## Fiber Concentration on Fermentation of Cleome Gynandra L Based on Storage Time and Solvent Change

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#### Abstract

Cleome gynandra L (CGL) or Maman plant, is the basic ingredient of Malay food in Riau Province, Indonesia. The young leaves and stems of CGL are processed into fermented food (Joruk Maman). It contains crude fiber and is useful for lowering blood cholesterol levels. However, effective storage of this CGL has not been carried out. This study wants to see the effect of storage time and solvent change on the fiber content of Joruk Maman. An experimental study applied a completely randomized design (CRD) using 5 groups and 2 repetitions. This sample of CGL leaves was taken from one seller in the Rokan Hilir market of Riau Province. The primary outcome was a difference to the number of fibers in Joruk Maman without solvent change (p = 0.001) and with solvent change (p = 0.001) based on the day group and there was no difference base on the temperature group. Secondary outcome was the difference in duration time to produce the highest fiber content at room temperature with the solvent change and not. The highest fiber content occurred at 5 days of storage at room temperature without solvent changing. Meanwhile, by changing the solvent, the fiber content would be optimal for 1-day of storage.

Keywords: Cleoma gynandra L; joruk maman; fiber; storage; solvent change.

Abbreviations: Cleoma Gynandra L (CGL)

#### **INTRODUCTION**

The World Health Organization (WHO) and the Food and Agriculture Organization (FAO) have recommended high consumption of fruits and vegetables to prevent various chronic diseases such as hypertension, coronary heart disease, and stroke. However, WHO and FAO emphasize consuming fresh fruits and vegetables to get high nutritional content (Endrizzi et al., 2009)

One of the examples is Cleoma which belongs to the Capparaceae family. The people of Rokan Hilir, Riau, Indonesia recognize the maman plant which originated from South Africa. Throughout Africa, the tender leave or young shoots, and often the flower are boiled and consumed as a potherb, delicious dish, stew, or side dish. The fresh leaves are used as an ingredient in other mashed foods, and the dried leaves are ground and put into weaning foods. However, boiling the leaves can reduce the vitamin C content by up to 81%, while drying the leaves reduces the vitamin content by up to 95% (Heever & Venter, 2007). This maman plant is used as a traditional antidiabetic, anti-agingg, anti-cancer, and cardiovascular disease prevention (Mishra et al., 2011).

This plant is often found as a wild plant and it grows anywhere. People in this village usually consume this plant as a fermented vegetable. This fermented product of the maman plant (Cleome gynandra L) is processed from young leaves and stems which are mixed with salt, rice, and warm water and then it is left for 2-3 days before the people consume it. The villagers of Bunga Tanjung Rohul and Tanah Putih Rohil usually call this food by the name of CGL Fermentation or joruk maman. Fermented products of the Maman plant are commonly consumed by the public, and fermented products can be consumed with or without rice (Saida, E, 2014).

Fermented products of Maman plant or CGL Fermentation contain crude fiber and Lactic Acid Bacteria (LAB) which function as probiotics. Dietary fiber has a very important function for health maintenance and prevention of various degenerative diseases such as diabetes, hypercholesterolemia, stroke, coronary heart disease, and obesity as well as digestive disorders such as constipation, hemorrhoids, and colon cancer (Winarti, 2010).

Based on a review (Institute of Medicine, 2005) the adequacy of total dietary fiber in adolescents and adults

is 14 g/1000 kcal. The highest crude fiber content was obtained in CGL Fermentation with the addition of 2% salt and 15% rice, which was 0.43 g per 100 g of fermentation. Maman has crude fiber content which can reduce cholesterol levels and also contains lactic acid bacteria. The content of lactic acid bacteria in Maman's fermentation with the addition of 5% salt and 10% rice had the highest result of 2.40 x 108 cfu/g (Lily Restusari, Muharni, 2019).

People can make Joruk maman, or they can purchase at traditional markets in Rokan Hulu and Rokan Hilir, Riau Indonesia. Traditionally, local people will replace the fermented soaked water with boiled water if CGL Fermentation is stored for a long time and the sensory taste change. Moreover, after they change the water, they leave it overnight and CGL Fermentationis ready to be consumed again with the same taste as the new CGL Fermentation produced.

#### MATERIALS AND METHODS

#### Study area

The determination of Maman in Sekeladi Village, Rokan Hilir Regency and determined at the Andalas University Herbarium (Figure 1).



Figure 1. Cleome gynandra L (Maman's plant) by the author on Sekeladi village, Rokan Hilir Regency.

