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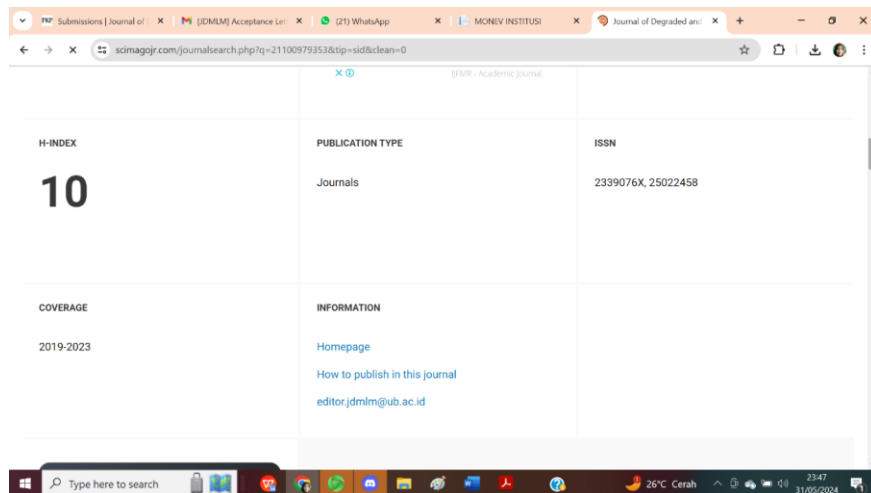
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Research Articles

**The Impact of Organic Fertilizer on the Growth, Rhizome Yield, and Secondary Metabolite Levels of Bangle Plant (*Zingiber montanum*)**

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**Abstract**

**Riwayat artikel:**

Diterima  
Diterima  
Diterbitkan

**Kata kunci:**

bangle  
hasil  
kemiringan lahan  
pengolahan tanah  
pertumbuhan

Indonesia is a major producer of spices in the world. Bangle is one of the traditional medicines that some people have heard has beneficial effects on the body. Although the bangle has a lot to offer in terms of content and advantages, there is still a huge gap in market demand, which is caused by the cultivation method's shortcomings. One way to boost the bangle plant's productivity is to provide manure. The goal of this study was to establish the ideal manure dosage for bangle plant growth, rhizome yield, and secondary metabolic antioxidant content. The study employed a Randomized Block Design (RAK), with one treatment consisting of organic fertilizer in the form of cow and chicken manure, and the levels were divided into seven, namely P0 (control), P1 (cow manure 20 tons/ha), P2 (cow manure 40 tons/ha), P3 (cow manure 60 tons/ha), P4 (chicken manure 20 tons/ha), P5 (40 tons/ha of chicken manure), and P6 (60 tons/ha of chicken manure), and then repeat four times. Data analysis in the form of qualitative and quantitative data. DMRT level 5% is further tested quantitatively using analysis of variance, while qualitative data is collected using the descriptive method. The study's findings

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indicate that a cow manure dose of 60 tons/ha is the best dose for plant growth, as measured by an average height increase of 42.78 cm, an increase in the number of leaves (116.65 pieces), and an increase in the number of tillers (14.45 pieces) at 18 weeks after planting. A dose of 60 tons/ha of chicken manure produced the best root length of 41.03 cm. The highest dry and wet weight yields, of about 179.75 g and 822 g, respectively, came from the rhizome weight of cow manure at a dose of 60 tons/ha. The highest secondary metabolic levels in each parameter are found in dry rhizomes (phenolic 202.79mg/L, flavonoid 181.91mg/L, and tannin 5406.33mg/L), and wet rhizomes (phenolic 178.56mg/L, flavonoid 104.39mg/L) with the highest tannin compound at around 4144.83mg/L in chicken manure dose of 40 tons/ha. According to the antioxidant results, providing chicken manure at a dose of 60 tons/ha resulted in very strong antioxidant results in each of the wet and dry rhizomes, which were respectively 9.52 ppm and 8.06 ppm.

**Keywords:** *organic fertilizer, bangle, secondary metabolism*

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## Introduction

One of the biggest producers of spices worldwide is Indonesia. Bangle is a traditional medicine that some people believe has beneficial properties for the body. Bangle (*Zingiber montanum*), a Zingiberaceae family member, has long been used in traditional medicine (Noviyanto et al., 2020). Bangle rhizome is aromatic, which distinguishes it from other Zingiberaceae family members. Bangle, in its physical form, is similar to other spices in the Zingiberaceae family (Fernandarisky et al., 2020). The bangle is rich in saponins, flavonoids, phenolic compounds, essential oils, tannins, steroids, triterpenoids, antioxidants, vitamin C, vitamin E, and carotene. According to research, Bangle rhizome extract has pharmacological activity as an antibacterial, laxative, pancreatic lipase inhibitor, and protects cells from oxidative stress caused by H<sub>2</sub>O<sub>2</sub> (Noviyanto et al., 2020).

Due to insufficient fertilization, pest control, soil management, and other cultivation techniques, bangle rhizome production frequently experiences a significant gap between the maximum and minimum yields or is said to frequently experience erratic fluctuations in yields. there aren't enough references on growing this plant. Bangle cultivation can yield fresh rhizomes weighing 10-20 tons per acre (Evizal, 2013). One very important cultivation technique is fertilization.

Other than its primary function as a stem, the rhizome may serve other purposes, the most

common of which is to serve as a storage site for certain metabolism (metabolic) products. Metabolic products can be used as drugs in some cases, but their content is sometimes uncertain and tends to be lower. Temperature, nutrition, water availability, and CO<sub>2</sub> levels in the atmosphere are all factors that can influence secondary metabolic levels (Amelia, 2015). The availability of complete and balanced nutrients that can be absorbed by plants is a factor that influences plant growth and production (Dewanto et al., 2017)

Giving the bangle plant organic fertilizer is one way to boost its own productivity. Organic fertilizers can be in the form of solid or liquid fertilizers made from dead plants, animal dung, animal parts, or other organic wastes that have undergone an engineering process. They can also be enriched with microorganisms or minerals (Permentan, 2011). Manure is one of the most commonly used organic fertilizers. Manure is processed livestock manure that is applied to agricultural land in order to improve soil fertility and structure. The nutrients in manure vary depending on the source.

According to Hartatik (2015), cow manure contains N (6 kg/ton), P (1.5 kg/ton), K (3 kg/ton), Ca (1.2 kg/ton), Mg (1 kg/ton), and S (0.9 kg/ton). Chicken manure itself contains N (15 kg/ton), P (7 kg/ton), K (8.9 kg/ton), Ca (3 kg/ton), Mg (8.8 kg/ton) and S (0.3 kg/ton). Animal manure contains a lot of nitrogen as well as metallic minerals like magnesium, potassium, and calcium. The primary benefit of manure is

that it preserves the physical structure of the soil, allowing roots to grow properly, as well as supporting the biological and chemical properties of the soil (Melsasail et al., 2019). Therefore, the purpose of fertilization is to replenish lost nutrients and increase the amount of nutrients available to plants, thereby increasing plant quality and quantity.

## Method

### Place and Time

The study was conducted from September 2021 to March 2022, lasting about 5 months. The research location was in the experimental garden of Mulawarman University Teluk Dalam, L2, Tenggarong Sebrang, East Kalimantan. The second place is in the Laboratory of Post-Harvest and Packaging of Agricultural Products, Faculty of Agriculture, Mulawarman University.

### Research Design

The study was designed using a Randomized Block Design (RAK), with one treatment using organic fertilizer in the form of cow and chicken manure. The levels were then divided into 7 groups, with P<sub>0</sub>, P<sub>1</sub>, P<sub>2</sub>, P<sub>3</sub>, P<sub>4</sub>, P<sub>5</sub>, and P<sub>6</sub> each receiving a different dose of each type of manure. The treatment was then repeated four times.

The following dosages are used:

- P<sub>0</sub>: Control (without fertilizer)
- P<sub>1</sub>: 20 ton/ha of cow manure
- P<sub>2</sub>: 40 ton/ha of cow manure
- P<sub>3</sub>: 60 ton/ha of cow manure
- P<sub>4</sub>: 20 ton/ha of chicken manure
- P<sub>5</sub>: 40 ton/ha of chicken manure
- P<sub>6</sub>: 60 ton/ha of chicken manure

The plot has a length of 6 m and a width of 1 m. Planting one seed per planting hole results in a spacing of approximately 50 x 100 cm. To prevent waterlogging and seedling rot, seedlings are planted in ditches with good drainage. In order to facilitate landfilling later, planting is done in the trench.

### Data Analysis

Observed data include: an increase in plant height; an increase in the number of leaves; an increase in the number of tillers; an increase in root length; an increase in the wet and dry weights of rhizomes; and an increase in secondary metabolic and antioxidant levels. Data will be collected every three weeks at 3, 6, 9, 12, 15, and 18 MST for height, additional number of leaves, and number of tillers. After the plants were harvested, wet weight, dry weight, root length, secondary metabolic, and antioxidant levels were measured. The analysis of variance (ANOVA) was used with a 5% confidence level. If the variance is obtained and the results show

that each treatment in each group is significantly different from each other, a 5% level DMRT test will be performed.

The results of the secondary metabolic level identification and antioxidant activity tests were analyzed qualitatively using descriptive methods. Comparing phytochemical compound levels in each treatment based on secondary metabolism levels. Antioxidant activity is assessed by comparing the IC<sub>50</sub> values in each treatment. We used spectrophotometry method for all of parameter of secondary metabolic level test.

## Results and Discussion

### Result

According to the results of an analysis of variance with an ANOVA table at the 5% level, the treatment of giving organic fertilizer in the form of chicken and cow manure is significantly different for height increase, number of tillers, the number of leaves increased, root length, wet weight of rhizomes, and dry weight of plant rhizomes. Only root length was affected significantly.

### Plant growth

Table (1) shows that when the plant is about 6 weeks old, a cow manure dose of 60 tons/ha produces the best results (on average about 26.16 cm), followed by chicken manure doses of 60 tons/ha, chicken 40 tons/ha, and cattle 40 tons/ha. In comparison to the previous week, the plant's age 9 weeks after planting showed a very rapid increase in plant height. At 9 mst, the best dose was 40 tons/ha of chicken manure with an average addition of about 32.45 cm. The best results were obtained at week 12, with a cow manure dose of 60 tons/ha yielding an average yield of 37.24 cm. Cow manure at 20 tons/ha and chicken manure at 20 tons/ha did not produce significantly different results than the control. The best dose at week 15 after planting was 60 tons/ha of cow manure, with an average depth of 40.47 cm, and 38.38 cm of chicken manure. Cow manure provided the highest dose, with an average of 42.78 cm at week 18 after planting.

### Number of Tillers

Table (2) shows that the highest number of tillers were found at a dose of 60 tons/ha of chicken manure, with an average of 1.65 at 6 weeks after planting. At the age of 9 mst, a control with an average tiller of about 2.55 produced results that were not significantly different from chicken manure 20 tons/ha. The best application this week is 60 tons/ha of cow manure with an average of 4.15 tillers. At 12 mst, the effect of manure treatment revealed that the best dose was 40 tons/ha chicken manure, with an average of 6.65. This best dose did not differ significantly

between chickens and cows manure at 60 tons/ha or chickens manure at 20 tons/ha. Cow and chicken manure at a dose of 60 tons/ha at 15 mst produced the highest yields, with cow yields of 9.50 and chicken yields of 9.00, respectively. At 18 weeks, the control did not differ significantly from the cow manure doses of 20 and 40 tons/ha. The highest dose was in cow manure at 60 tons/ha with an average of 14.45 tillers and chicken manure at 60 tons/ha with an average of 13.37 tillers.

#### Number of Leaves

Table (3) shows that after 6 weeks of treatment with a dose of 20 tons/ha of chicken manure, the results were not statistically significantly different from the control but significantly different from those other than the control. The best dose of cow manure, 60 tons/ha, was produced at 9 weeks of turbidity, with an average increase in the number of leaves of 37.95 strands. At 12 weeks, the control showed no significant difference from cow manure 20 and 40 tons/ha, as well as chicken manure 20 and 40 tons/ha. Cow manure 60 tons/ha with an average of 52.80 strands provided the best dose at 12 mst. The highest dose was found in cow manure at a rate of 60 tons/ha in week 15, with an average of 84.55. Cow manure 60 tons/ha with an average of 116.65 strands in the 18th week produced the best results with a significant difference from the control, but chicken manure 60 tons/ha with an average increase in the number of leaves 105.80.

#### Root length wet weight and Rhizome dry weight

Table (4) shows that the lowest yield in cow manure is 20 tons/ha with an average root length of 24.36 cm. The control yielded results that were not statistically different from cow and chicken manure at 20 tons/ha, as well as the highest dose. This result indicates that applying different doses of manure had no effect on the root length of the bangle plant in all treatments.

The chicken manure 20 tons/ha dosage of bangle rhizome had the lowest wet weight, with an average weight of 392.35g. While the control yielded results that were not significantly different from 20 tons/ha of cow and chicken manure as well as 40 tons/ha of chicken and cattle, the results from 40 tons/ha of chicken and cows also did not differ significantly from those from 60 tons/ha of chicken manure. Ha. Cow manure yielded the highest yield of 60 tons/ha with an average wet weight of rhizome of 822.00g and produced statistically significant differences for all treatments.

The lowest dry weight of bangle rhizome was shown by cow and chicken manure at a dose of 20 tons/ha, respectively 86.00g of cow manure and 78.25g of chicken manure. This result showed a lower weight compared to the control, with an average of 93.75g. The control yielded statistically insignificantly different results than chicken manure 20 and 40 tons/ha, cattle 20 and 40 tons/ha, and chicken 60 tons/ha, but yielded significantly different results than the best dose. The best dose is 60 tons/ha of cow manure with an average dry weight of 179.75g of rhizomes, followed by 60 tons/ha of chicken manure with an average of 135.75g

**Table 1.** Effect of Cow and Chicken Manure on Rhizome Height (in cm).

Manure Treatment (ton/ha)	Weeks after Planting (MST)				
	6	9	12	15	18
Control	11,38 a	18,61 a	24,68 a	20,41 a	18,82 a
Cow 20 ton/ha	17,46 b	20,26 ab	25,44 a	24,02 ab	28,51 b
Cow 40 ton/ha	20,51 c	27,95 b	29,81 ab	35,57 cd	37,19 c
Cow 60 ton/ha	26,16 e	31,18 b	37,24 c	40,47 d	42,78 c
Chicken 20 ton/ha	17,36 b	27,21 b	30,16 abc	29,15 bc	28,82 b
Chicken 40 ton/ha	22,84 cd	32,45 b	34,83 bc	37,61 cd	36,37 c
Chicken 60 ton/ha	25,68 de	32,17 b	36,84 bc	38,38 d	39,22 c

**Table 2.** Effect of Cow and Chicken Manure on Rhizome number of tillers (in weeks)

Manure Treatment (ton/ha)	Weeks after Planting (MST)				
	6	9	12	15	18
Control	0,8 a	2,55 a	3,95 a	5,80 a	8,60 a
Cow 20 ton/ha	0,7 b	3,45 b	5,20 b	6,60 a	10,40 ab
Cow 40 ton/ha	1,7 ac	3,40 b	5,05 ab	7,35 ab	11,55 b
Cow 60 ton/ha	1,6 c	4,15 b	6,10 bc	9,50 c	14,45 d
Chicken 20 ton/ha	1,3 bc	3,35 ab	5,70 bc	7,40 ab	11,05 b
Chicken 40 ton/ha	1,5 c	4,00 b	6,65 c	8,45 bc	12,15 bc
Chicken 60 ton/ha	1,65 c	3,75 b	6,05 bc	9,00 c	13,70 cd

**Table 3.** Effect of Cow and Chicken Manure on number of Rhizome Leaves (in weeks)

Manure Treatment (ton/ha)	Weeks after Planting (MST)				
	6	9	12	15	18
Control	4,70 a	19,60 a	35,80 a	52,25 a	77,55 a
Cow 20 ton/ha	5,55 a	25,00 ab	41,05 ab	55,65 a	83,80 a
Cow 40 ton/ha	9,65 b	28,25 abc	42,70 abc	66,00 ab	95,35 ab
Cow 60 ton/ha	10,05 b	37,95 c	52,80 c	84,55 c	116,65 c
Chicken 20 ton/ha	9,20 b	26,00 ab	40,25 ab	56,85 a	81,75 a
Chicken 40 ton/ha	9,65 b	30,90 bc	43,25 abc	67,05 bc	91,35 ab
Chicken 60 ton/ha	11,05 b	30,45 abc	50,75 bc	79,55 bc	105,80 bc

**Table 4.** Effect of Cow and Chicken Manure on Root Length and Rhizome Weight (in 18 week)

Manure Treatment (ton/ha)	Root Length (cm)	Rhizome wet weight (g)	Rhizome dry weight (g)
Control	31,90 ab	415,25 ab	93,75 ab
Cow 20 ton/ha	24,36 a	444,75 ab	86,00 a
Cow 40 ton/ha	35,36 b	530,00 bc	119,00 ab
Cow 60 ton/ha	36,50 b	822,00 d	179,75 c
Chicken 20 ton/ha	33,20 ab	393,25 a	78,25 a
Chicken 40 ton/ha	38,95 b	487,50 abc	99,75 ab
Chicken 60 ton/ha	41,03 b	618,75 c	134,75 bc

**Notes:** Based on Duncan's multiple-distance further test (DMRT) at a 5% significance level, values in the same columns that are followed by the same letter do not differ significantly.

**Table 5.** Secondary Metabolic Levels in Bangle Rhizome in Each Treatment (mg/L).

Condition	Treatment	Level (mg/L)		
		Phenolic	Flavonoid	Tanin
Fresh	Control	31,64	23,08	2034,83
	Cow 20 ton/ha	43,18	41,55	2991,50
	Cow 40 ton/ha	77,03	64,30	3151,50
	Cow 60 ton/ha	86,90	71,14	3198,17
	Chicken 20 ton/ha	97,67	74,79	4086,50
	Chicken 40 ton/ha	107,54	78,71	4144,83
	Chicken 60 ton/ha	178,56	104,39	3861,33
Dry	Control	124,97	38,44	1932,33
	Cow 20 ton/ha	133,56	67,23	2504,00
	Cow 40 ton/ha	148,44	68,76	3034,83
	Cow 60 ton/ha	181,38	99,43	4734,67
	Chicken 20 ton/ha	175,23	120,06	4903,00
	Chicken 40 ton/ha	198,69	156,10	5088,00
	Chicken 60 ton/ha	202,79	181,91	5406,33

**Note:** The number followed by yellow denotes the best outcome in each observation variable for the flavonoid test, tannin test, and phenolic test results.

**Table 6.** Antioxidant based on IC50 value

Antioxidant (IC50) (mg/L)	Fertilizer Dose ton/ha						
	Control	Cow			Chicken		
		20	40	60	20	40	60
Fresh Rhizome	53.58	48.55	47.36	47.14	32.73	27.42	9.52
Dry Rhizome	40.91	38.30	34.78	34.46	24.41	14.05	8.06

**Note:** The smaller the IC50 value, the stronger the antioxidant



### Secondary metabolic level

In accordance with the findings of the secondary metabolism test, the rhizome of the bangle plant contained phenolic, flavonoid, and tannin-containing active substances, and also steroid, alkaloid, terpenoid qualitatively. Different levels of the compounds involved in this secondary metabolism are evident in each fertilizer application. The results are shown in table (5). Different concentrations were produced by the concentration of active substances in the bangle rhizome. The aforementioned data demonstrate that, despite using the same amount of fertilizer, the active compound content of the bangle rhizome is highest when the rhizome is dry (low moisture content) as opposed to when the rhizome is still fresh.

The amount of chicken manure that produces the highest phenolic content in the rhizome under wet and dry conditions, correspondingly around 178.56 mg/L and 202.79 mg/L, is 60 tons/ha. A dose of 60 tons/ha of chicken manure also provides higher levels of active flavonoids and tannins than other doses. When compared to the fertilizer treatment, the control had the lowest active

compound content. When treated with cow manure, yields are lower than when treated with chicken manure at the same dose.

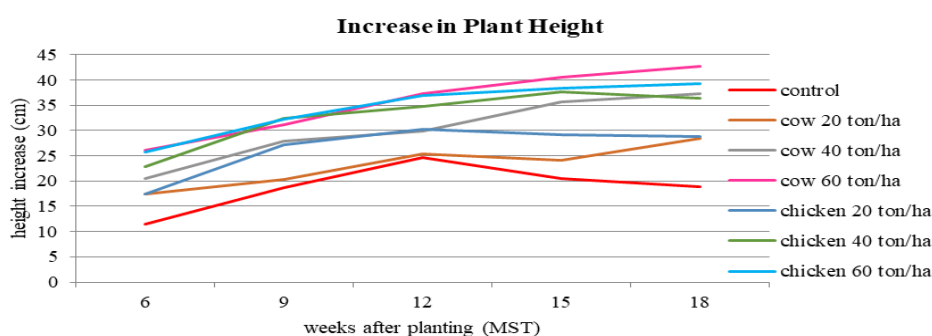
### Antioxidant

Antioxidant analysis of bangle rhizomes in table (6), using several samples based on manure dose and fresh and dry rhizome condition.  $IC_{50}$  value was obtained, which describes how well the sample can capture free radicals. The dose of 60 tons/ha of chicken manure was found to have the lowest  $IC_{50}$  value in the fresh sample, with a value of 9.52 ppm, while the control had the highest value, with an  $IC_{50}$  value of 53.58 ppm in fresh rhizome conditions.

The same quality was obtained when dry rhizome conditions were used for the analysis; specifically, the dose of 60 tons/ha of chicken manure produced the lowest  $IC_{50}$  value and the largest control. The plant's antioxidants are more potent the lower the  $IC_{50}$  value. Cow manure is always treated with an  $IC_{50}$  value higher than chicken manure.

### Discussion

#### Plant Height



According to the table 1, the average increase in plant height starting at 6,9,12,15, and 18 WAP produced mixed results that tended to fluctuate. At the age of 6 weeks, the treatment without fertilizer (control) continued to grow until week 12 when it reached its peak with an average height increase of about 24.68 cm, before adding less height the following week. In comparison to other treatments, the results from the fertilizer-free treatment had the lowest graph. In order for a plant to grow, nutrients must be obtained from the soil by the roots through their root hairs (Sudewi et al., 2022). Organic matter affects plant growth by influencing the physical, chemical, and biological properties of the soil ( Anwar, 2013). The more organic matter provided, the faster the plant will grow. Compared to chicken manure, cow manure typically produces better results. According to

(Hartatik et al., 2015) cow manure contains N (6 kg/ton), P (1.5 kg/ton), K (3.0 kg/ton), Ca (1.2 kg/ton), Mg (1.0 kg/ton), and S (0.9 kg/ton), and these nutrients can support the growth of Bangle plants, which have a long harvest period. Cow manure's graph tends to rise with each observation.

Chicken manure at all levels showed a significant increase at 9 weeks after planting, but at 12 weeks the increase in plant height slightly decreased in plant height. In chicken manure at a level of 20 tons/ha, the height increase was averaged at 30.16 cm after 12 weeks and decreased the following week. High nitrogen elements are found in chicken manure. Although the amount of nitrogen plants require is always higher than that of other nutrients, a deficiency or excess can hinder and disrupt plant growth (Raja et al., 2021). After planting, the growth rate of the

bangle plant accelerated between 2 and 5 months. As plants get older, their growth rate for height starts to slow down (Rosita et al., 2005).

According to some research, applying chicken manure always results in the best plant response in the first growing season. This is because chicken manure decomposes relatively quickly and has enough nutrients compared to other manures of the same weight (Hartatik et al., 2015). The graph shows that chicken manure tends to increase quickly at 6, 9, and 12 weeks of age, then declines as plant age increases. Large-scale application of chicken manure is thought to be less effective because the nutrients will exhaust quickly.

The same result was also shown by chicken manure at a dose of 40 tons/ha which decreased the addition of plant height, the maximum height increase at this dose was at 15 weeks with an average height increase of 37.61 cm. When compared to the other two doses of chicken manure, the results for the 60 ton/ha level of chicken manure were a little bit different. A graph that increases over the course of 18 weeks can be used to demonstrate this, but the increase in plant height is typically not too different from the previous week. Even though cow manure at a dose of 40 tons/ha initially produced fewer yields than chicken manure at a dose of 40 tons/ha, at the end of the observation at 18 weeks the increase in height was more apparent and might even have exceeded that of chicken manure at a dose of 40 tons/ha, which caused a decrease in plant height.

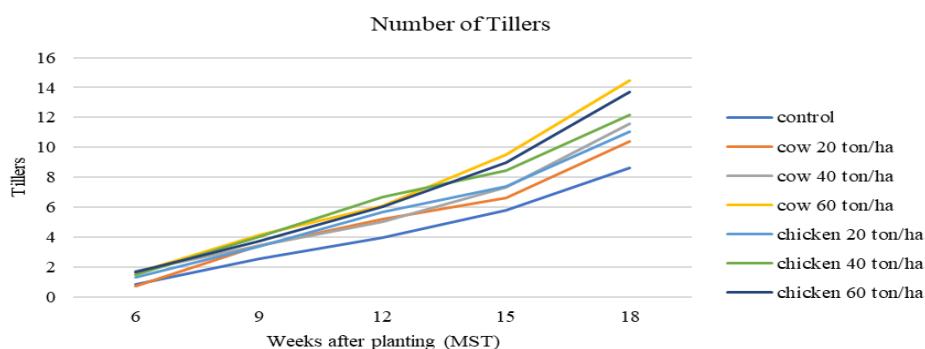
The addition of cow manure improves the physical properties of the soil. Improved soil physical

characteristics include things like increased permeability, total pore space, aggregate stability, volume weight, texture, color, temperature, and others (Sudewi et al., 2022). A dose of 60 tons/ha of chicken and cow manure has the tendency to produce steady, dependable results. Cow manure doses of 60 tons/ha tend to yield less than chicken manure doses of 40 and 60 tons/ha from the start of planting until the plant is 9 weeks old, but the yields increase the following week.

The plant's need for nutrients grows as it ages. If the nutrient requirements are not met and the nutrients are not readily available, plants may experience nutrient deficits at specific times. According to Rosita et al. (2005), bangle plants absorb N (0.06 - 3.07 g), P (0.01 - 0.53 g), and K at 2 to 10 months after planting in the canopy (0.10 to 2.25 g). N is the nutrient that is most required in the plant canopy itself. The primary nutrient for plants, nitrogen, is typically essential for the development and expansion of vegetative parts of plants, such as leaves, stems, and roots (Purba et al., 2021). A sufficient supply of plant N is indicated by high photosynthetic activity, good vegetative growth, and dark green plant colors (Nurhayati, 2021).

Due to the individual characteristics of each animal, which are influenced by the type of feed and the animal's age, each manure contains a different mix of nutrients (Nurjanah et al., 2020). Because each treatment dose of fertilizer has a different nutrient content, they all produce different yields and have different recommended doses. Due to their movement with crop yields, surface runoff, erosion, or evaporation, nutrients in the soil will gradually decrease over time (Lawenga et al., 2015).

Number of Tillers



The table of the total number of tillers from each level reveals results that increase with plant aging and are influenced by the quantity of fertilizer applied, as shown in the figure below. Plants without fertilizer developed more tillers every week, but the growth was typically modest. This slight increase resulted from the fact that

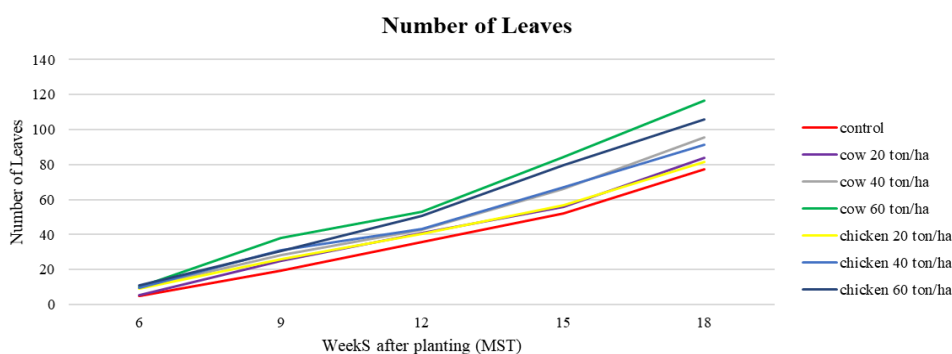
during the initial stages of planting, the products of photosynthesis were utilized for the vegetative development of plants.

In comparison to manure application, treatment without fertilizer produced the lowest yield. The graph above makes it easy to see that there was a noticeable increase

at plants that were 18 WAP of age. A plant needs nutrients for its physiological processes during growth and development. Plant growth and production will be subpar due to a lack of nutrients (Purba et al., 2021).

Functions of organic matter as a biological buffer so that the soil can supply plants with a balanced amount of nutrients (Hartatik et al., 2015). Loosening the topsoil, increasing water absorption and storage, and boosting soil fertility are all important functions of manure (Yulianto et al., 2021). A sudden rise in the number of tillers can result from the ease with which new shoots can emerge from loose, moist soil.

At the start of planting, there were typically fewer and nearly identical numbers of tillers in each treatment. The nutrients in this fertilizer are not readily available to plants, which is the cause of the slow plant growth at the **Number of Leaves**



An increasing number of leaves are produced each week as a result of the weekly application of cow and chicken manure. The table 3 shows that at about 12 weeks of age, the number of leaves increases significantly. Every week, the increase varies depending on the treatment. The number of leaves significantly increased with a dose of 60 tons/ha of cow manure, averaging 15–37 leaves every three weeks. The number of similar-looking leaves tends to increase when manure is applied in the same amount. Both cow and chicken manure at a dose of 20 tons/ha and chicken and cow manure at a dose of 40 tons/ha produced nearly identical results during planting.

In addition to the nutrients that plant's needs, manure also contains humic, fulvic acids, growth hormones, and other substances that promote plant growth and increase nutrient uptake by plants (Hartatik et al., 2015). The amount of photosynthesis is influenced by the number of leaves present, and plants with more leaves may produce heavier and bigger rhizomes as a result.

The number of leaves added is also affected by the number of shoots and plant height. The number of leaves will increase as the plant ages and grows taller, produces more leaves on a single stem, and produces more tillers.

start of the planting period. The extent of these materials' mineralization or decomposition has a significant impact on the nutrients' availability. Manure's low nutrient availability is partially caused by the presence of N, P, and other elements in complex compounds that are challenging to decompose (Hartatik et al., 2015).

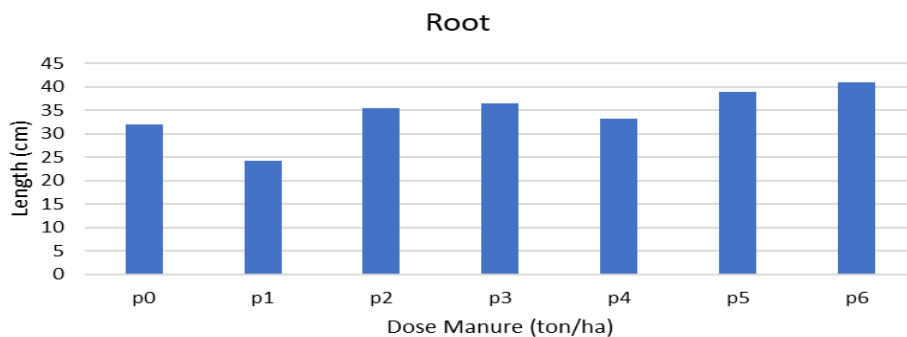
At 6 mst, all treatments tended to be similar and the differences between the tillers in each treatment tended to be minimal. Although cow manure at a dose of 60 tons/ha is the best dose with an average number of tillers of 14.45 tillers, chicken manure typically produces better results than cow manure at the same dose. In comparison to other manure doses, chicken manure 40 tons/ha at 12 mst produced the best results with 6.65 tillers.

The nutrients required for plant growth are present in sufficient amounts in manure. The related observation variables will be impacted by the food's growth quality and planting age.

In relation to the addition of the number of leaves, the most influential element is N. In comparison to other nutrients, nitrogen is required in sufficient amounts for plant growth. N makes up 40–50% of the dry weight of protoplasm, the living component of plants (Nurhayati, 2021). Since protein is the source of all plant enzymes, nitrogen participates in all enzymatic processes in plants. Additionally, nitrogen is one of the constituent elements of chlorophyll, the primary component of chloroplasts, and it contributes to improving the quality and quantity of the dry matter produced (Dr. Vladimir, 2021).

Fertilizer use and the amount of nutrients in the soil have a significant impact on how plants grow and develop. Nutrient uptake is restricted by nutrients in a minimum state (Purba et al., 2021). In terms of the addition of leaves, the treatment of plants without fertilizer differs significantly enough for each observation. In comparison to other treatments, plants without fertilizer produce the lowest yield.

## Root Length



The results of the lowest root length were cow manure dose of 20 tons/ha with an average root length of about 24.36 cm and lower than the control. With an average root length of 41.03 cm, chicken manure had the longest roots. When plants respond to water shortages by reducing the rate of transpiration in order to conserve water, the roots play a crucial role (Torey et al., 2013). Plant roots have a significant impact on overall plant growth and development. The failure of the root function will result in a complete change in the plant for the top (Nurhayati, 2021).

Manure has the ability to bind water in the soil. Because the soil around the roots in the deeper layers is still moist, the roots will continue to grow. Maximizing exposure to groundwater will encourage the growth of roots (Torey et al., 2013). Plant roots directly respond to the physical characteristics of the soil (Lawenga et al., 2015).

The table's data, which was derived from root length in chicken manure, generally produces better results than cow manure. The use of this fertilizer can loosen the soil, increase aeration, and increase the soil's capacity to hold water, all of which can improve the physical properties of the soil (Raja et al., 2021). Additionally, organic matter has the ability to control soil temperature, slow down phosphorus fixation, increase soil cation exchange capacity, and lessen the leaching of nutrients like potassium, calcium, and magnesium (Guimarães et al., 2019). Another environmental factor that has been shown to affect the nitrate absorption process is the temperature around the roots (Dr. Vladimir, 2021).

The initial analysis of the soil revealed that the soil's pH ranged from 3.86 to 4.86. Al elements are common in excessively acidic soil, and they can poison plants and bind phosphorus (P). Low pH soil can hinder plant growth by preventing the roots from properly absorbing nutrients. Giving chicken manure up to 5–25 tons/ha, as demonstrated by Tufail et al. (2014), shows that manure can raise the pH of soil from 5.0 to 5.8–6.4. By raising

pH, Al in exchangeable form will be reduced and nutrients will become more available to plants.

Nutrient uptake on the roots of bangle plants at 2-10 months after planting was as follows: N (0.01 - 0.52 g), P (0.002 - 0.15 g), and K (0.02 to 0.82 g) (Rosita et al., 2005). It was discovered that the roots of the bangle plant had more K buildup than N and P. K is primarily used to aid in the synthesis of proteins and carbohydrates. In the face of drought, illness, and pests, potassium gives plants strength (Purba et al., 2021). Organic fertilizers can help the soil's physical and chemical composition, which will facilitate root development. Up until the soil reaches its critical water potential, plant roots expand into moist soil and draw water (Solichatun et al., 2005). The looseness of the soil can promote root development. Strong roots will make it simpler for plants to absorb nutrients and water

### **Rhizome Wet Weight**

Plant biomass is a common parameter used to study plant growth. The fresh weight of the plant describes the water and moisture content of the plant. The plant will weigh more when wet the more fertile it is (Supandi, 2021). When plant nutrient requirements are met, yields will be optimal (Purba et al., 2021). The rhizome of the bangle plant is the part that is most advantageous for cultivation. One could also argue that this rhizome's wet weight is a crucial factor in determining how well bangle plant cultivation is going. The cultivation method is better and more productive the more weight of the wet rhizome that can be obtained.

Rosita (2005) found that giving 250 kg/ha of urea, 250 kg/ha of SP36, 250 kg/ha of KCl, and 20 tons/ha of manure resulted in a fresh bangle rhizome weight of 311.39 g/plant 5 months after planting. According to the results of the application of organic fertilizers, at the age of 18 weeks, cow manure at a dose of 60 tons/ha produced the highest wet weight of rhizomes, averaging about 822

grams/plant, and chicken manure at a dose of 20 tons/ha produced the lowest wet weight of rhizomes, an average of 393.25 g/plant. Manure increases crop yield and quality while also enhancing the chemistry, physical characteristics, and biological properties of the soil (Seker et al., 2011). The graph below demonstrates that the yield of fresh rhizomes increases with increasing manure dosage.

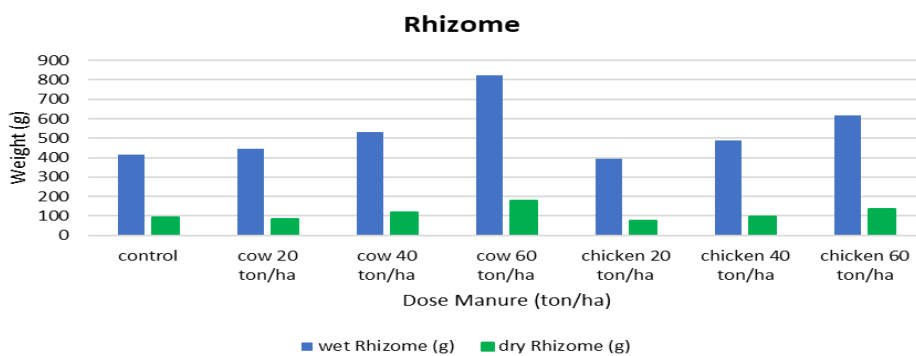
Hasil It is impossible to separate the good physical qualities of the soil from the yield of this fairly large plant. The physical condition of the soil must support plant growth in addition to a supply of nutrients that is adequate and balanced (Lawenga et al., 2015). These soil aggregates will keep the soil in a loose condition (Anwar, 2013). Cow manure will enhance the physical characteristics of the soil. Improved soil physical characteristics include things like increased permeability, total pore space, aggregate stability, volume weight, texture, color, temperature, and others (Sudewi et al., 2022).

Intensive tillage affects the physical properties of the soil. Low organic matter soils will have more severe damage to the soil's structure (Anwar, 2013). When the soil does not receive enough water and becomes dense and hard, soil damage is evident. Plant rhizomes won't be able to grow or spread out in compacted or hard soil. The ability to maintain loose soil conditions that are difficult to harden or compact increases with the amount of organic matter added.

Additionally, manure helps to improve soil structure, cation exchange capacity, and water resistance. Giving manure has the indirect effect of making it simpler to keep water in the soil (Yuliana et al., 2015). Since water availability plays a significant role in plant growth, water frequently restricts the growth and development of cultivated plants.

The plants will experience drought conditions if there is not enough water in the soil. Due to decreased primary metabolism, reduced leaf area, and decreased photosynthetic activity, drought stress can lower plant productivity (biomass). Smaller leaves grow as a result of a lack of water during the vegetative stage, which can reduce light absorption. Lack of water also inhibits the synthesis of chlorophyll and some enzymes, such as nitrate reductase, from working (Solichatun et al., 2005).

Organic substances in the soil may have physiological effects on plant growth that are direct or indirect (Syaiful Anwar, 2013). Compared to other types of manure, chicken manure contains a fair amount of P. This is due to the fact that chicken manure contains feed (Sudewi et al., 2022). Phosphorus aids in the growth of plant roots, photosynthesis, transfer respiration, cell division, and growth (Supandi, 2021). The number of cells increases more quickly when they divide quickly, which causes the rhizome to grow larger.



### Rhizome Dry Weight

The relationship between plant growth and the quantity and concentration of mineral nutrients in plant tissues is known as dry weight (Purba et al., 2021). Dry weight reflects plant nutritional status because it is affected by the rate of photosynthesis and respiration in each treatment. Based on the collected data, it was determined that applying chicken and cow manure at a dose of 20 tons/ha resulted in lower dry weight yields than the control. Additionally, the results from the 40 tons/ha

manure dose were not significantly different from the control. Additionally, the results from the 40 tons/ha manure dose were not significantly different from the control. Many nutrients, such as N, P, and K, build up in the canopy when the bangle plant is between 2 and 7 BST, according to Rosita et al (2005). This is the active vegetative formation stage.

Cow manure at a dose of 60 tons/ha demonstrated different results and produced significantly better outcomes than other doses and types of manure. The

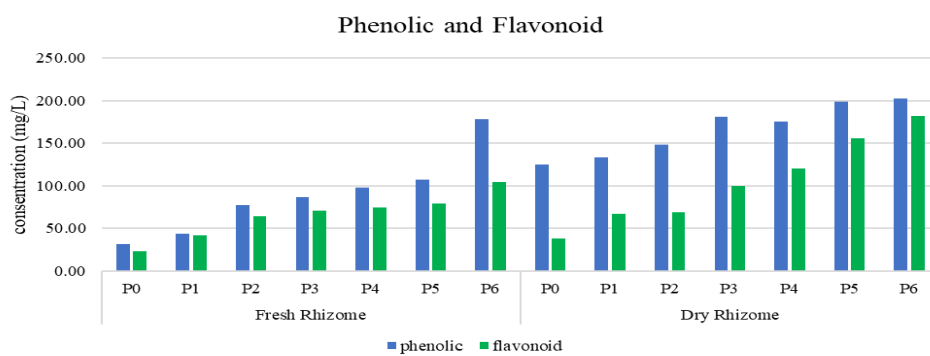
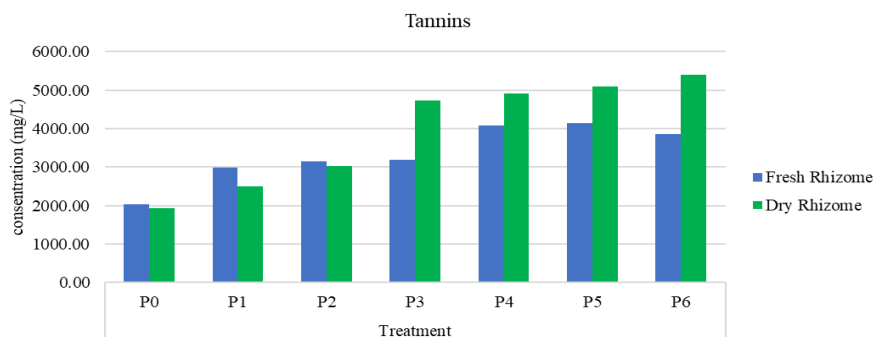
amount and timing of fertilizer applied can have an impact on crop yields, among other things. Organic matter plays a crucial role in soil health because it can create stable soil aggregates, increase soil fertility, and serve as a source of energy for organisms (Nurjanah et al., 2020).

Based on earlier studies on dry matter production and nutrient content in Bangle plant tissue, the N, P, and K amounts of nutrient uptake in dry rhizomes of Bangle plants aged 2 to 10 months were N (0.01 to 4.89 g), P (0.002 to 1.04 g), and K (0.01 to 2.34 g), respectively (Rosita et al., 2005). It is clear that as plants get older, they are able to absorb an increasing amount of N, P, and K nutrients. As can be seen, N is the nutrient that has accumulated in the rhizome in the greatest amounts.

The application of manure enhances the chemical, physical, and biological properties of the soil increase crop yield, and improves crop quality (Yolcu, Turan, et al., 2011). High organic matter soils have beneficial microorganisms that encourage the breakdown of organic matter and release inorganic nutrients that are then available for plant uptake (Seker, et al., 2011). Organic fertilizers can help to create ideal conditions in the soil for microorganisms that are beneficial to plants.

Chicken manure is an organic fertilizer with high nitrogen content, despite not being the best dose for bangle rhizome weight yield. As they ensure the best nutrient management for plants, such fertilizers should be used promptly to partially replace chemical fertilizers (Guimarães et al., 2019).

**Secondary metabolic levels (phenolic compounds, flavonoids and tannins)**



The results of laboratory analysis found that the positive bangle rhizome contains compounds in the form of phenolics, tannins and flavonoids. These findings are consistent with Amalia et al (2021), which found that the Bangle plant contains secondary metabolism in the form of alkaloids, phenolic compounds, flavonoids, saponins,

and triterpenoids. These compounds respond differently to the concentration of organic fertilizer applied with cow and chicken manure. The graph above shows the outcomes of these phytochemicals.

According to the graph above, the amount of tannin compounds will increase as manure dosage rises. As can be seen, just like with chicken manure, cow manure with three doses of 20, 40, and 60 tons always increased the tannin concentration in the rhizome as the dose increased. When compared to cow manure, applying chicken manure produces significantly better results. It is evident that using 20 tons/ha (P4) of chicken manure instead of 60 tons/ha (P4) of cow manure resulted in a higher concentration (P3).

Dry rhizomes produced a higher concentration of tannins than fresh rhizomes, which produced different results regarding tannin concentration. The concentration of tannins in fresh rhizomes increased non-significantly with the addition of a dose of cow manure, whereas the concentration of tannin compounds in fresh rhizomes decreased with an increase in the dose of cow manure. In comparison to the use of manure in fresh and dry rhizomes, the control provided the lowest tannin concentration.

Tannins are chemical substances that have an astringent and bitter flavor. These substances act as controlling substances in plant metabolism as well as important defenses against herbivores and pests that prey on plants (Julianto, 2019). Tannins are metabolically active substances with multiple uses, including as astringents, antibacterial agents for treating diarrhea, and antioxidants. Leather tanning is another industrial application for tannins (Amelia, 2015). Tannic acid is the type of tannin substance present in bangle plants. Diarrhea can be effectively treated with tannic acid. Additionally, tannic acid exhibits antimicrobial, antienzymatic, antioxidant, and antimutagenic properties (Hidjrawan Yusi, 2018)

In addition to increasing the fertilizer dose, applying organic fertilizer in the form of cow and chicken manure resulted in an increase in phenolic compounds and flavonoids. When compared to when the rhizome was fresh, the dry rhizome had a higher concentration of phenolic and flavonoid compounds. The content of active compounds in the *simplicia* is impacted by the drying process. Antioxidant activity is influenced by the total phenolic and flavonoid content (Amelia, 2015). Given that fresh samples are more prone to damage and experience a quicker loss in quality than dry samples, it is advised to use dry samples instead of fresh samples (Julianto, 2019).

The highest phenolic and flavonoid concentrations were found in both fresh and dry rhizomes when chicken

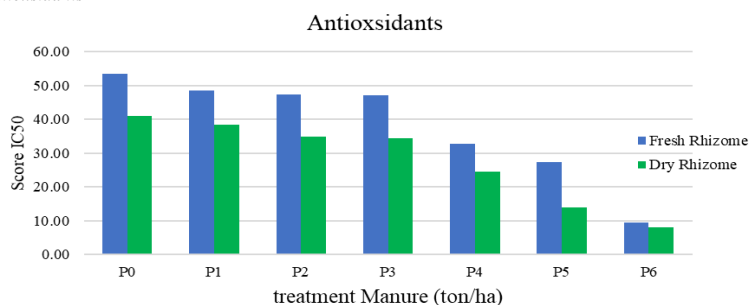
manure was applied at a dose of 60 tons/ha. The highest dose of cow manure, 60 tons/ha, resulted in phenolic compound concentrations of 86.90 mg/L in fresh rhizome and 181.38 mg/L in dry rhizome, while flavonoids were 71.14 mg/L in fresh rhizome and 99.43 mg/L in dry rhizome. Compared to chicken manure at the same dose, this result is smaller. A dose of 60 tons/ha of chicken manure resulted in phenolic compound concentrations in fresh rhizome of 178.56 mg/L and dry rhizome of 202.79 mg/L, while fresh rhizome flavonoids were 104.39 mg/L and dry rhizome flavonoids were 181.91 mg/L. In comparison to treatments with organic fertilizers, treatments without fertilizer produced lower phenolic and flavonoid concentrations.

