to white. It can be seen that the L* (Brightness) value of avocado seed flour is higher using the 100 °C 10 minute steam blanching technique, namely 58.54, this is in line with research by Saputro, et al., (2016), that with boiling using Water media can increase the brightness of the color of the material. This is because the longer the blanching takes, the brighten the color becomes. The color of the flour with a higher level of brightness is caused by the inactivation of the polyphenol oxidase enzyme during the blanching process (Jang et al., 2005).

The a* values in Table 2 range from 12.88 to 13.70, indicating the brightness level of green to red. The highest a* value was found in the 100 °C 10 minute steam blanching technique, namely +13.70, while the lowest a* value was found in the technique without soaking, namely +12.88. This means that the color of avocado seed flour, whether treated or not, is red.

The results of the b* values in Table 2 range from 27.55 to 28.18, with the value +b indicating the brightness level of the color from blue to yellow. The highest b* value was found in the 100 °C 10 minute steam blanching technique, namely +28.18, while the lowest b* value was found in the technique without soaking, namely +27.55. This means that avocado seed flour, whether after being treated or not, is yellow in color. According to Zuhrotun (2017), avocado seeds contain phenolic compounds which cause browning and also contain flavonoid pigments which produce a yellow color. Avocado seeds are known to contain phenolic compounds in the form of flavonoids.

Conclusion

Avocado seed processing techniques have a significant effect on tannin content, antioxidants and color, but have no significant effect on water content. The processing process produces the best characteristics of avocado seed flour obtained in the steam blanching treatment, namely with a water content of 8.14%, tannin content of 54.07 μg ATE/g, antioxidant activity of 56.29%.