This research type is an experimental study conducted to determine the storage time and solvent change to fiber content in CGL Fermentation by applying a completely randomized design using 1 treatment and 2 repetitions. Maman fermentation was stored at room temperature and then was taken aseptically every day on days 1, 2, 3, 4, and 5. On day 5, the solvent was replaced by boiled water with the same amount of water replaced. Then, it was continued the inspection of dietary fiber on days 1, 2, 3, 4, and 5 after the solvent was replaced.

The research tools used were analytical balance, Niemeyer, measuring cup, condenser, funnel, vacuum pump, oven, desiccator, heater, and watch glass. The materials used were Maman (Cleome gynandra L) leaves, 0.85% NaCl diluent solution, table salt (PN salt) and local rice-pandan wangi varieties, sulfuric acid, NaOH, Whatman filter paper, alcohol, micropipette tips, and distilled water

#### Procedures

This research was carried out by weighing 1-2 grams of fermented Maman samples and put into a 500 ml Erlenmeyer, and then it was added 50 ml of hot 1.25% H2SO4 and refluxed for 30 minutes. After that, it was added 50 ml of 3.25% NaOH and refluxed for 30 minutes. The heated sample was then filtered in hot condition with Whatman 42 filter paper whose weight was known.

After being filtered, the sample was washed with 50 ml of 1.25% H2SO4 and 50 ml of 36% alcohol, then the precipitate was dried in an oven at 105°C and weighed to a constant weight. This research was conducted at the Laboratory of Agricultural Polytechnic of Andalas University in Payakumbuh and the Microbiology Laboratory of the Health Polytechnic of the Ministry of Health Riau. The research time was approximately 6 months from July to November 2020. This research involved 1 alumnus as a field survey officer and 1 laboratory analyst.

#### Data analysis

The survey primarily collected the sample at the Rokan Hilir market on the same day, with direct contact with the seller. Passing the fermentation process, data was collected at the laboratory.

Data analysis of dietary fiber results was carried out using SPSS 25 as the statistic program. All variables were tested for normality, then if the variables were normally distributed, Anova Block Design was applied. To see the variation difference among treatments, it is followed by a posthoc test, namely LSD and Duncan to find out the real difference between treatments.

#### **RESULTS AND DISCUSSION**

Results explain fiber content in CGL fermentation (Joruk Maman) base on storage time and solvent change and difference fiber concentration (Figure 2-3; Table 1-9).

#### Fiber content of Joruk Maman

 Table 1. Description of fiber content of Joruk Maman base on storage time.

Storage	Average of Fiber Content (gr/100gr)		
Storage Time	Refrigerator	Room	
	Temperature	Temperature	
Day 1	0.46	0.51	
Day 2	0.56	0.58	
Day 3	1.69	1.41	
Day 4	1.62	1.69	
Day 5	1.65	1.74	

Note: Primary data sources

The graph of fiber content in CGL Fermentation which was stored at Refrigerator temperature and room temperature (Figure 2)



Figure 2. Fiber Content of Joruk Maman against Temperature and Storage Time.

The normality test for the effect of temperature and storage time on fiber content in CGL Fermentation are normally distributed. So it is continued with the Anova Block Design test. The results of the Anova Block Design test were shown at Table 2.

**Table 2.** The Analysis of Anova Block Design, Temperature Effect, and

 Storage Time on Fiber Content of Joruk maman.

Treatment Group	Average + SD	p-value
Storage Time		0.891
Refrigerator Temperature	1.19 + 0.48	
Room Temperature	1.18 + 0.48	
Storage Time		0.001*
Day 1	0.48 + 0.77	
Day 2	0.57 + 0.77	
Day 3	$1.55 \pm 0.77$	
Day 4	$1.65 \pm 0.77$	
Day 5	$1.69 \pm 0.77$	

Note: \* Significantly different in the Anova Block Design test ( $\alpha < 0.05$ )

The significance value of the block (Day 1,2,3,4,5) was 0.001<0.05. There was a difference between the day group on the number of fibers; the posthoc test could be continued. The results showed in Table 3-4.

Table 3. Post-Hoc LSD Test, Temperature Effect, and Storage Time on Fiber Content in Joruk maman.