In plants, flavonoids serve as pigments for the flowers, fruits, and roots, as well as occasionally as growth regulators and disease resistance (Julianto, 2019). Catechins are one class of flavonoid compounds present in bangle rhizomes (*catechins*). Catechins have antioxidant properties, and because they can stop the growth of viruses, bacteria, tumors, and fungi, they can also get rid of rotten and rancid odors (Aprilliza AM et al., 2021). Phenolic compounds are compounds that plants make in response to environmental stress. Phenolic compounds protect DNA from dimerization and damage by blocking UV-B rays and cell death (Hanin & Pratiwi, 2017). Gallic acid is the type of phenolic compound found in bangle rhizome. Gallic acid serves as an antibacterial, antiviral, analgesic, and antioxidant in medicine (Junaidi & Anwar, 2018).

The application of chicken manure resulted in a higher concentration of secondary metabolism because it had a relatively higher P nutrient content than other manures (Simanungkali et al., 2006). Phosphorus can be found in DNA, RNA, and the parts of nucleotides that provide metabolic energy (like ATP). The process of photosynthesis depends heavily on phosphorus. Stunted growth is one of phosphorus deficiency's signs (Khairuna, 2019). The amount of P<sub>2</sub>O<sub>5</sub> in cow manure is 0.2%, compared to 1.3% in chicken manure (Simanungkali et al., 2006). The concentration of secondary metabolism in the form of tannins, phenolic compounds, and flavonoids is influenced by the difference in the P nutrient content between these 2 types of manure. Environmental factors affect the levels of flavonoids and other phenolic compounds in plants, which vary among parts, tissues, and ages of plants. These include air temperature, nutrient availability, water availability, and atmospheric CO<sub>2</sub> concentrations (Amelia, 2015).

#### ***Antioxidants***

## Antioxidants



The antioxidant activity analysis produced different IC<sub>50</sub> values depending on the type of fertilizer used. The IC<sub>50</sub> decreases as the fertilizer

dose increases. A concentration known as IC<sub>50</sub> is capable of reducing 50% of DPPH free radicals. The greater the antioxidant activity, the lower the IC<sub>50</sub> value (Widyasanti et al., 2016). Antioxidants are compounds that can absorb or neutralize free radicals, thereby preventing certain diseases caused by free radicals (Parwata, 2016).

The treatment of cow manure, as shown in the graph, results in lower yields than chicken manure. The antioxidant activity of the dried rhizome samples was higher than that of the fresh rhizomes. The highest antioxidant activity was produced by cow manure at a dose of 60 tons/ha, with an IC<sub>50</sub> value of 9.52 ppm for fresh rhizome and 8.6 ppm for dry rhizome. At a dose of 20 tons/ha in both fresh and dry rhizome conditions, cow manure had a lower IC<sub>50</sub> value than chicken manure. For fresh rhizomes, the treatment without fertilizer produced an IC<sub>50</sub> value of around 53.58 ppm, and for dry rhizomes, it was around 40.91 ppm.

Widyasari et al (2016) claim that the antioxidant activity in bangle rhizome is incredibly powerful. Wartano et al (2016) classified antioxidants into five groups based on their IC<sub>50</sub> values: 50 very strong, 50-100 strong, 101-250 moderate, 251-500 weak, and >501 inactive (Wartono et al., 2021). Except for the treatment without fertilizer in wet rhizome conditions, the IC<sub>50</sub> value in bangle rhizome in all treatments gave a value of 50 and included a very potent antioxidant. The high secondary metabolic compounds found in the bangle rhizome are inextricably linked to the high antioxidant activity. Secondary plant metabolites like flavonoids and phenolics play a part in antioxidant activity. More phenolic compounds will have a higher level of antioxidant activity (Amelia, 2015).

## Conclusion

A cow manure dose of 60 tons/ha is the best dose for plant growth, with an average height increase of 42.78 cm, an increase in the number of leaves of 116.65 pieces, and an increase in the number of tillers of 14.45. A dose of 60 tons/ha of chicken manure produced the best root length of 41.03 cm.

The weight of the rhizomes revealed that the application of cow manure at a dose of 60 tons/ha resulted in the highest yields of dry weight and wet weight, which were about 179.75 g and 822 g, respectively.

A chicken manure dose of 60 tons/ha results in the highest secondary metabolic rate in each parameter, including dry rhizome (phenolic 202.79mg/L, flavonoid 181.91mg/L, and tannin 5406.33mg/L) and wet rhizome (phenolic 178.56mg/L, flavonoid 104.39mg/L), while a chicken manure dose of 40 tons/ha results in the highest tannin. Giving chicken manure at a dose of 60 tons/ha produced very strong antioxidant results at 9.52 ppm wet rhizome and 8.06 ppm dry rhizome, according to the antioxidant results.

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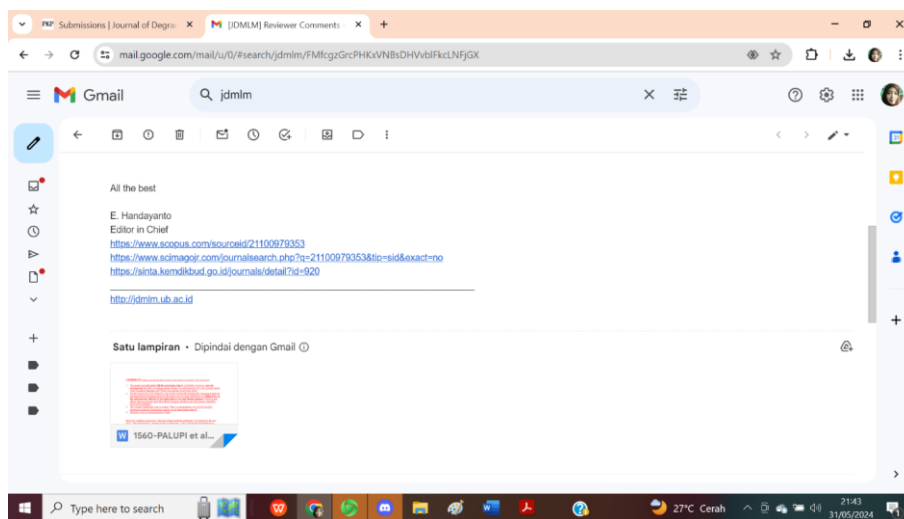
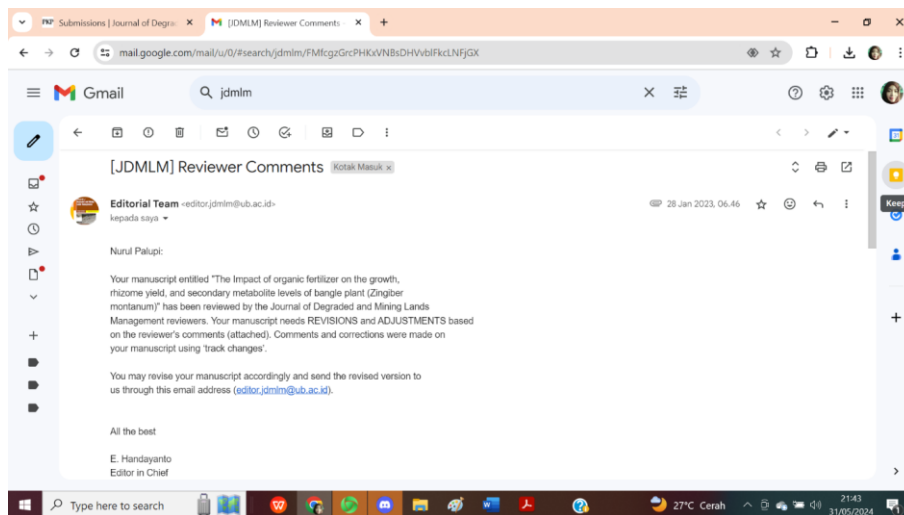
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## 2. Mendapat jawaban dari Editor JDMLM



Revisi 1 :

**COMMENTS:** (please see the detailed corrections and comments inserted in this manuscript)

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Open Access

1. This manuscript **only deals with the agronomic aspects of *Zingiber montanum*, not soil management** (the effect of adding organic fertilizer on soil properties) → is the soil used in this study considered degraded soil? What is the soil type used in this study?.
2. For this manuscript to fit within the scope of this journal (the management of degraded land/soil and management of mining land/soil), the authors have to **make adjustments by adding data on the soil properties affected by the application of cow and chicken manures**, which in turn affects plant growth and yield. The effects of organic fertilizers on soil properties should be discussed accordingly.
3. The rationale behind this study is unclear. What was the problem to be solved? *Zingiber montanum* production (agronomic aspects) or soil management aspects?
4. Methods are trivial (not presented in detail)

Please be careful in using tenses; when describing Methods and Results, you should use the past tense. The present tense is appropriate for accepted facts, such as the background information presented in the Introduction. In addition, you may use the present tense when you discuss your results and conclusions

**Recommendation: revisions and adjustments are required;** a substantial amount of work is necessary to raise this manuscript to a standard research article

## **The impact of organic fertilizer on the growth, rhizome yield, and secondary metabolite levels of *Zingiber montanum* grown on degraded soils → soil types? (Ultisols? Inceptisols?)**

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### **Abstract**

Indonesia is a major producer of spices in the world. Bangle (the local name of *Zingiber montanum*) is one of the traditional medicines that some people have heard has beneficial effects on the body. Although the bangle has a lot to offer in terms of content and advantages, there is still a huge gap in market demand, which is caused by the cultivation method's shortcomings. One way to boost the bangle plant's productivity is to provide manure. The goal of this study was to establish the ideal manure dosage for bangle plant growth, rhizome yield, and secondary metabolic antioxidant content. The study employed a Randomized Block Design (RAK), with one treatment consisting of organic fertilizer in the form of cow manure and chicken manure, and the levels were divided into seven, namely P0 (control), P1 (cow manure 20 t/ha), P2 (cow manure 40 tons/ha), P3 (cow manure 60 tons/ha), P4 (chicken manure 20 t/ha), P5 (40 t/ha of chicken manure), and P6 (60 t/ha of chicken manure), and then repeated four times. Results of this study indicated that a cow manure dose of 60 t/ha is the best dose for plant growth, as measured by an average height increase of 42.78 cm, an increase in the number of leaves (116.65 pieces), and an increase in the number of tillers (14.45 pieces) at 18 weeks after planting. A dose of 60 t/ha of chicken manure produced the best root length of 41.03 cm. The highest dry and wet weight yields, of about 179.75 g and 822 g, respectively, came from the rhizome weight of cow manure at a dose of 60 t/ha. The highest secondary

metabolic levels in each parameter were found in dry rhizomes (phenolic 202.79 mg/L, flavonoid 181.91 mg/L, and tannin 5406.33 mg/L), and wet rhizomes (phenolic 178.56 mg/L, flavonoid 104.39 mg/L) with the highest tannin compound at around 4144.83mg/L in chicken manure dose of 40 t/ha. According to the antioxidant results, providing chicken manure at a dose of 60 t/ha resulted in very strong antioxidant results in each of the wet and dry rhizomes, which were respectively 9.52 ppm and 8.06 ppm.

**Keywords:** *organic fertilizer, bangle, secondary metabolism*

## **Introduction → in this section, the existing problems of the soils used for growing Zingiber must be described**

One of the biggest producers of spices worldwide is Indonesia. Bangle (the local name of *Zingiber montanum*) is a traditional medicine that some people believe has beneficial properties for the body. Bangle (*Zingiber montanum*), a Zingiberaceae family member, has long been used in traditional medicine (Noviyanto et al., 2020). Bangle rhizome is aromatic, which distinguishes it from other Zingiberaceae family members. Bangle, in its physical form, is similar to other spices in the Zingiberaceae family (Fernandarisky et al., 2020). The bangle is rich in saponins, flavonoids, phenolic compounds, essential oils, tannins, steroids, triterpenoids, antioxidants, vitamin C, vitamin E, and carotene. According to Noviyanto et al. (2020), Bangle rhizome extract has pharmacological activity as an antibacterial, laxative, pancreatic lipase inhibitor, and protects cells from oxidative stress caused by H<sub>2</sub>O<sub>2</sub>.

Due to insufficient fertilization, pest control, soil management, and other cultivation techniques, bangle rhizome production frequently experiences a significant gap between the maximum and minimum yields or is said to frequently experience erratic fluctuations in yields. There are not enough references for growing this plant. Bangle cultivation can yield fresh rhizomes weighing 10-20 tons per acre (Evizal, 2013). One very important cultivation technique is fertilization.

Other than its primary function as a stem, the rhizome may serve other purposes, the most common of which is to serve as a storage site for certain metabolism (metabolic) products. Metabolic products can be used as drugs in some cases, but their content is sometimes uncertain and tends to be lower. Temperature, nutrition, water availability, and CO<sub>2</sub> levels in the atmosphere are all factors that can influence secondary metabolic levels (Amelia, 2015). The availability of complete and balanced nutrients that can be absorbed by plants is a factor that influences plant growth and production (Dewanto et al., 2017)

Giving the bangle plant organic fertilizer is one way to boost its own productivity. Organic fertilizers can be in the form of solid or liquid fertilizers made from dead plants, animal dung, animal parts, or other organic wastes that have undergone an engineering process. They can also be enriched with microorganisms or minerals (Permentan, 2011). Manure is one of the most commonly used organic fertilizers. Manure is processed livestock manure that is applied to agricultural land in order to improve soil fertility and structure. The nutrients in manure vary depending on the source.

According to Hartatik (2015), cow manure contains N (6 kg/t), P (1.5 kg/t), K (3 kg/t), Ca (1.2 kg/t), Mg (1 kg/t), and S (0.9 kg/t). Chicken manure itself contains N (15 kg/t), P (7 kg/t), K (8.9 kg/t), Ca (3 kg/t), Mg (8.8 kg/t) and S (0.3 kg/t). Animal manure contains a lot of nitrogen as well as metallic minerals like magnesium, potassium, and calcium. The primary benefit of manure is that it preserves the physical structure of the soil, allowing roots to grow properly, as well as supporting the biological and chemical properties of the soil (Melsasail et al., 2019). Therefore, the purpose of fertilization is to replenish lost nutrients and increase the amount of nutrients available to plants, thereby increasing plant quality and quantity.

## **Materials and Methods**

### ***Place and Time***

The study was conducted from September 2021 to March 2022, lasting about 5 months. The research location was in the experimental field of Mulawarman University Teluk Dalam, L2, Tenggarong Sebrang, East Kalimantan. The second place is in the Laboratory of Post-Harvest and Packaging of Agricultural Products, Faculty of Agriculture, Mulawarman University.

### ***Research Design***

The study was designed using a randomized block design with one treatment factor using organic fertilizer in the form of cow and chicken manure. The levels were then divided into 7 groups, with P0, P1, P2, P3, P4, P5, and P6 each receiving a different dose of each type of manure. Each treatment was repeated four times.

The following dosages were used:

P<sub>0</sub> : Control (without fertilizer)

P<sub>1</sub> : 20 t cow manure /ha  
P<sub>2</sub> : 40 t cow manure /ha  
P<sub>3</sub> : 60 t cow manure /ha  
P<sub>4</sub> : 20 t chicken manure /ha  
P<sub>5</sub> : 40 t chicken manure /ha  
P<sub>6</sub> : 60 t chicken manure /ha

The plot had a length of 6 m and a width of 1 m. Planting one seed per planting hole results in a spacing of approximately 50 x 100 cm. To prevent waterlogging and seedling rot, seedlings are planted in ditches with good drainage. In order to facilitate landfiling later, planting is done in the trench. → **characteristics of materials used for this study (soil, cow manure and chicken manure) have to be presented in this Materials and Methods section, as their characteristics are very important to support the discussion**

### Data Analysis

Plant height, number of leaves, and number of tillers were measured at 3, 6, 9, 12, 15, and 18 WAP (weeks after planting). After the plants were harvested, fresh weight, dry weight, root length, secondary metabolic, and antioxidant levels were measured. The data obtained were subjected to the analysis of variance (ANOVA) with a 5% confidence level, followed by a 5% level DMRT (Duncan Multiple Range Test) to detect significant differences among treatments. The results of the secondary metabolic level identification and antioxidant activity tests were analyzed qualitatively using the descriptive method. Comparing phytochemical compound levels in each treatment was based on secondary metabolism levels. Antioxidant activity was assessed by comparing the IC<sub>50</sub> values in each treatment. A spectrophotometry method was used for all of the parameters of the secondary metabolic level test.

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### Results

The results of an analysis of variance (ANOVA) at a 5% level showed that the treatments of giving organic fertilizers in the form of chicken and cow manure gave significant differences in plant height, number of tillers, number of leaves, root length, fresh weight of rhizomes, and dry weight of rhizomes. Only root length that was not affected significantly by the treatments.

#### Plant growth

Table 1 shows that when the plant was about 6 weeks old, the application of cow manure at a dose of 60 t/ha produced the best plant growth (26.16 cm), followed by chicken manure at a dose of 60 t/ha, chicken manure of 40 t/ha, and cow manure of 40 t/ha. In comparison to the previous week, the plant's age 9 weeks after planting showed a very rapid increase in plant height. At 9 weeks after planting, the best dose was 40 t/ha of chicken manure with an average plant height addition of about 32.45 cm. The best results were obtained at week 12, with a cow manure at a dose of 60 t/ha that yielded an average plant height of 37.24 cm. The application of cow manure at a dose of 20 t/ha and chicken manure at a dose of 20 t/ha did not produce significantly different plant height from the control. The best dose at 15 weeks after planting was 60 t cow manure /ha, with an average plant height of 40.47 cm, and 38.38 cm of chicken manure. The highest dose of cow manure yielded an average plant height of 42.78 cm at 18 weeks after planting.

#### Number of Tillers

Table 2 shows that the highest number of tillers was found at a dose of 60 t/ha of chicken manure, with an average of 1.65 tillers at 6 weeks after planting. At the age of 9 weeks after planting, a control with an average of about 2.55 tillers produced number of tillers that were not significantly different from chicken manure 20 t/ha. The best application was 60 t/ha of cow manure with an average of 4.15 tillers. At 12 weeks after planting, the effect of manure treatment revealed that the best dose was 40 t/ha chicken manure, with an average of 6.65 tillers. This best dose did not differ significantly between chickens and cows manure at 60 t/ha or chickens manure at 20 t/ha. Cow and chicken manure at a dose of 60 t/ha at 15 weeks after planting produced the highest yields, with cow manure yielded 9.50 tillers and chicken yielded 9.00 tillers, respectively. At 18 weeks, the control did not differ significantly from the cow manure doses of 20 and 40 t/ha. The highest number of tillers was observed for the treatment of cow manure at 60 t/ha with an average of 14.45 tillers and chicken manure at 60 t/ha with an average of 13.37 tillers.

#### Number of Leaves

Table 3 shows that after 6 weeks of treatment with a dose of 20 t chicken manure /ha, the number of leaves were not statistically significantly different from the control but significantly different from other treatments. The best

dose of 60 t/ha cow manure, was produced at around 9 weeks, with an average increase in the number of leaves of 37.95 leaves. At 12 weeks, the control showed no significant difference from cow manure of 20 and 40 t/ha, as well as chicken manure of 20 and 40 t/ha. Cow manure of 60 t/ha with an average of 52.80 leaves provided the best dose at 12 weeks after planting. The highest dose was found in cow manure at a rate of 60 t/ha in week 15, with an average of 84.55 leaves. The application of 60 t/ha cow manure produced the largest number of leaves of 116.65 leaves in the 18th week, that significantly difference from the control. However, the application of 60 t/ha chicken manure increased the number of leaves by 105.80 leaves.

#### Rhizome root length, wet weight and dry weight

Table 4 shows that the lowest root length of 24.36 cm was obtained by the application of 20 t/ha cow manure. The control yielded root length that was not statistically different from that yielded by the application of 20 t/ha cow and chicken manure as well as the highest dose. This result indicates that applying different doses of manure had no effect on the root length of the bangle plant.

The chicken manure dosage of 20 t/ha yielded the lowest bangle rhizome fresh weight of 392.35 g. While the control yielded fresh weight that was not significantly different from 20 t/ha and 40 t/ha cow and chicken manure. The fresh weight of the bangle rhizome due to the application of 40 t/ha chicken and cow manure also did not differ significantly from those from 60 t/ha chicken manure. The treatment of 60 t/ha cow manure yielded the highest rhizome fresh weight of 822.00 g that was significantly for all treatments.

The lowest dry weight of bangle rhizome was shown by the cow and chicken manure treatment at a dose of 20 t/ha, respectively 86.00 g for cow manure treatment and 78.25 g for chicken manure treatment. These values were lower than that of the control, with an average of 93.75 g. The control yielded statistically insignificant different rhizome dry weight with chicken manure at 20 and 40 t/ha, cow manure at 20 and 40 t/ha, and chicken manure at 60 t/ha, but yielded significantly different rhizome dry weight with the best dose of 60 t/ha cow manure with an average rhizome dry weight of 179.75 g, followed by 60 t chicken manure /ha with an average rhizome dry weight of 135.75 g

**Table 1.** Effect of Cow and Chicken Manure on Rhizome Height (in cm) → Tables have to be presented in Word format (generated from Excel format), not in JPEG format, as JPEG format cannot be edited.

Manure Treatment (ton/ha)	Weeks after Planting (MST)				
	6	9	12	15	18
Control	11,38 a	18,61 a	24,68 a	20,41 a	18,82 a
Cow 20 ton/ha	17,46 b	20,26 ab	25,44 a	24,02 ab	28,51 b
Cow 40 ton/ha	20,51 c	27,95 b	29,81 ab	35,57 cd	37,19 c
Cow 60 ton/ha	26,16 e	31,18 b	37,24 c	40,47 d	42,78 c
Chicken 20 ton/ha	17,36 b	27,21 b	30,16 abc	29,15 bc	28,82 b
Chicken 40 ton/ha	22,84 cd	32,45 b	34,83 bc	37,61 cd	36,37 c
Chicken 60 ton/ha	25,68 de	32,17 b	36,84 bc	38,38 d	39,22 c

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ton/ha is abbreviated to t/ha

**Table 2.** Effect of Cow and Chicken Manure on Rhizome number of tillers (in weeks) → Tables have to be presented in Word format (generated from Excel format), not in JPEG format, as JPEG format cannot be edited.

Manure Treatment (ton/ha)	Weeks after Planting (MST)				
	6	9	12	15	18
Control	0,8 a	2,55 a	3,95 a	5,80 a	8,60 a
Cow 20 ton/ha	0,7 b	3,45 b	5,20 b	6,60 a	10,40 ab
Cow 40 ton/ha	1,7 ac	3,40 b	5,05 ab	7,35 ab	11,55 b
Cow 60 ton/ha	1,6 c	4,15 b	6,10 bc	9,50 c	14,45 d
Chicken 20 ton/ha	1,3 bc	3,35 ab	5,70 bc	7,40 ab	11,05 b
Chicken 40 ton/ha	1,5 c	4,00 b	6,65 c	8,45 bc	12,15 bc
Chicken 60 ton/ha	1,65 c	3,75 b	6,05 bc	9,00 c	13,70 cd

**Table 3.** Effect of Cow and Chicken Manure on number of Rhizome Leaves (in weeks) → Tables have to be presented in Word format (generated from Excel format), not in JPEG format, as JPEG format cannot be edited.

Manure Treatment (ton/ha)	Weeks after Planting (MST)				
	6	9	12	15	18
Control	4,70 a	19,60 a	35,80 a	52,25 a	77,55 a
Cow 20 ton/ha	5,55 a	25,00 ab	41,05 ab	55,65 a	83,80 a
Cow 40 ton/ha	9,65 b	28,25 abc	42,70 abc	66,00 ab	95,35 ab
Cow 60 ton/ha	10,05 b	37,95 c	52,80 c	84,55 c	116,65 c
Chicken 20 ton/ha	9,20 b	26,00 ab	40,25 ab	56,85 a	81,75 a
Chicken 40 ton/ha	9,65 b	30,90 bc	43,25 abc	67,05 bc	91,35 ab
Chicken 60 ton/ha	11,05 b	30,45 abc	50,75 bc	79,55 bc	105,80 bc

**Table 4.** Effect of Cow and Chicken Manure on Root Length and Rhizome Weight (in 18 week) → Tables have to be presented in Word format (generated from Excel format), not in JPEG format, as JPEG format cannot be edited.

Manure Treatment (ton/ha)	Root Length (cm)	Rhizome wet weight (g)	Rhizome dry weight (g)
Control	31,90 ab	415,25 ab	93,75 ab
Cow 20 ton/ha	24,36 a	444,75 ab	86,00 a
Cow 40 ton/ha	35,36 b	530,00 bc	119,00 ab
Cow 60 ton/ha	36,50 b	822,00 d	179,75 c
Chicken 20 ton/ha	33,20 ab	393,25 a	78,25 a
Chicken 40 ton/ha	38,95 b	487,50 abc	99,75 ab
Chicken 60 ton/ha	41,03 b	618,75 c	134,75 bc

**Notes:** Based on Duncan Multiple Range Test (DMRT) at a 5% significance level, values in the same columns that are followed by the same letter do not differ significantly.

**Table 5.** Secondary Metabolic Levels in Bangle Rhizome in Each Treatment (mg/L). → Tables have to be presented in Word format (generated from Excel format), not in JPEG format, as JPEG format cannot be edited.

Condition	Treatment	Level (mg/L)		
		Phenolic	Flavonoid	Tanin
Fresh	Control	31,64	23,08	2034,83
	Cow 20 ton/ha	43,18	41,55	2991,50
	Cow 40 ton/ha	77,03	64,30	3151,50
	Cow 60 ton/ha	86,90	71,14	3198,17
	Chicken 20 ton/ha	97,67	74,79	4086,50
	Chicken 40 ton/ha	107,54	78,71	4144,83
	Chicken 60 ton/ha	178,56	104,39	3861,33
Dry	Control	124,97	38,44	1932,33
	Cow 20 ton/ha	133,56	67,23	2504,00
	Cow 40 ton/ha	148,44	68,76	3034,83
	Cow 60 ton/ha	181,38	99,43	4734,67
	Chicken 20 ton/ha	175,23	120,06	4903,00
	Chicken 40 ton/ha	198,69	156,10	5088,00
	Chicken 60 ton/ha	202,79	181,91	5406,33

**Note:** The number followed by yellow denotes the best outcome in each observation variable for the flavonoid, tannin, and phenolic contents.

**Table 6.** Antioxidant based on IC<sub>50</sub> value → Tables have to be presented in Word format (generated from Excel format), not in JPEG format, as JPEG format cannot be edited.

Antioxidant (IC <sub>50</sub> ) (mg/L)	Fertilizer Dose ton/ha						
	Control	Cow			Chicken		
		20	40	60	20	40	60
Fresh Rhizome	53.58	48.55	47.36	47.14	32.73	27.42	9.52
Dry Rhizome	40.91	38.30	34.78	34.46	24.41	14.05	8.06

**Note:** The smaller the IC<sub>50</sub> value, the stronger the antioxidant

### Secondary metabolic level

In accordance with the findings of the secondary metabolism test, the rhizome of the bangle plant contained phenolic, flavonoid, and tannin-containing active substances, and also steroids, alkaloids, and terpenoids qualitatively. Different levels of the compounds involved in this secondary metabolism are evident in each organic fertilizer application (Table 5). Different concentrations were produced by the concentration of active substances in the bangle rhizome. The data mentioned above demonstrated that, despite using the same amount of fertilizer, the active compound content of the bangle rhizome was highest when the rhizome was dry (low moisture content) as opposed to when the rhizome was still fresh. The amount of chicken manure that produced the highest phenolic content in the rhizome under fresh and dry conditions, correspondingly around 178.56 mg/L and 202.79 mg/L, was 60 t/ha. A dose of 60 t/ha of chicken manure also yielded higher levels of active flavonoids and tannins than other doses. When compared to the fertilizer treatment, the control had the lowest active compound content. When treated with cow manure, yields were lower than when treated with chicken manure at the same dose.

### Antioxidant

Results of antioxidant analysis of bangle rhizomes using several samples based on manure dose and fresh and dry rhizome condition are shown Table 6. IC<sub>50</sub> value obtained described how well the sample captured free radicals. The dose of 60 t chicken manure s/ha was found to have the lowest IC<sub>50</sub> value of 9.52 ppm in the fresh sample, , while the control had the highest IC<sub>50</sub> value of 53.58 ppm in fresh rhizome conditions. The same quality was obtained when dry rhizome conditions were used for the analysis; specifically, the dose of 60 t chicken manure /ha chicken manure produced the lowest IC<sub>50</sub> value and the largest was in the control. The plant's antioxidants are more potent the lower the IC<sub>50</sub> value. Cow manure is always treated with an IC<sub>50</sub> value higher than chicken manure.

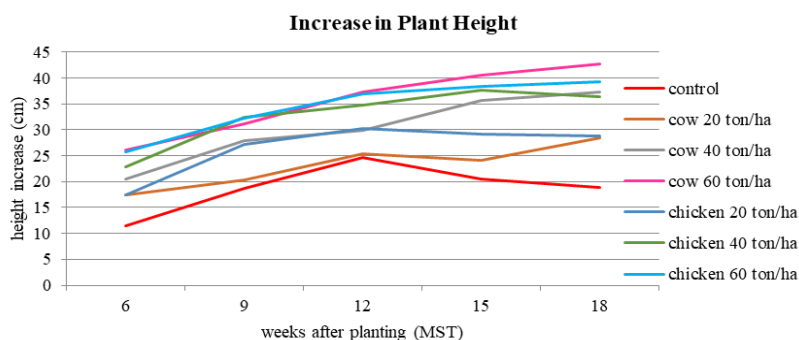
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## Discussion

### Plant Height

According to the Table 1, the average increase in plant height starting at 6,9,12,15, and 18 WAP tended to fluctuate. At the age of 6 weeks, the treatment without fertilizer (control) continued to grow until week 12 when it reached its peak with an average height increase of about 24.68 cm, before the delining height at the following weeks.. In order for a plant to grow, nutrients must be obtained from the soil by the roots through their root hairs (Sudewi et al., 2022). Organic matter affects plant growth by influencing the physical, chemical, and biological properties of the soil ( Anwar, 2013). The more organic matter is provided, the faster the plant will grow. Compared to chicken manure, cow manure typically produces better plant growth . According to ((Hartatik et al. ( 2015), cow manure contain N (6 kg/t), P (1.5 kg/t), K (3.0 kg/t), Ca (1.2 kg/t), Mg (1.0 kg/t), and S (0.9 kg/t), and these nutrients can support the growth of Bangle plants, which have a long harvest period.





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The application of chicken manure at all levels significantly increased plant height at 9 weeks after planting, but at 12 weeks, the plant height slightly decreased. In the 20 t/ha chicken manure treatment, the plant height was in average of 30.16 cm at 12 weeks after planting and decreased in the following weeks. High nitrogen elements are found in chicken manure → **how many %?**. Although the amount of nitrogen required by plants is always higher than other nutrients, a deficiency or excess of nitrogen can hinder and disrupt plant growth (Raja et al., 2021). After planting, the growth rate of the bangle plant accelerated between 2 and 5 months. As plants get older, their growth rate for height starts to slow down (Rosita et al., 2005).

According to Hartatik et al. (2015), applying chicken manure always results in the best plant response in the first growing season. This is because chicken manure decomposes relatively quickly and has enough nutrients compared to other manures of the same weight. Table 1 shows that the application of chicken manure tended to increase plant height rapidly at 6, 9, and 12 weeks of age, then declined as plant age increased. Large-scale application of chicken manure is thought to be less effective because the nutrients will exhaust quickly.

The same result was also shown by the application of chicken manure at a dose of 40 t/ha which decreased the plant height, the maximum height increase at this dose was at 15 weeks with an average height increase of 37.61 cm. When compared to the other two doses of chicken manure, the plant height for the 60 t/ha chicken manure was different. The plant height increase over 18 weeks demonstrates this, but the increase in plant height was typically not too different from the previous weeks. Even though the application of cow manure at a dose of 40 t/ha initially produced less yields than that produced by the application of chicken manure at a dose of 40 t/ha, at the end of the observation at 18 weeks the increase in height was more apparent and might even have exceeded that of the chicken manure at a dose of 40 t/ha, which caused a decrease in plant height.

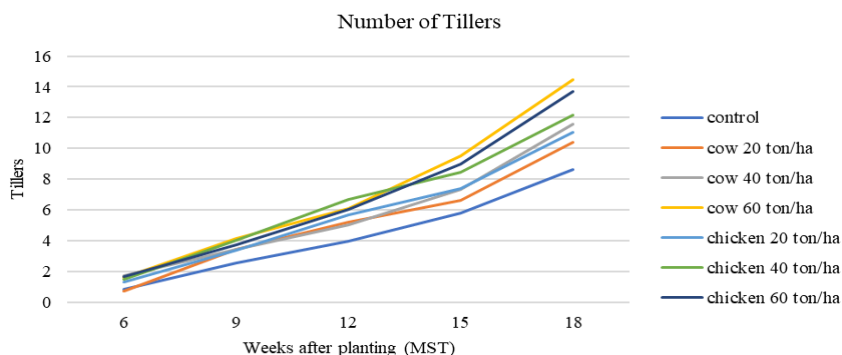
The addition of cow manure improves soil permeability, total pore space, aggregate stability, bulk density, texture, color, and temperature (Sudewi et al., 2022). A dose of 60 t/ha of chicken and cow manure had the tendency to produce steady, dependable results. The application of cow manure at a dose of 60 t/ha tended to yield less than chicken manure at doses of 40 and 60 t/ha from the start of planting until the plant was 9 weeks old, but the yields increased in the following weeks.

The plant's need for nutrients grows as it ages. If the nutrient requirements are not met and the nutrients are not readily available, plants may experience nutrient deficits at specific times. According to Rosita et al. (2005), bangle plants absorb N (0.06 - 3.07 g), P (0.01 - 0.53 g), and K (0.10 to 2.25 g) at 2 to 10 months after planting in the canopy. N is the nutrient that is most required in the plant canopy itself. The primary nutrient for plants, nitrogen, is typically essential for the development and expansion of vegetative parts of plants, such as leaves, stems, and roots (Purba et al., 2021). A sufficient supply of plant N is indicated by high photosynthetic activity, good vegetative growth, and dark green plant colors (Nurhayati, 2021).

Due to the individual characteristics of each animal, which are influenced by the type of feed and the animal's age, each manure contains a different mix of nutrients (Nurjanah et al., 2020). Because each treatment dose of fertilizer has a different nutrient content, they all produce different yields and have different recommended doses. Due to their movement with crop yields, surface runoff, erosion, or evaporation, nutrients in the soil will gradually decrease over time (Lawenga et al., 2015).

#### Number of Tillers

The table of the total number of tillers from each level reveals results that increase with plant aging and are influenced by the quantity of fertilizer applied, as shown in Table 2. Plants without fertilizer developed more tillers every week, but the growth was typically modest. This slight increase resulted from the fact that during the initial stages of planting, the products of photosynthesis were utilized for the vegetative development of plants.



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In comparison to manure application, treatment without fertilizer produced the lowest yield. Data presented in Table 2 show that there was a noticeable increase in the number of tillers at 18 WAP of age. A plant needs nutrients for its physiological processes during growth and development. Plant growth and production will be subpar due to a lack of nutrients (Purba et al., 2021).

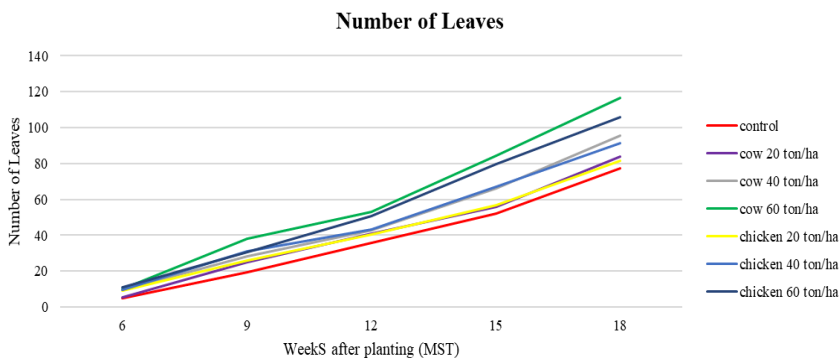
Functions of organic matter as a biological buffer so that the soil can supply plants with a balanced amount of nutrients (Hartatik et al., 2015). Loosening the topsoil, increasing water absorption and storage, and boosting soil fertility are all important functions of manure (Yulianto et al., 2021). A sudden rise in the number of tillers can result from the ease with which new shoots can emerge from loose, moist soil.

At the start of planting, there were typically fewer and nearly identical numbers of tillers in each treatment. The nutrients in this fertilizer are not readily available to plants, which is the cause of the slow plant growth at the start of the planting period. The extent of these materials' mineralization or decomposition has a significant impact on the nutrients' availability. Manure's low nutrient availability is partially caused by the presence of N, P, and other elements in complex compounds that are challenging to decompose (Hartatik et al., 2015).

At 6 weeks after planting, all treatments tended to be similar and the differences between the tillers in each treatment tended to be minimal. Although cow manure at a dose of 60 t/ha was the best dose with an average number of tillers of 14.45 tillers, chicken manure typically produced better results than cow manure at the same dose. In comparison to other manure doses, chicken manure 40 t/ha at 12 weeks after planting produced the best results with 6.65 tillers.

#### **Number of Leaves**

An increasing number of leaves are produced each week as a result of the weekly application of cow and chicken manure. Table 3 show that at about 12 weeks of age, the number of leaves increased significantly. Every week, the increase varied depending on the treatment. The number of leaves significantly increased with a dose of 60 t/ha of cow manure, averaging 15–37 leaves every three weeks. Similarly, the number of leaves tended to increase when manure was applied in the same amount. Both cow and chicken manure at a dose of 20 t/ha and 40 t/ha produced a nearly identical number of leaves during plant growth.



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In addition to the nutrients that plant's needs, manure also contains humic, fulvic acids, growth hormones, and other substances that promote plant growth and increase nutrient uptake by plants (Hartatik et al., 2015). The amount of photosynthesis is influenced by the number of leaves present, and plants with more leaves may produce heavier and bigger rhizomes as a result.

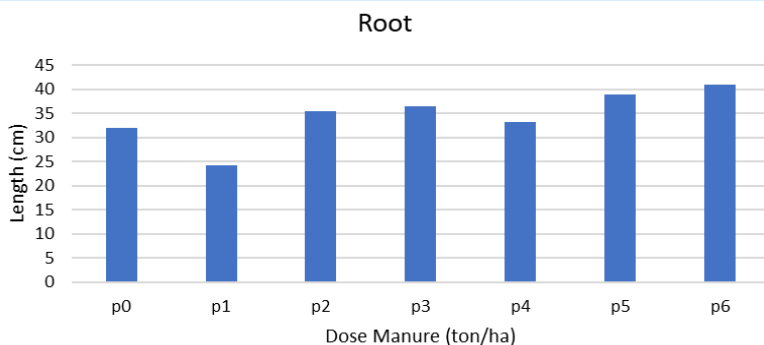
The number of leaves added is also affected by the number of shoots and plant height. The number of leaves will increase as the plant ages and grows taller, produces more leaves on a single stem, and produces more tillers. The nutrients required for plant growth are present in sufficient amounts in manure. The related observation variables will be impacted by the food's growth quality and planting age.

In relation to the addition of the number of leaves, the most influential element is N. In comparison to other nutrients, nitrogen is required in sufficient amounts for plant growth. N makes up 40–50% of the dry weight of protoplasm, the living component of plants (Nurhayati, 2021). Since protein is the source of all plant enzymes, nitrogen participates in all enzymatic processes in plants. Additionally, nitrogen is one of the constituent elements of chlorophyll, the primary component of chloroplasts, and it contributes to improving the quality and quantity of the dry matter produced (Vladimir, 2021).--> this reference cannot be traced electronically

Fertilizer use and the amount of nutrients in the soil have a significant impact on how plants grow and develop. Nutrient uptake is restricted by nutrients in a minimum state (Purba et al., 2021). In terms of the addition of leaves, the treatment of plants without fertilizer differs significantly enough for each observation. In comparison to other treatments, plants without fertilizer produce the lowest yield.

#### Root Length

The lowest root length was observed for the cow manure dose of 20 ts/ha treatment with an average root length of about 24.36 cm which was lower than the control. With an average root length of 41.03 cm, the application of chicken manure yielded the longest roots. When plants respond to water shortages by reducing the rate of transpiration to conserve water, the roots play a crucial role (Torey et al., 2013). Plant roots have a significant impact on overall plant growth and development. The failure of the root function will result in a complete change in the plant for the top (Nurhayati, 2021).



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Manure can bind water in the soil. Because the soil around the roots in the deeper layers is still moist, the roots will continue to grow. Maximizing exposure to groundwater will encourage the growth of roots (Torey et al., 2013). Plant roots directly respond to the physical characteristics of the soil (Lawenga et al., 2015).

Data presented in Table 4 show that the application of chicken manure yielded better root length than cow manure. The use of organic fertilizers can loosen the soil, increase aeration, and increase the soil's capacity to hold water, all of which can improve the physical properties of the soil (Raja et al., 2021). Additionally, organic matter has the ability to control soil temperature, slow down phosphorus fixation, increase soil cation exchange capacity, and lessen the leaching of nutrients like potassium, calcium, and magnesium (Guimarães et al., 2019). Another environmental factor that has been shown to affect the nitrate absorption process is the temperature around the roots (Vladimir, 2021). → this reference cannot be traced; please replace it with others

The initial analysis of the soil revealed that the pH ranged from 3.86 to 4.86. Al is common excessively in acidic soil, and it can poison plants and bind phosphorus (P). Low pH soil can hinder plant growth by preventing the roots from properly absorbing nutrients. Giving chicken manure up to 5–25 t/ha, as demonstrated by Tufail et al. (2014), could raise the pH of soil from 5.0 to 5.8–6.4. By raising pH, Al in the exchangeable form will be reduced, and nutrients will become more available to plants.

According to Rosita et al. (2005), nutrient uptake on the roots of bangle plants at 2-10 months after planting was as follows: N (0.01 - 0.52 g), P (0.002 - 0.15 g), and K (0.02 to 0.82 g). It was discovered that the roots of the bangle plant had more K buildup than N and P. K is primarily used to aid in the synthesis of proteins and carbohydrates. In the face of drought, illness, and pests, potassium gives plants strength (Purba et al., 2021). Organic fertilizers can help the soil's physical and chemical composition, which will facilitate root development. Up until the soil reaches its critical water potential, plant roots expand into moist soil and draw water (Solichatun et al., 2005). The looseness of the soil can promote root development. Strong roots will make it simpler for plants to absorb nutrients and water

#### ***Rhizome fresh Weight***

Plant biomass is a common parameter used to study plant growth. The fresh weight of the plant describes the water and moisture content of the plant. The plant will weigh more when wet the more fertile it is (Supandi, 2021). When plant nutrient requirements are met, yields will be optimal (Purba et al., 2021). The rhizome of the bangle plant is the part that is most advantageous for cultivation. One could also argue that this rhizome's fresh weight is a crucial factor in determining how well bangle plant cultivation is going. The cultivation method is better and more productive the more weight of the wet rhizome can be obtained.

Rosita (2005) found that giving 250 kg/ha of urea, 250 kg/ha of SP36, 250 kg/ha of KCl, and 20 tons/ha of manure resulted in a fresh bangle rhizome weight of 311.39 g/plant 5 months after planting. According to the results of the application of the organic fertilizers at the age of 18 weeks, cow manure at a dose of 60 t/ha produced the highest fresh weight of rhizomes, averaging 822 g/plant, and chicken manure at a dose of 20 t/ha produced the lowest fresh weight of rhizomes, averaging 393.25 g/plant. Manure increases crop yield and quality while also enhancing the chemistry, physical characteristics, and biological properties of the soil (Seker et al., 2011). Data presented in Table 4 demonstrate that the yield of fresh weight of rhizomes increased with increasing manure dosage.

The physical condition of the soil must support plant growth in addition to a supply of adequate and balanced nutrients (Lawenga et al., 2015). These soil aggregates will keep the soil in a loose condition (Anwar, 2013). Cow manure will enhance the physical characteristics of the soil. Improved soil physical characteristics include things like increased permeability, total pore space, aggregate stability, bulk density, texture, color, and temperature (Sudewi et al., 2022).

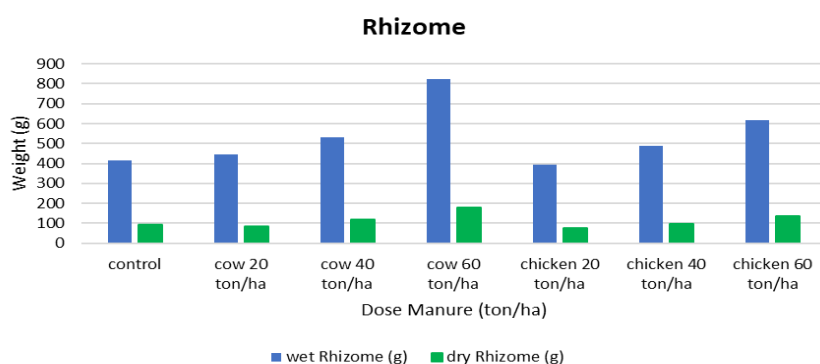
Intensive tillage affects the physical properties of the soil. Low organic matter soils will have more severe damage to the soil's structure (Anwar, 2013). When the soil does not receive enough water and becomes dense and hard, soil damage is evident. Plant rhizomes will not be able to grow or spread out in compacted or hard soil. The ability to maintain loose soil conditions that are difficult to harden or compact increases with the amount of organic matter added.

Additionally, manure helps to improve soil structure, cation exchange capacity, and water resistance. Giving manure has the indirect effect of making it simpler to keep water in the soil (Yuliana et al., 2015). Since water availability plays a significant role in plant growth, water frequently restricts the growth and development of cultivated plants.

The plants will experience drought conditions if there is not enough water in the soil. Due to decreased primary metabolism, reduced leaf area, and decreased photosynthetic activity, drought stress can lower plant productivity (biomass). Smaller leaves grow as a result of a lack of water during the vegetative stage, which can reduce light absorption. Lack of water also inhibits the synthesis of chlorophyll and some enzymes, such as nitrate reductase, from working (Solichatun et al., 2005).

Organic substances in the soil may have physiological effects on plant growth that are direct or indirect (Syaiful Anwar, 2013). Compared to other types of manure, chicken manure contains a fair amount of P. This is due to the

fact that chicken manure contains feed (Sudewi et al., 2022). Phosphorus aids in the growth of plant roots, photosynthesis, transfer respiration, cell division, and growth (Supandi, 2021). The number of cells increases more quickly when they divide quickly, which causes the rhizome to grow larger.