I	Post-Hoc LSD Test	Average Difference (gr/100gr)	Р
Day 1	Day 2	-0.0850	0.476
	Day 3	-1.0650*	0.001
	Day 4	-1.1700*	0.000
	Day 5	-1.2100*	0.000
Day 2	Day 1	0.0850	0.476
	Day 3	-0.9800*	0.001
	Day 4	-1.0850*	0.001
	Day 5	-1.1250*	0.000
Day 3	Day 1	1.0650*	0.001
	Day 2	0.9800*	0.001
	Day 4	-0.1050	0.387
	Day 5	-0.1450	0.252
Day 4	Day 1	1.1700*	0.000
	Day 2	1.0850*	0.001
	Day 3	0.1050	0.387
	Day 5	-0.0400	0.731
Day 5	Day 1	1.2100*	0.000
	Day 2	1.1250*	0.000
	Day 3	0.1450	0.252
	Day 4	0.0400	0.731

Note: \* Significantly different in Post-Hoc LSD Test (p<0.05).

Based on the post-hoc LSD test, the different mean pairs are on Days 1, 2, 3, 4, and 5 pairs. The number of differences in the average fiber is highest on Day 5 and Day 1 which is 1.21.

 Table 4. Temperature Effect, and Storage Time on Fiber Content in Joruk maman.

Treatment	Average Fiber Content (gr	/100gr)
Storage Time		
Day 1	0.48a	
Day 2	0.57a	
Day 3	1.55b	
Day 4	1.65b	
Day 5	1.69b	

Note: Numbers followed by the same lowercase letters in one column show no significant difference in (p < 0.05).

# The fiber content in Joruk maman with a solvent change towards temperature and storage time

**Table 5.** Description of Fiber Content in CGL Fermentation with Solvent Change towards Temperature and Storage Time.

Storago	Average Fiber Content (gr/100gr)		
Storage Time	<b>Refrigerator</b> <b>Temperature</b>	Room Temperature	
Day 1	1.79	1.89	
Day 2	1.65	1.57	
Day 3	1.24	1.11	
Day 4	1.11	1.08	
Day 5	1.1	1.04	

Note: Primary data sources.

The fiber content in CGL Fermentation which was stored at refrigerator temperature and at room temperature with solvent change also describe of Figure 3.



Figure 3. Fiber Content in CGL Fermentation with Changes in Solvents against Temperature and Storage Time.

The normality test for the temperature effect and storage time on fiber content in CGL Fermentation with Solvent Change were normally distributed, so it was continued with the Anova Block Design test. The results are in Table 6.

 Table 6. Description of Anova Block Design, Temperature Effect, and

 Storage Time on Fiber Content in CGL Fermentation with Solvent Change.

Treatment Group	Average $\pm$ SD	p-value
Storage Temperature		0.359
Refrigerator Temperature	1.37 <u>+</u> 0.27	
Room Temperature	1.33 <u>+</u> 0.27	
Storage Time		0.001*
Day 1	1.84 <u>+</u> 0.43	
Day 2	1.61 <u>+</u> 0.43	
Day 3	1.17 <u>+</u> 0.43	
Day 4	1.09 <u>+</u> 0.43	
Day 5	1.07 <u>+</u> 0.43	

Note: \*Significantly different in the Anova Block Design test  $(\alpha < 0.05)$ 

The Anova Block Design test showed the significant value of the block (Day 1,2,3,4,5) was 0.001 < 0.05. There was a difference between the Day group with the fibers content; the LSD post hoc test was shown in table 7 and table 8.

 Table 7. Description of The LSD Test, Temperature Effect, and Storage Time on Fiber Content in Joruk Maman.

Post-Ho	c LSD Test	Average Difference (gr/100gr)	Р
Day 1	Day 2	0.2300*	0.020
	Day 3	$0.6650^{*}$	0.000
	Day 4	$0.7450^{*}$	0.000
	Day 5	$0.7700^{*}$	0.000
Day 2	Day 1	-0.2300*	0.020
	Day 3	$0.4350^{*}$	0.002
	Day 4	$0.5150^{*}$	0.001
	Day 5	$0.5400^{*}$	0.001
Day 3	Day 1	-0.6650*	0.000
	Day 2	-0.4350*	0.002
	Day 4	0.0800	0.260
	Day 5	0.1050	0.160
Day 4	Day 1	-0.7450*	0.000
	Day 2	$-0.5150^{*}$	0.001
	Day 3	-0.0800	0.260
	Day 5	0.0250	0.703
Day 5	Day 1	-0.7700*	0.000
	Day 2	-0.5400*	0.001
	Day 3	-0.1050	0.160
	Day 4	-0.0250	0.703

Note: \*Significantly different in the LSD Post-Hoc Test (p<0.05)

Based on table 7, we know the pairs of average differences are on the 1st, 2nd, 3rd, 4th, and 5th -day pairs. Thus, the number of differences in the average fiber is highest on Day 1 and Day 5 which is 0.77

 Table 8. Description of The Duncan's Test Analysis of Temperature Effect

 and Storage Time on Fiber Content in CGL Fermentation with Solvent

 Change.