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#### Rhizome Dry Weight

The relationship between plant growth and the quantity and concentration of mineral nutrients in plant tissues is known as dry weight (Purba et al., 2021). Dry weight reflects a plant nutritional status because it is affected by the rate of photosynthesis and respiration in each treatment. Based on the collected data, it was determined that applying chicken and cow manure at a dose of 20 t/ha resulted in lower dry weight yields than the control. Additionally, the results from the application of 40 t/ha manure dose were not significantly different from the control. Many nutrients, such as N, P, and K, build up in the canopy when the bangle plant is between 2 and 7 months after planting to (Rosita et al., 2005). This is the active vegetative formation stage.

The application of cow manure at a dose of 60 t/ha demonstrated different results and produced significantly better outcomes than other doses of chicken manure. The amount and timing of fertilizer applied can impact crop yields, among other things. Organic matter plays a crucial role in soil health because it can create stable soil aggregates, increase soil fertility, and serve as a source of energy for organisms (Nurjanah et al., 2020).

Rosita et al. (2005), reported that the N, P, and K amounts of nutrient uptake in dry rhizomes of Bangle plants aged 2 to 10 months were N (0.01 to 4.89 g), P (0.002 to 1.04 g), and K (0.01 to 2.34 g), respectively. It is clear that as plants get older, they are able to absorb an increasing amount of N, P, and K nutrients. As can be seen, N is the nutrient that has accumulated in the rhizome in the greatest amounts.

The application of manure enhances the chemical, physical, and biological properties of the soil, increase crop yield, and improves crop quality (Yolcu, Turan et al., 2011). High organic matter soils have beneficial microorganisms that encourage the breakdown of organic matter and release inorganic nutrients that are then available for plant uptake (Seker et al., 2011). Organic fertilizers can help to create ideal conditions in the soil for microorganisms that are beneficial to plants.

Chicken manure is an organic fertilizer with high nitrogen content, despite not being the best dose for bangle rhizome weight yield. As they ensure the best nutrient management for plants, such fertilizers should be used promptly to partially replace chemical fertilizers (Guimarães et al., 2019).

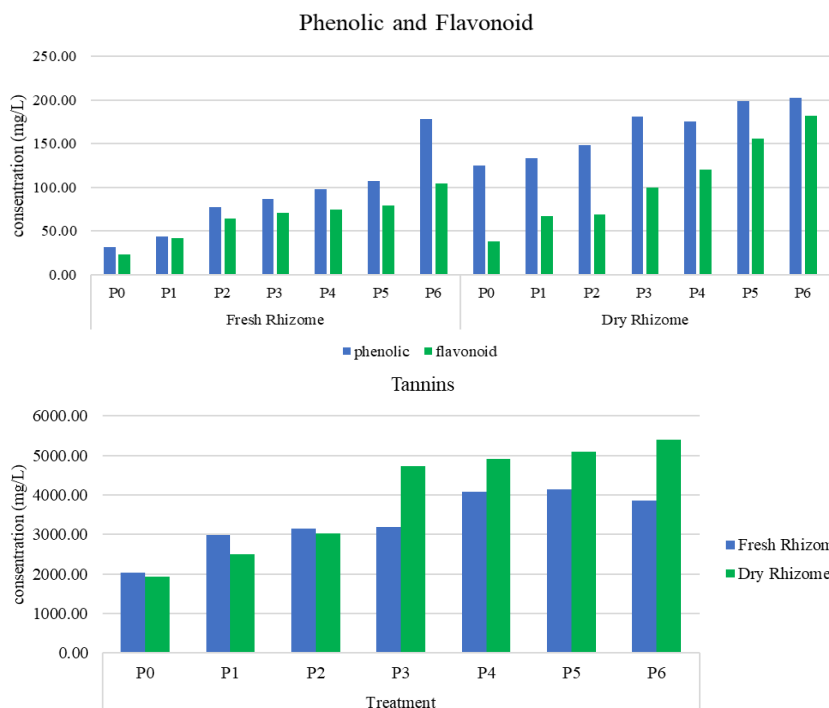
#### Secondary metabolic levels (phenolic compounds, flavonoids and tannins)

The results of laboratory analysis found that the positive bangle rhizome contains compounds in the form of phenolics, tannins and flavonoids. These findings are consistent with Amalia et al. (2021), which found that the Bangle plant contains secondary metabolism in the form of alkaloids, phenolic compounds, flavonoids, saponins, and triterpenoids. These compounds respond differently to the concentration of organic fertilizer applied in the form of cow and chicken manures. Table 5 shows the outcomes of these phytochemicals.

According to Table 5, the amount of tannin compounds increased as manure dosage rose. The application of chicken manure or cow manure with three doses of 20, 40, and 60 t always increased the tannin concentration in the rhizome as the dose increased. When compared to cow manure, applying chicken manure produced significantly better results. It is evident that using 20 t/ha of chicken manure instead of 60 t/ha of cow manure resulted in a higher tannin concentration.

Dry rhizomes produced a higher concentration of tannins than fresh rhizomes, which produced different results regarding tannin concentration. The concentration of tannins in fresh rhizomes increased non-significantly with the addition of cow manure, whereas the concentration of tannin compounds in fresh rhizomes decreased with the

increase in the dose of cow manure. In comparison to the use of manure that increased fresh and dry rhizomes, the control provided the lowest tannin concentration.



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Tannins are chemical substances that have an astringent and bitter flavor. These substances act as controlling substances in plant metabolism as well as important defenses against herbivores and pests that prey on plants (Julianto, 2019). Tannins are metabolically active substances with multiple uses, including as astringents, antibacterial agents for treating diarrhea, and antioxidants. Leather tanning is another industrial application for tannins (Amelia, 2015). Tanad acid is the type of tannin substance present in bangle plants. Diarrhea can be effectively treated with tannic acid. Additionally, tannic acid exhibits antimicrobial, antienzymatic, antioxidant, and antimutagenic properties (Hidjrawan Yusi, 2018)

In addition to increasing the fertilizer dose, applying organic fertilizer in the form of cow and chicken manure resulted in an increase in phenolic compounds and flavonoids. When compared to when the rhizome was fresh, the dry rhizome had a higher concentration of phenolic and flavonoid compounds. The content of active compounds in the simplicia is impacted by the drying process. Antioxidant activity is influenced by the total phenolic and flavonoid content (Amelia, 2015). Given that fresh samples are more prone to damage and experience a quicker loss in quality than dry samples, it is advised to use dry samples instead of fresh samples (Julianto, 2019).

The highest phenolic and flavonoid concentrations were found in both fresh and dry rhizomes when chicken manure was applied at a dose of 60 t/ha. The application of the highest dose of cow manure of 60 t/ha, resulted in phenolic compound concentrations of 86.90 mg/L in the fresh rhizome and 181.38 mg/L in the dry rhizome, while flavonoids were 71.14 mg/L in fresh rhizome and 99.43 mg/L in dry rhizome. Compared to chicken manure at the same dose, this result is smaller. A dose of 60 t/ha of chicken manure resulted in phenolic compound concentrations in the fresh rhizome of 178.56 mg/L and dry rhizome of 202.79 mg/L, while fresh rhizome flavonoids were 104.39 mg/L and dry rhizome flavonoids were 181.91 mg/L. In comparison to treatments with organic fertilizers, treatments without fertilizer produced lower phenolic and flavonoid concentrations.

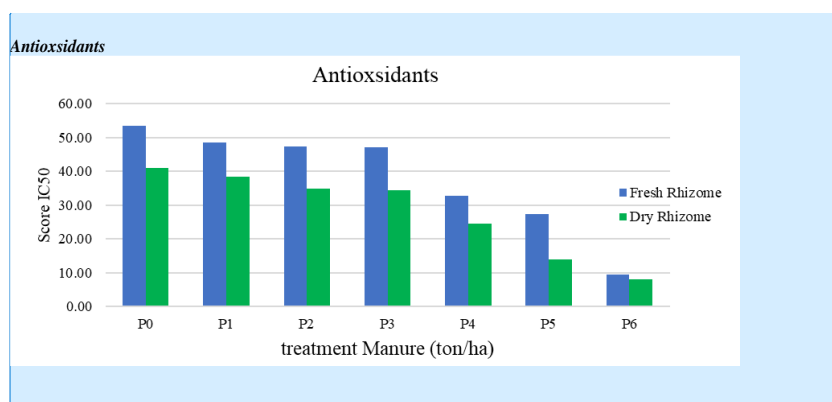
In plants, flavonoids serve as pigments for the flowers, fruits, and roots, as well as occasionally as growth regulators and disease resistance (Julianto, 2019). Catechins are one class of flavonoid compounds present in bangle rhizomes (*catechins*). Catechins have antioxidant properties, and because they can stop the growth of

viruses, bacteria, tumors, and fungi, they can also get rid of rotten and rancid odors (Aprilliza et al., 2021). Phenolic compounds are compounds that plants make in response to environmental stress. Phenolic compounds protect DNA from dimerization and damage by blocking UV-B rays and cell death (Hanin and Pratiwi, 2017). Gallic acid is the type of phenolic compound found in bangle rhizome. Gallic acid serves as an antibacterial, antiviral, analgesic, and antioxidant in medicine (Junaidi and Anwar, 2018).

The application of chicken manure resulted in a higher concentration of secondary metabolism because it had a relatively higher P nutrient content than other manures (Simanungkali et al., 2006). Phosphorus can be found in DNA, RNA, and the parts of nucleotides that provide metabolic energy (like ATP). The process of photosynthesis depends heavily on phosphorus. Stunted growth is one of phosphorus deficiency's signs (Khairuna, 2019). The amount of  $P_2O_5$  in cow manure is 0.2%, compared to 1.3% in chicken manure (Simanungkali et al., 2006). The concentration of secondary metabolism in the form of tannins, phenolic compounds, and flavonoids is influenced by the difference in the P nutrient content between these two types of manures. Environmental factors affect the levels of flavonoids and other phenolic compounds in plants, which vary among parts, tissues, and ages of plants. These include air temperature, nutrient availability, water availability, and atmospheric  $CO_2$  concentrations (Amelia, 2015).

### Antioxidants

The antioxidant activity analysis produced different  $IC_{50}$  values depending on the type of organic fertilizer used (Table 6). The  $IC_{50}$  decreased as the fertilizer dose increased. A concentration known as  $IC_{50}$  is capable of reducing 50% of DPPH free radicals. The greater the antioxidant activity, the lower the  $IC_{50}$  value (Widyasanti et al., 2016). Antioxidants are compounds that can absorb or neutralize free radicals, thereby preventing certain diseases caused by free radicals (Parwata, 2016).



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The treatments of cow manure, as shown in Table 6 resulted in lower yields than chicken manure. The antioxidant activity of the dried rhizome samples was higher than that of the fresh rhizomes. The highest antioxidant activity was produced by cow manure at a dose of 60 t/ha, with an  $IC_{50}$  value of 9.52 ppm for fresh rhizome and 8.6 ppm for dry rhizome. At a dose of 20 t/ha in both fresh and dry rhizome conditions, cow manure had a lower  $IC_{50}$  value than chicken manure. For fresh rhizomes, the treatment without fertilizer produced an  $IC_{50}$  value of around 53.58 ppm, and for dry rhizomes, it was around 40.91 ppm.

Widyasari et al. (2016) claimed that the antioxidant activity in bangle rhizomes is incredibly powerful. Wartano et al. (2016) classified antioxidants into five groups based on their  $IC_{50}$  values: 50 very strong, 50-100 strong, 101-250 moderate, 251-500 weak, and >501 inactive. Except for the treatment without fertilizer in fresh rhizome conditions, the  $IC_{50}$  value in bangle rhizome in all treatments gave a value of 50 and included a very potent antioxidant. The high secondary metabolic compounds found in the bangle rhizome are inextricably linked to the high antioxidant activity. Secondary plant metabolites like flavonoids and phenolics play a part in antioxidant activity. More phenolic compounds will have a higher level of antioxidant activity (Amelia, 2015).

### Conclusion

A cow manure dose of 60 t/ha is the best dose for plant growth, with an average height increase of 42.78 cm, an increase in the number of leaves of 116.65 pieces, and an increase in the number of tillers of 14.45. A dose of 60

t/ha of chicken manure produced the best root length of 41.03 cm. The weight of the rhizomes revealed that the application of cow manure at a dose of 60 t/ha resulted in the highest yields of dry weight and wet weight, which were about 179.75 g and 822 g, respectively. A chicken manure dose of 60 t/ha results in the highest secondary metabolic rate in each parameter, including dry rhizome (phenolic 202.79mg/L, flavonoid 181.91mg/L, and tannin 5406.33mg/L) and wet rhizome (phenolic 178.56mg/L, flavonoid 104.39mg/L), while a chicken manure dose of 40 t/ha results in the highest tannin. Giving chicken manure at a dose of 60 t/ha produced very strong antioxidant results at 9.52 ppm wet rhizome and 8.06 ppm dry rhizome, according to the antioxidant results.

#### Acknowledgements

Please insert acknowledgements to those who funded and assisted in implementing this study/research. It would be better if we got used to acknowledging anyone who has helped us

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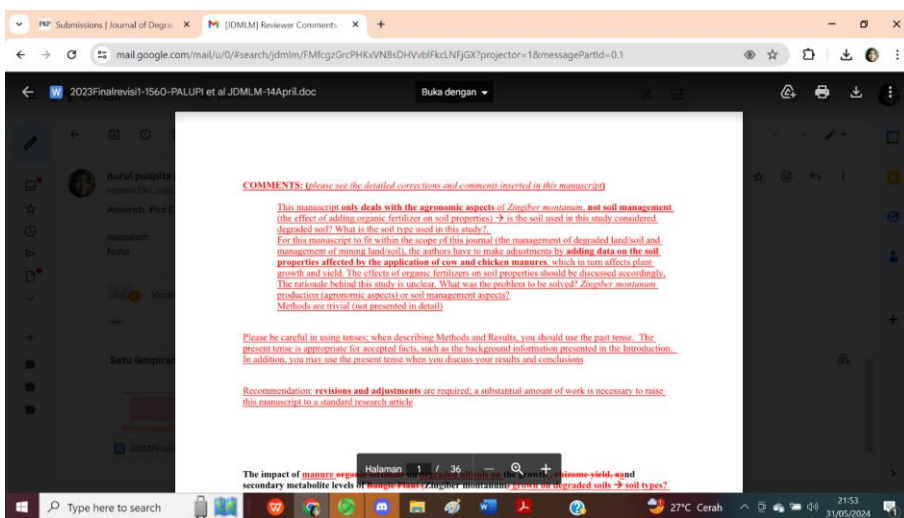
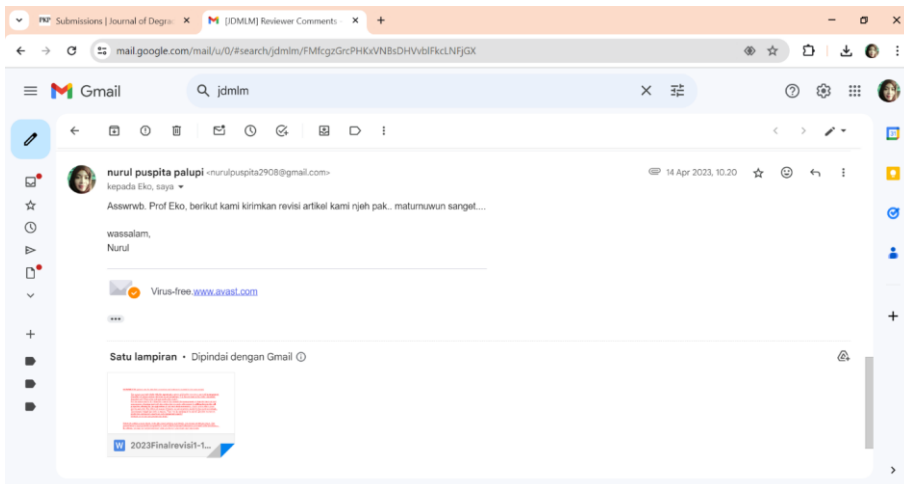
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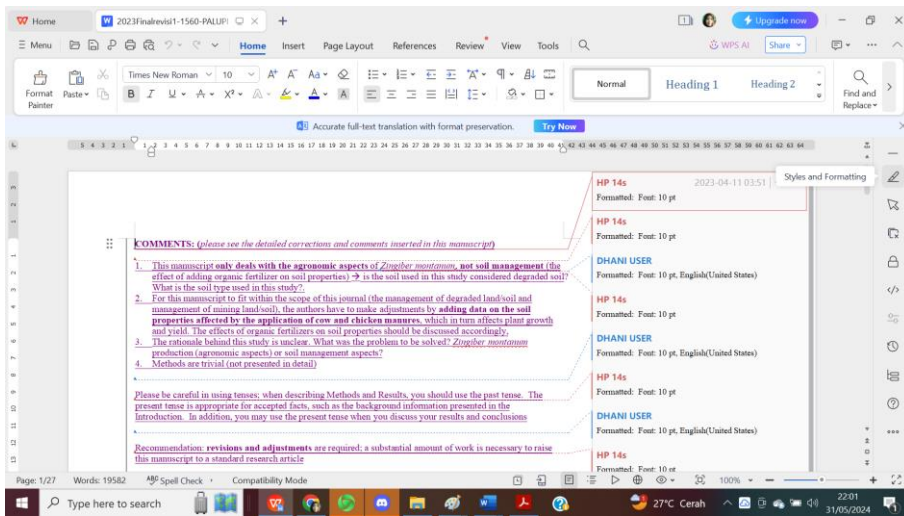
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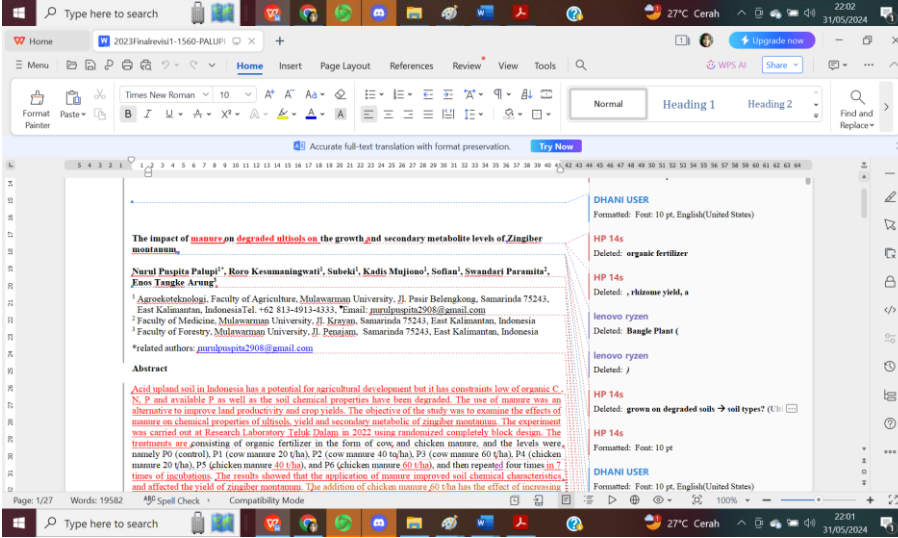
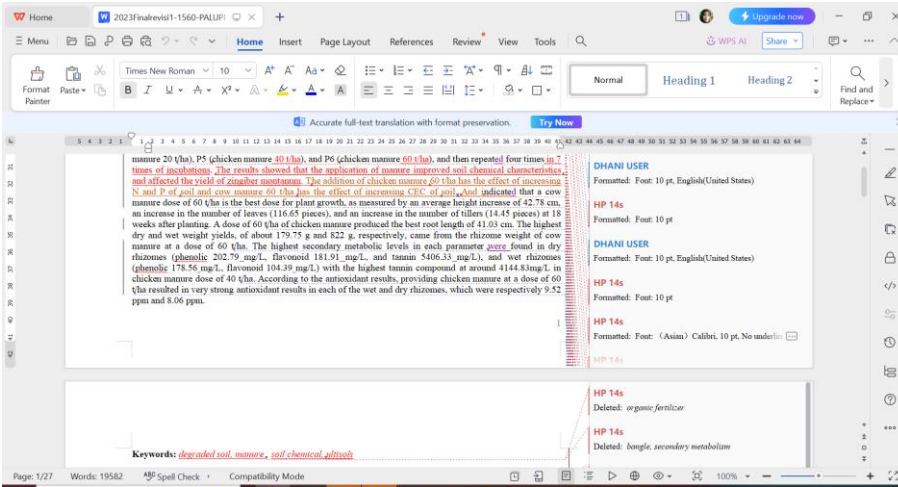


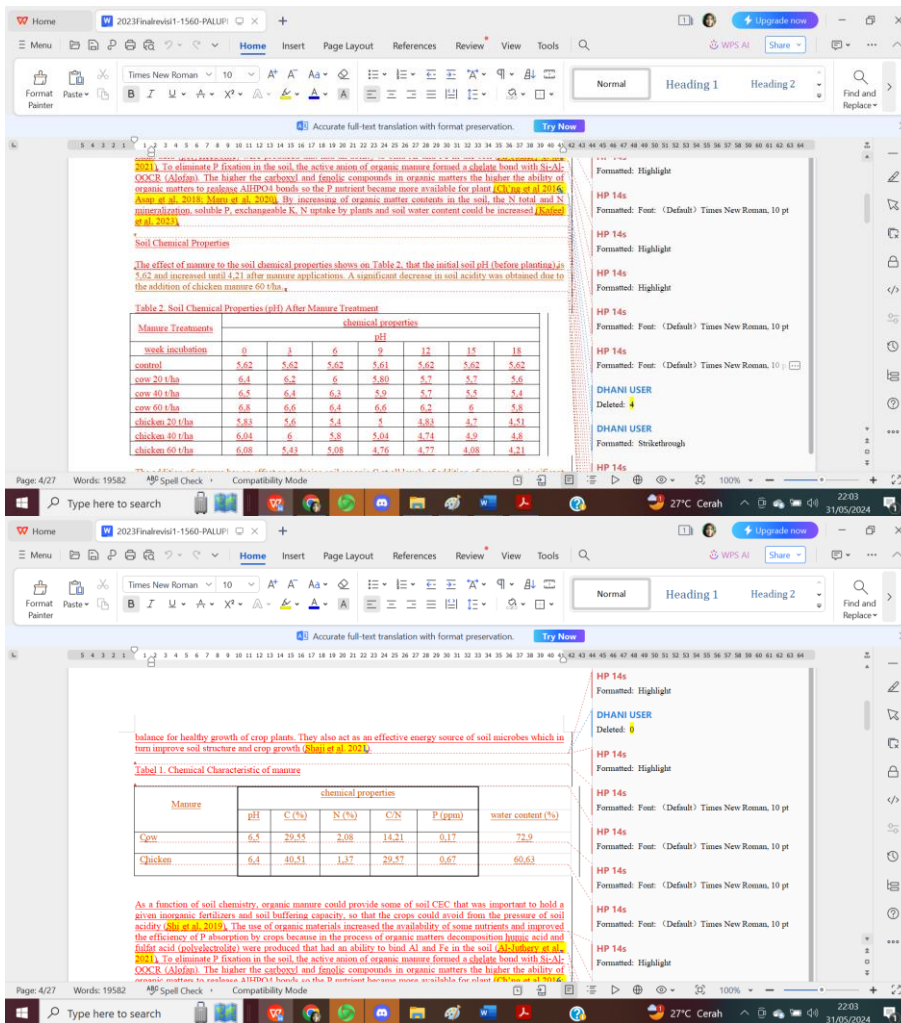
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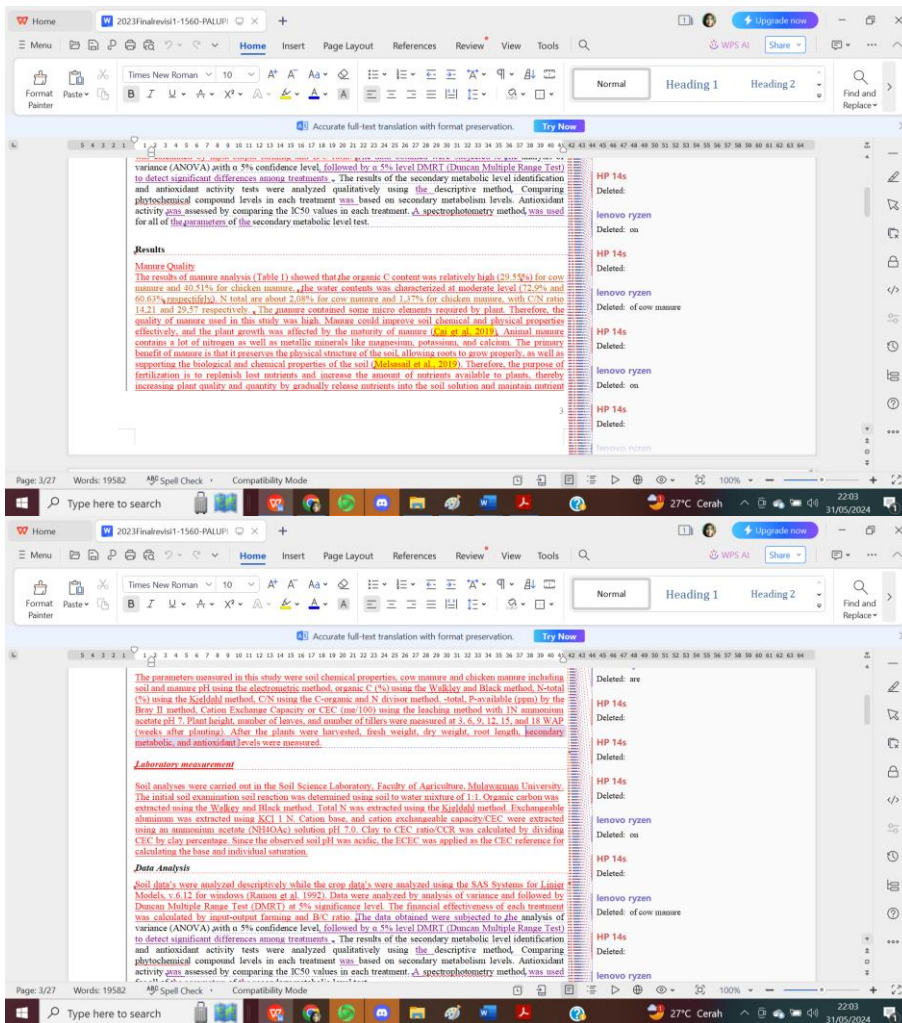
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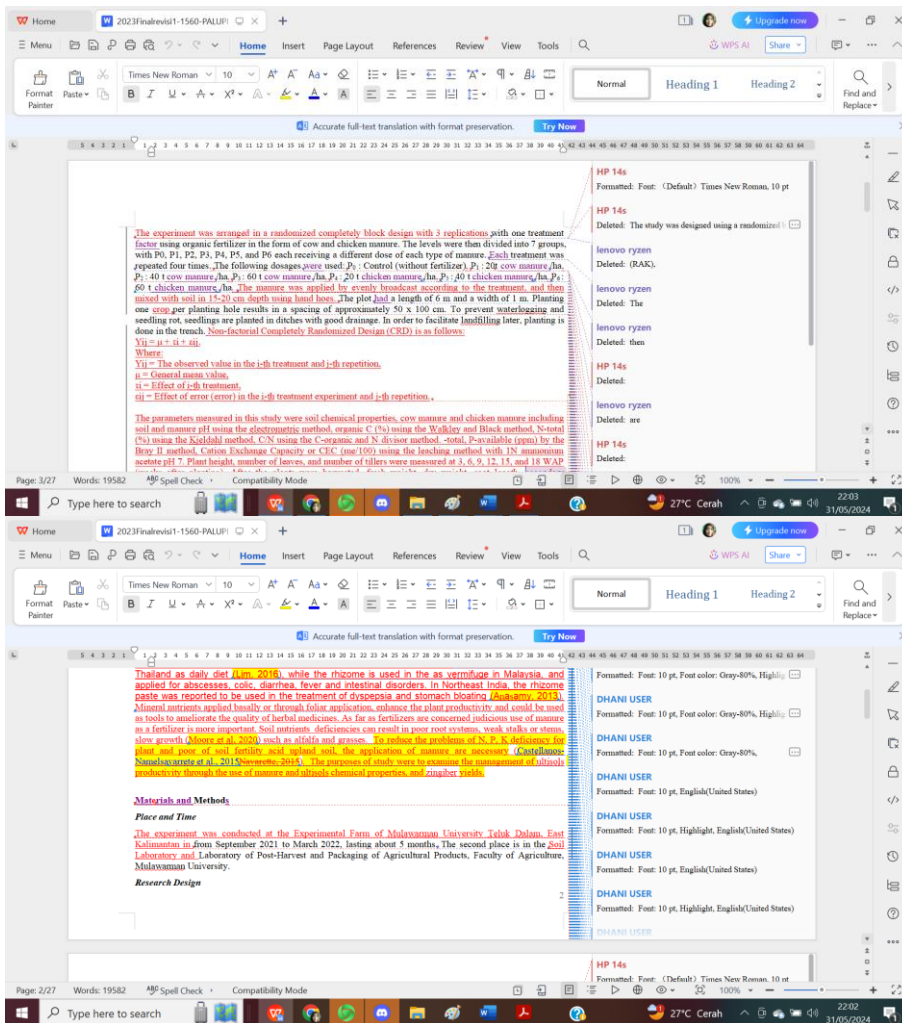




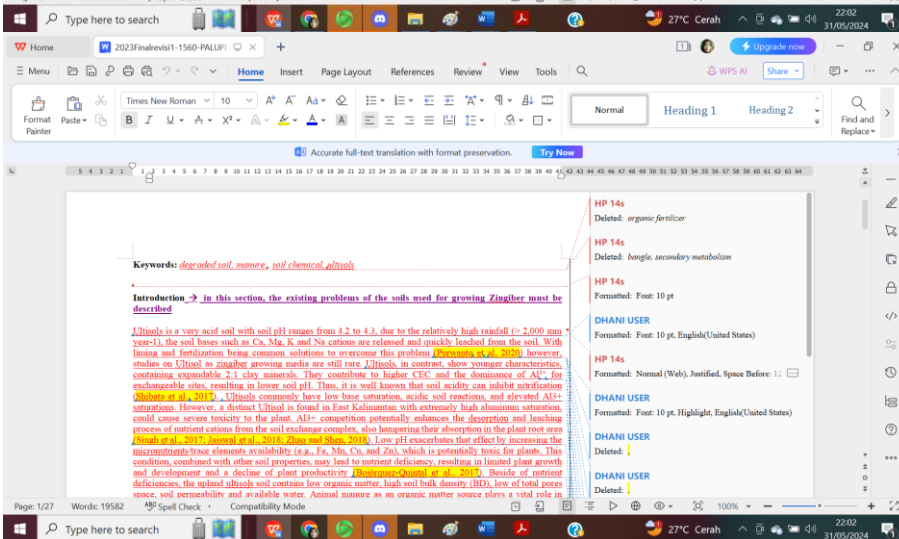
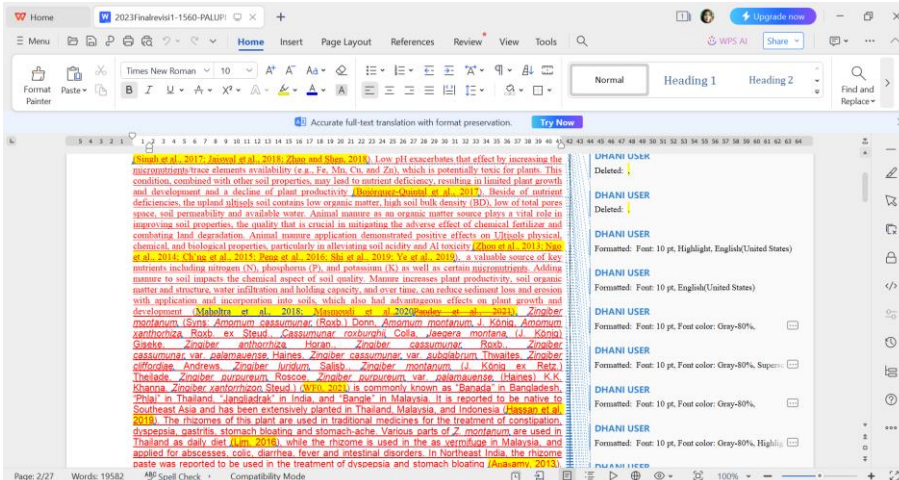








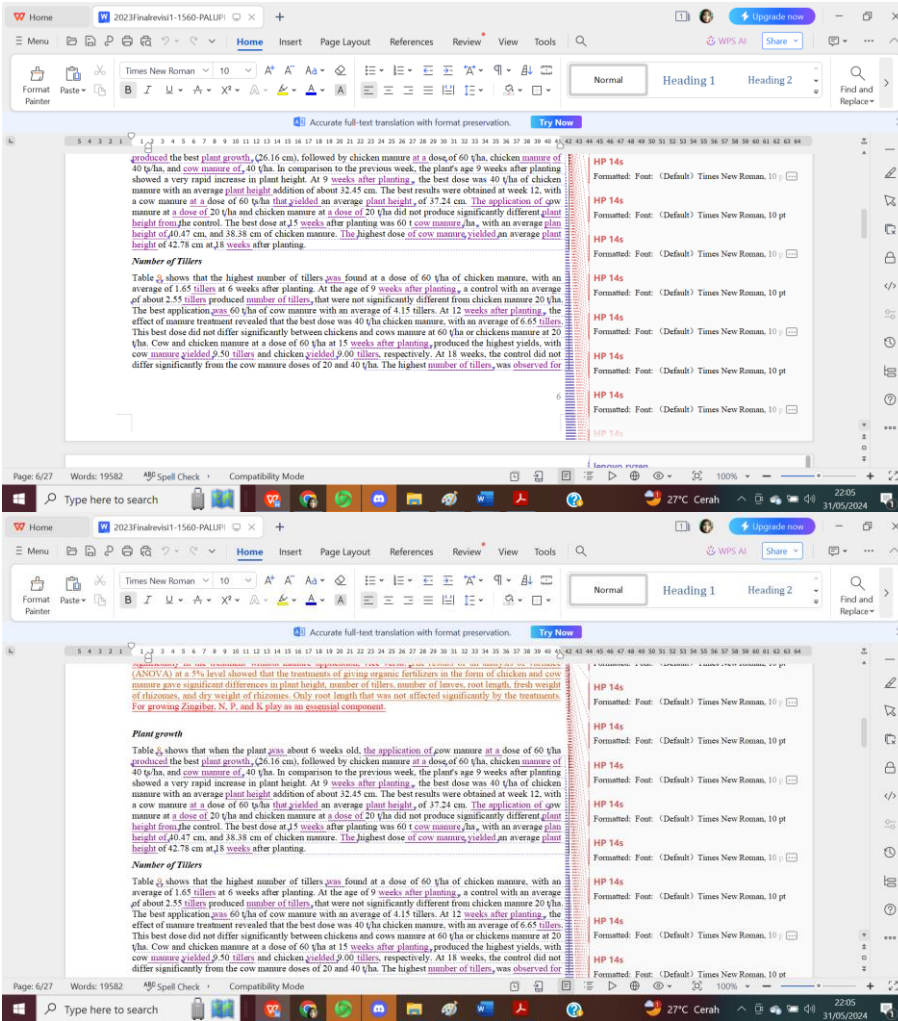
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Table 7. Soil Chemical Properties (CEC) After Manure Treatment

Manure Treatments	chemical properties						
	CEC(meq/100g)						
week incubation	0	3	6	9	12	15	18
control	14.33	14.33	14.32	14.33	14.34	14.33	14.33
cow 20 t/ha	19.15	19.15	19.17	19.17	19.18	19.18	19.19
cow 40 t/ha	19.35	19.36	19.36	19.37	19.37	19.38	19.38
cow 60 t/ha	20.45	20.45	20.46	20.5	20.51	20.55	20.56
chicken 20 t/ha	18.52	18.52	18.53	18.54	18.57	18.58	18.4
chicken 40 t/ha	18.38	18.38	18.4	18.41	18.43	18.44	18.45
chicken 60 t/ha	19	19.12	19.14	19.15	19.18	19.20	19.22

The nutrient added to the soil with low CEC could not be held and easily lost. This condition was reflected to the increasing of soil organic C contents in the treatment with manure application and it did not increase significantly in the treatment without manure application, vice versa. The results of an analysis of variance (ANOVA) at a 5% level showed that the treatments of giving organic fertilizers in the form of chicken and cow manure gave significant differences in plant height, number of tillers, number of leaves, root length, fresh weight of rhizomes, and dry weight of rhizomes. Only root length that was not affected significantly by the treatments. For growing Zingiber, N, P, and K play as an essential component.

**Plant growth**  
Table 6 shows that when the plant was about 6 weeks old, the application of cow manure at a dose of 60 t/ha

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The results of this study indicate that the addition of all levels of manure applications provides an increase in cation exchange capacity. The highest increase in cation exchange capacity was obtained due to the addition of cow manure 60 t/ha as shown on Table 7. Cation exchange in the soil occurs due to the presence of a negative charge from the soil colloid which adsorbs cations in an exchangeable form (Munier et al., 2020). Soils with high CEC are able to absorb and provide nutrients better than soils with low CEC. Because these elements are in the soil adsorption complex, these nutrients are not easily lost or washed away by water. Mineralization of soil organic fractions provides limited supplies of N, S, and macronutrients, while mineral dissolution and surface exchange reactions resupply P, K, Ca, Mg, and micronutrients. Nutrient mobility in soil influences ion transport to plant roots, evaluation of nutrient availability to plants, and ultimately nutrient management decisions (Fahim, 2020).

chicken 60 t/ha	15.68	15.68	15.68	15.69	15.7	15.72	15.73
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Manure input at all application levels has an effect in the form of increasing the P content in the soil along with the increase in incubation time. The highest increase in P in the soil was obtained due to the addition of chicken manure 60 t/ha as shown in Table 6.

**Table 6. Soil Chemical Properties (available P) After Manure Treatment**

Manure Treatments	chemical properties							
	week incubation	0	3	6	9	12	15	18
control		10.52	10.52	10.52	10.52	10.51	10.52	10.52
cow 20 t/ha		0.35	0.36	0.36	0.37	0.37	0.38	0.38
cow 40 t/ha		0.4	0.4	0.42	0.43	0.44	0.45	0.45
cow 60 t/ha		0.44	0.44	0.45	0.47	0.48	0.48	0.49
chicken 20 t/ha		12.78	12.78	12.8	12.81	12.9	12.92	12.93
chicken 40 t/ha		13.42	13.44	13.47	13.48	13.49	13.49	13.5

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The C/N ratio of organic matter is the ratio between the amount of elemental carbon (C) to the amount of elemental nitrogen (N) presented in an organic matter. Good manure must have a C/N ratio <20 (Munier-Corrat **et al.**, 2019). From this study it can be seen that the C/N of soil organics ranged from 2.00 to 11.91 and decreased to 2.05 after 18 weeks of incubation (Table 5).

**Table 5. Soil Chemical Properties (C/N) After Manure Treatment**

Manure Treatments	chemical properties							
	week incubation	0	3	6	9	12	15	18
control		1.29	1.29	1.29	1.29	1.29	1.20	1.21
cow 20 t/ha		10.38	8.00	5.20	4.00	3.11	2.42	2.11
cow 40 t/ha		11.91	11.55	10.00	7.00	5.68	5.63	4.81
cow 60 t/ha		10.77	9.13	8.44	8.25	7.71	6.89	5.65
chicken 20 t/ha		2.00	1.83	1.68	1.58	1.35	1.00	0.86
chicken 40 t/ha		2.10	2.00	1.86	1.27	1.09	0.94	0.74
chicken 60 t/ha		4.90	4.45	2.97	2.63	2.42	2.16	2.05

Manure input at all application levels has an effect in the form of increasing the P content in the soil along with the increase in incubation time. The highest increase in P in the soil was obtained due to the addition of chicken manure 60 t/ha as shown in Table 6.

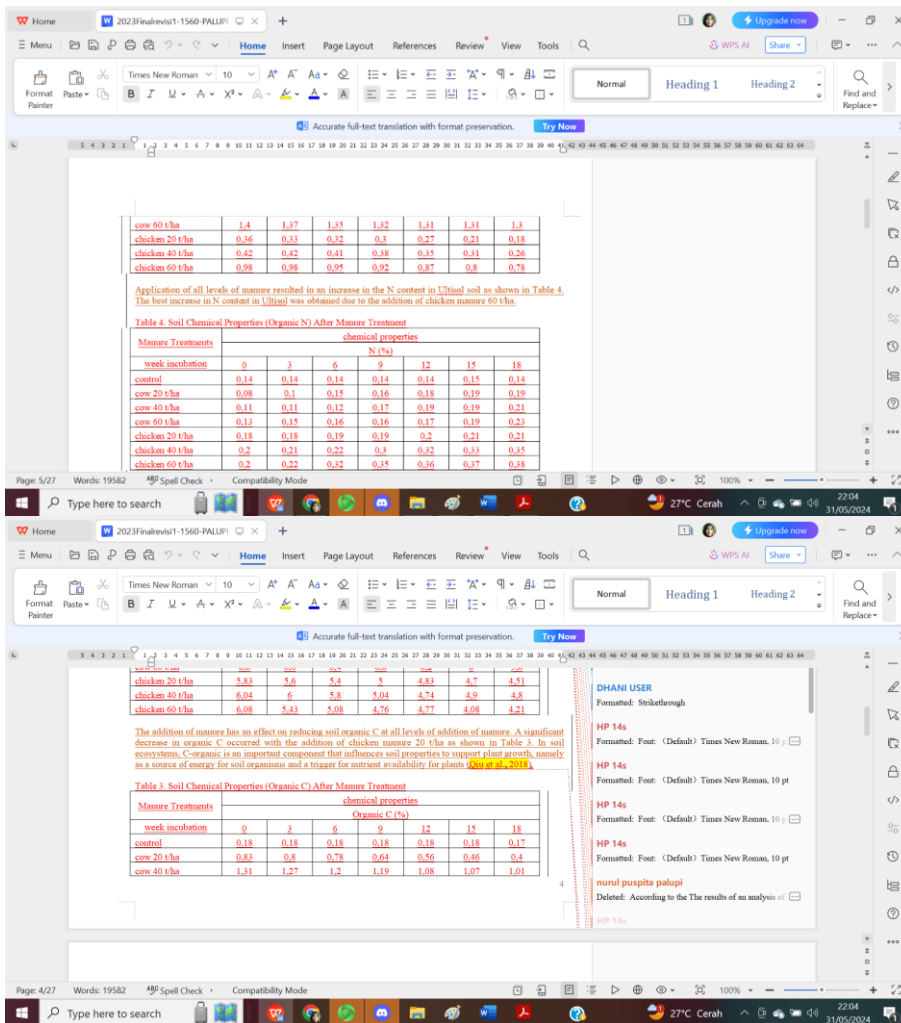
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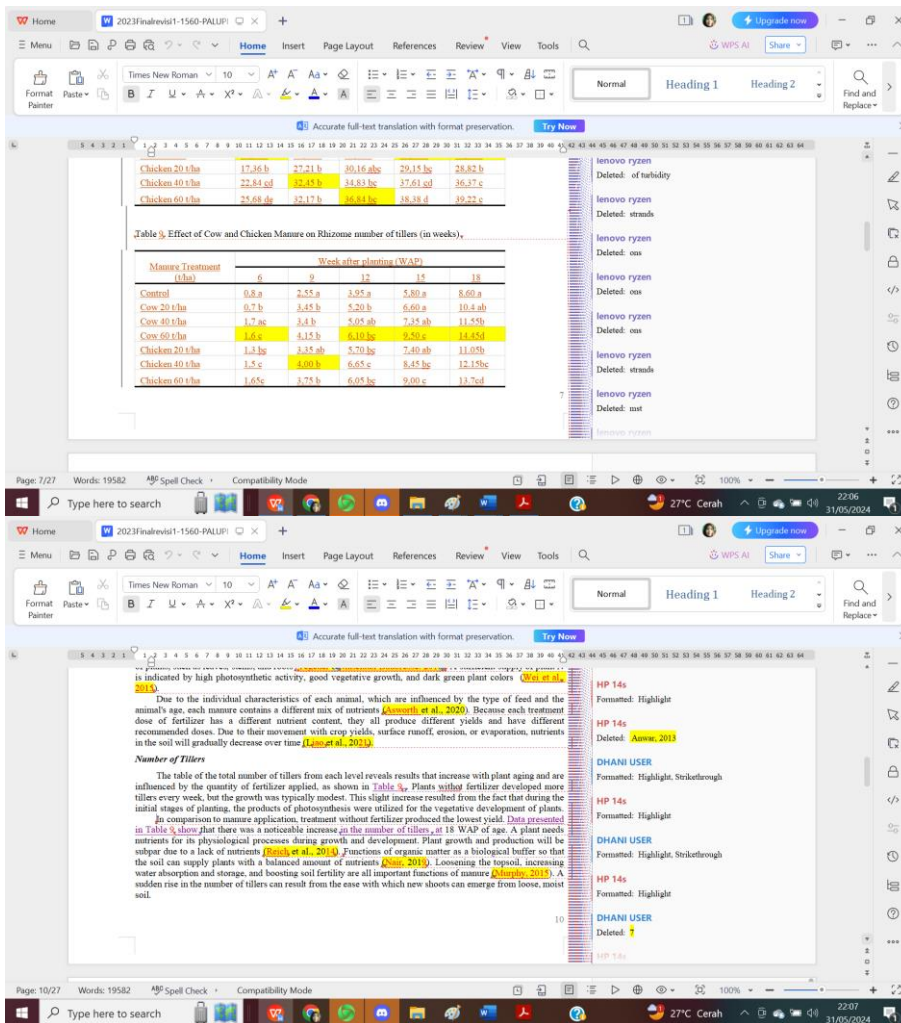
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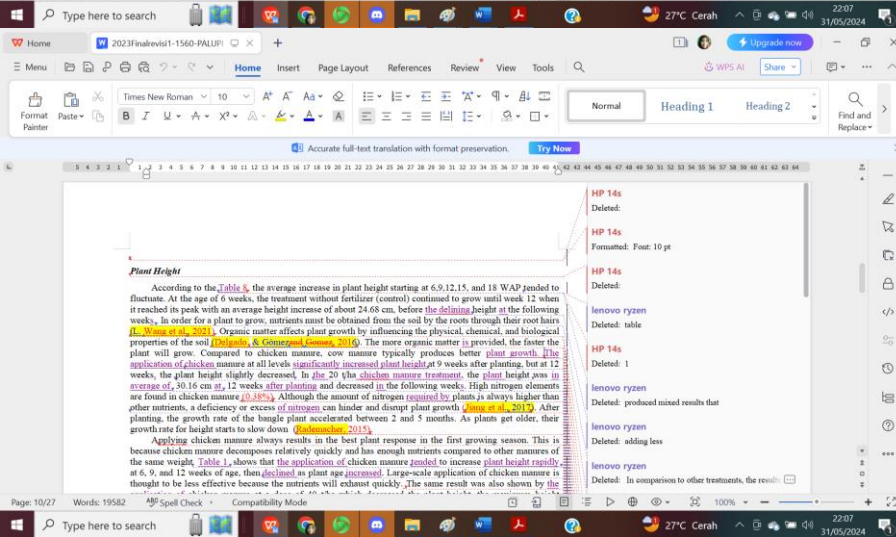
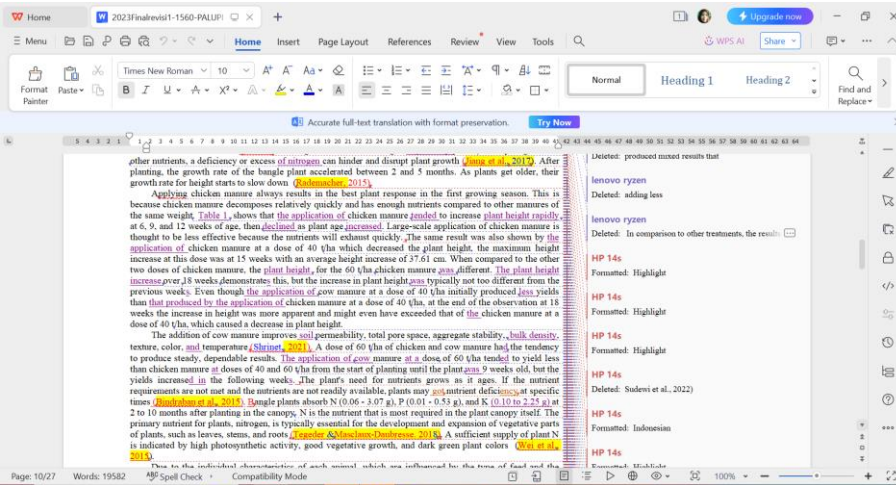
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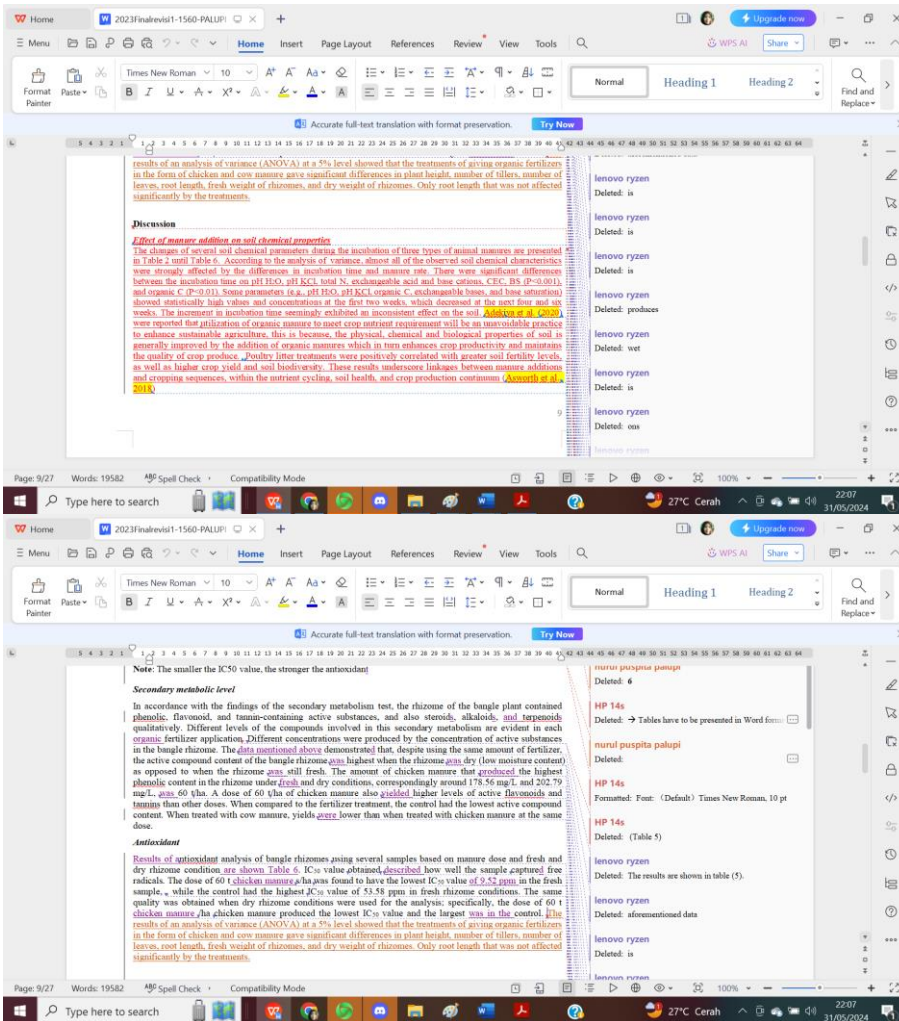
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**Table 13. Antioxidant based on IC50 value**

Antioxidant (IC50) (mg/L)	Control	Manure Treatment (t/ha)					
		20	40	60	20	40	60
Fresh rhizome	52.58	48.55	47.56	47.14	22.23	27.42	9.92
Dry rhizome	40.91	38.30	34.78	34.46	24.41	14.05	3.02

Note: The smaller the IC50 value, the stronger the antioxidant

**Secondary metabolic level**

In accordance with the findings of the secondary metabolism test, the rhizome of the bangle plant contained phenolic, flavonoid, and tannin-containing active substances, and also steroids, alkaloids, and saponins metabolites. Different levels of the compounds involved in this secondary metabolism are available in each

Condition

Condition	Manure Treatment (t/ha)	Level (mg/L)		
		Phenolic	Flavonoid	Tannin
Fresh	Control	31.64	21.08	2034.83
	Cow 20 t/ha	43.18	41.55	2091.50
	Cow 40 t/ha	77.03	64.30	3151.50
	Cow 60 t/ha	86.90	71.14	3198.17
	Chicken 20 t/ha	97.67	74.79	4086.50
	Chicken 40 t/ha	107.54	78.71	4144.83
Dry	Control	124.97	38.44	1932.33
	Cow 20 t/ha	133.56	67.23	2504.00
	Cow 40 t/ha	148.44	68.76	3034.83
	Cow 60 t/ha	181.38	99.43	4734.67
	Chicken 20 t/ha	175.23	120.06	4903.00

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Table 11. Effect of Cow and Chicken Manure on Root Length and Rhizome Weight (in 18 week).

Manure Treatment (t/ha)	Root length (cm)	Rhizome wet weight (g)	Rhizome dry weight (g)
Control	31.90 ab	415.25 ab	93.75 ab
Cow 20 t/ha	24.36 a	444.75 ab	86.00 a
Cow 40 t/ha	35.36 b	530.00 bc	119.00 ab
Cow 60 t/ha	36.50 b	522.00 d	175.75 c
Chicken 20 t/ha	33.20 ab	393.25 a	78.25 a
Chicken 40 t/ha	38.95 b	487.50 abc	99.75 ab
Chicken 60 t/ha	41.03 b	618.75 c	134.75 bc

Notes: Based on Duncan Multiple Range Test (DMRT) at a 5% significance level, values in the same column that are followed by the same letter do not differ significantly.

Table 12. Secondary Metabolic Levels in Bangle Rhizome in Each Treatment (mg/L).

Condition	Manure Treatment (t/ha)	Level mg/L		
		Phenolic	Flavonoid	Tannin
Control		11.64	23.08	2034.83
Cow 20 t/ha		43.18	41.55	2991.50

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Manure Treatment (ton/ha)	g
Control	4.70 a
Cow 20 ton/ha	5.55 a
Cow 40 ton/ha	9.65 b
Cow 60 ton/ha	10.05 b
Chicken 20 ton/ha	9.20 b
Chicken 40 ton/ha	9.65 b
Chicken 60 ton/ha	11.05 b

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Table 13. Effect of Cow and Chicken Manure on number of Rhizome Leaves (in weeks).

Manure Treatment (t/ha)	Week after planting (WAP)				
	6	9	12	15	18
Control	4.70 a	19.60 a	35.80 a	52.25 a	77.55 a
Cow 20 t/ha	5.55 a	25.00 ab	41.05 ab	55.65 a	83.80 a
Cow 40 t/ha	9.65 b	28.25 abc	42.70 abc	66.00 ab	95.15 ab
Cow 60 t/ha	10.05 b	37.95 c	52.80 c	84.55 c	116.65 c
Chicken 20 t/ha	9.20 b	26.00 ab	40.25 ab	56.85 a	81.75 a
Chicken 40 t/ha	9.65 b	30.90 bc	43.25 abc	67.85 bc	91.35 a
Chicken 60 t/ha	11.05 b	30.45 abc	50.75 bc	79.45 bc	105.80 bc

Table 14. Effect of Cow and Chicken Manure on Root Length and Rhizome Weight (in 18 week).

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Manure Treatment (ton/ha)	g
Control	0.8 a
Cow 20 ton/ha	0.7 b
Cow 40 ton/ha	1.7 ac
Cow 60 ton/ha	1.6 c
Chicken 20 ton/ha	1.3 bc
Chicken 40 ton/ha	1.5 c
Chicken 60 ton/ha	1.65 c

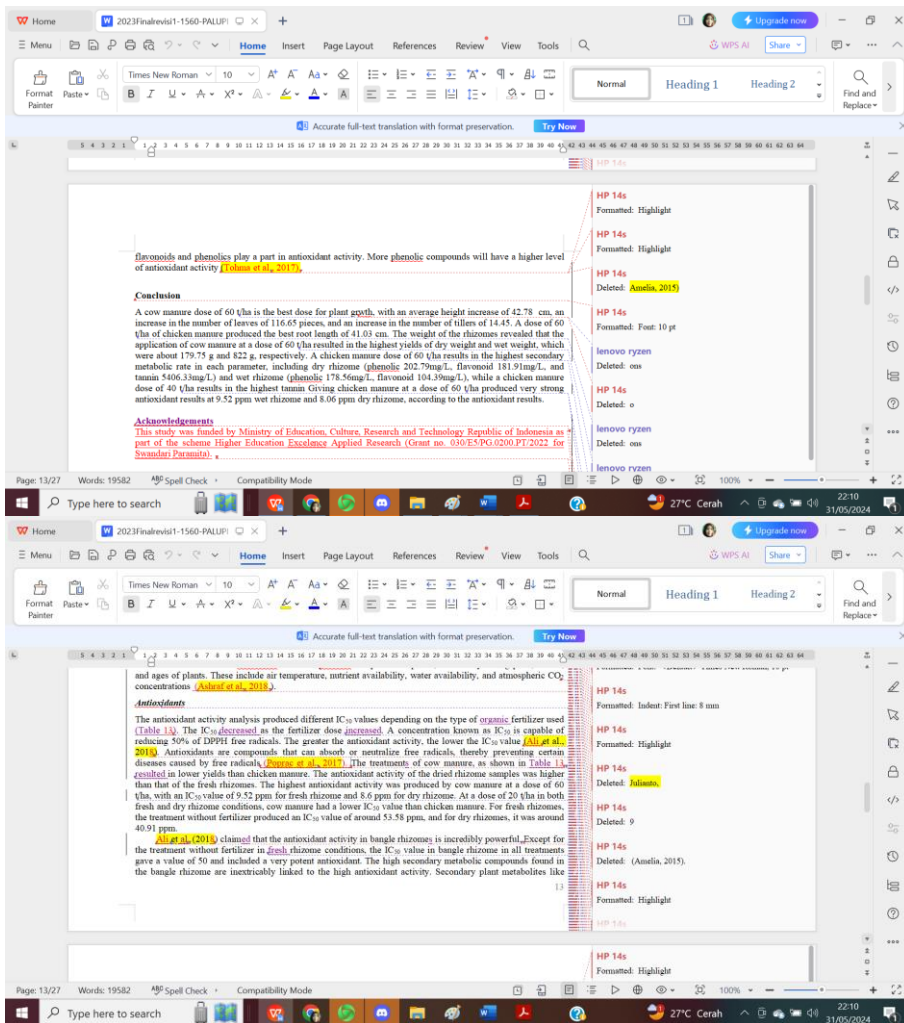
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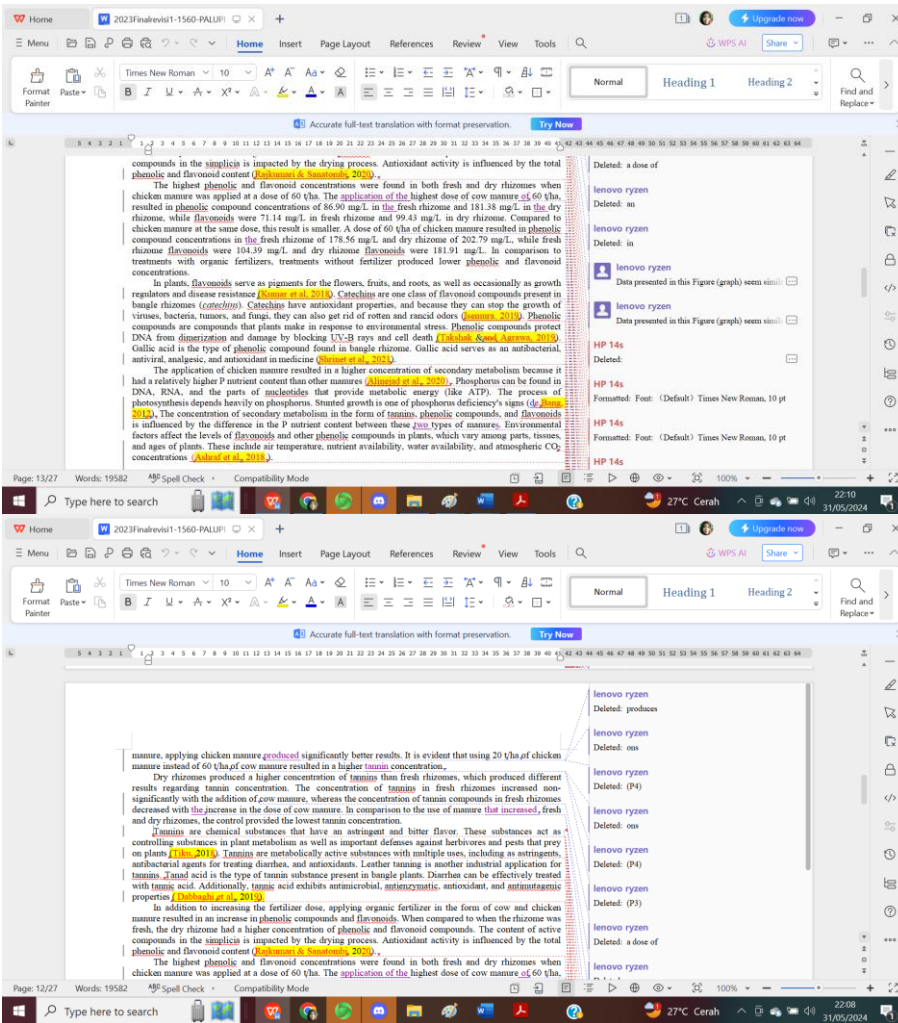
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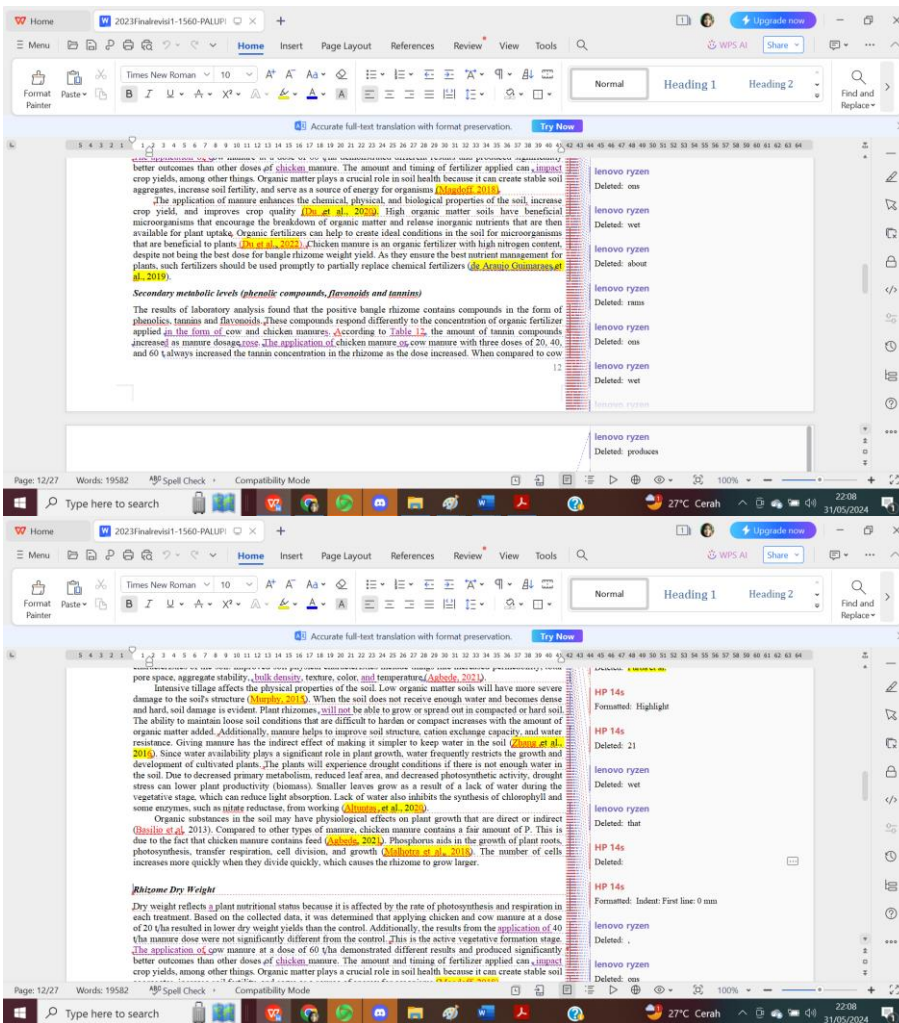
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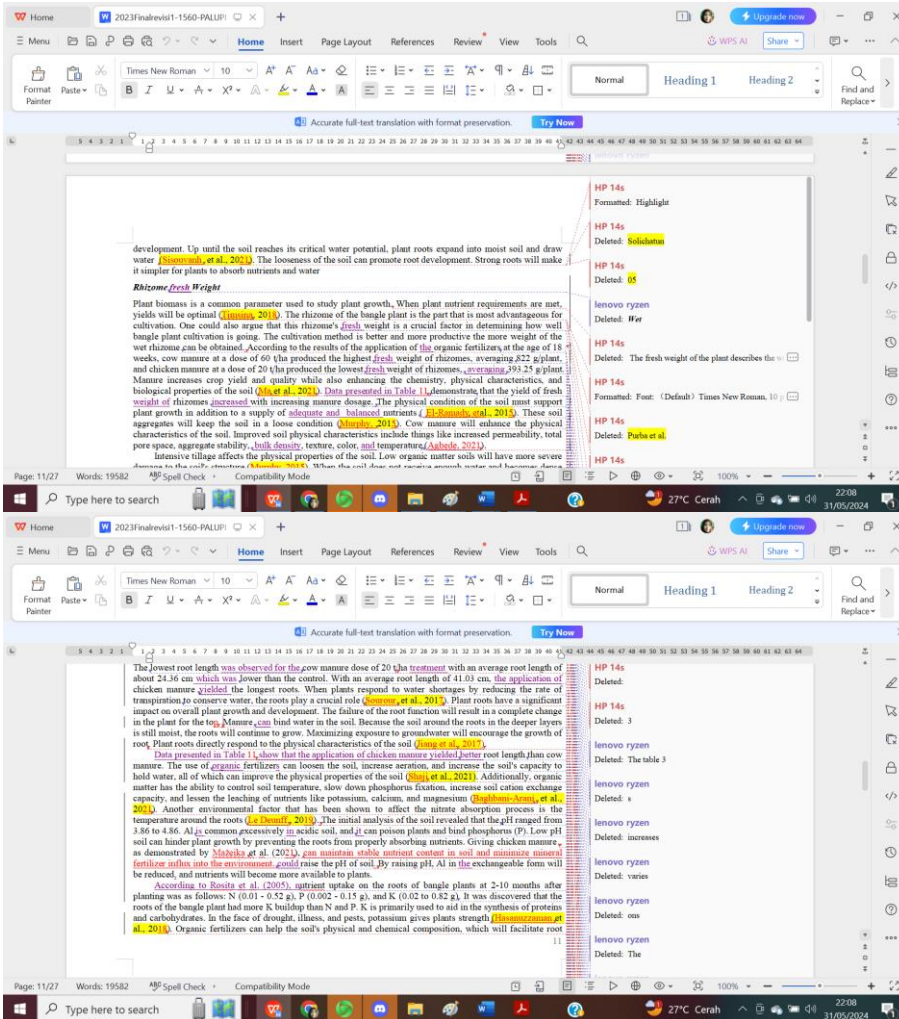
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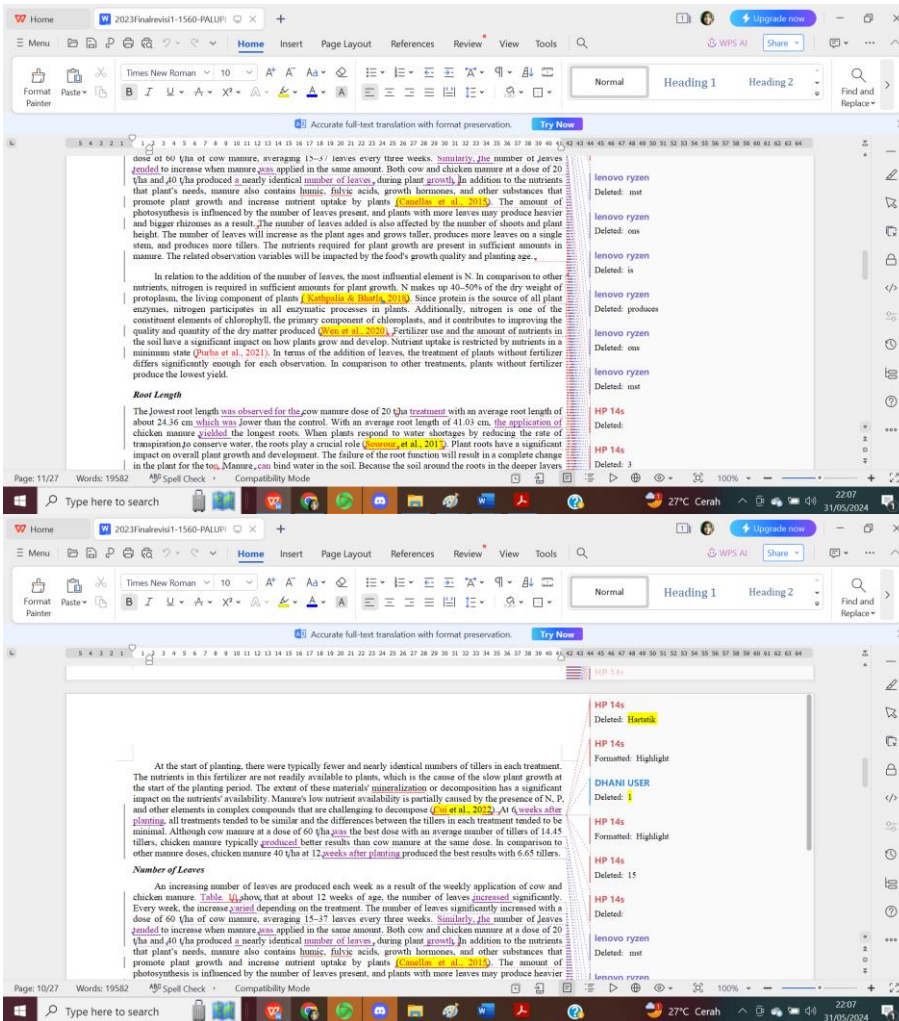
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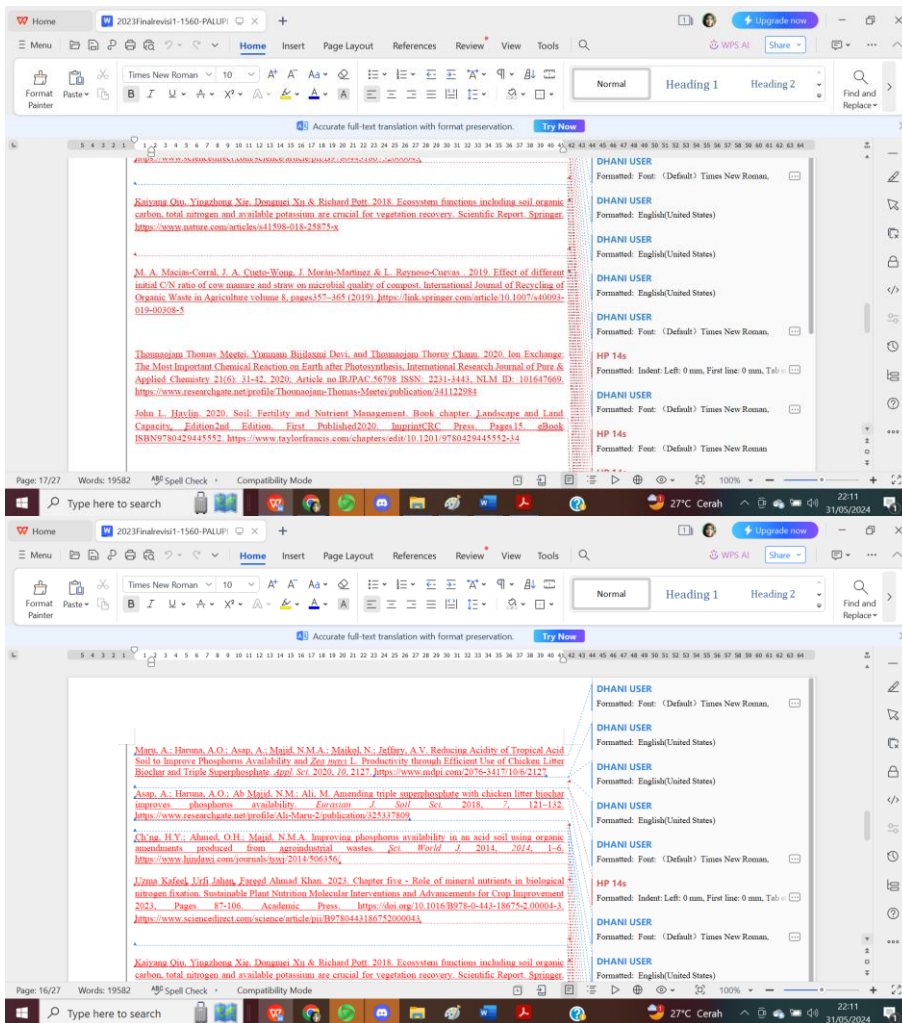






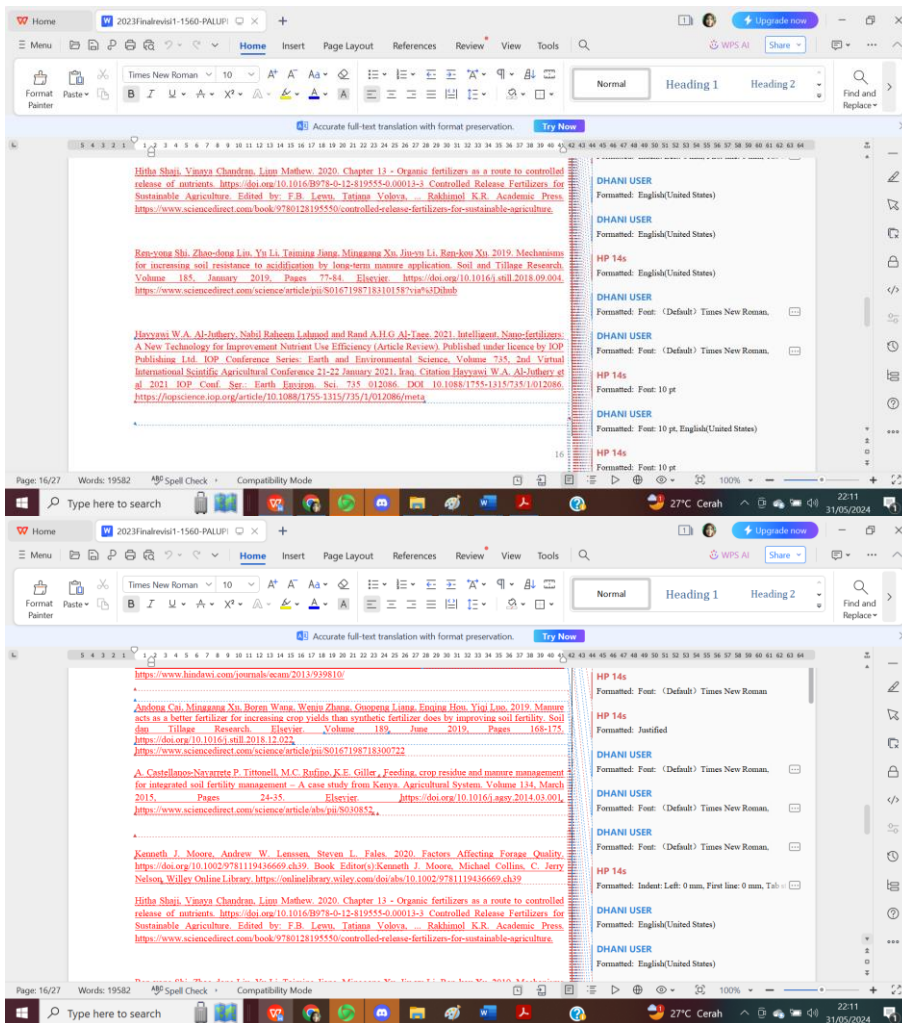






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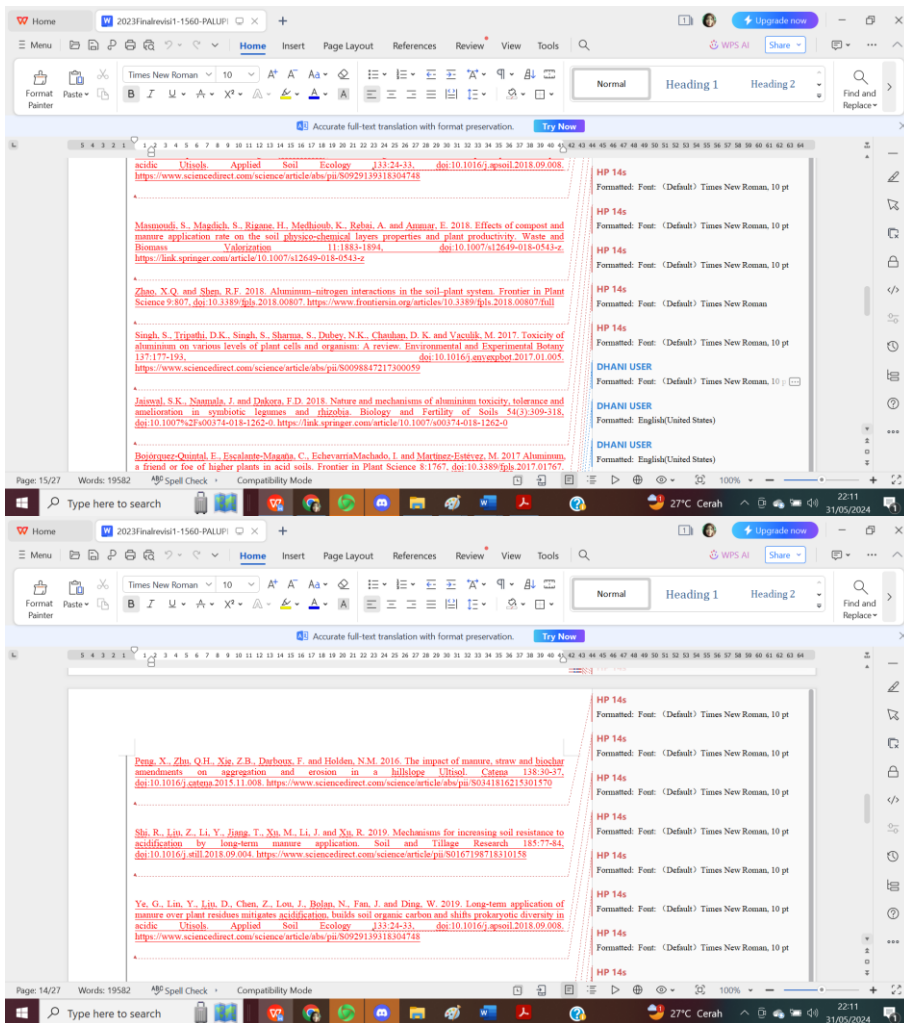
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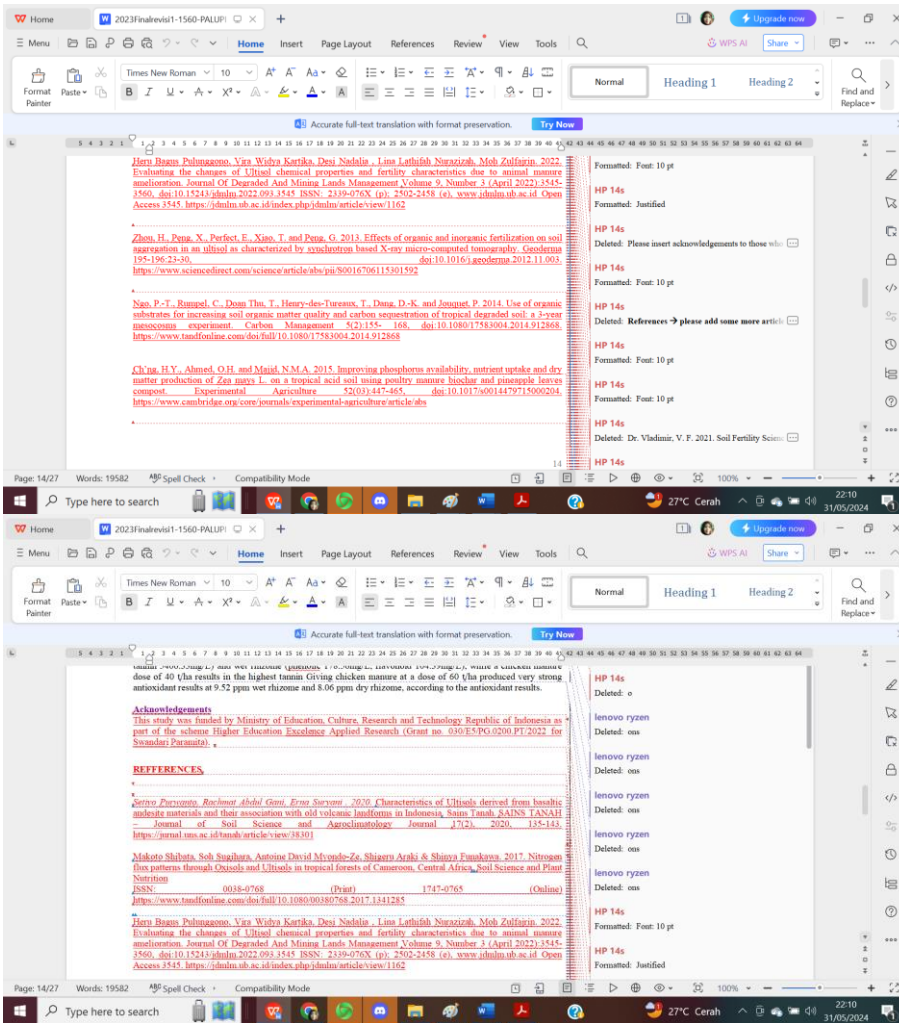
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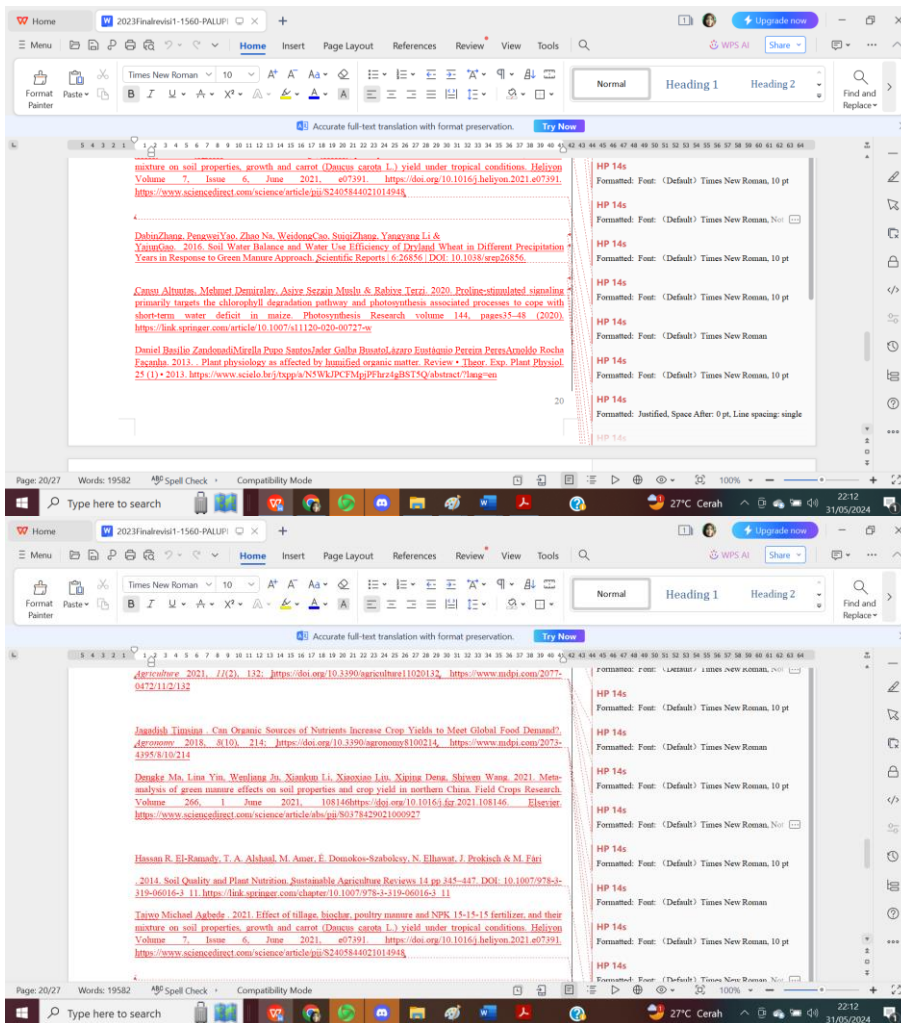
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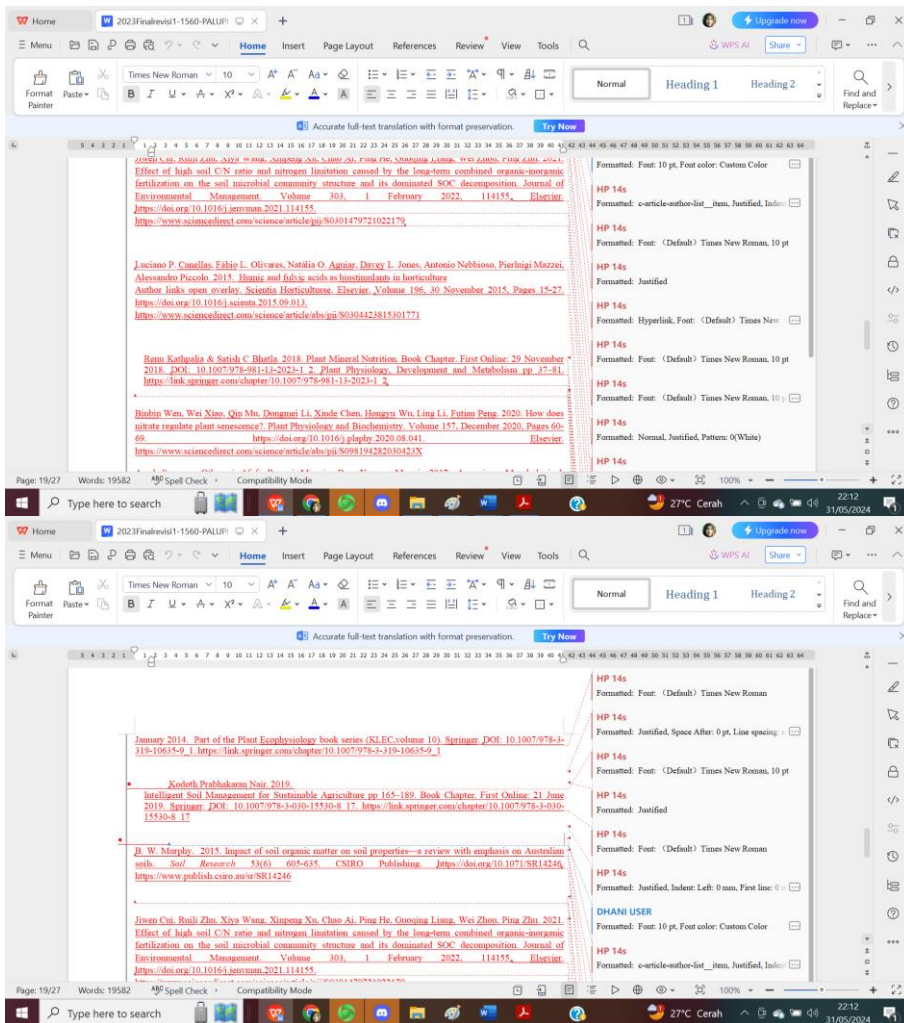
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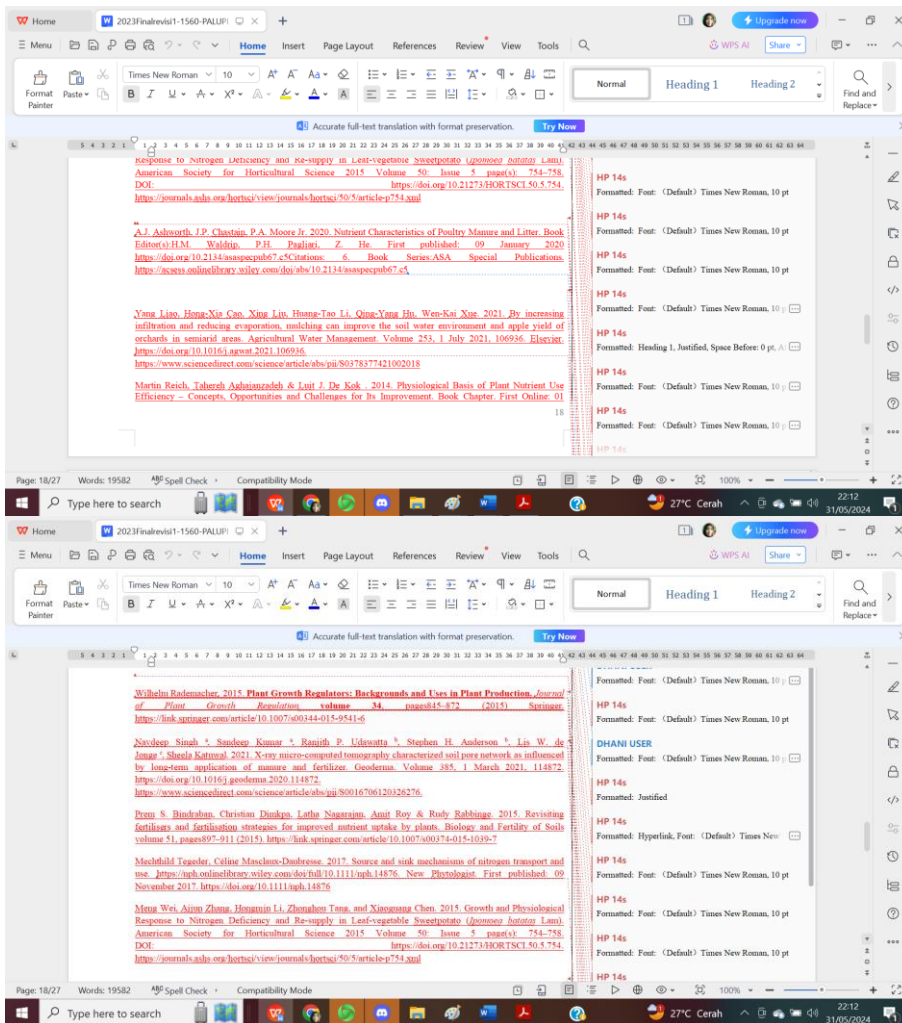


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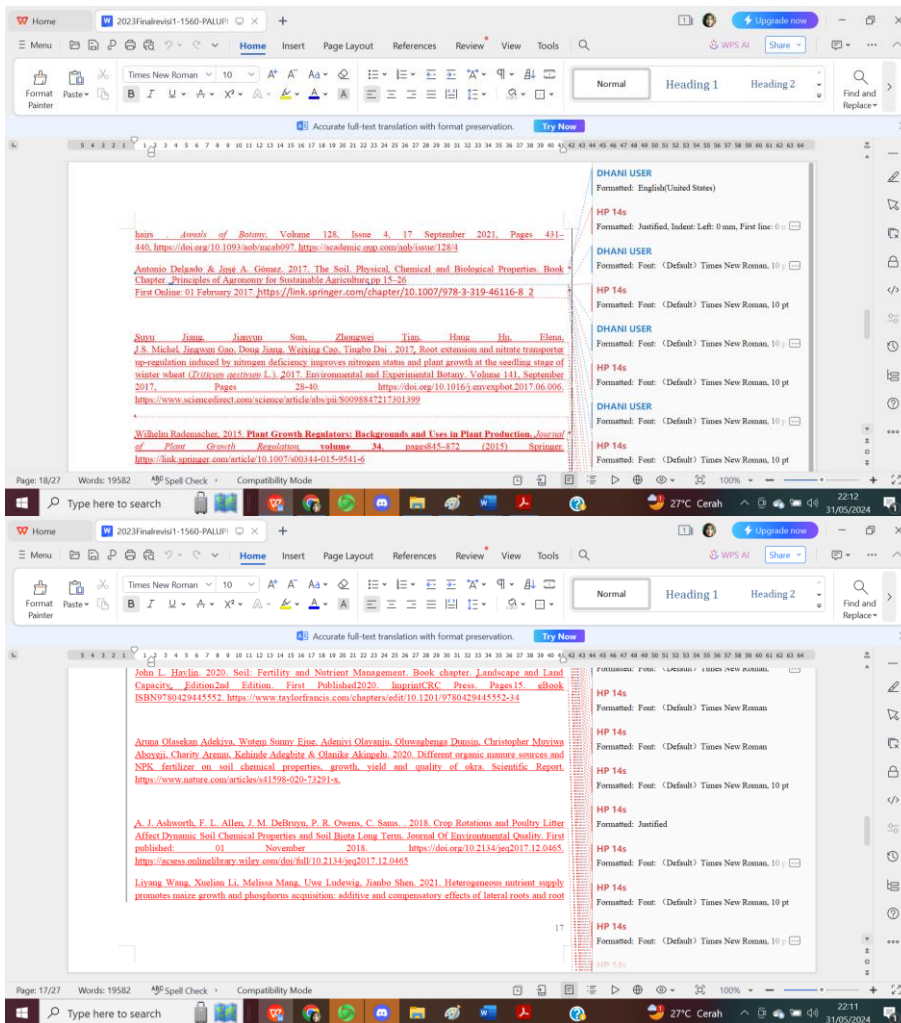