Treatment	Average Fiber Content (gr/100gr)
Storage Time	
Day 1	1.84 <sup>c</sup>
Day 2	1.61 <sup>b</sup>
Day 3	$1.17^{a}$
Day 4	1.09 <sup>a</sup>
Day 5	1.07 <sup>a</sup>

Note: Numbers followed by the same lowercase letters in one column show no significant difference in (p < 0.05)

The difference in fiber content in CGL Fermentation before and after dissolving, the paired t-test can be seen in Table 9.

 Table 9. Paired T-Test of Fiber Concentration in CGL Fermentation

 Before and After Dissolving.

Solvent Change	Mean ± Std. Deviation	P-Value	
Before	$1.19\pm0.57$	0.572	
After	$1.35\pm0.33$	0.372	

Note: There is a significant difference in the paired sample t-test ( $\propto < 0.05$ )

There is no difference in fiber concentration of CGL fermentation before and after solvent changes

#### Discussion

The fiber content in CGL Fermentation at room temperature ranged from 0.51 to 1.74 g/100 g, and Fiber content in CGL Fermentation at refrigerator temperature ranged from 0.46-1.69 g/100g. Both at room temperature and refrigerator temperature, the lowest fiber content was obtained on the first day, namely 0.51 g/100 g and 0.46 g/100g respectively; and the highest fiber content was obtained on the fifth day, namely 1.69 g/100 g and 1.74 g/100g. During the fermentation process, there were bacteria that played an important role, namely Acetobacter xylinum which has the ability to convert ethanol into vinegar. However, these bacteria required oxygen in the process. Crude fiber it self was the result of sugar reform in the fermentation medium by the activity of Acetobacter xylinum (Saleh, 2011) (Putriana & Siti Aminah, 2013).

There are not many research results that report fiber content related to temperature in this fermented food of the Malay tribe, named Joruk Maman. However, there are similar studies that examine the crude fiber content of the leaves and seeds of the rubber tree (Hevea Brasiliense). It is known that the crude fiber content of leaves and seeds of rubber trees (Hevea Brasiliense) after a 2-day fermentation period is much lower than the 5-day and 8-day fermentation periods. However, at 34 and 44 degrees Celsius, the crude fiber content is higher than the 2 and 5-day fermentation period. This result occurs because at high temperature, enzyme activity is reduced due to enzyme damage, and the water content of the substrate becomes much reduced. As a result, the raw fiber is converted into sugar. On the first day of the fermentation period, the addition of incubation time will reduce the crude fiber content of the leaves and seeds of the rubber plant (Hevea Brasiliense). The decrease in crude fiber from a temperature of 24-34 degrees Celsius is higher than the decrease in crude fiber from a temperature of 34-44. Similar results are also obtained at the 5 and 8-Day fermentation periods. These changes are closely related to the growth of molds and cellulase enzyme activity. With increasing incubation time, some cellulase enzymes can break down higher crude fiber; so that, the fiber content decreases (Syahruddin et al., 2016).

The research that we have done has obtained that the fiber content in CGL Fermentation stored at Refrigerator temperature compared to room temperature without any changes had the same graphic pattern. On Day 3 of storing CGL Fermentation at both temperatures, the fiber content increased which was quite high. The block significance value (Day 1,2,3,4,5) was different among Day groups on the number of fibers. While the different mean pairs were on Day 1, 2, 3, 4, and 5 pairs, where the highest number of fiber average differences occurs on Day 5 and Day 1, namely 1.21. Storage time on Day 1 (0.48 gr/100gr) had the smallest significant difference compared to other treatments, and storage on Day 5 (1.69 gr/100gr) was significantly different from other treatments. The final results showed that the fiber content of CGL Fermentation with a combination of 5-Day fermentation treatment at room temperature was the best treatment because it had the highest fiber content.

During the fermentation process, the ability of lactic acid bacteria (LAB) continued to increase in order to produce sufficient lactic acid to loosen the bonds of lignocellulose and lignohemicellulose which are components of crude fiber. Thus, the increase of crude fiber was in line with the fermentation time. The pH condition that was not optimal in LAB could not produce lactic acid in sufficient quantities to loosen lignocellulosic and lignohemicellulose bonds; so LAB had not caused changes in the crude fiber content of the silage. Therefore, at the beginning of storage, the amount of LAB in CGL Fermentation was less; so that, the lactic acid produced was also less to produce crude fiber.