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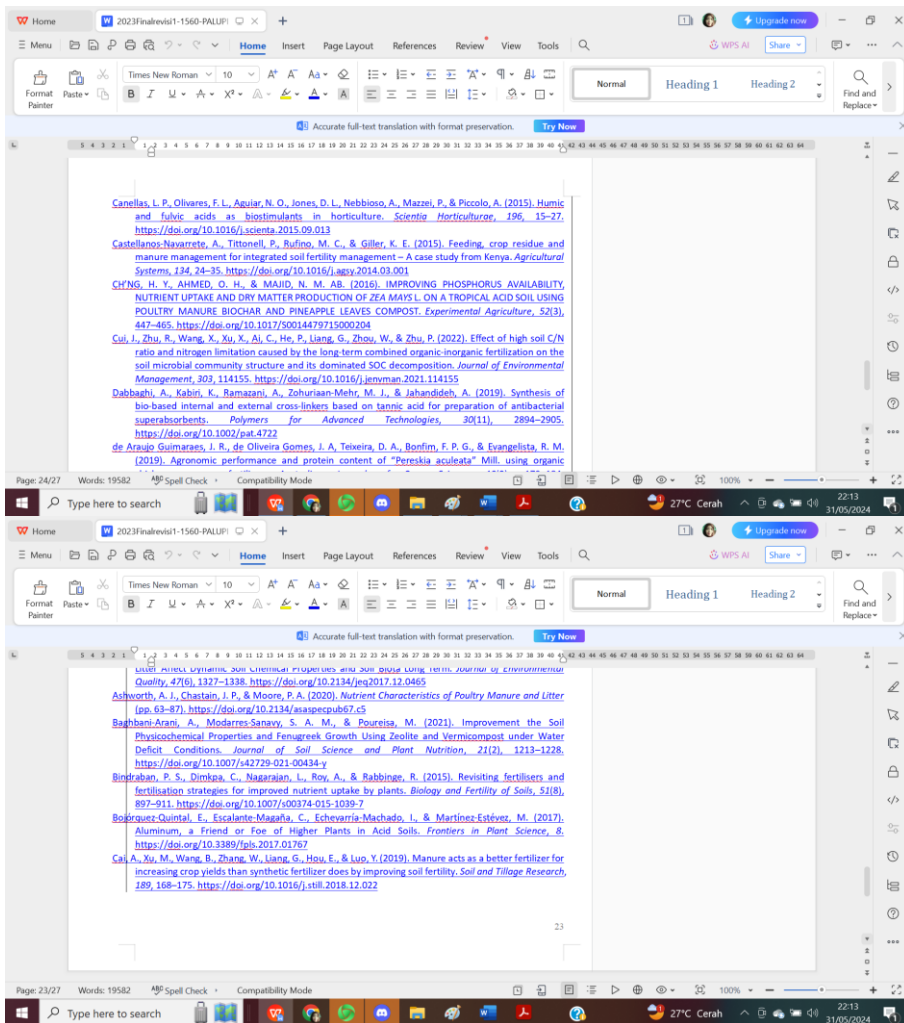


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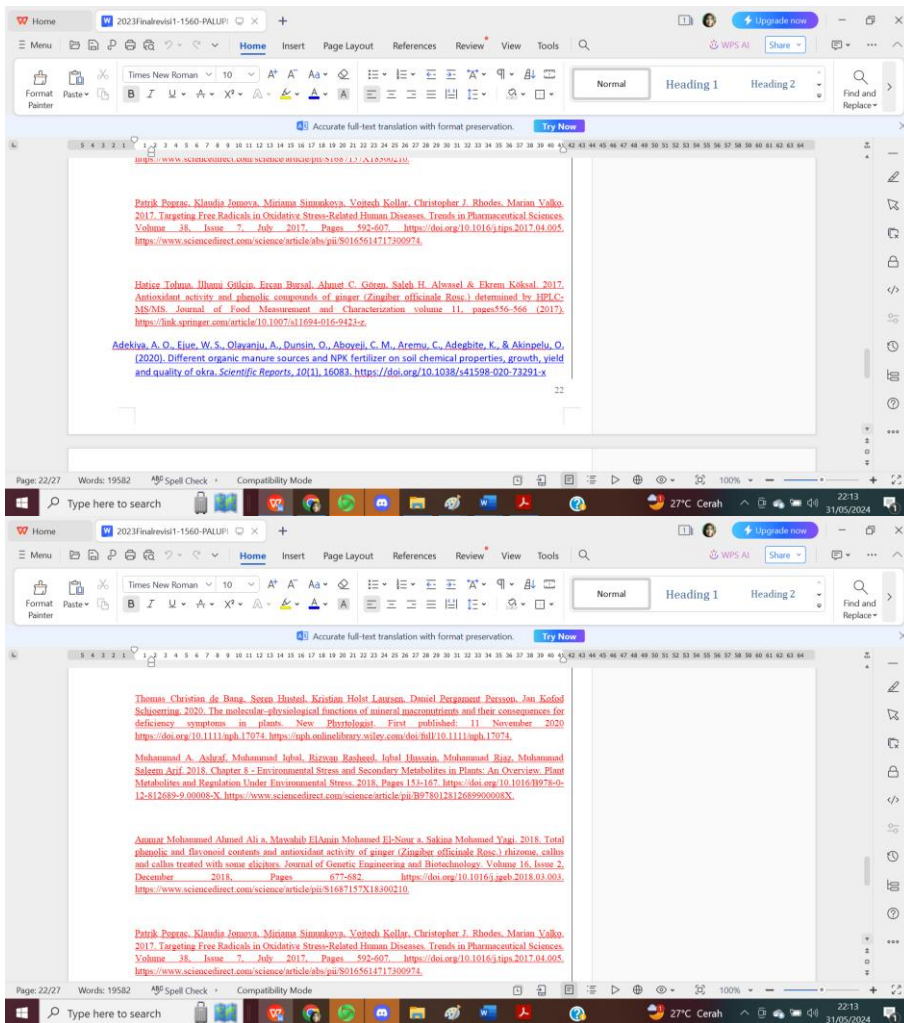


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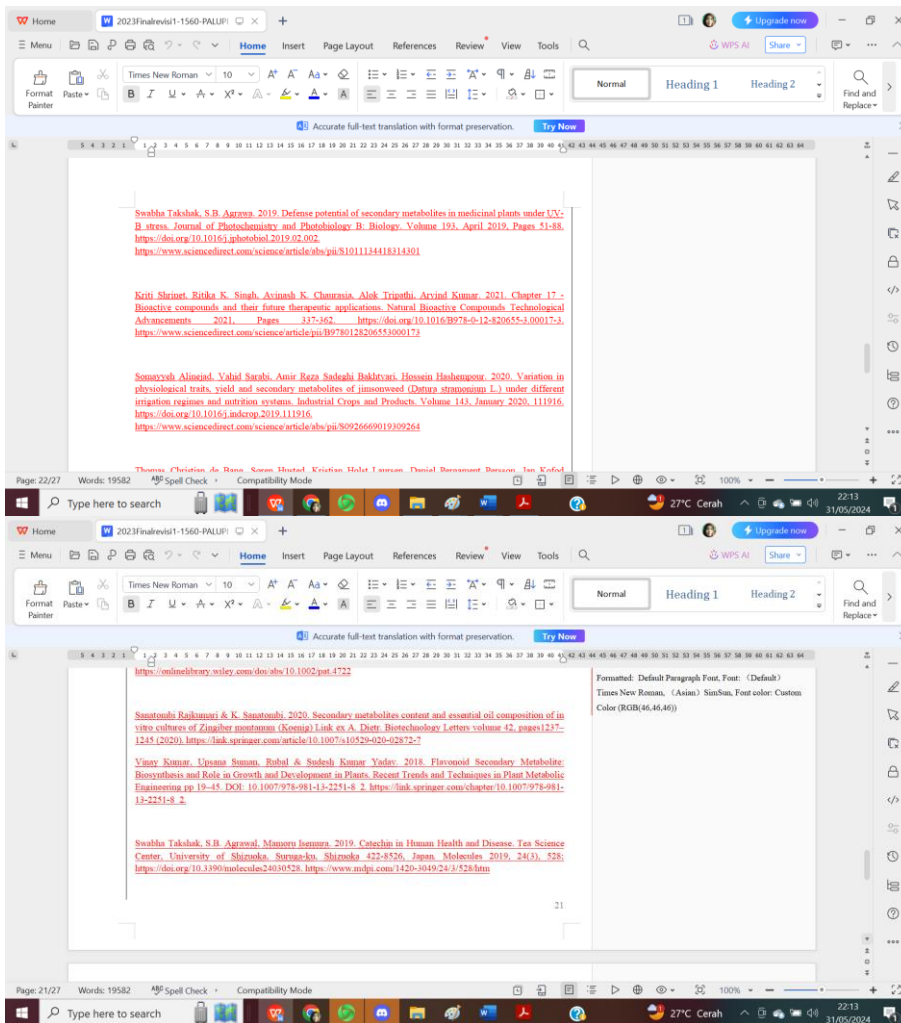


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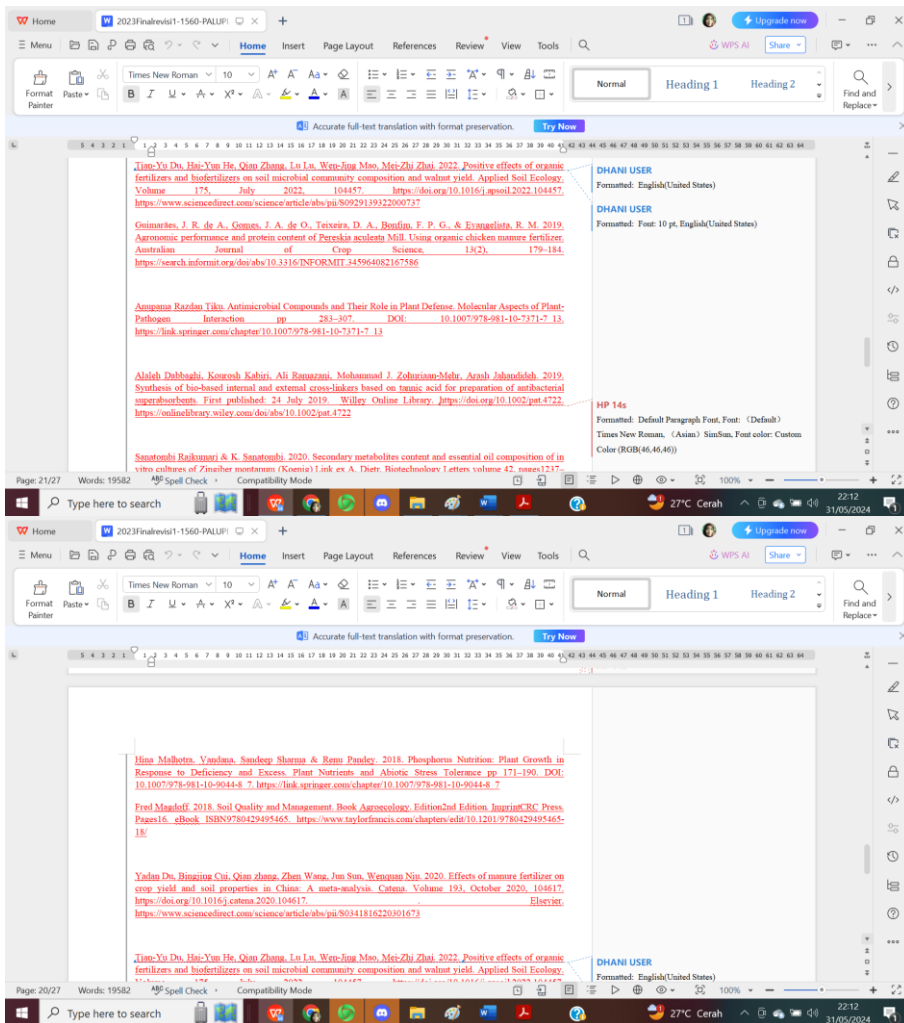


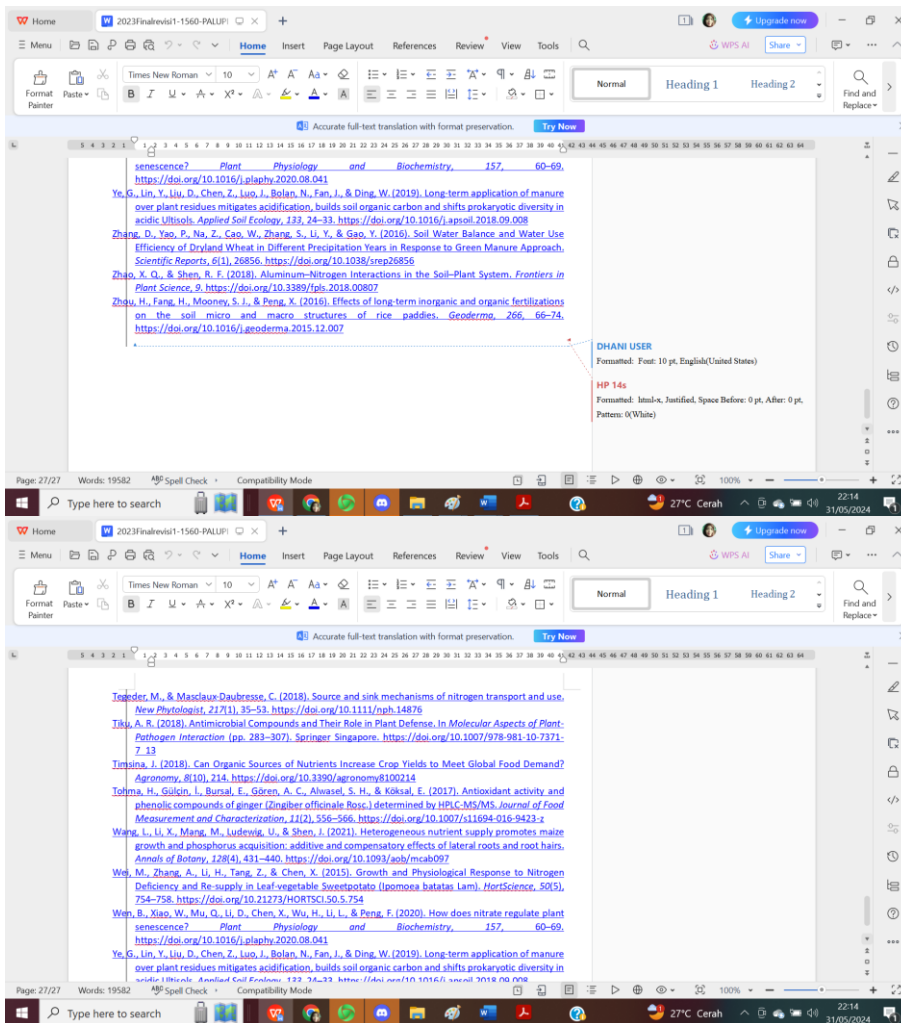


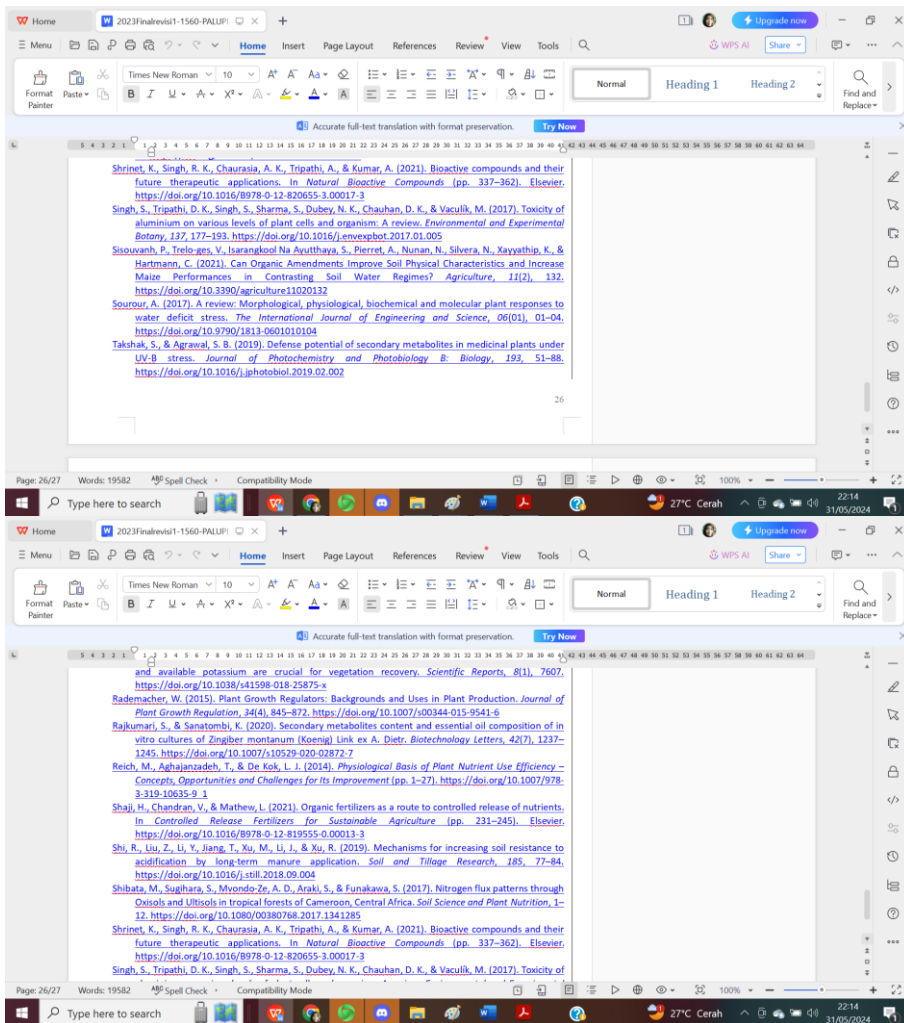
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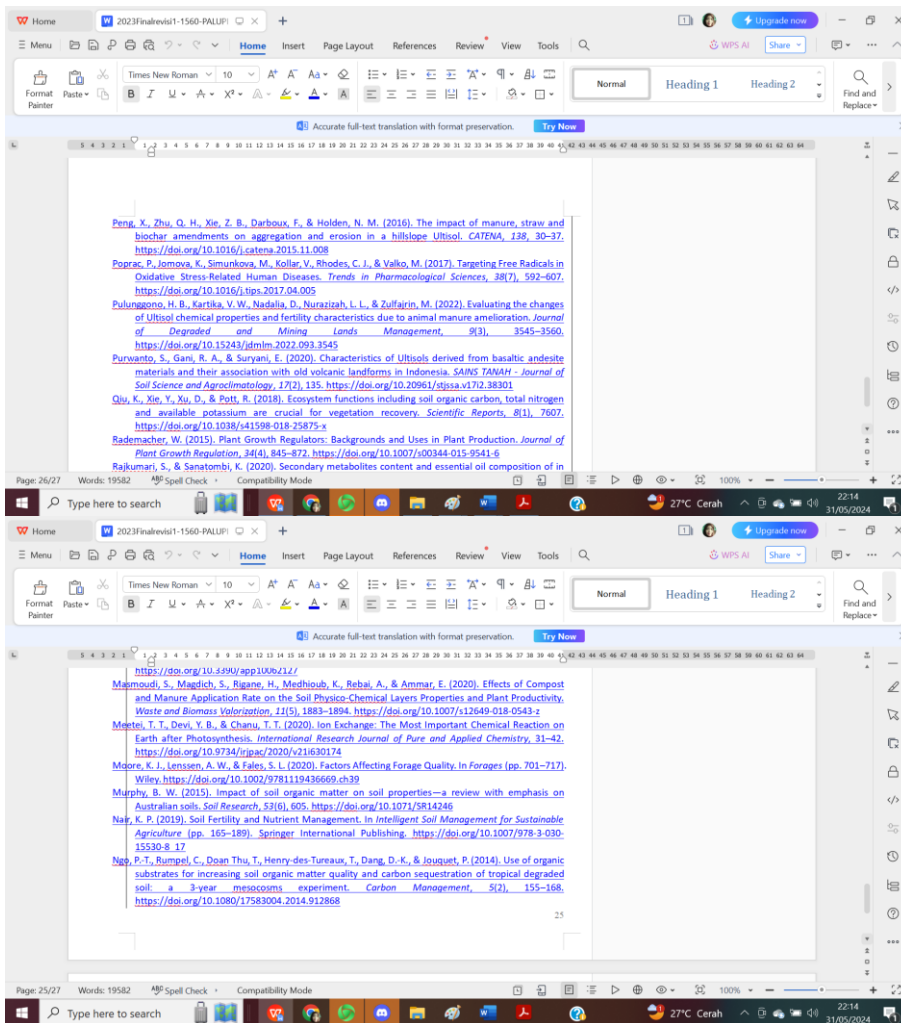




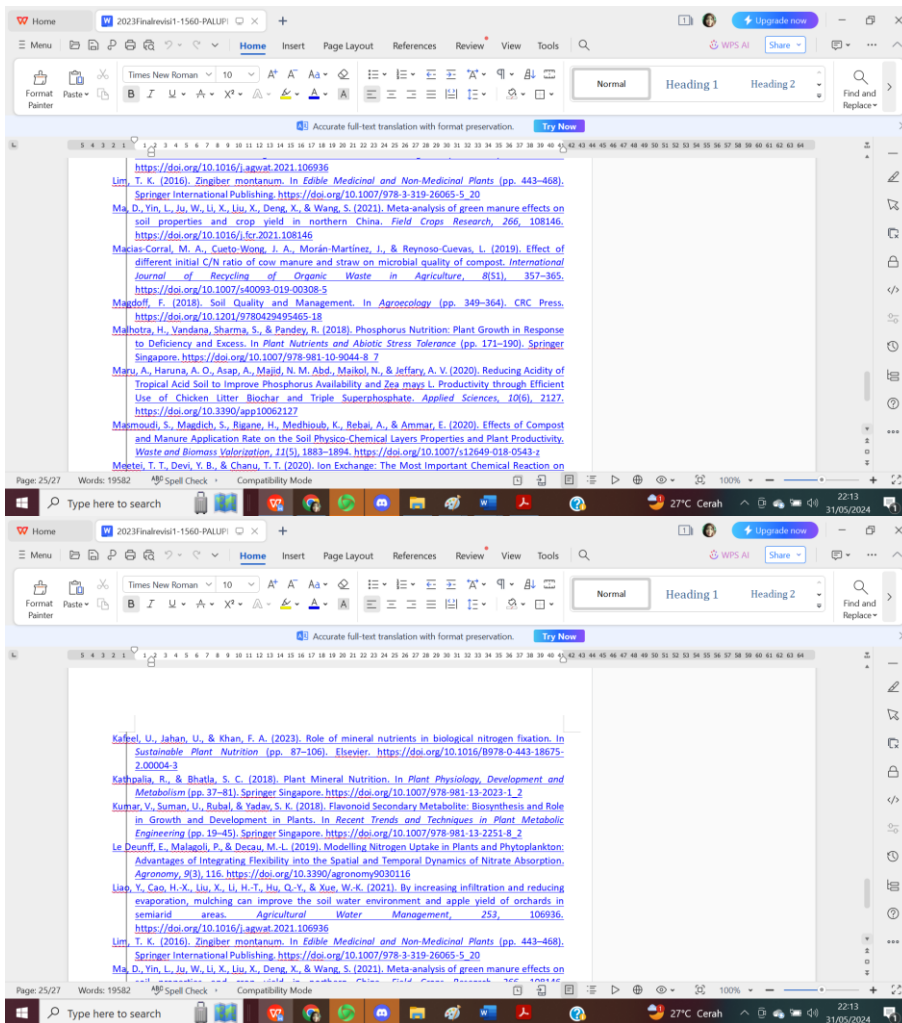


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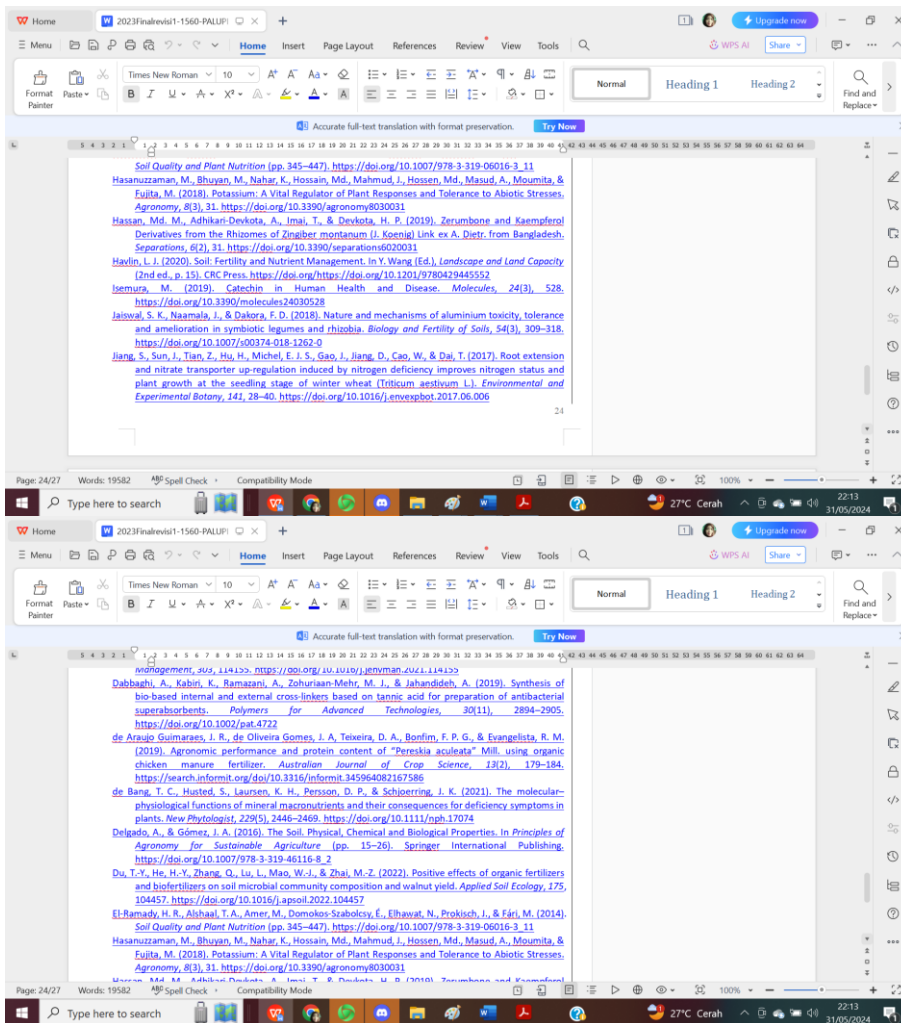




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Setelah dilakukan revisi :

**COMMENTS:** (please see the detailed corrections and comments inserted in this manuscript)

5. This manuscript **only deals with the agronomic aspects of *Zingiber montanum*, not soil management** (the effect of adding organic fertilizer on soil properties) → is the soil used in this study considered degraded soil? What is the soil type used in this study?
6. For this manuscript to fit within the scope of this journal (the management of degraded land/soil and management of mining land/soil), the authors have to **make adjustments by adding data on the soil properties affected by the application of cow and chicken manures**, which in turn affects plant growth and yield. The effects of organic fertilizers on soil properties should be discussed accordingly,

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7. The rationale behind this study is unclear. What was the problem to be solved? *Zingiber montanum* production (agronomic aspects) or soil management aspects?
8. Methods are trivial (not presented in detail)

Please be careful in using tenses; when describing Methods and Results, you should use the past tense. The present tense is appropriate for accepted facts, such as the background information presented in the Introduction. In addition, you may use the present tense when you discuss your results and conclusions

**Recommendation: revisions and adjustments are required:** a substantial amount of work is necessary to raise this manuscript to a standard research article

### **The impact of manure on degraded ultisols on the growth and secondary metabolite levels of *Zingiber montanum***

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#### **Abstract**

Acid upland soil in Indonesia has a potential for agricultural development but it has constraints low of organic C, N, P and available P as well as the soil chemical properties have been degraded. The use of manure was an alternative to improve land productivity and crop yields. The objective of the study was to examine the effects of manure on chemical properties of ultisols, yield and secondary metabolic of zingiber montanum. The experiment was carried out at Research Laboratory Teluk Dalam in 2022 using randomized completely block design. The treatments are consisting of organic fertilizer in the form of cow and chicken manure, and the levels were namely P0 (control), P1 (cow manure 20 t/ha), P2 (cow manure 40 t/ha), P3 (cow manure 60 t/ha), P4 (chicken manure 20 t/ha), P5 (chicken manure 40 t/ha), and P6 (chicken manure 60 t/ha), and then repeated four times in 7 times of incubations. The results showed that the application of manure improved soil chemical characteristics and affected the yield of zingiber montanum. The addition of chicken manure 60 t/ha has the effect of increasing N and P of soil and cow manure 60 t/ha has the effect of increasing CEC of soil. And indicated that a cow manure dose of 60 t/ha is the best dose for plant growth, as measured by an average height increase of 42.78 cm, an increase in the number of leaves (116.65 pieces), and an increase in the number of tillers (14.45 pieces) at 18 weeks after planting. A dose of 60 t/ha of chicken manure produced the best root length of 41.03 cm. The highest dry and wet weight yields, of about 179.75 g and 822 g, respectively, came from the rhizome weight of cow manure at a dose of 60 t/ha. The highest secondary metabolic levels in each parameter were found in dry rhizomes (phenolic 202.79 mg/L, flavonoid 181.91 mg/L, and tannin 5406.33 mg/L), and wet rhizomes (phenolic 178.56 mg/L, flavonoid 104.39 mg/L) with the highest tannin compound at around 4144.83 mg/L in chicken manure dose of 40 t/ha. According to the antioxidant results, providing chicken manure at a dose of 60 t/ha resulted in very strong antioxidant results in each of the wet and dry rhizomes, which were respectively 9.52 ppm and 8.06 ppm.

**Keywords:** *degraded soil, manure, soil chemical, ultisols*

**Introduction** → in this section, the existing problems of the soils used for growing *Zingiber* must be described

Ultisols is a very acid soil with soil pH ranges from 4.2 to 4.3, due to the relatively high rainfall (> 2,000 mm year<sup>-1</sup>), the soil bases such as Ca, Mg, K and Na cations are released and quickly leached from the soil. With

liming and fertilization being common solutions to overcome this problem (Purwanto, et.al, 2020) however, studies on Ultisol as zingiber growing media are still rare. Ultisols, in contrast, show younger characteristics, containing expandable 2:1 clay minerals. They contribute to higher CEC and the dominance of  $Al^{3+}$  for exchangeable sites, resulting in lower soil pH. Thus, it is well known that soil acidity can inhibit nitrification (Shibata et al. 2017). Ultisols commonly have low base saturation, acidic soil reactions, and elevated  $Al^{3+}$  saturations. However, a distinct Ultisol is found in East Kalimantan with extremely high aluminum saturation, could cause severe toxicity to the plant.  $Al^{3+}$  competition potentially enhances the desorption and leaching process of nutrient cations from the soil exchange complex, also hampering their absorption in the plant root area (Singh et al., 2017; Jaiswal et al., 2018; Zhao and Shen, 2018). Low pH exacerbates that effect by increasing the micronutrients/trace elements availability (e.g., Fe, Mn, Cu, and Zn), which is potentially toxic for plants. This condition, combined with other soil properties, may lead to nutrient deficiency, resulting in limited plant growth and development and a decline of plant productivity (Bojórquez-Quintal et al., 2017). Beside of nutrient deficiencies, the upland ultisols soil contains low organic matter, high soil bulk density (BD), low of total pores space, soil permeability and available water. Animal manure as an organic matter source plays a vital role in improving soil properties, the quality that is crucial in mitigating the adverse effect of chemical fertilizer and combating land degradation. Animal manure application demonstrated positive effects on Ultisols physical, chemical, and biological properties, particularly in alleviating soil acidity and Al toxicity (Zhou et al., 2013; Ngo et al., 2014; Ch'ng et al., 2015; Peng et al., 2016; Shi et al., 2019; Ye et al., 2019), a valuable source of key nutrients including nitrogen (N), phosphorus (P), and potassium (K) as well as certain micronutrients. Adding manure to soil impacts the chemical aspect of soil quality. Manure increases plant productivity, soil organic matter and structure, water infiltration and holding capacity, and over time, can reduce sediment loss and erosion with application and incorporation into soils, which also had advantageous effects on plant growth and development (Masmoudi et al., 2018; Pandey et al., 2021). *Zingiber montanum* (Syns: *Amomum cassumunar* (Roxb.) Donn, *Amomum montanum* J. König, *Amomum xanthorhiza* Roxb. ex Steud., *Cassumunar roxburghii* Colla, *Jaegera montana* (J. König) Giseke, *Zingiber anthorrhiza* Horan., *Zingiber cassumunar* Roxb., *Zingiber cassumunar* var. *palamaense* Haines, *Zingiber cassumunar* var. *subglabrum* Thwaites, *Zingiber cliffordiae* Andrews, *Zingiber luridum* Salisb., *Zingiber montanum* (J. König ex Retz.) Theilade, *Zingiber purpureum* Roscoe, *Zingiber purpureum* var. *palamaense* (Haines) K.K. Khanna, *Zingiber xanthorrhizon* Steud.) (WF0, 2021) is commonly known as “Banada” in Bangladesh, “Phlai” in Thailand, “Janliadrak” in India, and “Bangle” in Malaysia. It is reported to be native to Southeast Asia and has been extensively planted in Thailand, Malaysia, and Indonesia (Hassan et al. 2019). The rhizomes of this plant are used in traditional medicines for the treatment of constipation, dyspepsia, gastritis, stomach bloating and stomach-ache. Various parts of *Z. montanum* are used in Thailand as daily diet (Lim, 2016), while the rhizome is used in the as vermifuge in Malaysia, and applied for abscesses, colic, diarrhea, fever and intestinal disorders. In Northeast India, the rhizome paste was reported to be used in the treatment of dyspepsia and stomach bloating (Anasamy, 2013). Mineral nutrients applied basally or through foliar application, enhance the plant productivity and could be used as tools to ameliorate the quality of herbal medicines. As far as fertilizers are concerned judicious use of manure as a fertilizer is more important. Soil nutrients deficiencies can result in poor root systems, weak stalks or stems, slow growth (Moore et al. 2020) such as alfalfa and grasses. To reduce the problems of N, P, K deficiency for plant and poor of soil fertility acid upland soil, the application of manure are necessary (Navarette, 2015). The purposes of study were to examine the management of ultisols productivity through the use of manure and ultisols chemical properties, and zingiber yields.

## Materials and Methods

### Place and Time

The experiment was conducted at the Experimental Farm of Mulawarman University Teluk Dalam, East Kalimantan in from September 2021 to March 2022, lasting about 5 months. The second place is in the Soil Laboratory and Laboratory of Post-Harvest and Packaging of Agricultural Products, Faculty of Agriculture, Mulawarman University.

### Research Design

The experiment was arranged in a randomized completely block design with 3 replications with one treatment factor using organic fertilizer in the form of cow and chicken manure. The levels were then divided into 7 groups, with P<sub>0</sub>, P<sub>1</sub>, P<sub>2</sub>, P<sub>3</sub>, P<sub>4</sub>, P<sub>5</sub>, and P<sub>6</sub> each receiving a different dose of each type of manure. Each treatment was repeated four times. The following dosages were used: P<sub>0</sub> : Control (without fertilizer), P<sub>1</sub> : 20t cow manure /ha, P<sub>2</sub> : 40 t cow manure /ha, P<sub>3</sub> : 60 t cow manure /ha, P<sub>4</sub> : 20 t chicken manure /ha, P<sub>5</sub> : 40 t chicken manure /ha, P<sub>6</sub> : 60 t chicken manure /ha. The manure was applied by evenly broadcast according to the treatment, and then mixed with soil in 15-20 cm depth using hand hoes. The plot had a length of 6 m and a width of 1 m. Planting one crop per planting hole results in a spacing of approximately 50 x 100 cm. To prevent waterlogging and seedling rot,

seedlings are planted in ditches with good drainage. In order to facilitate landfiling later, planting is done in the trench. Non-factorial Completely Randomized Design (CRD) is as follows:

$$Y_{ij} = \mu + \tau_i + \epsilon_{ij}$$

Where:

$Y_{ij}$  = The observed value in the i-th treatment and j-th repetition,

$\mu$  = General mean value,

$\tau_i$  = Effect of i-th treatment,

$\epsilon_{ij}$  = Effect of error (error) in the i-th treatment experiment and j-th repetition.

The parameters measured in this study were soil chemical properties, cow manure and chicken manure including soil and manure pH using the electrometric method, organic C (%) using the Walkley and Black method, N-total (%) using the Kjeldahl method, C/N using the C-organic and N divisor method. -total, P-available (ppm) by the Bray II method, Cation Exchange Capacity or CEC (me/100) using the leaching method with 1N ammonium acetate pH 7. Plant height, number of leaves, and number of tillers were measured at 3, 6, 9, 12, 15, and 18 WAP (weeks after planting). After the plants were harvested, fresh weight, dry weight, root length, secondary metabolic, and antioxidant levels were measured.

### Laboratory measurement

Soil analyses were carried out in the Soil Science Laboratory, Faculty of Agriculture, Mulawarman University. The initial soil examination soil reaction was determined using soil to water mixture of 1:1. Organic carbon was extracted using the Walkley and Black method. Total N was extracted using the Kjeldahl method. Exchangeable aluminum was extracted using KCl 1 N. Cation base, and cation exchangeable capacity/CEC were extracted using an ammonium acetate (NH<sub>4</sub>OAc) solution pH 7.0. Clay to CEC ratio/CCR was calculated by dividing CEC by clay percentage. Since the observed soil pH was acidic, the ECEC was applied as the CEC reference for calculating the base and individual saturation.

### Data Analysis

Soil data's were analyzed descriptively while the crop data's were analyzed using the SAS Systems for Linier Models, v.6.12 for windows (Ramon et al. 1992). Data were analyzed by analysis of variance and followed by Duncan Multiple Range Test (DMRT) at 5% significance level. The financial effectiveness of each treatment was calculated by input-output farming and B/C ratio. The data obtained were subjected to the analysis of variance (ANOVA) with a 5% confidence level, followed by a 5% level DMRT (Duncan Multiple Range Test) to detect significant differences among treatments. The results of the secondary metabolic level identification and antioxidant activity tests were analyzed qualitatively using the descriptive method. Comparing phytochemical compound levels in each treatment was based on secondary metabolism levels. Antioxidant activity was assessed by comparing the IC<sub>50</sub> values in each treatment. A spectrophotometry method was used for all of the parameters of the secondary metabolic level test.

## Results

### Manure Quality

The results of manure analysis (Table 1) showed that the organic C content was relatively high (29.55%) for cow manure and 40.51% for chicken manure, the water contents was characterized at moderate level (72,9% and 60.63% respectively). N total are about 2,08% for cow manure and 1,37% for chicken manure, with C/N ratio 14,21 and 29,57 respectively. The manure contained some micro elements required by plant. Therefore, the quality of manure used in this study was high. Manure could improve soil chemical and physical properties effectively, and the plant growth was affected by the maturity of manure (Cai et al. 2019). Animal manure contains a lot of nitrogen as well as metallic minerals like magnesium, potassium, and calcium. The primary benefit of manure is that it preserves the physical structure of the soil, allowing roots to grow properly, as well as supporting the biological and chemical properties of the soil (Melsasail et al., 2019). Therefore, the purpose of fertilization is to replenish lost nutrients and increase the amount of nutrients available to plants, thereby increasing plant quality and quantity by gradually release nutrients into the soil solution and maintain nutrient balance for healthy growth of crop plants. They also act as an effective energy source of soil microbes which in turn improve soil structure and crop growth (Shaji et al. 2020).

Tabel 1. Chemical Characteristic of manure

Manure	chemical properties
--------	---------------------

Commented [12]: how did you analyze them? (what methods?)

	pH	C (%)	N (%)	C/N	P (ppm)	water content (%)
Cow	6,5	29,55	2,08	14,21	0,17	72,9
Chicken	6,4	40,51	1,37	29,57	0,67	60,63

As a function of soil chemistry, organic manure could provide some of soil CEC that was important to hold a given inorganic fertilizers and soil buffering capacity, so that the crops could avoid from the pressure of soil acidity (Shi et al, 2019). The use of organic materials increased the availability of some nutrients and improved the efficiency of P absorption by crops because in the process of organic matters decomposition humic acid and fulfat acid (polyelectrolite) were produced that had an ability to bind Al and Fe in the soil (Al-Juthery et al, 2021). To eliminate P fixation in the soil, the active anion of organic manure formed a chelate bond with Si-Al-OOCR (Alofan). The higher the carboxyl and fenolic compounds in organic matters the higher the ability of organic matters to realease AlHPO4 bonds so the P nutrient became more available for plant (Ch'ng et al 2014; Asap et al, 2018; Maru et al, 2020). By increasing of organic matter contents in the soil, the N total and N mineralization, soluble P, exchangeable K, N uptake by plants and soil water content could be increased (Kafeel et al, 2023).

#### Soil Chemical Properties

The effect of manure to the soil chemical properties shows on Table 2, that the initial soil pH (before planting) is 5,62 and increased until 4,21 after manure applications. A significant decrease in soil acidity was obtained due to the addition of chicken manure 60 t/ha.

Table 2. Soil Chemical Properties (pH) After Manure Treatment

Manure Treatments	chemical properties						
	pH						
week incubation	0	3	6	9	12	15	18
control	5,62	5,62	5,62	5,61	5,62	5,62	5,62
cow 20 t/ha	6,4	6,2	6	5,80	5,7	5,7	5,6
cow 40 t/ha	6,5	6,4	6,3	5,9	5,7	5,5	5,4
cow 60 t/ha	6,8	6,6	6,4	6,6	6,2	6	5,8
chicken 20 t/ha	5,83	5,6	5,4	5	4,83	4,7	4,51
chicken 40 t/ha	6,04	6	5,8	5,04	4,74	4,9	4,8
chicken 60 t/ha	6,08	5,43	5,08	4,76	4,77	4,08	4,21

The addition of manure has an effect on reducing soil organic C at all levels of addition of manure. A significant decrease in organic C occurred with the addition of chicken manure 20 t/ha as shown in Table 3. In soil ecosystems, C-organic is an important component that influences soil properties to support plant growth, namely as a source of energy for soil organisms and a trigger for nutrient availability for plants (Qiu et al 2018).

Table 3. Soil Chemical Properties (Organic C) After Manure Treatment

Manure Treatments	chemical properties						
	Organic C (%)						
week incubation	0	3	6	9	12	15	18
control	0,18	0,18	0,18	0,18	0,18	0,18	0,17
cow 20 t/ha	0,83	0,8	0,78	0,64	0,56	0,46	0,4
cow 40 t/ha	1,31	1,27	1,2	1,19	1,08	1,07	1,01
cow 60 t/ha	1,4	1,37	1,35	1,32	1,31	1,31	1,3
chicken 20 t/ha	0,36	0,33	0,32	0,3	0,27	0,21	0,18
chicken 40 t/ha	0,42	0,42	0,41	0,38	0,35	0,31	0,26
chicken 60 t/ha	0,98	0,98	0,95	0,92	0,87	0,8	0,78

Application of all levels of manure resulted in an increase in the N content in Ultisol soil as shown in Table 4. The best increase in N content in Ultisol was obtained due to the addition of chicken manure 60 t/ha.

Table 4. Soil Chemical Properties (Organic N) After Manure Treatment

Manure Treatments	chemical properties						
	N (%)						
week incubation	0	3	6	9	12	15	18
control	0,14	0,14	0,14	0,14	0,14	0,15	0,14
cow 20 t/ha	0,08	0,1	0,15	0,16	0,18	0,19	0,19
cow 40 t/ha	0,11	0,11	0,12	0,17	0,19	0,19	0,21
cow 60 t/ha	0,13	0,15	0,16	0,16	0,17	0,19	0,23
chicken 20 t/ha	0,18	0,18	0,19	0,19	0,2	0,21	0,21
chicken 40 t/ha	0,2	0,21	0,22	0,3	0,32	0,33	0,35
chicken 60 t/ha	0,2	0,22	0,32	0,35	0,36	0,37	0,38

The C/N ratio of organic matter is the ratio between the amount of elemental carbon (C) to the amount of elemental nitrogen (N) presented in an organic matter. Good manure must have a C/N ratio <20 (Macias-Corral et al 2019). From this study it can be seen that the C/N of soil organics ranged from 2,00 to 11,91 and decreased to 2,05 after 18 weeks of incubation (Table 5)

Table 5. Soil Chemical Properties (C/N) After Manure Treatment

Manure Treatments	chemical properties						
	C/N						
week incubation	0	3	6	9	12	15	18
control	1,29	1,29	1,29	1,29	1,29	1,20	1,21
cow 20 t/ha	10,38	8,00	5,20	4,00	3,11	2,42	2,11
cow 40 t/ha	11,91	11,55	10,00	7,00	5,68	5,63	4,81
cow 60 t/ha	10,77	9,13	8,44	8,25	7,71	6,89	5,65
chicken 20 t/ha	2,00	1,83	1,68	1,58	1,35	1,00	0,86
chicken 40 t/ha	2,10	2,00	1,86	1,27	1,09	0,94	0,74
chicken 60 t/ha	4,90	4,45	2,97	2,63	2,42	2,16	2,05

Manure input at all application levels has an effect in the form of increasing the P content in the soil along with the increase in incubation time. The highest increase in P in the soil was obtained due to the addition of chicken manure 60 t/ha as shown in Table 6.

Table 6. Soil Chemical Properties (available P) After Manure Treatment

Manure Treatments	chemical properties						
	P (ppm)						
week incubation	0	3	6	9	12	15	18
control	10,52	10,52	10,52	10,52	10,51	10,52	10,52
cow 20 t/ha	0,35	0,36	0,36	0,37	0,37	0,38	0,38
cow 40 t/ha	0,4	0,4	0,42	0,43	0,44	0,45	0,45
cow 60 t/ha	0,44	0,44	0,45	0,47	0,48	0,48	0,49
chicken 20 t/ha	12,78	12,78	12,8	12,81	12,9	12,92	12,93
chicken 40 t/ha	13,42	13,44	13,47	13,48	13,49	13,49	13,5
chicken 60 t/ha	15,68	15,68	15,68	15,69	15,7	15,72	15,73

The results of this study indicate that the addition of all levels of manure applications provides an increase in cation exchange capacity. The highest increase in cation exchange capacity was obtained due to the addition of cow manure 60 t/ha as shown on Table 7. Cation exchange in the soil occurs due to the presence of a negative



charge from the soil colloid which adsorbs cations in an exchangeable form (Meetei et al. 2020). Soils with high CEC are able to absorb and provide nutrients better than soils with low CEC. Because these elements are in the soil adsorption complex, these nutrients are not easily lost or washed away by water. Mineralization of soil organic fractions provides limited supplies of N, S, and micronutrients, while mineral dissolution and surface exchange reactions resupply P, K, Ca, Mg, and micronutrients. Nutrient mobility in soil influences ion transport to plant roots, evaluation of nutrient availability to plants, and ultimately nutrient management decisions (Havlin, 2020).

Table 7. Soil Chemical Properties (CEC) After Manure Treatment

Manure Treatments	chemical properties						
	CEC (me/100g)						
week incubation	0	3	6	9	12	15	18
control	14,33	14,33	14,32	14,33	14,34	14,33	14,33
cow 20 t/ha	19,15	19,15	19,17	19,17	19,18	19,18	19,19
cow 40 t/ha	19,35	19,36	19,36	19,37	19,37	19,38	19,38
cow 60 t/ha	20,45	20,45	20,46	20,5	20,51	20,55	20,56
chicken 20 t/ha	18,32	18,32	18,33	18,34	18,37	18,38	18,4
chicken 40 t/ha	18,38	18,38	18,4	18,41	18,43	18,44	18,45
chicken 60 t/ha	19	19, 12	19, 14	19, 15	19, 18	19, 20	19, 22

The nutrient added to the soil with low CEC could not be held and easily lost. This condition was reflected to the increasing of soil organic C contents in the treatment with manure application and it did not increase significantly in the treatment without manure application, vice versa. The results of an analysis of variance (ANOVA) at a 5% level showed that the treatments of giving organic fertilizers in the form of chicken and cow manure gave significant differences in plant height, number of tillers, number of leaves, root length, fresh weight of rhizomes, and dry weight of rhizomes. Only root length that was not affected significantly by the treatments. For growing Zingiber, N, P, and K play as an essential component.

#### Plant growth

Table 8 shows that when the plant was about 6 weeks old, the application of cow manure at a dose of 60 t/ha produced the best plant growth (26.16 cm), followed by chicken manure at a dose of 60 t/ha, chicken manure of 40 ts/ha, and cow manure of 40 t/ha. In comparison to the previous week, the plant's age 9 weeks after planting showed a very rapid increase in plant height. At 9 weeks after planting, the best dose was 40 t/ha of chicken manure with an average plant height addition of about 32.45 cm. The best results were obtained at week 12, with a cow manure at a dose of 60 ts/ha that yielded an average plant height of 37.24 cm. The application of cow manure at a dose of 20 t/ha and chicken manure at a dose of 20 t/ha did not produce significantly different plant height from the control. The best dose at 15 weeks after planting was 60 t cow manure /ha, with an average plant height of 40.47 cm, and 38.38 cm of chicken manure. The highest dose of cow manure yielded an average plant height of 42.78 cm at 18 weeks after planting.

#### Number of Tillers

Table 9 shows that the highest number of tillers was found at a dose of 60 t/ha of chicken manure, with an average of 1.65 tillers at 6 weeks after planting. At the age of 9 weeks after planting, a control with an average of about 2.55 tillers produced number of tillers that were not significantly different from chicken manure 20 t/ha. The best application was 60 t/ha of cow manure with an average of 4.15 tillers. At 12 weeks after planting, the effect of manure treatment revealed that the best dose was 40 t/ha chicken manure, with an average of 6.65 tillers. This best dose did not differ significantly between chickens and cows manure at 60 t/ha or chickens manure at 20 t/ha. Cow and chicken manure at a dose of 60 t/ha at 15 weeks after planting produced the highest yields, with cow manure yielded 9.50 tillers and chicken yielded 9.00 tillers, respectively. At 18 weeks, the control did not differ significantly from the cow manure doses of 20 and 40 t/ha. The highest number of tillers was observed for the treatment of cow manure at 60 t/ha with an average of 14.45 tillers and chicken manure at 60 t/ha with an average of 13.37 tillers.

#### Number of Leaves

Table 10 shows that after 6 weeks of treatment with a dose of 20 t chicken manure /ha , the number of leaves were not statistically significantly different from the control but significantly different from other treatments. The best dose of 60 t/ha cow manure, was produced at around 9 weeks, with an average increase in the number of leaves of 37.95 leaves. At 12 weeks, the control showed no significant difference from cow manure of 20 and 40 t/ha, as well as chicken manure of 20 and 40 t/ha. Cow manure of 60 t/ha with an average of 52.80 leaves provided the best dose at 12 weeks after planting . The highest dose was found in cow manure at a rate of 60 t/ha in week 15, with an average of 84.55 leaves. The application of 60 t/ha cow manure produced the largest number of leaves of 116.65 leaves in the 18th week, that significantly difference from the control. However, the application of 60 t/ha chicken manure increased the number of leaves by 105.80 leaves.

#### Rhizome root length, wet weight and dry weight

Table 11 shows that the lowest root length of 24.36 cm was obtained by the application of 20 t/ha cow manure. The control yielded root length that was not statistically different from that yielded by the application of 20 t/ha cow and chicken manure as well as the highest dose. This result indicates that applying different doses of manure had no effect on the root length of the bangle plant. The chicken manure dosage of 20 t/ha yielded the lowest bangle rhizome fresh weight of 392.35 g. While the control yielded fresh weight that was not significantly different from 20 t/ha and 40 t/ha cow and chicken manure. The fresh weight of the bangle rhizome due to the application of 40 t/ha chicken and cow manure also did not differ significantly from those from 60 t/ha chicken manure. The treatment of 60 t/ha cow manure yielded the highest rhizome fresh weight of 822.00 g that was significantly for all treatments. The lowest dry weight of bangle rhizome was shown by the cow and chicken manure treatment at a dose of 20 t/ha, respectively 86.00 g for cow manure treatment and 78.25 g for chicken manure treatment. These values were lower than that of the control, with an average of 93.75 g. The control yielded statistically insignificantly different rhizome dry weight with chicken manure at 20 and 40 t/ha, cow manure at 20 and 40 ts/ha, and chicken manure at 60 t/ha, but yielded significantly different rhizome dry weight r with the best dose of 60 t/ha cow manure with an average rhizome dry weight of 179.75 g, followed by 60 t chicken manure /ha with an average rhizome dry weight of 135.75 g

**Table 8.** Effect of Cow and Chicken Manure on Plant Height (in cm)

Manure Treatment (t/ha)	Week after planting (WAP)				
	6	9	12	15	18
Control	11,38 a	18,61 a	24,68 a	20,41 a	18,82 a
Cow 20 t/ha	17,46 b	20,26 ab	25,44 a	24,02 ab	28,51 b
Cow 40 t/ha	20,51 c	27,95 b	29,81 ab	35,57 cd	37,19 c
Cow 60 t/ha	26,16 e	31,18 b	37,24 c	40,47 d	42,78 c
Chicken 20 t/ha	17,36 b	27,21 b	30,16 abc	29,15 bc	28,82 b
Chicken 40 t/ha	22,84 cd	32,45 b	34,83 bc	37,61 cd	36,37 c
Chicken 60 t/ha	25,68 de	32,17 b	36,84 bc	38,38 d	39,22 c

**Commented [13]:** What is the different between Rhizome height and plant height?

**Commented [14R13]:** We mean Table 8 is Plant height not rhizome height

**Table 9.** Effect of Cow and Chicken Manure on Rhizome number of tillers (in weeks)

Manure Treatment (t/ha)	Week after planting (WAP)				
	6	9	12	15	18
Control	0,8 a	2,55 a	3,95 a	5,80 a	8.60 a
Cow 20 t/ha	0,7 b	3,45 b	5,20 b	6,60 a	10.4 ab
Cow 40 t/ha	1,7 ac	3,4 b	5,05 ab	7,35 ab	11.55b
Cow 60 t/ha	1,6 c	4,15 b	6,10 bc	9,50 c	14.45d
Chicken 20 t/ha	1,3 bc	3,35 ab	5,70 bc	7,40 ab	11.05b
Chicken 40 t/ha	1,5 c	4,00 b	6,65 c	8,45 bc	12.15bc
Chicken 60 t/ha	1,65c	3,75 b	6,05 bc	9,00 c	13.7cd

**Table 10.** Effect of Cow and Chicken Manure on number of Rhizome Leaves (in weeks)

Manure Treatment (t/ha)	Week after planting (WAP)				
	6	9	12	15	18
Control	4,70 a	19,60 a	35,80 a	52,25 a	77,55 a
Cow 20 t/ha	5,55 a	25,00 ab	41,05 ab	55,65 a	83,80 a
Cow 40 t/ha	9,65 b	28,25 abc	42,70 abc	66,00 ab	95,35 ab
Cow 60 t/ha	10,05b	37,95 c	52,80 c	84,55 c	116,65 c
Chicken 20 t/ha	9,20 b	26,00 ab	40,25 ab	56,85 a	81,75 a
Chicken 40 t/ha	9,65 b	30,90 bc	43,25 abc	67,05 bc	91,35 a
Chicken 60 t/ha	11,05b	30,45 abc	50,75 bc	79,55 bc	105,80 bc

**Table 11.** Effect of Cow and Chicken Manure on Root Length and Rhizome Weight (in 18 week)

Manure Treatment (t/ha)	Root lenght (cm)	Rhizome wet weight (g)	Rhizome dry weight (g)
Control	31,90 ab	415,25 ab	93,75 ab
Cow 20 t/ha	24,36 a	444,75 ab	86,00 a
Cow 40 t/ha	35,36 b	530,00 bc	119,00 ab
Cow 60 t/ha	36,50 b	822,00 d	179,75 c
Chicken 20 t/ha	33,20 ab	393,25 a	78,25 a
Chicken 40 t/ha	38,95 b	487,50 abc	99,75 ab
Chicken 60 t/ha	41,03 b	618,75 c	134,75 bc

**Notes:** Based on Duncan Multiple Range Test (DMRT) at a 5% significance level, values in the same columns that are followed by the same letter do not differ significantly.

**Table 12.** Secondary Metabolic Levels in Bangle Rhizome in Each Treatment (mg/L).

Condition	Manure Treatment (t/ha)	Level mg/L		
		Phenolic	Flavonoid	Tannin
Fresh	Control	31.64	23.08	2034.83
	Cow 20 t/ha	43.18	41.55	2991.50
	Cow 40 t/ha	77.03	64.30	3151.50
	Cow 60 t/ha	86.90	71.14	3198.17
	Chicken 20 t/ha	97.67	74.79	4086.50
	Chicken 40 t/ha	107.54	78.71	4144.83
	Chicken 60 t/ha	178.56	104.39	3861.33
Dry	Control	124.97	38.44	1932.33
	Cow 20 t/ha	133.56	67.23	2504.00
	Cow 40 t/ha	148.44	68.76	3034.83
	Cow 60 t/ha	181.38	99.43	4734.67
	Chicken 20 t/ha	175.23	120.06	4903.00

Chicken 40 t/ha	198.69	156.10	5088.00
Chicken 60 t/ha	202.79	181.91	5406.33

**Note:** The number followed by yellow denotes the best outcome in each observation variable for the flavonoid, tannin, and phenolic contents.

**Tabel 13.** Antioxidant based on IC50 value

Antioxidant (IC50) (mg/L)	Manure Treatment (t/ha)						
	Control	Cow			Chicken		
		20	40	60	20	40	60
Fresh rhizome	53.58	48.55	47.36	47.14	32.73	27.42	9.52
Dry rhizome	40.91	38.30	34.78	34.46	24.41	14.05	8.06

**Note:** The smaller the IC50 value, the stronger the antioxidant

#### Secondary metabolic level

In accordance with the findings of the secondary metabolism test, the rhizome of the bangle plant contained phenolic, flavonoid, and tannin-containing active substances, and also steroids, alkaloids, and terpenoids qualitatively. Different levels of the compounds involved in this secondary metabolism are evident in each organic fertilizer application. Different concentrations were produced by the concentration of active substances in the bangle rhizome. The data mentioned above demonstrated that, despite using the same amount of fertilizer, the active compound content of the bangle rhizome was highest when the rhizome was dry (low moisture content) as opposed to when the rhizome was still fresh. The amount of chicken manure that produced the highest phenolic content in the rhizome under fresh and dry conditions, correspondingly around 178.56 mg/L and 202.79 mg/L, was 60 t/ha. A dose of 60 t/ha of chicken manure also yielded higher levels of active flavonoids and tannins than other doses. When compared to the fertilizer treatment, the control had the lowest active compound content. When treated with cow manure, yields were lower than when treated with chicken manure at the same dose.

#### Antioxidant

Results of antioxidant analysis of bangle rhizomes using several samples based on manure dose and fresh and dry rhizome condition are shown Table 6. IC<sub>50</sub> value obtained described how well the sample captured free radicals. The dose of 60 t chicken manure s/ha was found to have the lowest IC<sub>50</sub> value of 9.52 ppm in the fresh sample, , while the control had the highest IC<sub>50</sub> value of 53.58 ppm in fresh rhizome conditions. The same quality was obtained when dry rhizome conditions were used for the analysis; specifically, the dose of 60 t chicken manure /ha chicken manure produced the lowest IC<sub>50</sub> value and the largest was in the control. The results of an analysis of variance (ANOVA) at a 5% level showed that the treatments of giving organic fertilizers in the form of chicken and cow manure gave significant differences in plant height, number of tillers, number of leaves, root length, fresh weight of rhizomes, and dry weight of rhizomes. Only root length that was not affected significantly by the treatments.

#### Discussion

##### Effect of manure addition on soil chemical properties

The changes of several soil chemical parameters during the incubation of three types of animal manures are presented in Table 2 until Table 6. According to the analysis of variance, almost all of the observed soil chemical characteristics were strongly affected by the differences in incubation time and manure rate. There were significant differences between the incubation time on pH H<sub>2</sub>O, pH KCl, total N, exchangeable acid and base cations, CEC, BS (P<0.001), and organic C (P<0.01). Some parameters (e.g., pH H<sub>2</sub>O, pH KCl, organic C, exchangeable bases, and base saturation) showed statistically high values and concentrations at the first two weeks, which decreased at the next four and six weeks. The increment in incubation time seemingly exhibited an inconsistent effect on the soil. [Adekya et al. 2020](#) were reported that utilization of organic manure to meet crop nutrient requirement will be an unavoidable practice to enhance sustainable agriculture, this is because, the physical, chemical and biological properties of soil is generally improved by the addition of organic manures which in turn enhances crop productivity and maintains the quality of crop produce. Poultry litter treatments were positively correlated with greater soil fertility levels, as well as higher crop yield and soil biodiversity. These results underscore linkages between manure additions and cropping sequences, within the nutrient cycling, soil health, and crop production continuum ([Asworth et al. 2018](#))

### Plant Height

According to the Table 8, the average increase in plant height starting at 6,9,12,15, and 18 WAP tended to fluctuate. At the age of 6 weeks, the treatment without fertilizer (control) continued to grow until week 12 when it reached its peak with an average height increase of about 24.68 cm, before the delining height at the following weeks.. In order for a plant to grow, nutrients must be obtained from the soil by the roots through their root hairs (Wang et al., 2021). Organic matter affects plant growth by influencing the physical, chemical, and biological properties of the soil (Delgado and Gomez, 2017). The more organic matter is provided, the faster the plant will grow. Compared to chicken manure, cow manure typically produces better plant growth. [The application of chicken manure at all levels significantly increased plant height at 9 weeks after planting, but at 12 weeks, the plant height slightly decreased. In the 20 t/ha chicken manure treatment, the plant height was in average of 30.16 cm at 12 weeks after planting and decreased in the following weeks. High nitrogen elements are found in chicken manure (0.38%). Although the amount of nitrogen required by plants is always higher than other nutrients, a deficiency or excess of nitrogen can hinder and disrupt plant growth (Jiang et al 2021). After planting, the growth rate of the bangle plant accelerated between 2 and 5 months. As plants get older, their growth rate for height starts to slow down (Rademacher, 2015).

Applying chicken manure always results in the best plant response in the first growing season. This is because chicken manure decomposes relatively quickly and has enough nutrients compared to other manures of the same weight. Table 1 shows that the application of chicken manure tended to increase plant height rapidly at 6, 9, and 12 weeks of age, then declined as plant age increased. Large-scale application of chicken manure is thought to be less effective because the nutrients will exhaust quickly. The same result was also shown by the application of chicken manure at a dose of 40 t/ha which decreased the plant height, the maximum height increase at this dose was at 15 weeks with an average height increase of 37.61 cm. When compared to the other two doses of chicken manure, the plant height for the 60 t/ha chicken manure was different. The plant height increase over 18 weeks demonstrates this, but the increase in plant height was typically not too different from the previous weeks. Even though the application of cow manure at a dose of 40 t/ha initially produced less yields than that produced by the application of chicken manure at a dose of 40 t/ha, at the end of the observation at 18 weeks the increase in height was more apparent and might even have exceeded that of the chicken manure at a dose of 40 t/ha, which caused a decrease in plant height.

The addition of cow manure improves soil permeability, total pore space, aggregate stability, bulk density, texture, color, and temperature (Singh, 2021). A dose of 60 t/ha of chicken and cow manure had the tendency to produce steady, dependable results. The application of cow manure at a dose of 60 t/ha tended to yield less than chicken manure at doses of 40 and 60 t/ha from the start of planting until the plant was 9 weeks old, but the yields increased in the following weeks. The plant's need for nutrients grows as it ages. If the nutrient requirements are not met and the nutrients are not readily available, plants may get nutrient deficiency at specific times (Bindraban et al., 2015). Bangle plants absorb N (0.06 - 3.07 g), P (0.01 - 0.53 g), and K (0.10 to 2.25 g) at 2 to 10 months after planting in the canopy. N is the nutrient that is most required in the plant canopy itself. The primary nutrient for plants, nitrogen, is typically essential for the development and expansion of vegetative parts of plants, such as leaves, stems, and roots (Tegeader and Masclaux-Daubresse, 2017). A sufficient supply of plant N is indicated by high photosynthetic activity, good vegetative growth, and dark green plant colors (Wei et al., 2015).

Due to the individual characteristics of each animal, which are influenced by the type of feed and the animal's age, each manure contains a different mix of nutrients (Asworth et al., 2020). Because each treatment dose of fertilizer has a different nutrient content, they all produce different yields and have different recommended doses. Due to their movement with crop yields, surface runoff, erosion, or evaporation, nutrients in the soil will gradually decrease over time (Liao et al., 2021).

### Number of Tillers

The table of the total number of tillers from each level reveals results that increase with plant aging and are influenced by the quantity of fertilizer applied, as shown in Table 9. Plants without fertilizer developed more tillers every week, but the growth was typically modest. This slight increase resulted from the fact that during the initial stages of planting, the products of photosynthesis were utilized for the vegetative development of plants. In comparison to manure application, treatment without fertilizer produced the lowest yield. Data presented in Table 9 show that there was a noticeable increase in the number of tillers at 18 WAP of age. A plant needs nutrients for its physiological processes during growth and development. Plant growth and production will be subpar due to a lack of nutrients (Reich et al., 2014). Functions of organic matter as a biological buffer so that the soil can supply plants with a balanced amount of nutrients (Nair, 2019). Loosening the topsoil, increasing water absorption and storage, and boosting soil fertility are all important functions of manure (Murphy, 2015). A sudden rise in the number of tillers can result from the ease with which new shoots can emerge from loose, moist soil.

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At the start of planting, there were typically fewer and nearly identical numbers of tillers in each treatment. The nutrients in this fertilizer are not readily available to plants, which is the cause of the slow plant growth at the start of the planting period. The extent of these materials' mineralization or decomposition has a significant impact on the nutrients' availability. Manure's low nutrient availability is partially caused by the presence of N, P, and other elements in complex compounds that are challenging to decompose (Cui et al., 2021). At 6 weeks after planting, all treatments tended to be similar and the differences between the tillers in each treatment tended to be minimal. Although cow manure at a dose of 60 t/ha was the best dose with an average number of tillers of 14.45 tillers, chicken manure typically produced better results than cow manure at the same dose. In comparison to other manure doses, chicken manure 40 t/ha at 12 weeks after planting produced the best results with 6.65 tillers.

#### Number of Leaves

An increasing number of leaves are produced each week as a result of the weekly application of cow and chicken manure. Table 10 shows that at about 12 weeks of age, the number of leaves increased significantly. Every week, the increase varied depending on the treatment. The number of leaves significantly increased with a dose of 60 t/ha of cow manure, averaging 15–37 leaves every three weeks. Similarly, the number of leaves tended to increase when manure was applied in the same amount. Both cow and chicken manure at a dose of 20 t/ha and 40 t/ha produced a nearly identical number of leaves during plant growth. In addition to the nutrients that plant's needs, manure also contains humic, fulvic acids, growth hormones, and other substances that promote plant growth and increase nutrient uptake by plants (Canellas et al., 2015). The amount of photosynthesis is influenced by the number of leaves present, and plants with more leaves may produce heavier and bigger rhizomes as a result. The number of leaves added is also affected by the number of shoots and plant height. The number of leaves will increase as the plant ages and grows taller, produces more leaves on a single stem, and produces more tillers. The nutrients required for plant growth are present in sufficient amounts in manure. The related observation variables will be impacted by the food's growth quality and planting age.

In relation to the addition of the number of leaves, the most influential element is N. In comparison to other nutrients, nitrogen is required in sufficient amounts for plant growth. N makes up 40–50% of the dry weight of protoplasm, the living component of plants (Kathalia & Bhatla, 2018). Since protein is the source of all plant enzymes, nitrogen participates in all enzymatic processes in plants. Additionally, nitrogen is one of the constituent elements of chlorophyll, the primary component of chloroplasts, and it contributes to improving the quality and quantity of the dry matter produced (Wen et al., 2020). Fertilizer use and the amount of nutrients in the soil have a significant impact on how plants grow and develop. Nutrient uptake is restricted by nutrients in a minimum state (Purba et al., 2021). In terms of the addition of leaves, the treatment of plants without fertilizer differs significantly enough for each observation. In comparison to other treatments, plants without fertilizer produce the lowest yield.

#### Root Length

The lowest root length was observed for the cow manure dose of 20 t/ha treatment with an average root length of about 24.36 cm which was lower than the control. With an average root length of 41.03 cm, the application of chicken manure yielded the longest roots. When plants respond to water shortages by reducing the rate of transpiration to conserve water, the roots play a crucial role (Sourour et al., 2017). Plant roots have a significant impact on overall plant growth and development. The failure of the root function will result in a complete change in the plant for the top. Manure can bind water in the soil. Because the soil around the roots in the deeper layers is still moist, the roots will continue to grow. Maximizing exposure to groundwater will encourage the growth of root. Plant roots directly respond to the physical characteristics of the soil (Jiang et al., 2017).

Data presented in Table 11 show that the application of chicken manure yielded better root length than cow manure. The use of organic fertilizers can loosen the soil, increase aeration, and increase the soil's capacity to hold water, all of which can improve the physical properties of the soil (Shaji et al., 2021). Additionally, organic matter has the ability to control soil temperature, slow down phosphorus fixation, increase soil cation exchange capacity, and lessen the leaching of nutrients like potassium, calcium, and magnesium (Baghbani-Arani et al., 2021). Another environmental factor that has been shown to affect the nitrate absorption process is the temperature around the roots (Le Deunff, 2019). The initial analysis of the soil revealed that the pH ranged from 3.86 to 4.86. Al is common excessively in acidic soil, and it can poison plants and bind phosphorus (P). Low pH soil can hinder plant growth by preventing the roots from properly absorbing nutrients. Giving chicken manure, as demonstrated by Mažeika et al. (2021), can maintain stable nutrient content in soil and minimize mineral fertilizer influx into the environment. could raise the pH of soil. By raising pH, Al in the exchangeable form will be reduced, and nutrients will become more available to plants.

According to Rosita et al. (2005), nutrient uptake on the roots of bangle plants at 2-10 months after planting was as follows: N (0.01 - 0.52 g), P (0.002 - 0.15 g), and K (0.02 to 0.82 g). It was discovered that the roots of the bangle plant had more K buildup than N and P. K is primarily used to aid in the synthesis of proteins and carbohydrates. In the face of drought, illness, and pests, potassium gives plants strength (Hasanuzzaman et al.,

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2018). Organic fertilizers can help the soil's physical and chemical composition, which will facilitate root development. Up until the soil reaches its critical water potential, plant roots expand into moist soil and draw water (Sisouvanh et al., 2021). The looseness of the soil can promote root development. Strong roots will make it simpler for plants to absorb nutrients and water

#### **Rhizome fresh Weight**

Plant biomass is a common parameter used to study plant growth. When plant nutrient requirements are met, yields will be optimal (Timsina, 2018). The rhizome of the bangle plant is the part that is most advantageous for cultivation. One could also argue that this rhizome's fresh weight is a crucial factor in determining how well bangle plant cultivation is going. The cultivation method is better and more productive the more weight of the wet rhizome can be obtained. According to the results of the application of the organic fertilizers at the age of 18 weeks, cow manure at a dose of 60 t/ha produced the highest fresh weight of rhizomes, averaging 822 g/plant, and chicken manure at a dose of 20 t/ha produced the lowest fresh weight of rhizomes, averaging 393.25 g/plant. Manure increases crop yield and quality while also enhancing the chemistry, physical characteristics, and biological properties of the soil (Ma et al., 2021). Data presented in Table 11 demonstrate that the yield of fresh weight of rhizomes increased with increasing manure dosage. The physical condition of the soil must support plant growth in addition to a supply of adequate and balanced nutrients (El-Ramady et al., 2015). These soil aggregates will keep the soil in a loose condition (Murphy, 2015). Cow manure will enhance the physical characteristics of the soil. Improved soil physical characteristics include things like increased permeability, total pore space, aggregate stability, bulk density, texture, color, and temperature (Agbede, 2021).

Intensive tillage affects the physical properties of the soil. Low organic matter soils will have more severe damage to the soil's structure (Murphy, 2015). When the soil does not receive enough water and becomes dense and hard, soil damage is evident. Plant rhizomes will not be able to grow or spread out in compacted or hard soil. The ability to maintain loose soil conditions that are difficult to harden or compact increases with the amount of organic matter added. Additionally, manure helps to improve soil structure, cation exchange capacity, and water resistance. Giving manure has the indirect effect of making it simpler to keep water in the soil (Zhang et al., 2016). Since water availability plays a significant role in plant growth, water frequently restricts the growth and development of cultivated plants. The plants will experience drought conditions if there is not enough water in the soil. Due to decreased primary metabolism, reduced leaf area, and decreased photosynthetic activity, drought stress can lower plant productivity (biomass). Smaller leaves grow as a result of a lack of water during the vegetative stage, which can reduce light absorption. Lack of water also inhibits the synthesis of chlorophyll and some enzymes, such as nitrate reductase, from working (Altuntaş et al., 2020).

Organic substances in the soil may have physiological effects on plant growth that are direct or indirect (Basilio et al, 2013). Compared to other types of manure, chicken manure contains a fair amount of P. This is due to the fact that chicken manure contains feed (Agbede, 2021). Phosphorus aids in the growth of plant roots, photosynthesis, transfer respiration, cell division, and growth (Malhotra et al, 2018). The number of cells increases more quickly when they divide quickly, which causes the rhizome to grow larger.

#### **Rhizome Dry Weight**

Dry weight reflects a plant nutritional status because it is affected by the rate of photosynthesis and respiration in each treatment. Based on the collected data, it was determined that applying chicken and cow manure at a dose of 20 t/ha resulted in lower dry weight yields than the control. Additionally, the results from the application of 40 t/ha manure dose were not significantly different from the control. This is the active vegetative formation stage. The application of cow manure at a dose of 60 t/ha demonstrated different results and produced significantly better outcomes than other doses of chicken manure. The amount and timing of fertilizer applied can impact crop yields, among other things. Organic matter plays a crucial role in soil health because it can create stable soil aggregates, increase soil fertility, and serve as a source of energy for organisms (Magdoff, 2018).

The application of manure enhances the chemical, physical, and biological properties of the soil, increase crop yield, and improves crop quality (Du et al., 2020). High organic matter soils have beneficial microorganisms that encourage the breakdown of organic matter and release inorganic nutrients that are then available for plant uptake. Organic fertilizers can help to create ideal conditions in the soil for microorganisms that are beneficial to plants (Du et al 2022). Chicken manure is an organic fertilizer with high nitrogen content, despite not being the best dose for bangle rhizome weight yield. As they ensure the best nutrient management for plants, such fertilizers should be used promptly to partially replace chemical fertilizers (Guimarães et al., 2019).

#### **Secondary metabolic levels (phenolic compounds, flavonoids and tannins)**

The results of laboratory analysis found that the positive bangle rhizome contains compounds in the form of phenolics, tannins and flavonoids. These compounds respond differently to the concentration of organic fertilizer applied in the form of cow and chicken manures. According to Table 12, the amount of tannin compounds

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increased as manure dosage rose. The application of chicken manure or cow manure with three doses of 20, 40, and 60 t always increased the tannin concentration in the rhizome as the dose increased. When compared to cow manure, applying chicken manure produced significantly better results. It is evident that using 20 t/ha of chicken manure instead of 60 t/ha of cow manure resulted in a higher tannin concentration.

Dry rhizomes produced a higher concentration of tannins than fresh rhizomes, which produced different results regarding tannin concentration. The concentration of tannins in fresh rhizomes increased non-significantly with the addition of cow manure, whereas the concentration of tannin compounds in fresh rhizomes decreased with the increase in the dose of cow manure. In comparison to the use of manure that increased fresh and dry rhizomes, the control provided the lowest tannin concentration.

Tannins are chemical substances that have an astringent and bitter flavor. These substances act as controlling substances in plant metabolism as well as important defenses against herbivores and pests that prey on plants (Tiku, 2018). Tannins are metabolically active substances with multiple uses, including as astringents, antibacterial agents for treating diarrhea, and antioxidants. Leather tanning is another industrial application for tannins. Tannic acid is the type of tannin substance present in bangle plants. Diarrhea can be effectively treated with tannic acid. Additionally, tannic acid exhibits antimicrobial, antienzymatic, antioxidant, and antimutagenic properties (Dabbaghi et al., 2019).

In addition to increasing the fertilizer dose, applying organic fertilizer in the form of cow and chicken manure resulted in an increase in phenolic compounds and flavonoids. When compared to when the rhizome was fresh, the dry rhizome had a higher concentration of phenolic and flavonoid compounds. The content of active compounds in the *simplicia* is impacted by the drying process. Antioxidant activity is influenced by the total phenolic and flavonoid content (Rajkumari & Sanatombi, 2020).

The highest phenolic and flavonoid concentrations were found in both fresh and dry rhizomes when chicken manure was applied at a dose of 60 t/ha. The application of the highest dose of cow manure of 60 t/ha, resulted in phenolic compound concentrations of 86.90 mg/L in the fresh rhizome and 181.38 mg/L in the dry rhizome, while flavonoids were 71.14 mg/L in fresh rhizome and 99.43 mg/L in dry rhizome. Compared to chicken manure at the same dose, this result is smaller. A dose of 60 t/ha of chicken manure resulted in phenolic compound concentrations in the fresh rhizome of 178.56 mg/L and dry rhizome of 202.79 mg/L, while fresh rhizome flavonoids were 104.39 mg/L and dry rhizome flavonoids were 181.91 mg/L. In comparison to treatments with organic fertilizers, treatments without fertilizer produced lower phenolic and flavonoid concentrations.

In plants, flavonoids serve as pigments for the flowers, fruits, and roots, as well as occasionally as growth regulators and disease resistance (Kumar et al., 2018). Catechins are one class of flavonoid compounds present in bangle rhizomes (*catechins*). Catechins have antioxidant properties, and because they can stop the growth of viruses, bacteria, tumors, and fungi, they can also get rid of rotten and rancid odors (Isemura, 2019). Phenolic compounds are compounds that plants make in response to environmental stress. Phenolic compounds protect DNA from dimerization and damage by blocking UV-B rays and cell death (Takshak and Agrawa, 2019). Gallic acid is the type of phenolic compound found in bangle rhizome. Gallic acid serves as an antibacterial, antiviral, analgesic, and antioxidant in medicine (Shrinet et al., 2021).

The application of chicken manure resulted in a higher concentration of secondary metabolism because it had a relatively higher P nutrient content than other manures (Alinejad et al., 2020). Phosphorus can be found in DNA, RNA, and the parts of nucleotides that provide metabolic energy (like ATP). The process of photosynthesis depends heavily on phosphorus. Stunted growth is one of phosphorus deficiency's signs (De Bang, 2020). The concentration of secondary metabolism in the form of tannins, phenolic compounds, and flavonoids is influenced by the difference in the P nutrient content between these two types of manures. Environmental factors affect the levels of flavonoids and other phenolic compounds in plants, which vary among parts, tissues, and ages of plants. These include air temperature, nutrient availability, water availability, and atmospheric CO<sub>2</sub> concentrations (Ashraf et al., 2018).

#### **Antioxidants**

The antioxidant activity analysis produced different IC<sub>50</sub> values depending on the type of organic fertilizer used (Table 13). The IC<sub>50</sub> decreased as the fertilizer dose increased. A concentration known as IC<sub>50</sub> is capable of reducing 50% of DPPH free radicals. The greater the antioxidant activity, the lower the IC<sub>50</sub> value (Ali et al., 2018). Antioxidants are compounds that can absorb or neutralize free radicals, thereby preventing certain diseases caused by free radicals (Poprac et al 2017). The treatments of cow manure, as shown in Table 13 resulted in lower yields than chicken manure. The antioxidant activity of the dried rhizome samples was higher than that of the fresh rhizomes. The highest antioxidant activity was produced by cow manure at a dose of 60 t/ha, with an IC<sub>50</sub> value of 9.52 ppm for fresh rhizome and 8.6 ppm for dry rhizome. At a dose of 20 t/ha in both fresh and dry rhizome conditions, cow manure had a lower IC<sub>50</sub> value than chicken manure. For fresh rhizomes, the treatment without fertilizer produced an IC<sub>50</sub> value of around 53.58 ppm, and for dry rhizomes, it was around 40.91 ppm.

Ali et al. (2018) claimed that the antioxidant activity in bangle rhizomes is incredibly powerful. Except for the treatment without fertilizer in fresh rhizome conditions, the IC<sub>50</sub> value in bangle rhizome in all treatments gave



a value of 50 and included a very potent antioxidant. The high secondary metabolic compounds found in the bangle rhizome are inextricably linked to the high antioxidant activity. Secondary plant metabolites like flavonoids and phenolics play a part in antioxidant activity. More phenolic compounds will have a higher level of antioxidant activity (Tohma et al, 2017).

### Conclusion

A cow manure dose of 60 t/ha is the best dose for plant growth, with an average height increase of 42.78 cm, an increase in the number of leaves of 116.65 pieces, and an increase in the number of tillers of 14.45. A dose of 60 t/ha of chicken manure produced the best root length of 41.03 cm. The weight of the rhizomes revealed that the application of cow manure at a dose of 60 t/ha resulted in the highest yields of dry weight and wet weight, which were about 179.75 g and 822 g, respectively. A chicken manure dose of 60 t/ha results in the highest secondary metabolic rate in each parameter, including dry rhizome (phenolic 202.79mg/L, flavonoid 181.91mg/L, and tannin 5406.33mg/L) and wet rhizome (phenolic 178.56mg/L, flavonoid 104.39mg/L), while a chicken manure dose of 40 t/ha results in the highest tannin. Giving chicken manure at a dose of 60 t/ha produced very strong antioxidant results at 9.52 ppm wet rhizome and 8.06 ppm dry rhizome, according to the antioxidant results.

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**Table 81. Effect of Cow and Chicken Manure on Plant Rhizome Height (in cm)**

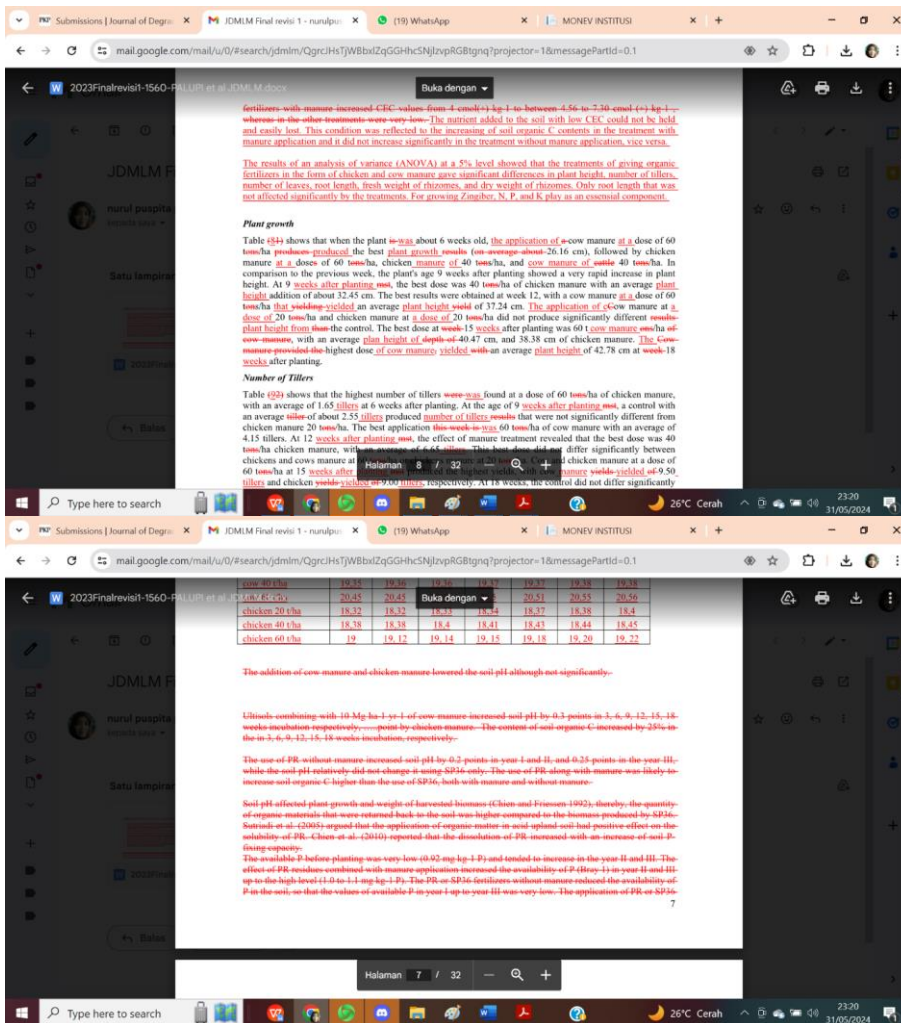
Manure Treatment (t/ha)	Week after planting (WAP)				
	6	9	12	15	18
Control	11.28 a	18.61 a	24.65 a	20.41 a	18.82 a
Cow 20 t/ha	17.46 b	20.26 ab	25.84 a	24.02 ab	28.51 b
Cow 40 t/ha	20.51 c	27.85 b	29.81 ab	35.57 cd	37.19 c
Cow 60 t/ha	26.16 c	31.18 b	37.24 c	40.47 d	42.78 c
Chicken 20 t/ha	17.36 b	27.21 b	30.16 abc	29.15 bc	28.82 b
Chicken 40 t/ha	22.84 cd	32.45 bc	34.83 bc	37.61 cd	36.37 c
Chicken 60 t/ha	25.68 de	32.17 b	36.94 bc	38.38 c	39.22 c

**Table 82. Effect of Cow and Chicken Manure on Plant Rhizome Length (in cm)**

Manure Treatment (t/ha)	Weeks after Planting (MST)				
	6	9	12	15	18
Control	11.28 a	18.61 a	24.65 a	20.41 a	18.82 a
Cow 20 t/ha	17.46 b	20.26 ab	25.84 a	24.02 ab	28.51 b
Cow 40 t/ha	20.51 c	27.85 b	29.81 ab	35.57 cd	37.19 c
Cow 60 t/ha	26.16 c	31.18 b	37.24 c	40.47 d	42.78 c

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**Table 6. Soil Chemical Properties (available P) After Manure Treatment**

Manure Treatments	chemical properties						
	P (ppm)						
week incubation	0	3	6	9	12	15	18
control	10.52	10.52	10.52	10.52	10.51	10.52	10.52
cow 20 t/ha	9.35	9.36	9.36	9.37	9.37	9.38	9.38
cow 40 t/ha	0.4	0.4	0.42	0.43	0.44	0.45	0.45
cow 60 t/ha	0.44	0.44	0.45	0.47	0.48	0.48	0.49
chicken 20 t/ha	12.78	12.78	12.8	12.81	12.9	12.95	12.93
chicken 40 t/ha	13.42	13.44	13.47	13.48	13.49	13.49	13.5
chicken 60 t/ha	15.68	15.68	15.68	15.69	15.7	15.72	15.73

The results of this study indicate that the addition of all levels of manure applications provides an increase in cation exchange capacity. The highest increase in cation exchange capacity was obtained due to the addition of cow manure 60 t/ha as shown on Table 7. Cation exchange in the soil occurs due to the presence of a negative charge from the soil colloid which adsorbs cations in an exchangeable form (Meead et al. 2020). Soils with high CEC are able to absorb and provide nutrients better than soils with low CEC. Because these elements are in the soil adsorption complex, these nutrients are not easily lost or washed away by water. Mineralization of soil organic fractions provides limited supplies of N, S, and micronutrients, while mineral dissolution and surface exchange reactions (especially P, K, Ca, Mg, and micronutrients). Nutrient mobility in soil influences ion transport to plant roots, evaluation of nutrient availability to plants, and ultimately nutrient management decisions (Hafid, 2020).

**Table 7. Soil Chemical Properties (CEC/TK) After Manure Treatment**

Manure Treatments	chemical properties						
	CEC/TK (meq/100g)						
week incubation	0	3	6	9	12	15	18
control	14.3	14.33	14.33	14.33	14.33	14.33	14.33
cow 20 t/ha	19.7	19.7	19.7	19.7	19.7	19.7	19.7

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**Table 4. Soil Chemical Properties (Organic N)**

Manure Treatments	chemical properties						
	N (%)						
week incubation	0	3	6	9	12	15	18
control	0.14	0.14	0.14	0.14	0.14	0.15	0.14
cow 20 t/ha	0.08	0.1	0.15	0.16	0.18	0.19	0.19
cow 40 t/ha	0.11	0.11	0.12	0.17	0.19	0.19	0.21
cow 60 t/ha	0.13	0.15	0.16	0.16	0.17	0.19	0.23
chicken 20 t/ha	0.18	0.18	0.19	0.19	0.2	0.21	0.21
chicken 40 t/ha	0.2	0.21	0.22	0.3	0.32	0.33	0.35
chicken 60 t/ha	0.2	0.22	0.32	0.35	0.36	0.37	0.38

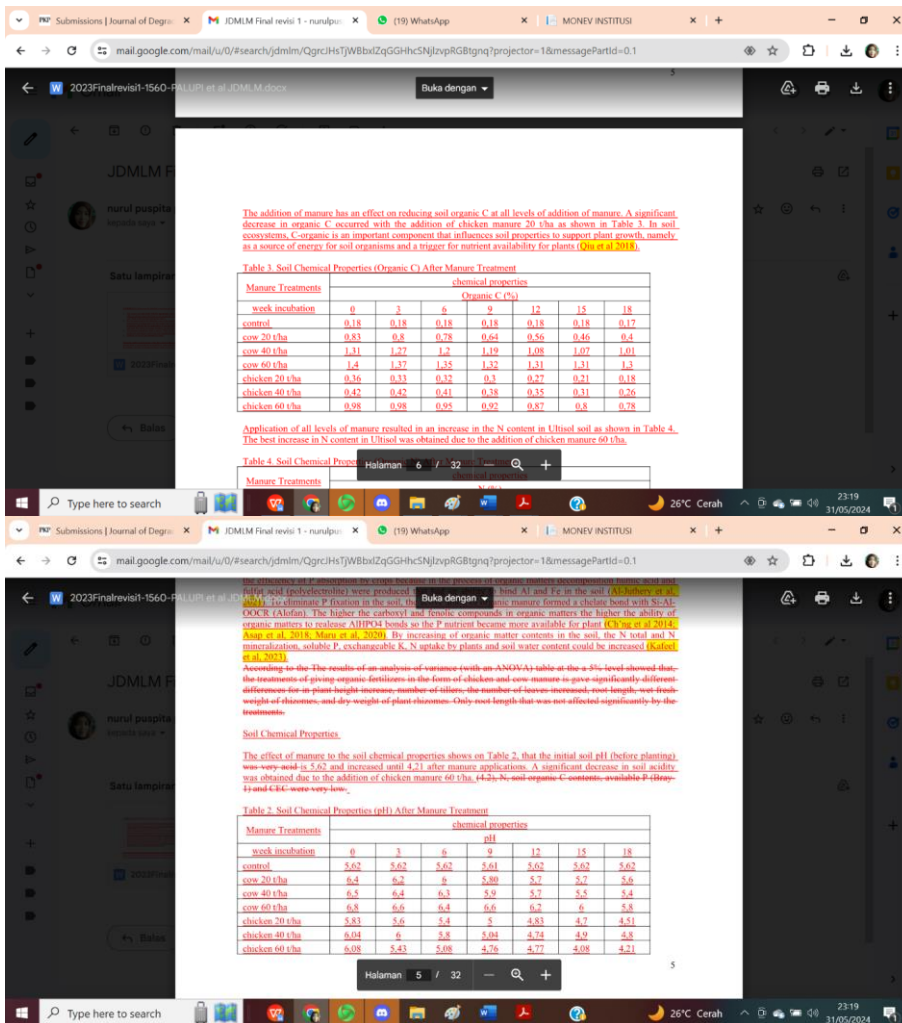
The CN ratio of organic matter is the ratio between the amount of elemental carbon (C) to the amount of elemental nitrogen (N) presented in an organic matter. Good manure must have a CN ratio <20 (Majumdar et al. 2010). From this study it can be seen that the CN of soil organics ranged from 2.00 to 11.91 and decreased to 2.05 after 18 weeks of incubation (Table 5).