Another finding of fiber content in CGL fermentation based on temperature and storage time on solvent change was a decrease in fiber content of CGL Fermentation from the first day to the fifth day; the decrease reached 0.85 g/100 g. Meanwhile, at refrigerator temperature, the fiber content of CGL FermentationDay 1 to Day 5 decreased to 0.69 g/100g. Based on the graph, it can be seen that on Day 3 the fiber content decreased quite high. There was a difference between the Day groups in the number of fibers. The different mean pairs were on the 1st, 2nd, 3rd, 4th, 5th Day pairs. The highest number of fiber average differences on Day 1 and Day 5 was 0.77. This finding is in line with olive pomace fermentation with simultaneous production of gallic acid, where the fermentation changes the chemical composition of olive pomace so that crude fiber decreases by 8.56% (Fathy et al., 2018).

The crude fiber content on the first day continued to decrease significantly to the fifth day until it reached the lowest fiber content. This is inversely proportional to the fiber content of CGL fermentation without solvent change. This decrease in fiber content can occur because it is closely related to the constituent components of the fiber, especially lignin. High lignin will make it difficult for microorganisms (bacteria) to degrade the material, so fiber content is decreased to low. The fiber content of CGL Fermentation with the combination of 1 Day fermentation period treatment at room temperature was the best treatment because of the highest fiber content in this treatment. The research results are different from other studies related to fiber in nata de cassava which states that the length of nata de cassava fermentation causes Acetobacter xylinum bacteria to work depending on differences in the number of nutrients to meet their needs. If the number of nutrients is sufficient, then the amount of cellulose formed is also big, whereas if the number of nutrients is not sufficient, the growth of Acetobacter xylinum bacteria is inhibited as a result, a small amount of cellulose is produced (Putriana & Siti Aminah, 2013).

The average value of the highest fiber content was obtained in the nata de cassava product on the 7th day of fermentation, which was 94.31 mg. The duration of data de cassava fermentation did not affect the fiber content of the data formed. This was because on Day 7 of fermentation, the Acetobacter xylinum bacteria were in an exponential phase because the Acetobacter xylinum bacteria concealed as much extracellular polymerase enzymes to arrange glucose polymers into cellulose; so that, more data matrix was produced in this phase. The 9th and 11th fermentation times decreased because Acetobacter xylinum bacteria were in a slow growth phase because the availability of nutrients had decreased. The 13th fermentation time increased because more data matrix was produced in this phase.

There is no explanation regarding the solvent change in the nata fermentation, this is certainly different from the conditions of the CGL Fermentationfermentation that we did. In the CGL Fermentationfermentation process, we used two groups, namely without solvent change and using solvent change. It turned out that the fermentation of CGL Fermentationwhich was replaced by a solvent (using solvent change) was not significantly different. This could be triggered by the previous initial conditions, where the fiber content of CGL Fermentation before and after being given dissolving did not have a much different numerical difference. Meanwhile, the highest CGL Fermentationfiber content occurred on Day 5. This could be due to a significant increase in the size of the cellulose fraction compared to the raw material, which was caused by the loosening of the cell walls and release of cellulose, and a reduction in the size of the hemicellulose and pectin fractions, such as the research on sauerkraut.

One of the more important functional properties of dietary fiber is its water-holding capacity which reflects the fiber's ability to swell. The results obtained that the ability of cabbage fiber to bind water depended on the cultivar and the length of storage time of fermented cabbage. The rehydration capacity of dried sauerkraut increased with storage time. The increase, compared to fresh cabbage, was an average of 22% after 10 days from the end of fermentation; after 30 and 90 Days were at the same level with an average value of 35%. This difference in water absorption probably arises from changes in the fractional composition of the fiber, namely from the increase in the size of the cellulose fraction observed during the storage of sauerkraut (Elkner, Krystyna, Kosson, 2009).

#### CONCLUSIONS

There was a different the number of fibers in CGL fermentation without solvent change and with solvent change based on the Day group. The highest fiber content production has 5 days duration, without solvent change and at room temperature. Meanwhile with solvent change, the optimal fiber content at 1 day and at room temperature. Regarding the length of storage in CGL fermentation, it has the potential to become a probiotic product.

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*Authors' Contributions:* Lily Restusari designed the study. Ayu Komalasari carried out the laboratory work. Alinea Dwi Elisanti analyzed the data. Lily Restusari and Alinea Dwi Elisanti wrote the manuscript. All authors read and approved the final version of the manuscript

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