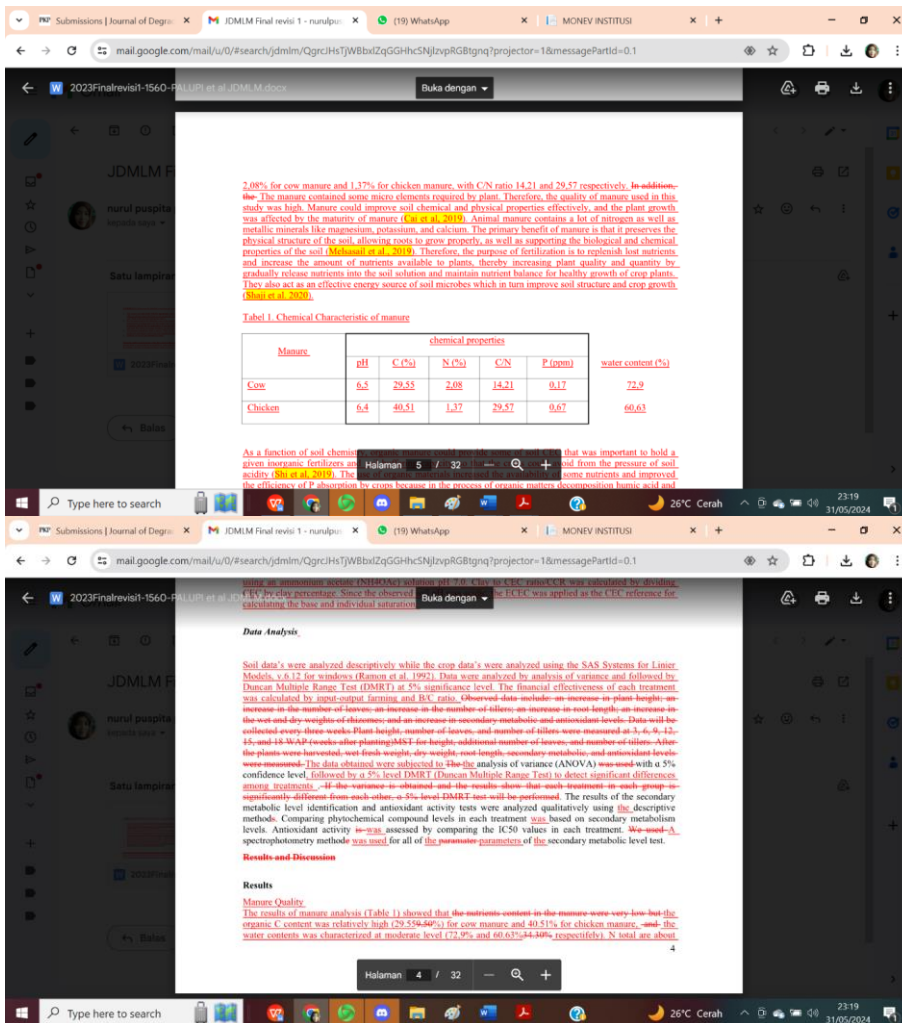
**Table 5. Soil Chemical Properties (CN) After Manure Treatment**

Manure Treatments	chemical properties						
	CN						
week incubation	0	3	6	9	12	15	18
control	1.29	1.29	1.29	1.29	1.29	1.20	1.21
cow 20 t/ha	10.38	8.00	5.20	4.00	3.11	2.42	2.11
cow 40 t/ha	11.91	11.55	10.00	7.00	5.68	5.63	5.81
cow 60 t/ha	10.77	9.13	8.44	8.25	7.71	6.89	5.65
chicken 20 t/ha	2.00	1.81	1.68	1.58	1.35	1.00	0.86
chicken 40 t/ha	2.10	2.00	1.86	1.27	1.09	0.94	0.74
chicken 60 t/ha	4.90	4.45	2.97	2.63	2.42	2.16	2.05

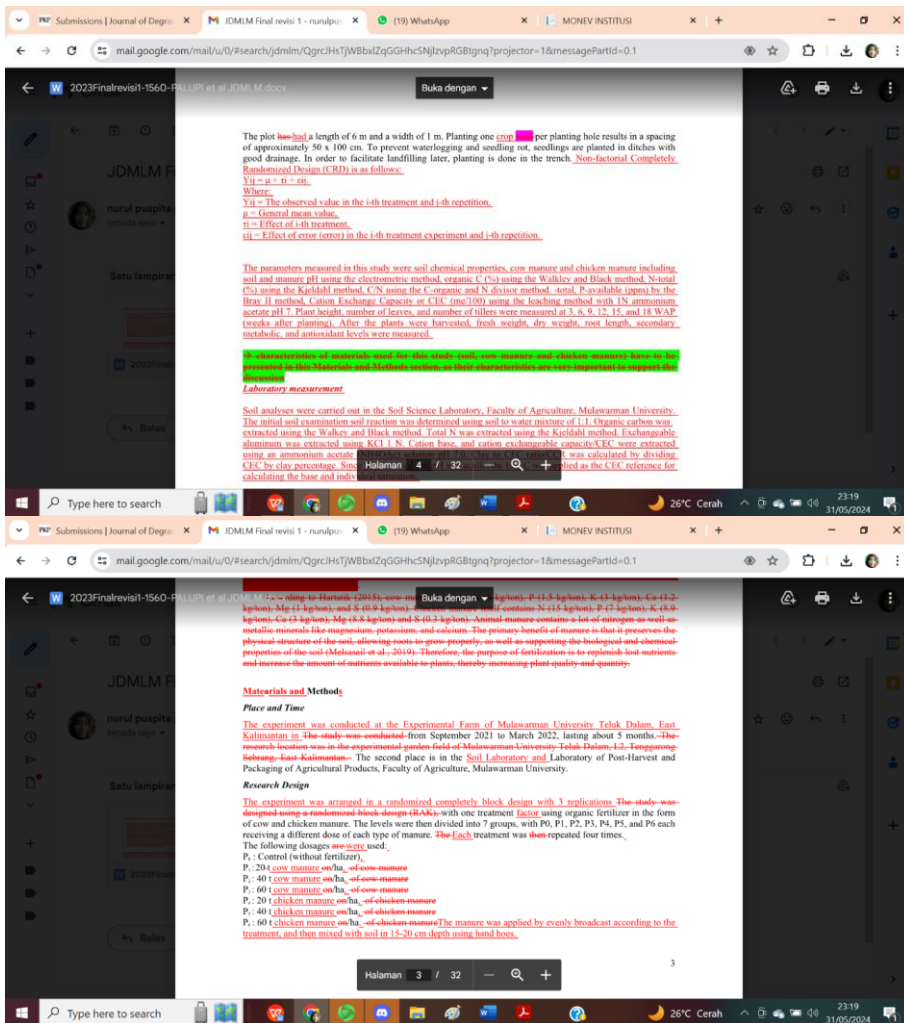
Manure input at all application levels increases the content of organic matter in the soil alone with the increase in incubation time. The highest increase in organic matter content in the soil alone with the addition of chicken manure 60 t/ha as shown in Table 5.

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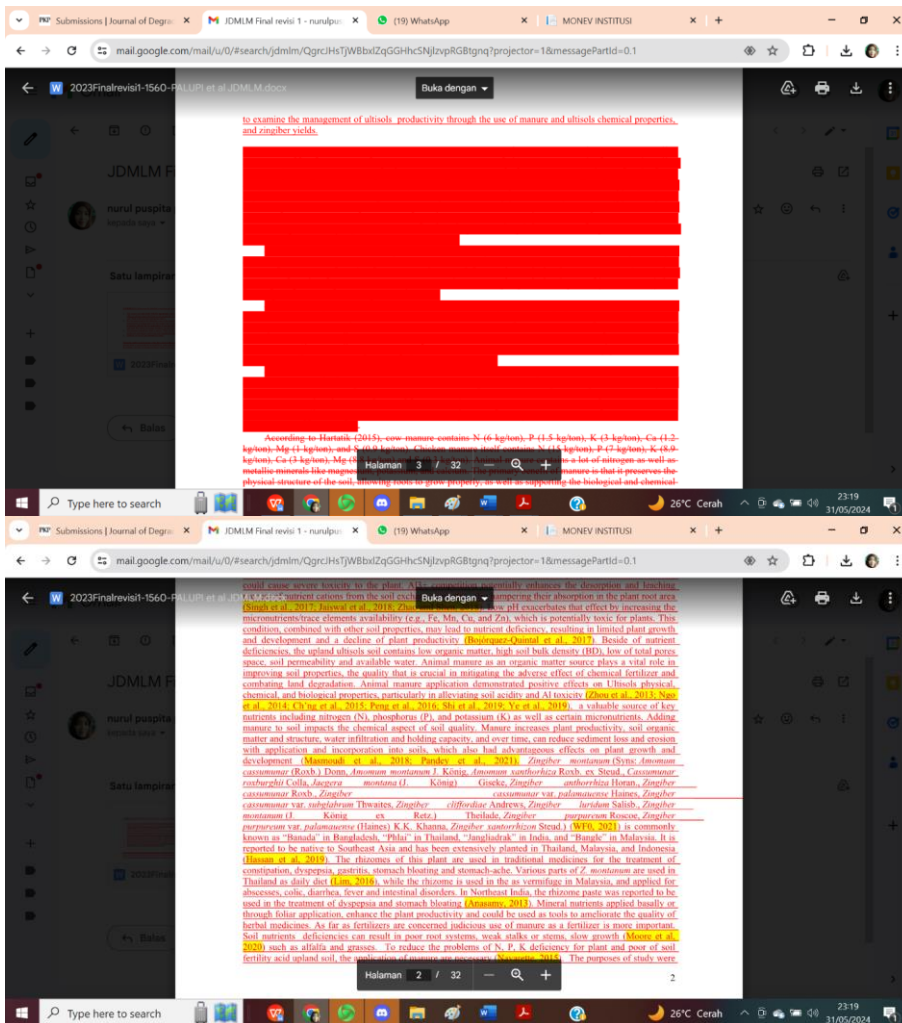




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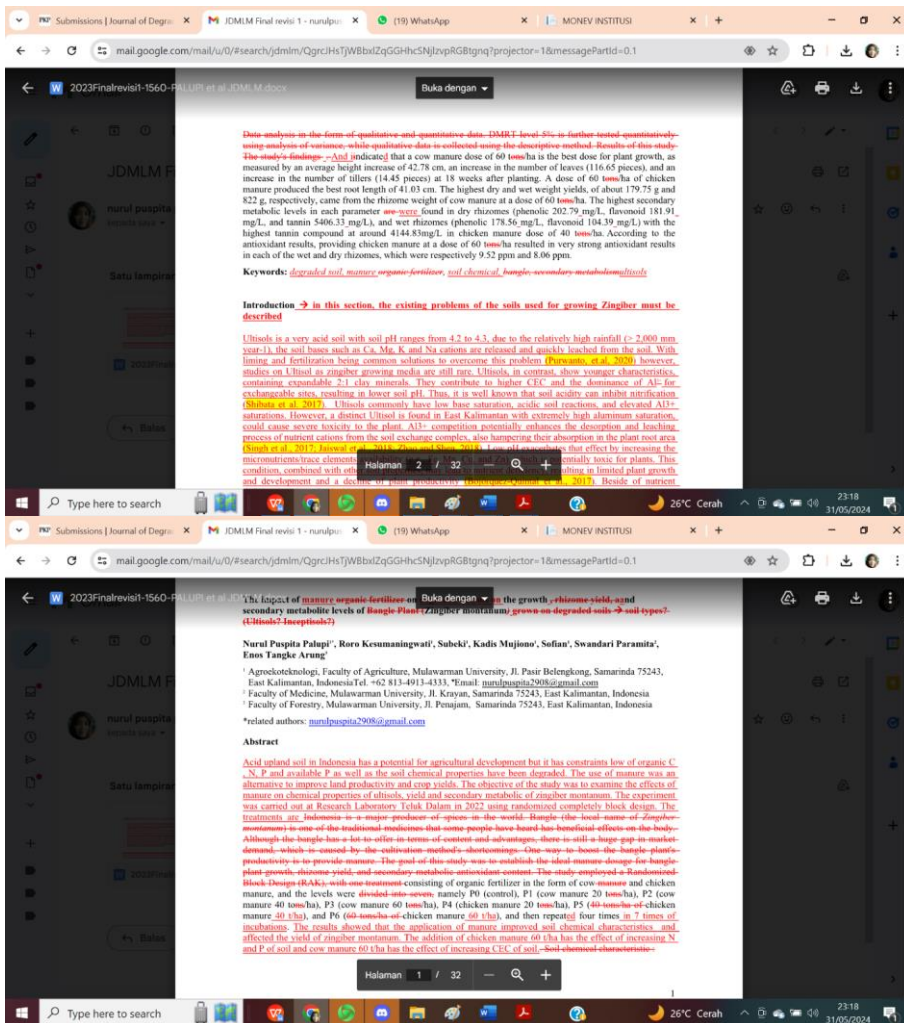


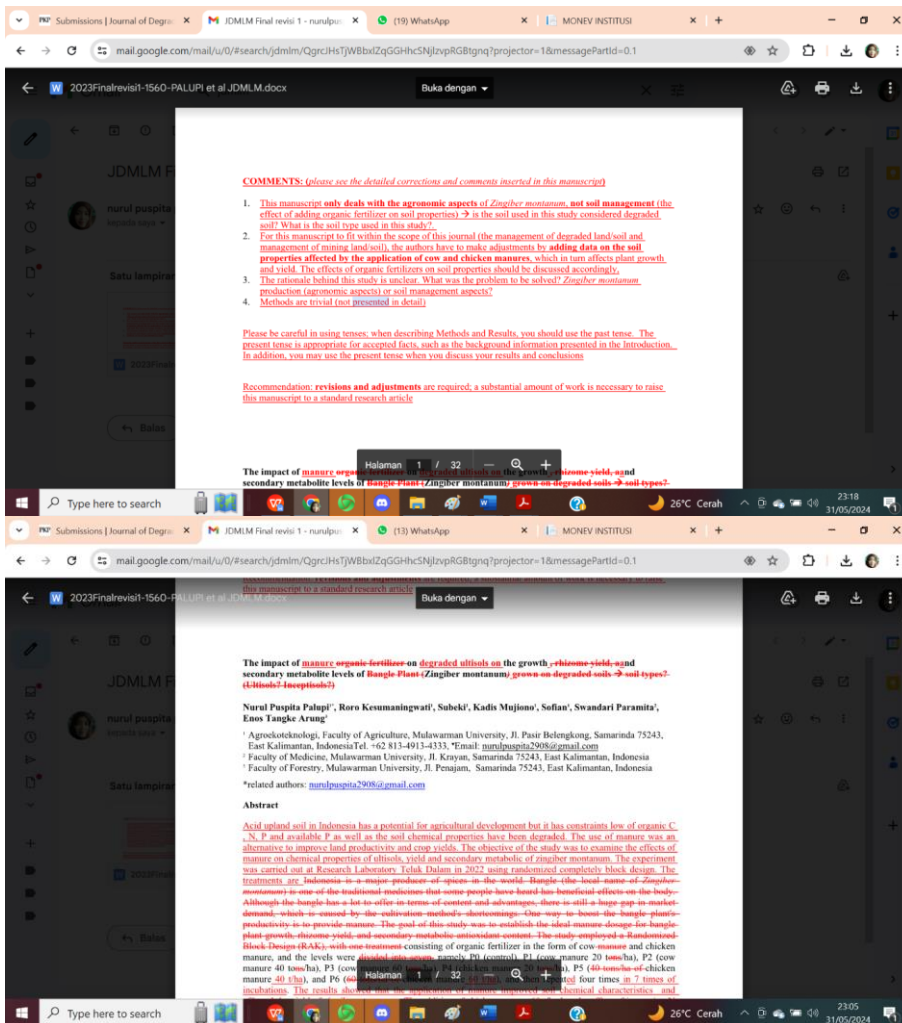
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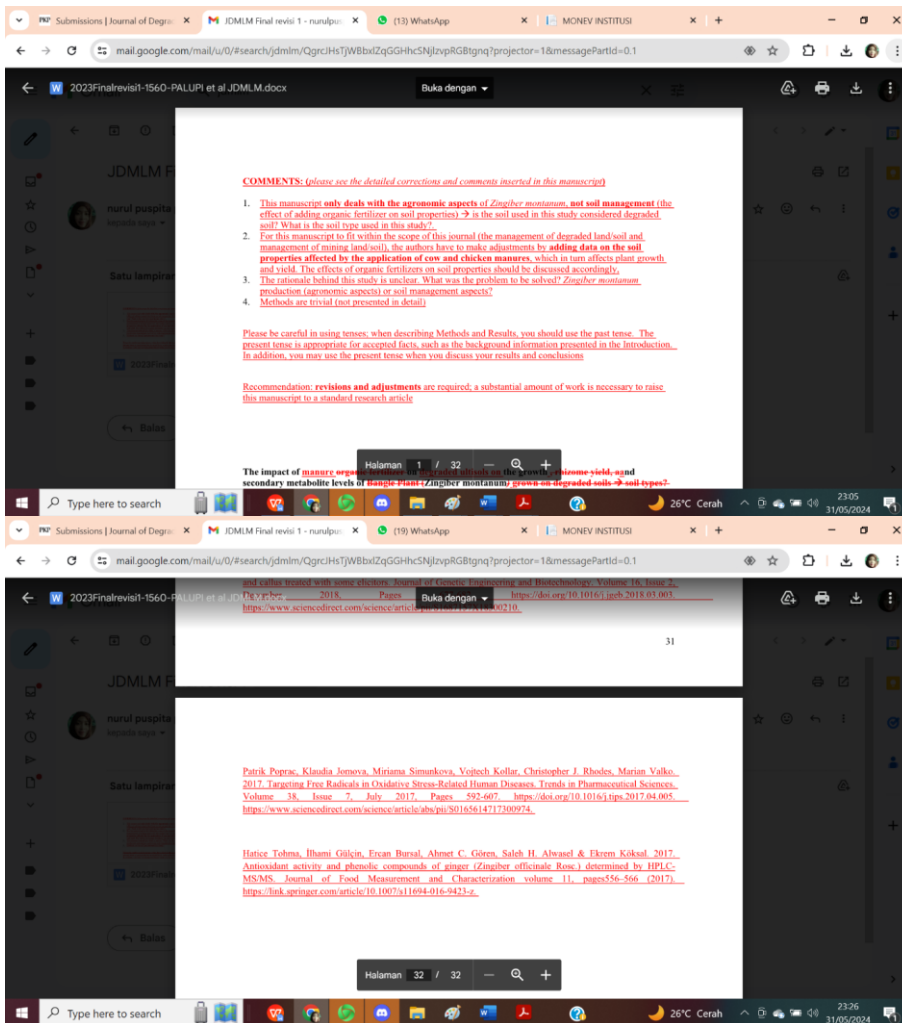
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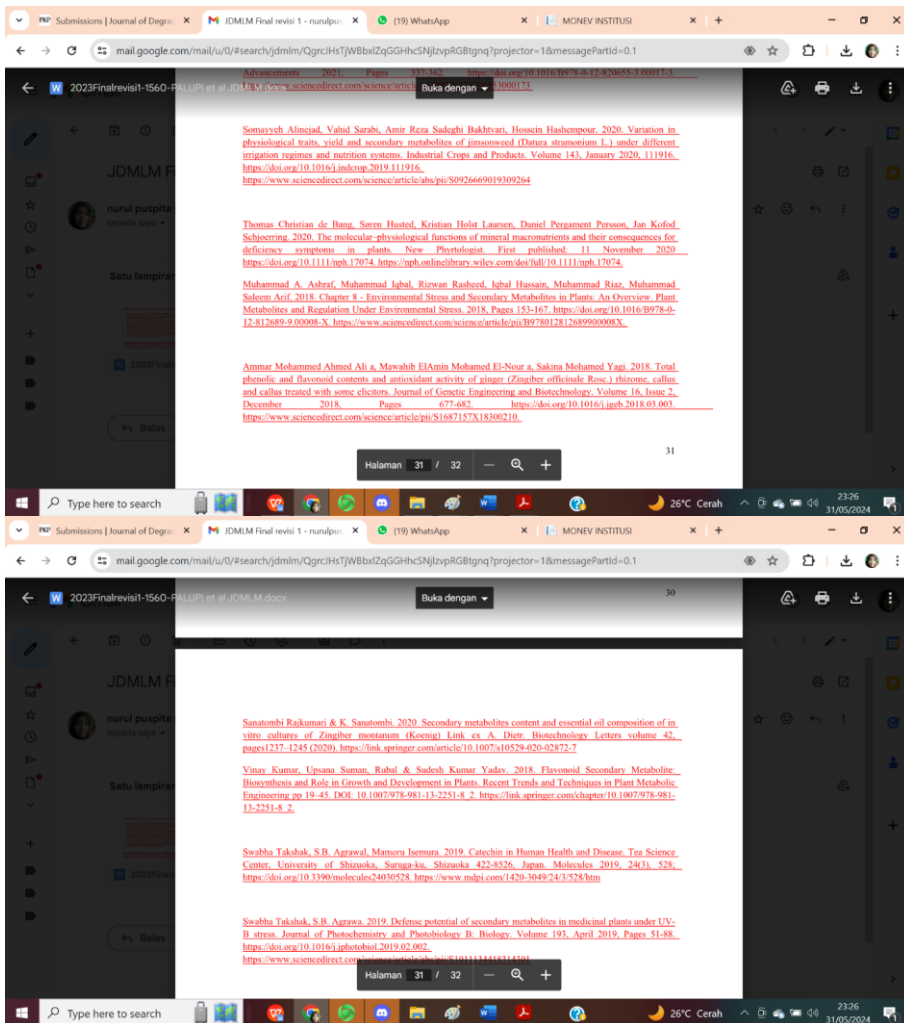




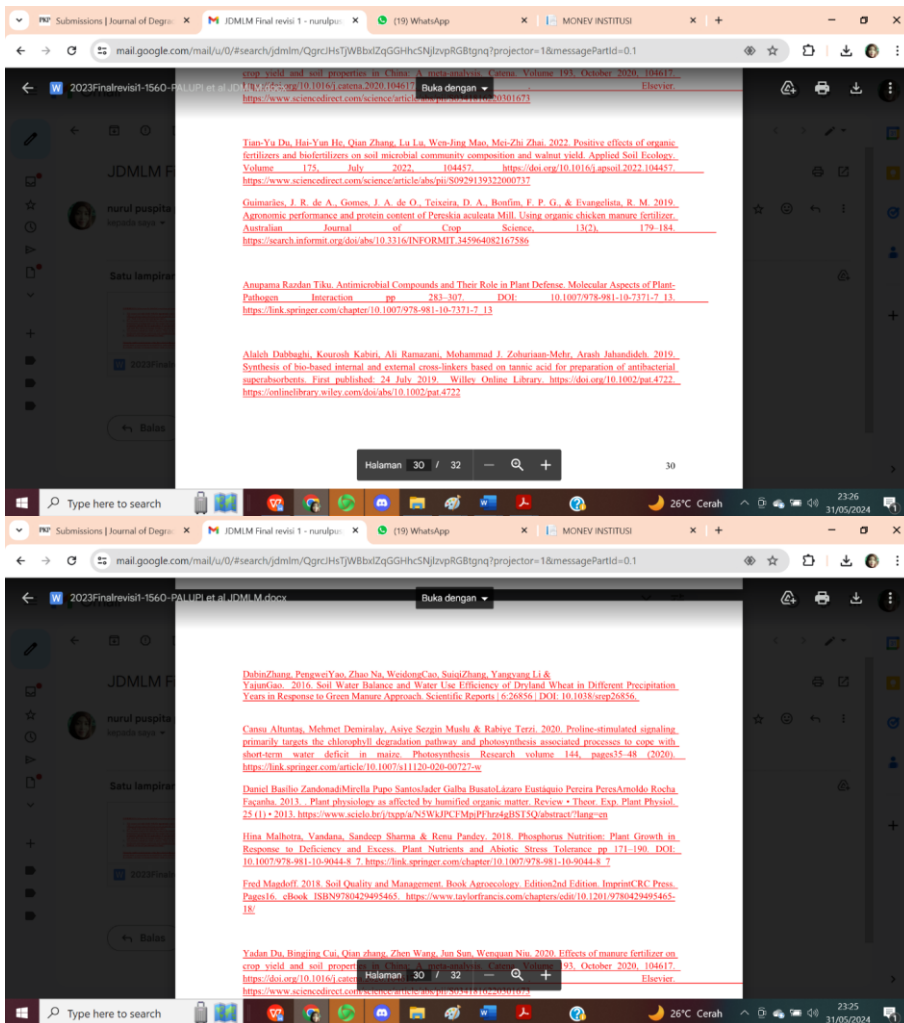
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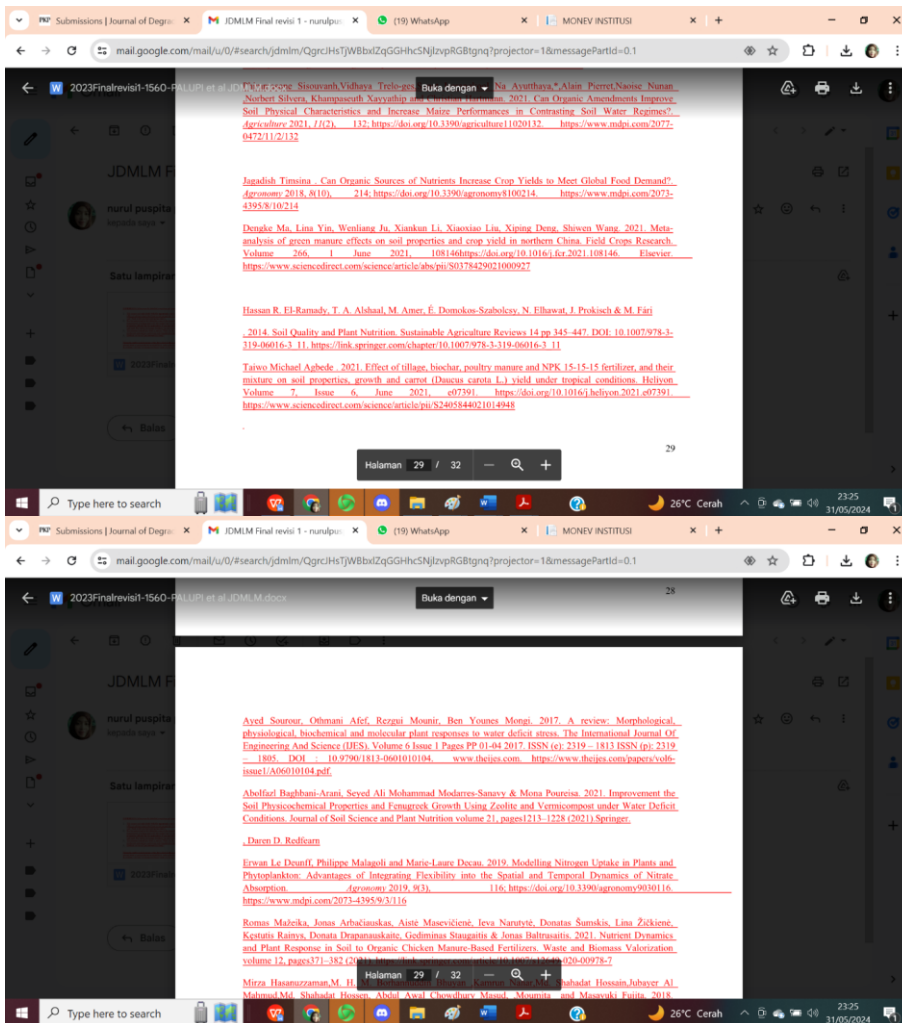
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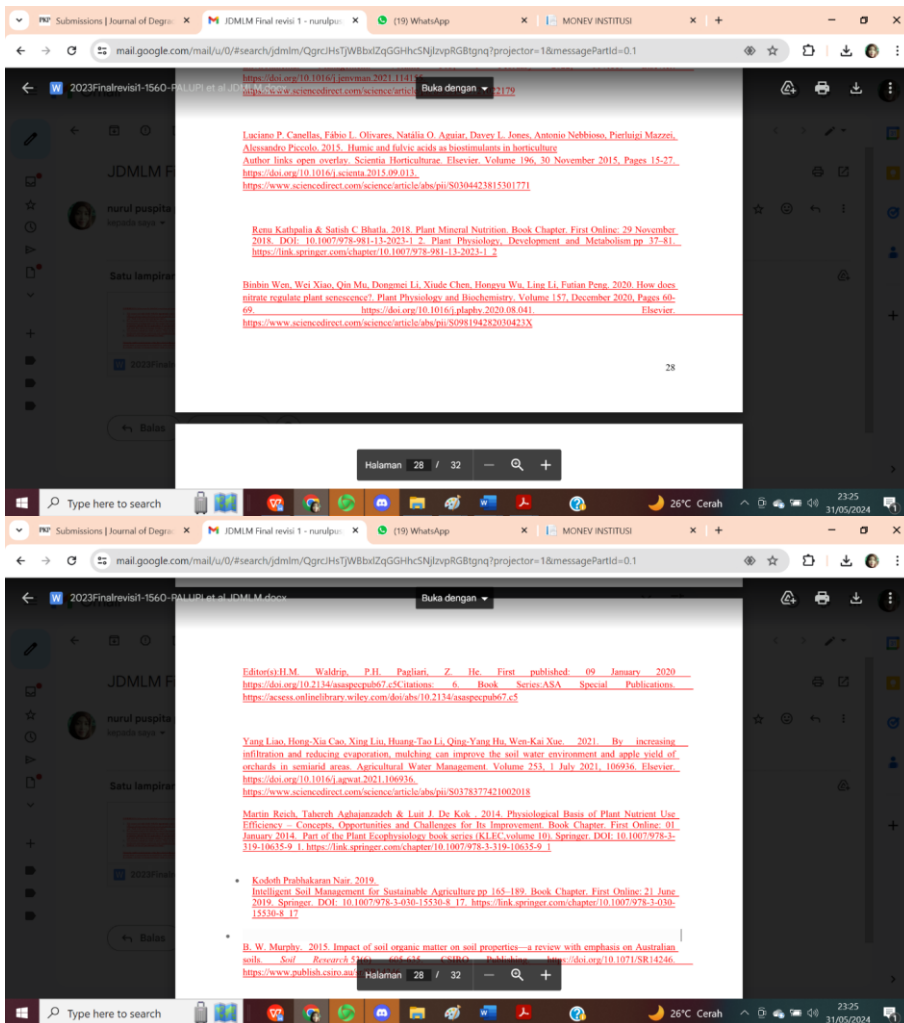
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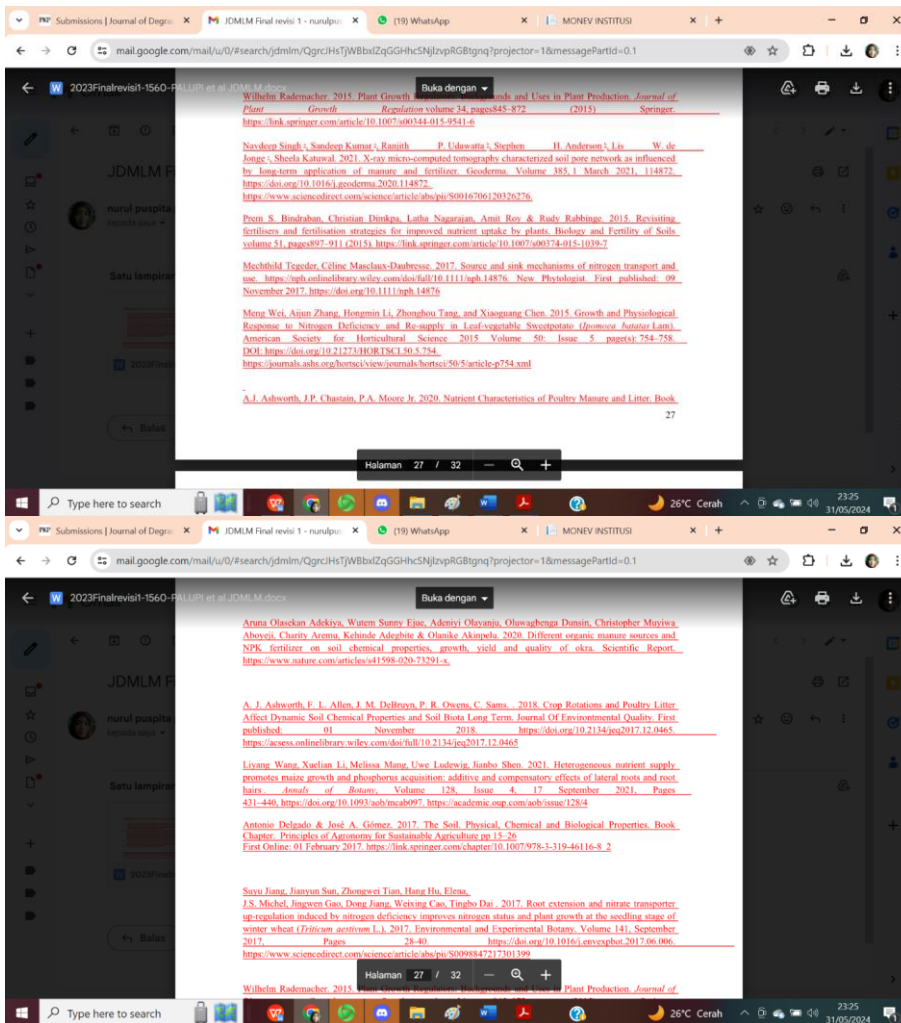
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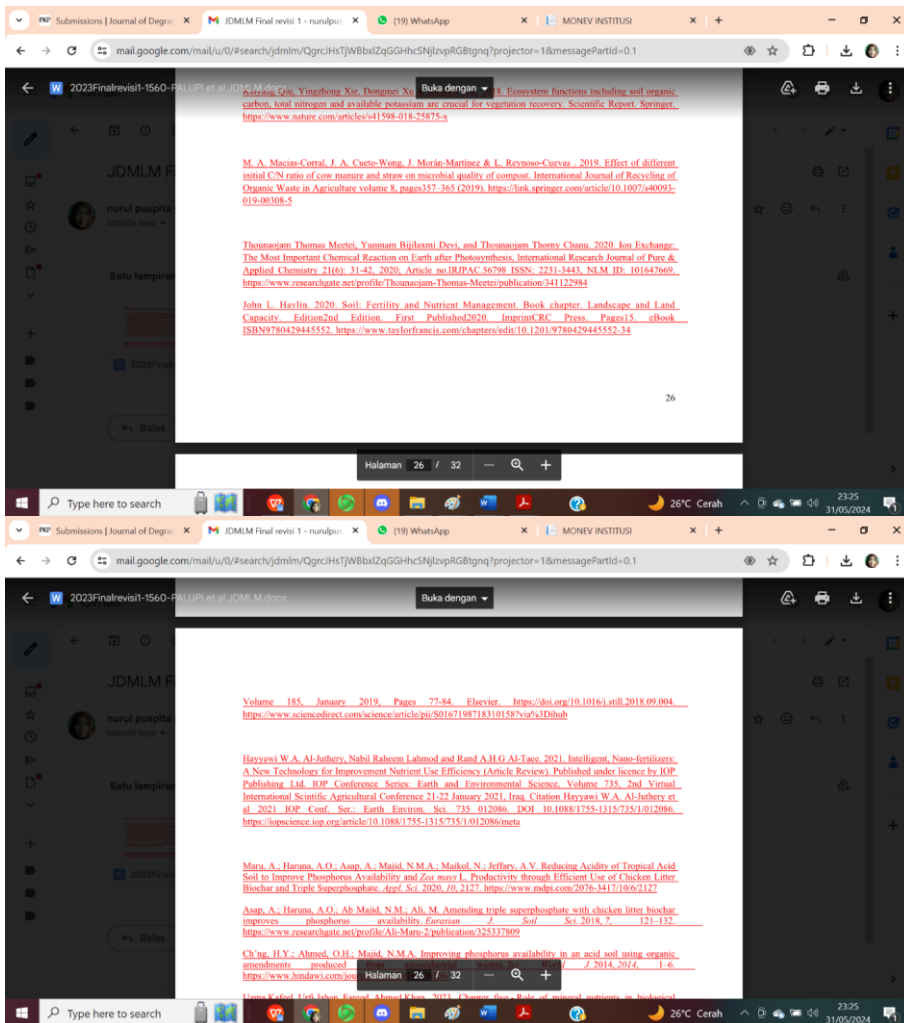


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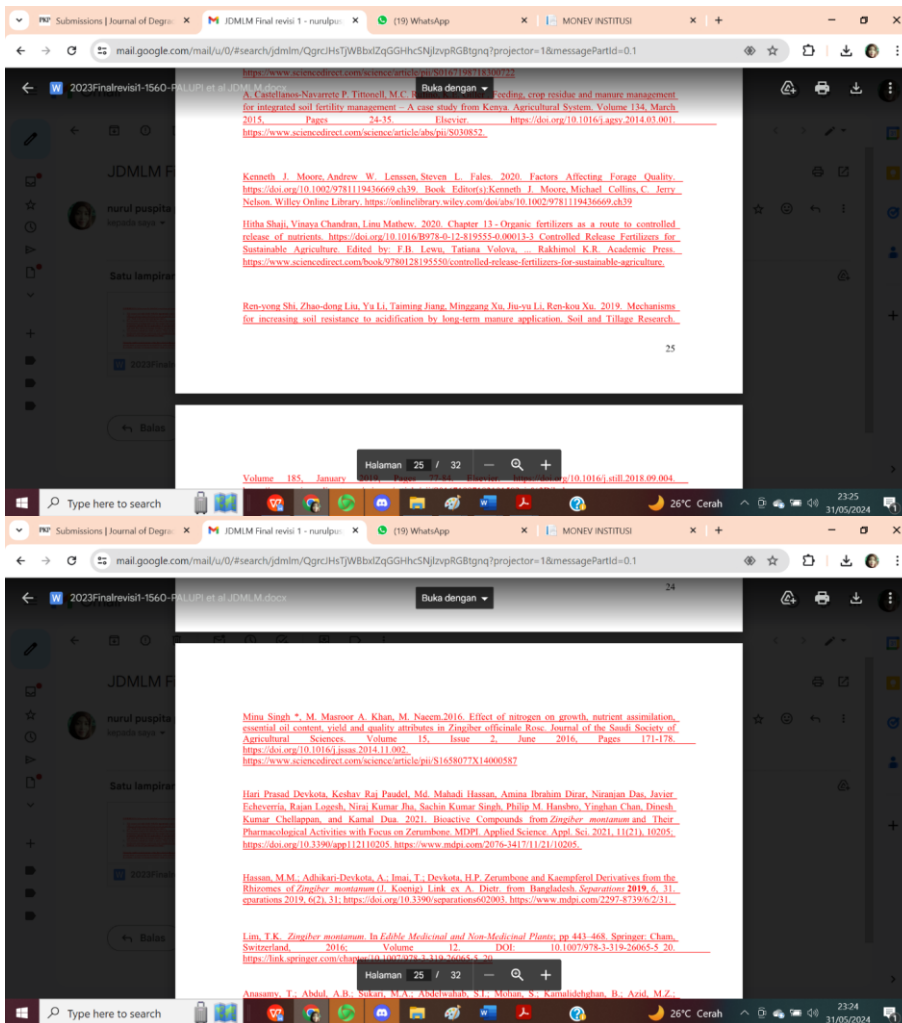


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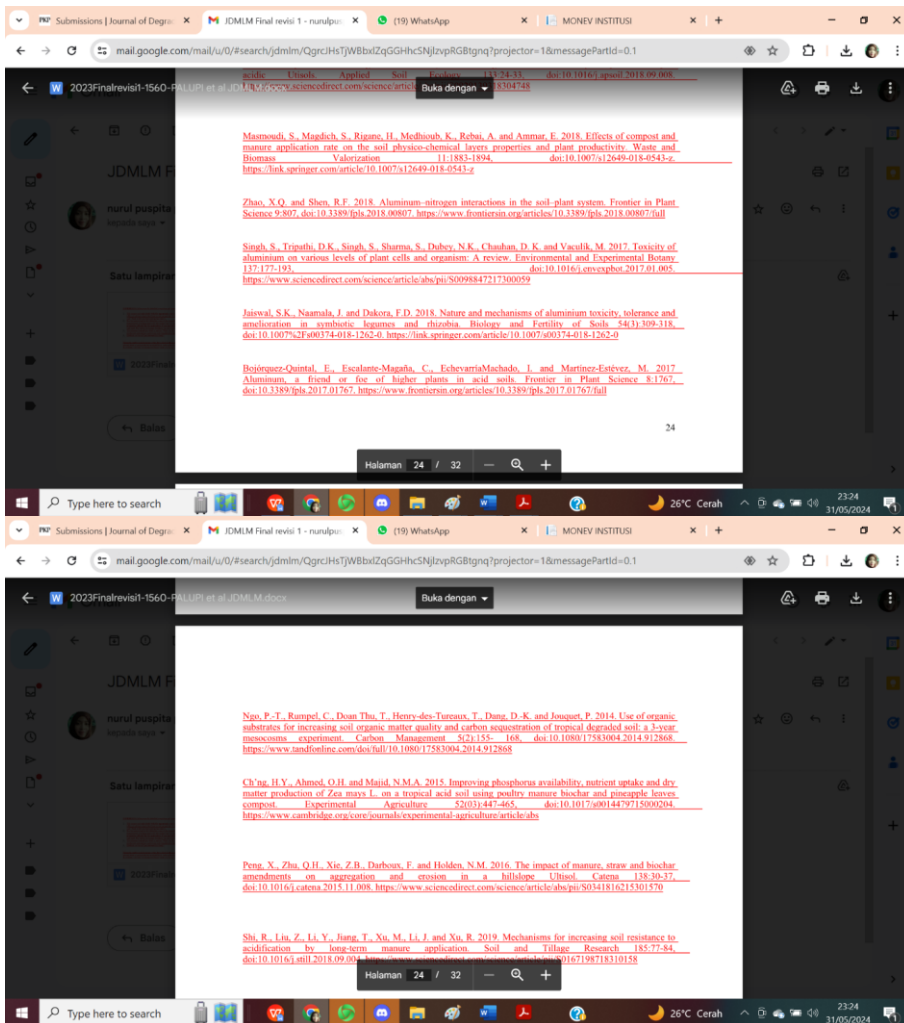




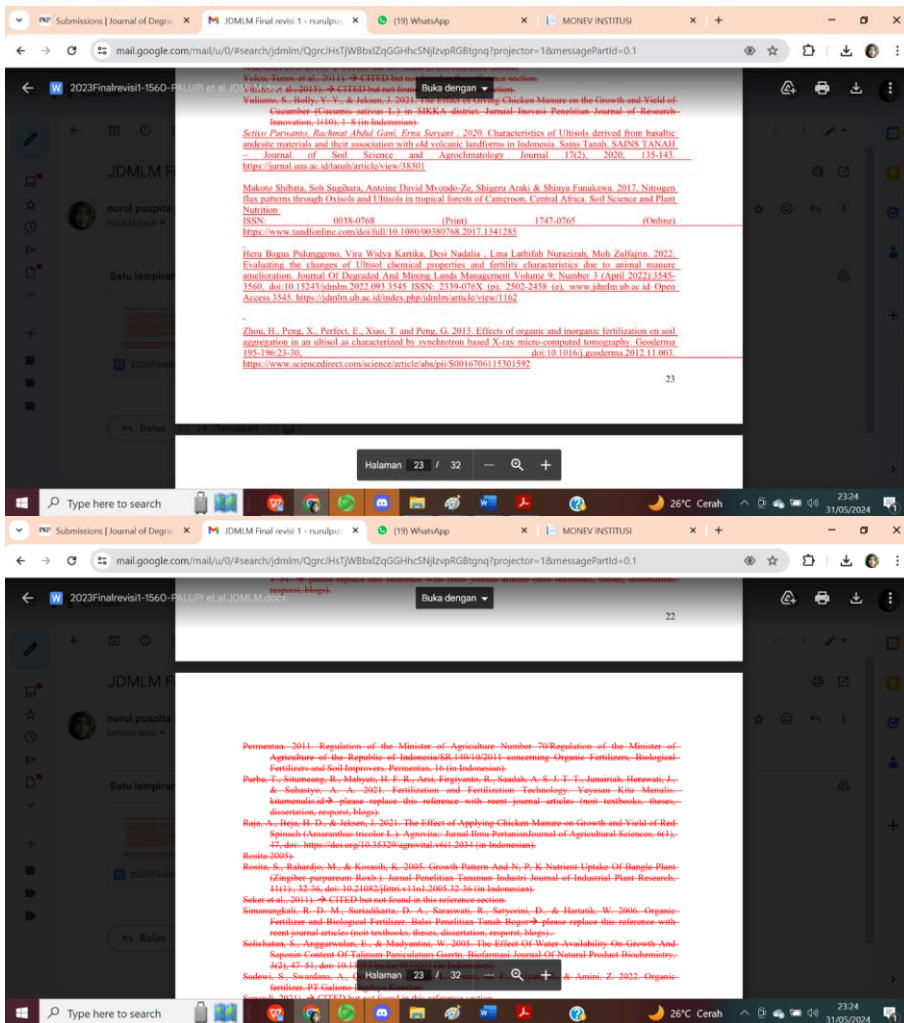
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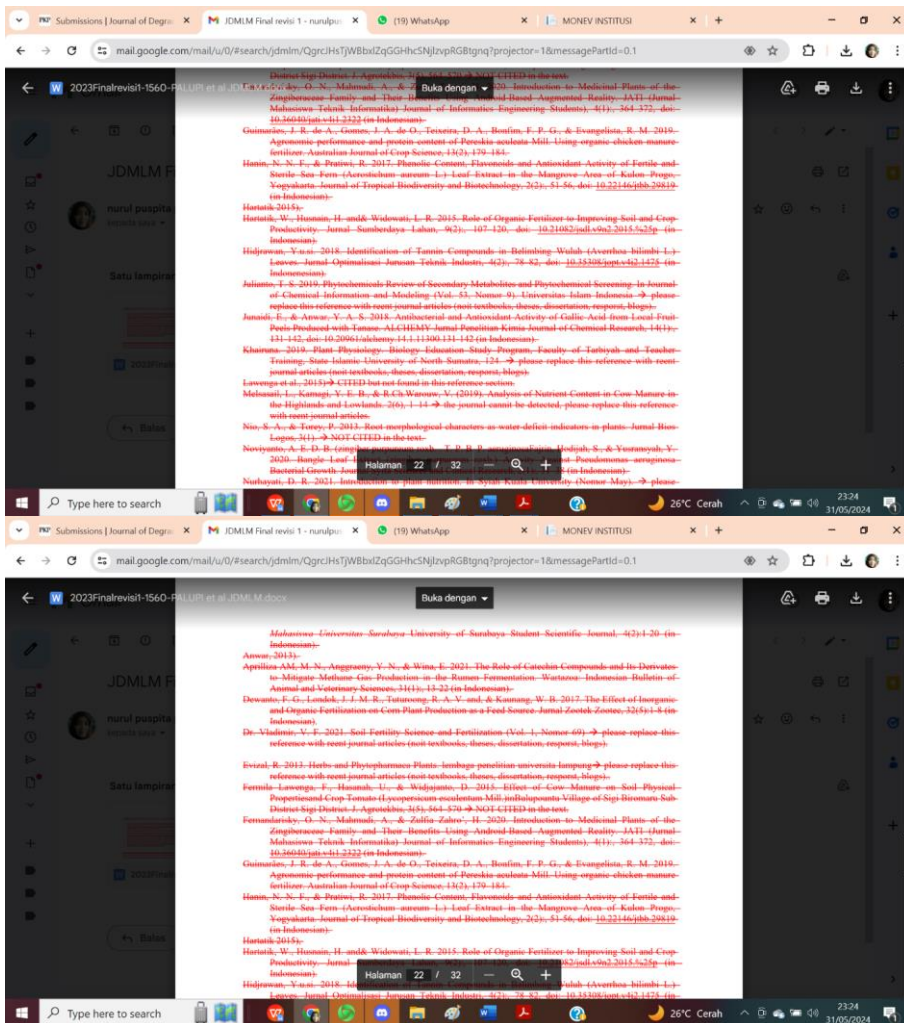
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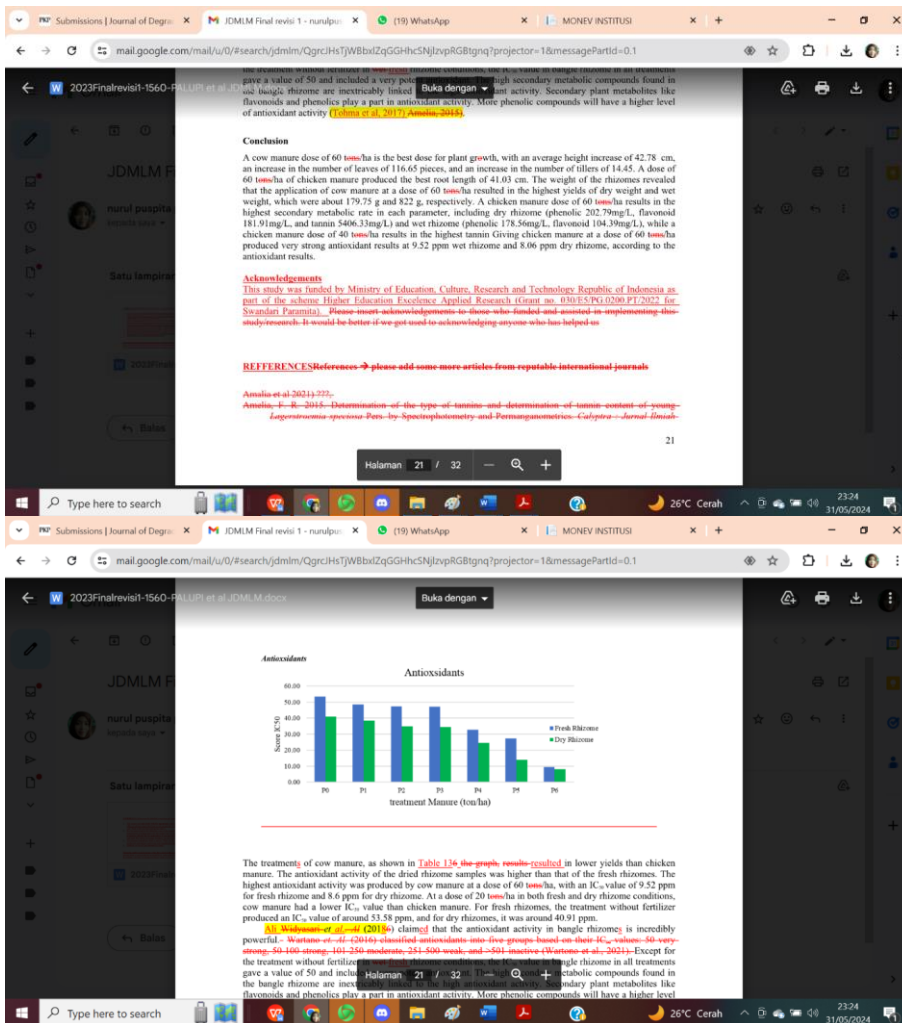
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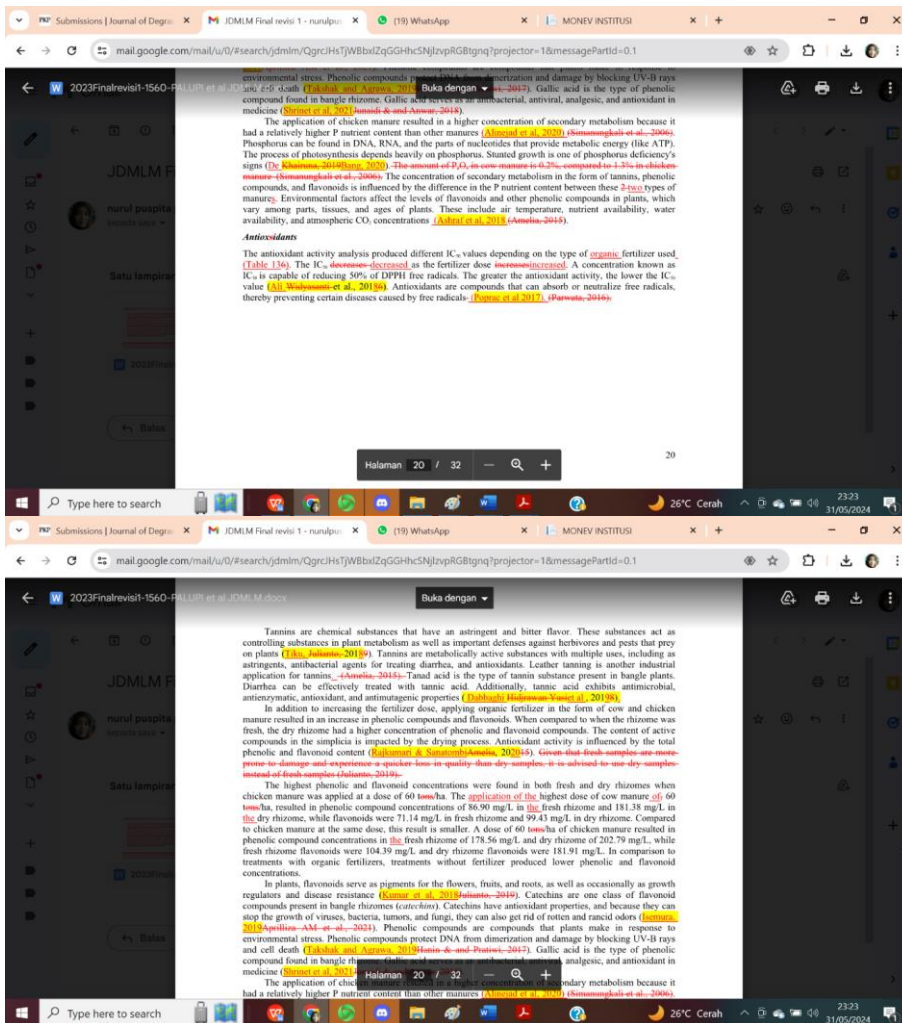


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rhizomes decreased with the increase in the dose of cow manure. In comparison to the use of manure that increased the concentration of tannin in fresh and dry rhizomes, the control provided the lowest tannin concentration.

Buka dengan

**Phenolic and Flavonoid**

Treatment	Fresh Rhizome (mg/L)	Dry Rhizome (mg/L)
P0	~20	~20
P1	~30	~30
P2	~40	~40
P3	~50	~50
P4	~60	~60
P5	~70	~70
P6	~80	~80

**Tannins**

Treatment	Fresh Rhizome (mg/L)	Dry Rhizome (mg/L)
P0	~2000	~2000
P1	~2500	~2500
P2	~3000	~3000
P3	~3500	~3500
P4	~4000	~4000
P5	~4500	~4500
P6	~5000	~5000

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is clear that as plants get older, they are able to absorb an increasing amount of N, P, and K nutrients. As can be seen, N is the nutrient that has accumulated in the rhizome in the greatest amount.

The application of manure enhances the chemical, physical, and biological properties of the soil, increase crop yield, and improves crop quality (D. Kurniawan, et al., 2023). High organic matter soils have beneficial microorganisms that encourage the breakdown of organic matter and release inorganic nutrients that are then available for plant uptake (Sobri, et al., 2014). Organic fertilizers can help to create ideal conditions in the soil for microorganisms that are beneficial to plants (Fu, et al., 2022).

Chicken manure is an organic fertilizer with high nitrogen content, despite not being the best dose for bangle rhizome weight yield. As they ensure the best nutrient management for plants, such fertilizers should be used promptly to partially replace chemical fertilizers (Gumarica et al., 2019).

**Secondary metabolic levels (phenolic compounds, flavonoids and tannins)**

The results of laboratory analysis found that the positive bangle rhizome contains compounds in the form of phenolics, tannins and flavonoids. These findings are consistent with (Rahmawati, et al., 2023), which found that the bangle plant contains secondary metabolites in the form of alkaloids, phenolic compounds, flavonoids, saponins, and terpenoids. These compounds respond differently to the concentration of organic fertilizer applied with in the form of cow and chicken manure. Table 5 The graph above shows the outcomes of these phytochemicals.

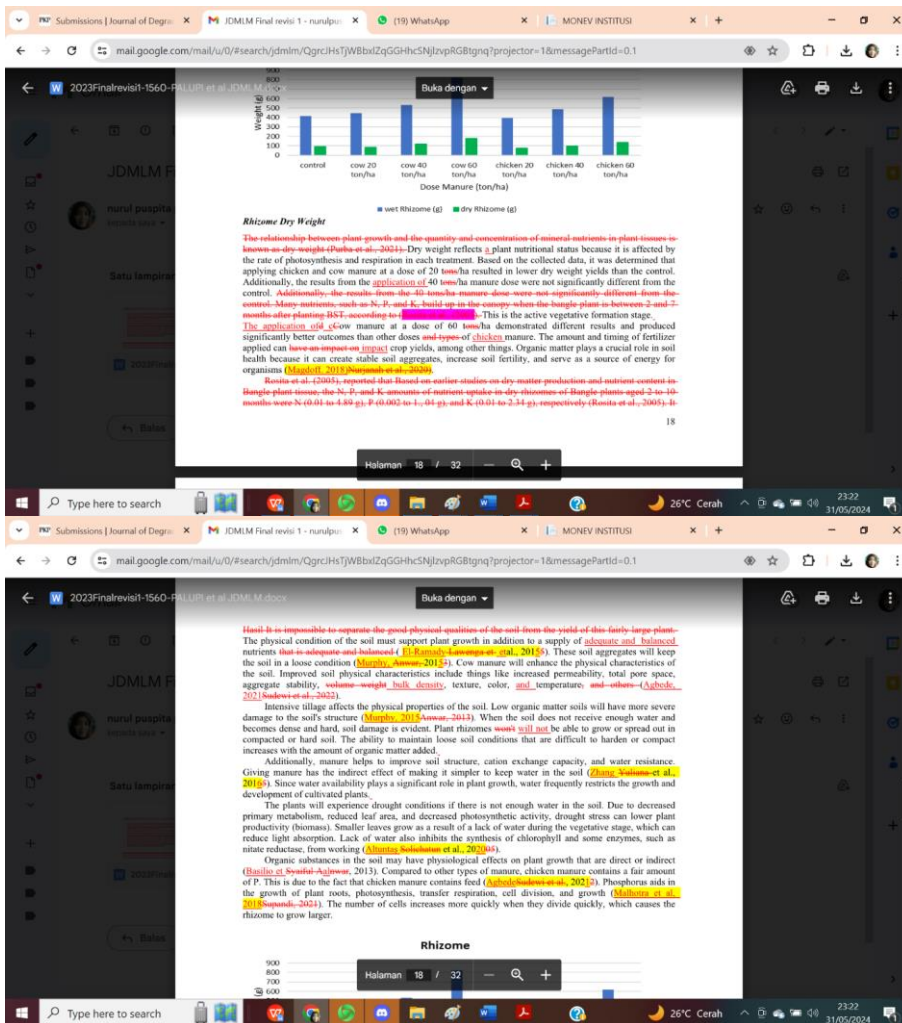
According to Table 12 (see graph above, the amount of tannin compounds will increase) as manure disappears rose. As can be seen, just like with The application of chicken manure on cow manure with three doses of 20, 40, and 60 tons always increased the tannin concentration in the rhizome as the dose increased. When compared to cow manure, applying chicken manure produced a significantly better result. It is evident that using 20 tons/ha (P4) of chicken manure instead of 60 tons/ha (P6) of cow manure resulted in a higher tannin concentration (P4).

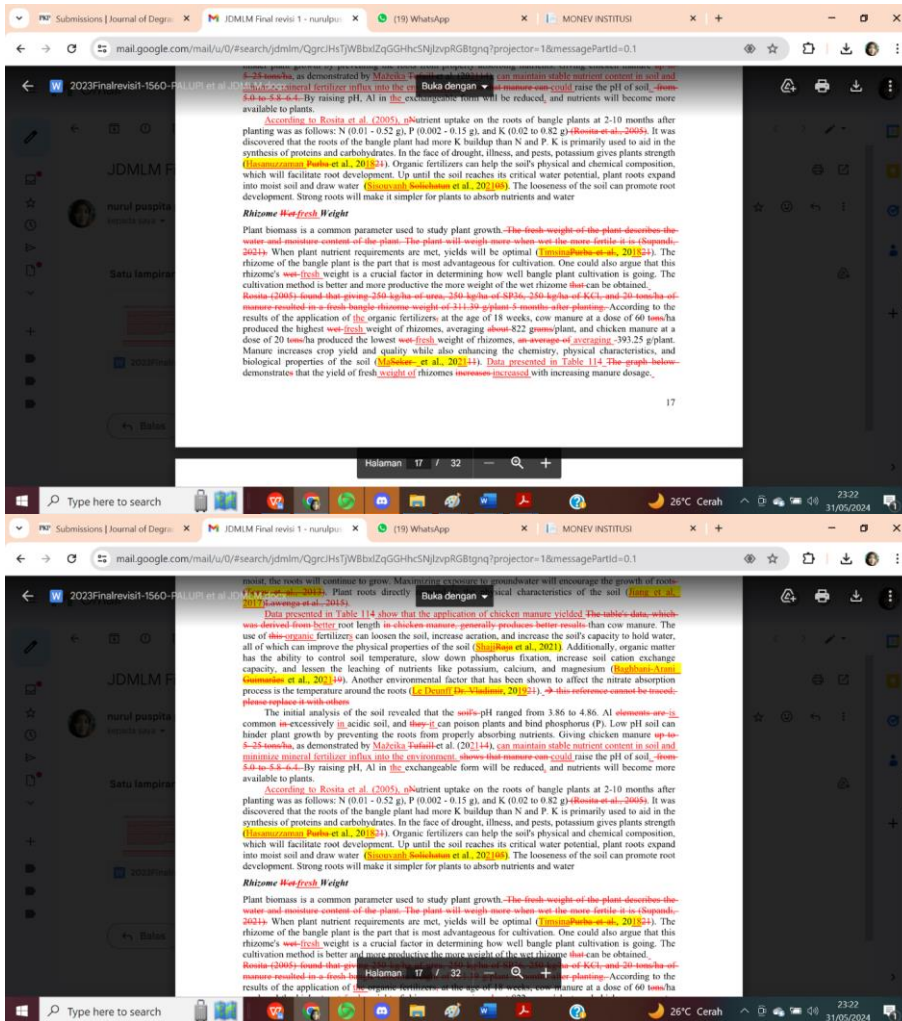
Dry rhizomes produced a higher concentration of tannins than fresh rhizomes, which produced different results regarding tannin concentration. The concentration of tannins in fresh rhizomes increased non-significantly with the addition of a dose of cow manure, whereas the concentration of tannin compounds in fresh rhizomes decreased with the increase in the dose of cow manure. In comparison to the use of manure that increased in fresh and dry rhizomes, the control provided the lowest tannin concentration.

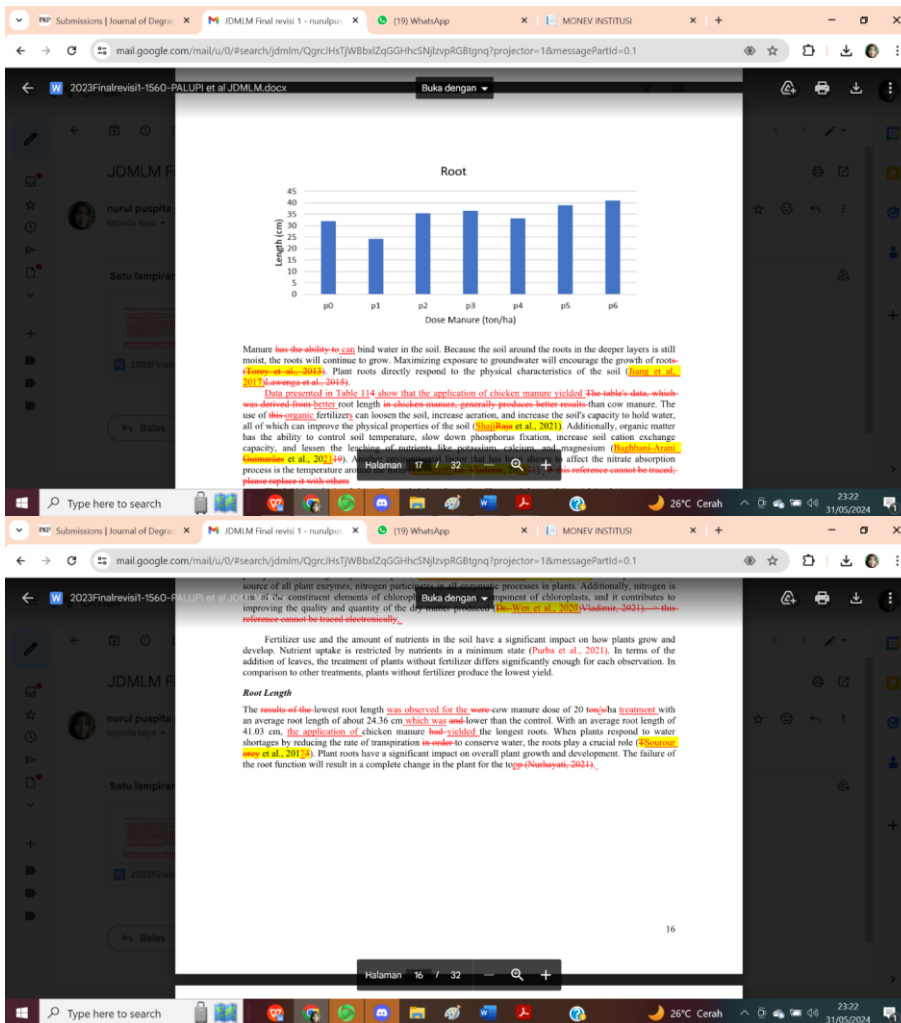
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In addition to the nutrients that plant's needs, manure also contains humic, fulvic acids, growth hormones, and other substances that promote plant growth and increase nutrient uptake by plants (Gudimov et al., 2014; Masita et al., 2014). The amount of photosynthesis is influenced by the number of leaves present, and plants with more leaves may produce heavier and bigger rhizomes as a result.

The number of leaves added is also affected by the number of shoots and plant height. The number of leaves will increase as the plant ages and grows taller, produces more leaves on a single stem, and produces more tillers. The nutrients required for plant growth are present in sufficient amounts in manure. The related observation variables will be impacted by the dose of manure quality and quantity.

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In comparison to manure application, treatment without fertilizer produced the lowest yield. Data accounted in Table 92 show that 20 tons/ha of chicken 40 tons/ha that there was a noticeable increase in the number of tiller plants that were at 18 WAP of age. A plant needs nutrients for its physiological processes during growth and development. Plant growth and production will be subpar due to a lack of nutrients (Eckstein et al., 2014).

Functions of organic matter as a biological buffer so that the soil can supply plants with a balanced amount of nutrients (Hartono et al., 2019). Loosening the topsoil, increasing water absorption and storage, and boosting soil fertility are all important functions of manure (Baskoro et al., 2021; Ganesa, 2013). A sudden rise in the number of tillers can result from the ease with which new shoots can emerge from loose, moist soil.

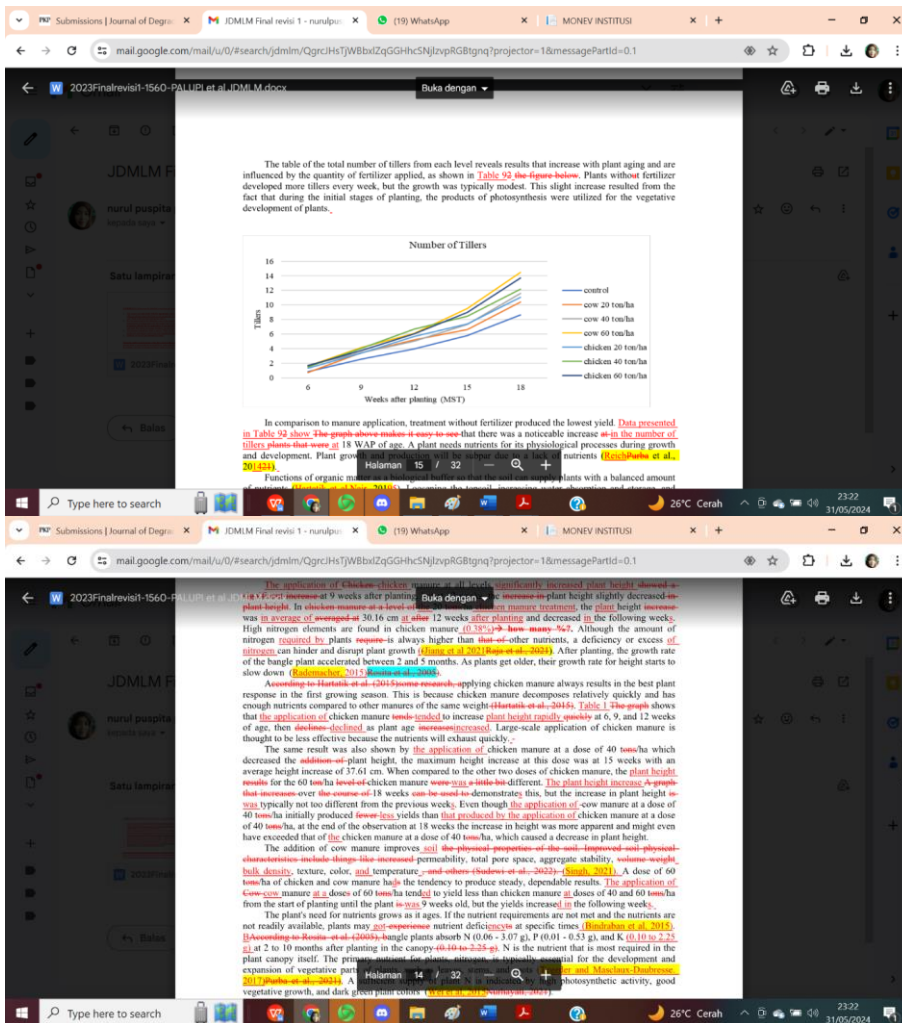
At the start of planting, there were typically fewer and nearly identical numbers of tillers in each treatment. The nutrients in this fertilizer are not readily available to plants, which is the cause of the slow plant growth at the start of the planting period. The extent of these materials' mineralization or decomposition has a significant impact on the nutrients' availability. Manure's low nutrient availability is partially caused by the presence of N, P, and other elements in complex compounds that are challenging to decompose (Hawinko et al., 2021; Iqbal, 2015).

At 6 week after planting, all treatments tended to be similar and the differences between the tillers in each treatment tended to be minimal. Although cow manure at a dose of 60 tons/ha was the best dose with an average number of tillers of 14.45 tillers, chicken manure typically produced better results than cow manure at the same dose. In comparison to other manure doses, chicken manure 40 tons/ha at 12 week after planting produced the best results with 6.65 tillers.

**Number of Leaves**

An increasing number of leaves are produced each week as a result of the weekly application of cow and chicken manure. Table 143 shows that at about 12 weeks of age, the number of leaves increased significantly. Every week, the number of leaves significantly increased with 37 leaves every three weeks. Similarly, the number of leaves increased with 37 leaves every three weeks. manure applied in the same amount. Both cow and chicken manure at a dose of 50 tons/ha and chicken manure at a dose of 40

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Weeks after planting (MST)	control	cow 20 ton/ha	cow 40 ton/ha	chicken 20 ton/ha	chicken 40 ton/ha	chicken 60 ton/ha
6	10	15	18	20	22	25
9	12	18	22	25	28	32
12	15	22	28	32	35	38
15	18	25	32	35	38	42
18	20	28	35	38	42	45

The application of Chicken-chicken manure at all levels significantly increased plant height showed a significant increase at 9 weeks after planting, but at 12 weeks, the increase in plant height slightly decreased in plant height. In chicken manure level of 20 ton/ha chicken manure treatment, the plant height increase was in average of averaged. High nitrogen elements are required by plants.

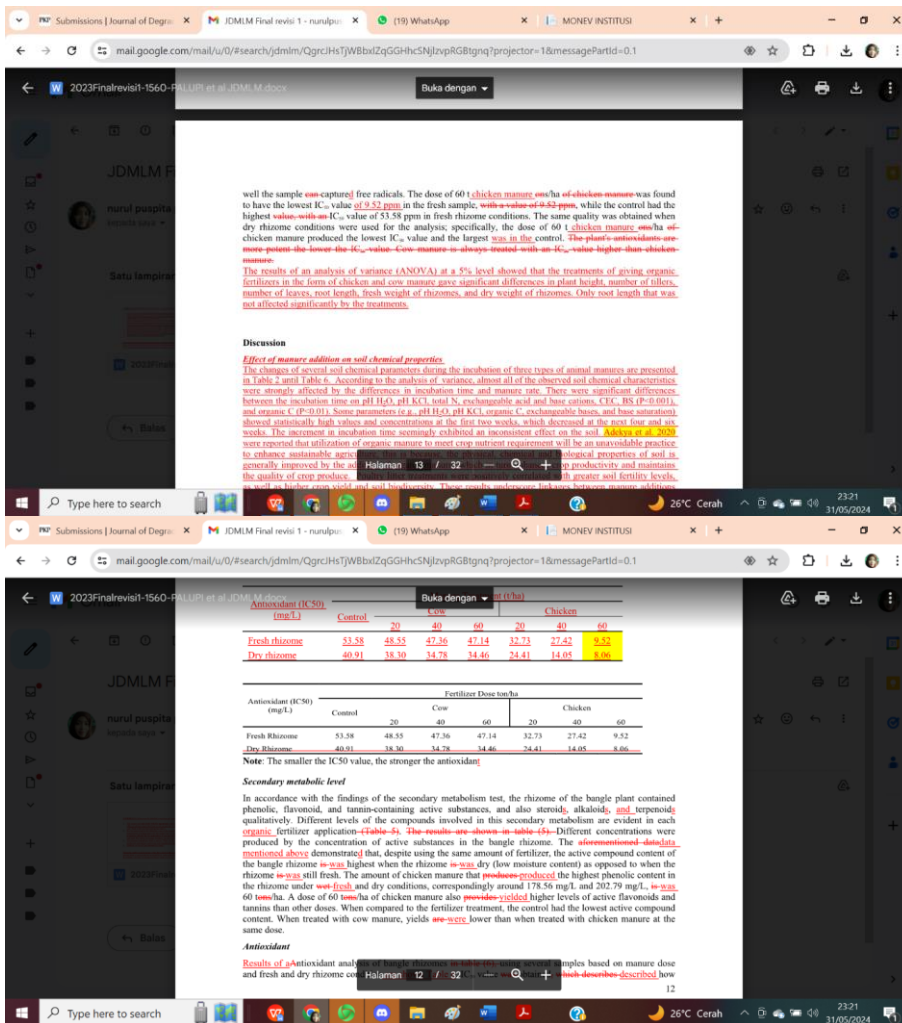
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Plant Height

According to the table Table S4, the average increase in plant height starting at 6,9,12,15, and 18 WAP produced mixed results that tended to fluctuate. At the age of 6 weeks, the treatment without fertilizer (control) continued to grow until week 12 when it reached its peak with an average height increase of about 24.68 cm, before declining, adding less height at the following weeks. In comparison to other treatments, the results from the fertilizer-free treatment had the lowest graph. In order for a plant to grow, nutrients must be obtained from the soil by the roots through their root hairs (Sudrajat et al., 2021; Siswani et al., 2022). Organic matter affects plant growth by influencing the physical, chemical, and biological properties of the soil (Rahman, 2010; Rahmat, 2010). The more organic matter is provided, the faster the plant will grow. Compared to chicken manure, cow manure typically produces better plant growth results. According to (Hartono et al., 2018), cow manure contains N (16-18%), P (4-6%), K (12-14%), Ca (1-2%), Mg (1-2%), and S (1-2%). These nutrients can support the growth of Bangko plants, which have a long harvest period. Cow manure's graph tends to rise with each observation.

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Condition	Treatment	Level (mg/L)		
		Phenolic	Flavonoid	Tannin
Fresh	Control	31,64	23,08	2934,83
	Cow 20 ton/ha	43,18	41,55	2991,50
	Cow 40 ton/ha	77,03	64,30	3151,50
	Cow 60 ton/ha	86,90	71,14	3198,17
	Chicken 20 ton/ha	97,67	74,79	4046,50
	Chicken 40 ton/ha	107,54	78,71	4144,83
Chicken 60 ton/ha	178,56	104,20	3661,33	
Dry	Control	124,97	38,44	1932,33
	Cow 20 ton/ha	133,56	67,23	2504,00
	Cow 40 ton/ha	148,44	68,76	3034,83
	Cow 60 ton/ha	181,38	99,43	4734,67
	Chicken 20 ton/ha	175,23	120,06	4903,00
	Chicken 40 ton/ha	198,69	156,10	5088,00
Chicken 60 ton/ha	207,79	181,91	5406,11	

Note: The number followed by yellow denotes the best outcome in each observation variable for the flavonoid, test, tannin test, and phenolic test results contents.

Table 116. Antioxidant based on IC50 value → Tables have to be presented in Word format (generated from Excel format), not in JPEG format, as JPEG format cannot be edited.

Antioxidant (IC50) (mg/L)	Control	Manure Treatment (t/ha)
Fresh rhizome	31,64	107,54
Dry	124,97	175,23

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Condition	Treatment	Phenolic	Flavonoid	Tannin
Fresh	Control	31,64	23,08	2934,83
	Cow 20 t/ha	43,18	41,55	2991,50
	Cow 40 t/ha	77,03	64,30	3151,50
	Cow 60 t/ha	86,90	71,14	3198,17
	Chicken 20 t/ha	97,67	74,79	4046,50
	Chicken 40 t/ha	107,54	78,71	4144,83
Dry	Control	124,97	38,44	1932,33
	Cow 20 t/ha	133,56	67,23	2504,00
	Cow 40 t/ha	148,44	68,76	3034,83
	Cow 60 t/ha	181,38	99,43	4734,67
	Chicken 20 t/ha	175,23	120,06	4903,00
	Chicken 40 t/ha	198,69	156,10	5088,00
Chicken 60 t/ha	207,79	181,91	5406,11	

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Manure Treatment (ton/ha)	Root Length (cm)	Rhizome wet weight (g)	Rhizome dry weight (g)
Control	31.90 ab	415.25 ab	93.75 ab
Cow 20 ton/ha	24.36 a	444.75 ab	86.00 a
Cow 40 ton/ha	35.36 b	530.00 bc	119.00 ab
Cow 60 ton/ha	36.50 b	422.00 ab	179.75 c
Chicken 20 ton/ha	33.20 ab	393.25 a	78.25 a
Chicken 40 ton/ha	38.95 b	487.50 abc	99.75 ab
Chicken 60 ton/ha	41.03 b	618.75 c	134.75 bc

Notes: Based on Duncan's Multiple Range Test, multiple-comparison-further-test (DMRT) at a 5% significance level, values in the same columns that are followed by the same letter do not differ significantly.

Table 125. Secondary Metabolic Levels in Bangle Rhizome in Each Treatment (mg/L). → Tables have to be presented in Word-format (generated from Excel-format), not in JPEG-format, as JPEG-format cannot be edited.

Condition	Manure Treatment (t/ha)	Level mg/L		
		Phenolic	Flavonoid	Tannin
Control		31.64	23.08	2034.83
Cow 20 t/ha		43.18	41.55	2991.50
Cow 40 t/ha		77.03	64.30	3151.50
Chicken 20 t/ha				3198.17
Chicken 40 t/ha				4086.50

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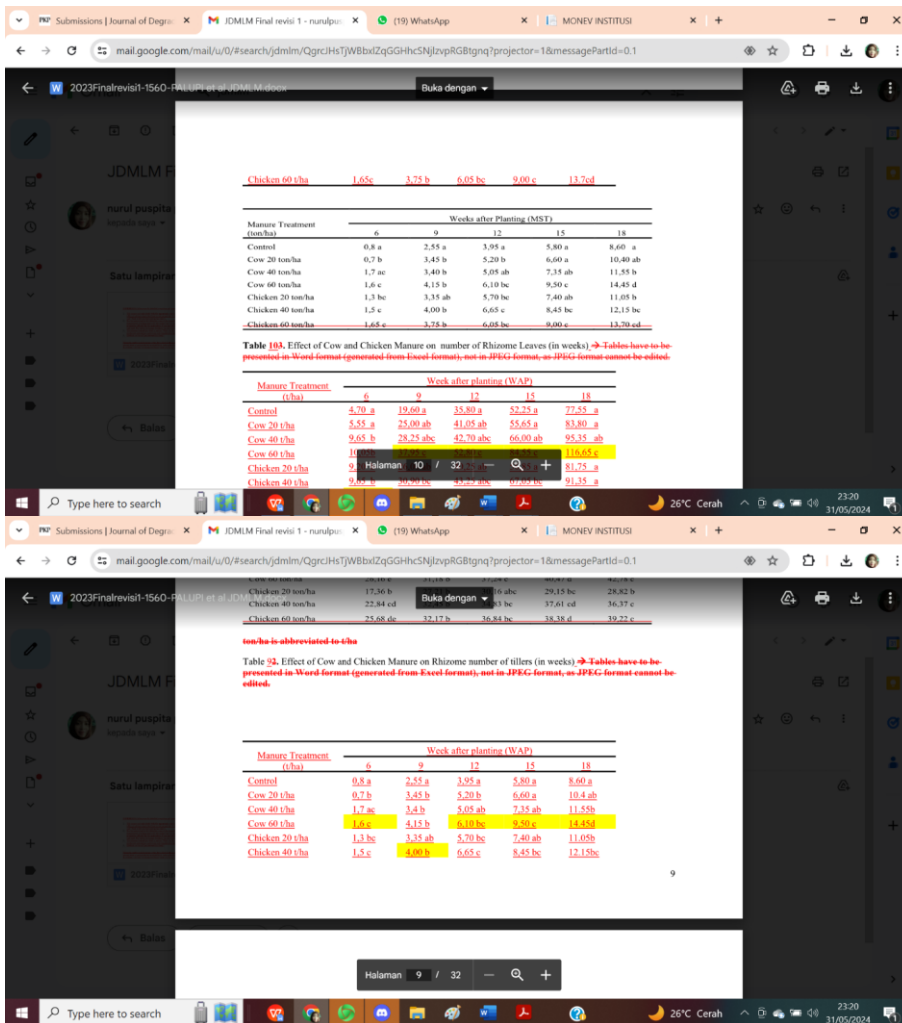
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Manure Treatment (ton/ha)	Root Length (cm)	Rhizome wet weight (g)	Rhizome dry weight (g)
Control	31.90 ab	415.25 ab	93.75 ab
Cow 20 t/ha	24.36 a	444.75 ab	86.00 a
Cow 40 t/ha	35.36 b	530.00 bc	119.00 ab
Cow 60 t/ha	36.50 b	422.00 ab	179.75 c
Chicken 20 t/ha	33.20 ab	393.25 a	78.25 a
Chicken 40 t/ha	38.95 b	487.50 abc	99.75 ab
Chicken 60 t/ha	41.03 b	618.75 c	134.75 bc

Table 114. Effect of Cow and Chicken Manure on Root Length and Rhizome Weight (in 15 week). → Tables have to be presented in Word-format (generated from Excel-format), not in JPEG-format, as JPEG-format cannot be edited.

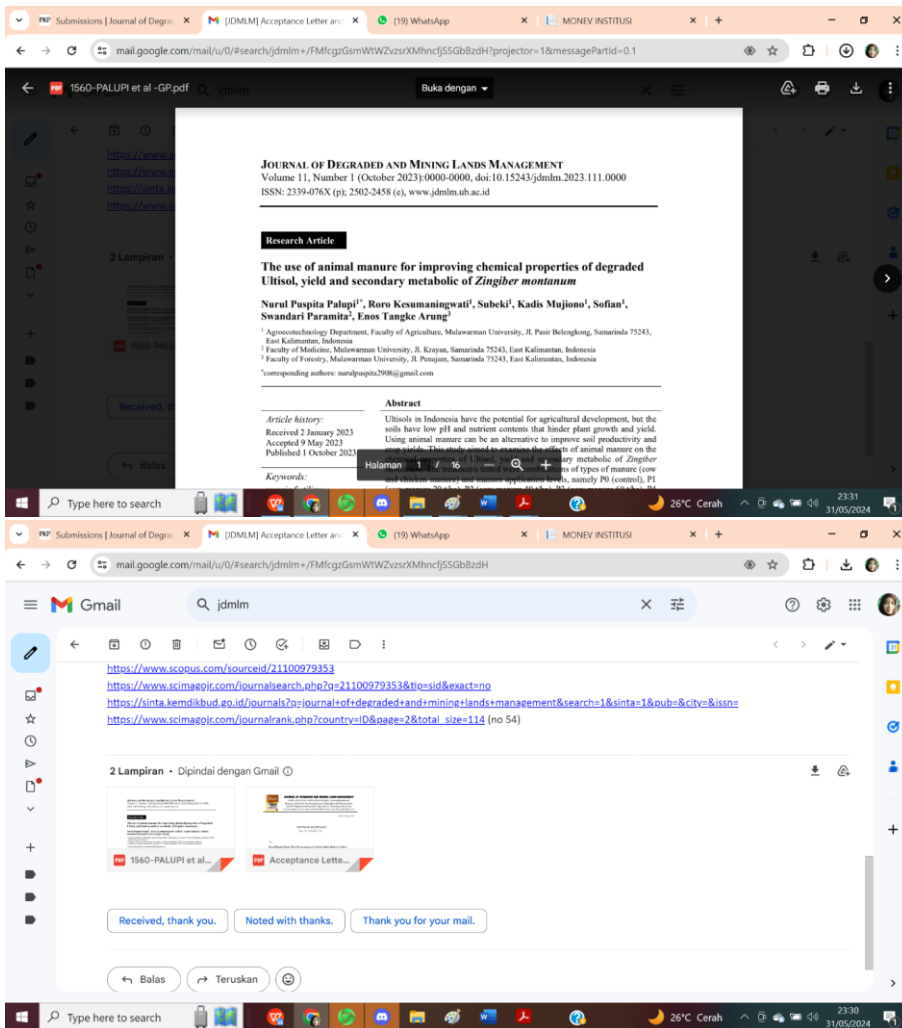
Manure Treatment (t/ha)	Root length (cm)	Rhizome wet weight (g)	Rhizome dry weight (g)
Cow 20 t/ha	24.36 a	444.75 ab	86.00 a
Cow 40 t/ha	35.36 b	530.00 bc	119.00 ab
Cow 60 t/ha	36.50 b	422.00 ab	179.75 c
Chicken 20 t/ha	33.20 ab	393.25 a	78.25 a
Chicken 40 t/ha	38.95 b	487.50 abc	99.75 ab

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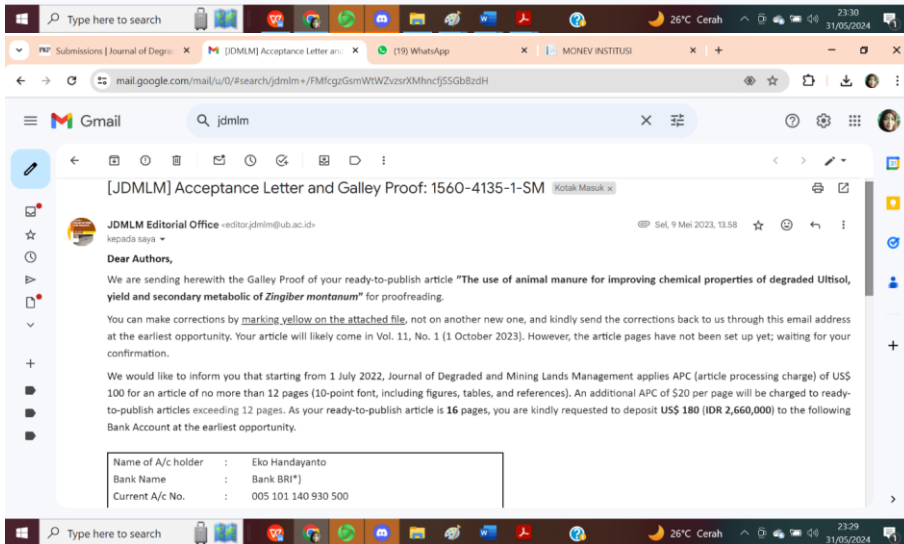
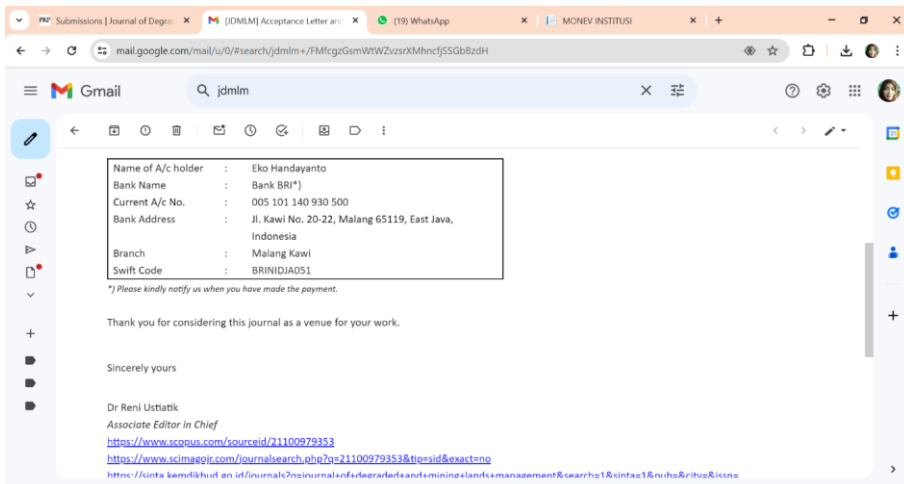


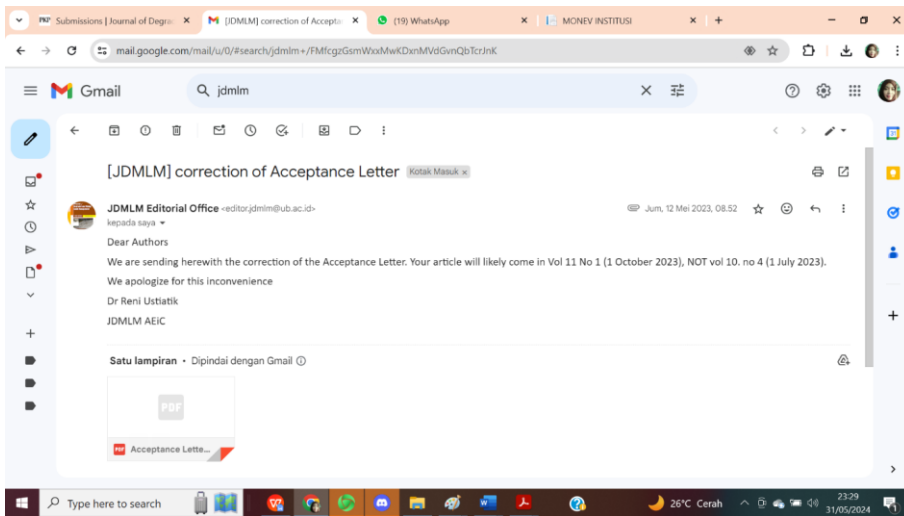
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Ultisols in Indonesia have the potential for agricultural development, but the soils have low pH and nutrient contents that hinder plant growth and yield. Using animal manure can be an alternative to improve soil productivity and crop yields. This study aimed to examine the effects of animal manure on the chemical properties of Ultisol, yield and secondary metabolic of *Zingiber montanum*. The treatments tested were combinations of types of manure (cow and chicken manure) and manure application levels, namely P0 (control), P1 (cow manure 20 t/ha), P2 (cow manure 40 t/ha), P3 (cow manure 60 t/ha), P4 (chicken manure 20 t/ha), P5 (chicken manure 40 t/ha), and P6 (chicken manure 60 t/ha). The results showed that the application of chicken manure of 60 t/ha increased N and P contents of the soil, and the application of cow manure of 60 t/ha increased soil cation exchange capacity. The application of cow manure of 60 t/ha gave the highest plant height, the number of leaves, and the number of at 18 weeks after planting, while the application of chicken manure dose of 60 t/ha produced the longest plant roots. The highest fresh and dry rhizome weight was observed for the 60 t/ha cow manure treatment. The highest secondary metabolic levels in each parameter were found in dry rhizomes (phenolic, flavonoid, and tannin) and fresh rhizomes (phenolic and flavonoid), with the highest tannin compound in the treatment of 40 t chicken manure/ha. Applying chicken manure at a dose of 60 t/ha resulted in a very strong antioxidant yield in fresh and dry rhizome.

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**Introduction**  
*Zingiber montanum* Roxb. (WFO, 2021) is commonly known as "Banada" in Bangladesh, "Phlai" in Thailand, "Angladrak" in India, and "Bangle" in Malaysia and Indonesia, is extensively planted in Thailand, Malaysia, and Indonesia (Hassan et al., 2019). This plant is commonly used in traditional medicines to treat constipation, dyspepsia, gastritis, stomach bloating and stomach ache. Various parts of *Z. montanum* are used in Thailand as a daily diet (Lim, 2016), while the rhizome is used as a vermifuge in Malaysia and applied for abscesses, colic, diarrhea, fever and intestinal disorders. In Northeast India, rhizome paste was reported to be used to treat dyspepsia and stomach bloating (Anasamy, 2013). In East Kalimantan, Indonesia, the productivity of this plant, commonly cultivated in the area dominated by the Ultisol soil order, is still low due to the low fertility of the soil.

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**Introduction**  
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Ultisol contains expandable 2:1 clay minerals that contribute to high cation exchange capacity (CEC) and the dominance of Al<sup>3+</sup> in exchangeable sites, resulting in low pH. It is well-known that soil acidity can inhibit nitrification (Shibata et al., 2017). A August Ultisol is found in East Kalimantan, Indonesia. September 2021 to March 2022. The second place was in the Soil Laboratory and Laboratory of Post-Harvest and Packaging of Agricultural Products, Faculty of Agriculture, Mulawarman University. The chemical compositions of the soil, cow manure, and chicken manure used in this study are presented in

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Ulnisol contains expandable 2:1 clay minerals that contribute to high cation exchange capacity (CEC) and the dominance of Al<sup>3+</sup> in exchangeable sites, resulting in low pH. It is well-known that soil acidity can inhibit nitrification (Shabata et al., 2017). A distinct Ulnisol is found in East Kalimantan with extremely high aluminum saturation, which can cause severe toxicity to the plant. Al<sup>3+</sup> enhances the desorption and leaching of nutrient cations from the soil exchange complex, hampering their absorption in the plant root area (Singh et al., 2017; Jaiswal et al., 2018; Zaso and Shen, 2018). Low pH exacerbates that effect by increasing the micronutrients/trace elements availability (e.g., Fe, Mn, Cu, and Zn), which is potentially toxic for plants. This condition and other soil properties may lead to nutrient deficiency, resulting in limited plant growth and development, including poor root systems, weak stalks or stems, and declining plant productivity (Bojorquez-Quintal et al., 2017; Moore et al., 2020). Besides nutrient deficiencies, the upland Ulnisol soil contains low organic matter, high soil bulk density, low total pores space, soil permeability and available water.

Liming and fertilization are the common solutions to this problem (Purwanto et al., 2020). However, most farmers cannot afford to buy lime and fertilizer to improve soil fertility to increase the productivity of the *Z. montanum* they cultivate. Alternatively, farmers use animal manure as a source of organic matter to improve soil fertility and production of *Z. montanum*. Several research workers demonstrated the positive effects of animal manure on the physical, chemical, and biological properties

September 2021 to March 2022. The second place was in the Soil Laboratory and Laboratory of Post-Harvest and Packaging of Agricultural Products, Faculty of Agriculture, Mulawarman University. The chemical compositions of the soil, cow manure, and chicken manure used in this study are presented in Table 1.

**Research design**

The dosages of manure applied were P<sub>0</sub> (control; without fertilizer), P<sub>1</sub> (cow manure 20 t/ha), P<sub>2</sub> (cow manure 40 t/ha), P<sub>3</sub> (cow manure 60 t/ha), P<sub>4</sub> (chicken manure 20 t/ha), P<sub>5</sub> (chicken manure 40 t/ha), and P<sub>6</sub> (chicken manure 60 t/ha). The seven treatments were arranged in a completely randomized block design with three replications with one treatment factor using organic fertilizers (cow manure and chicken manure).

Table 1. Chemical composition of the soil, cow manure, and chicken manure used in this study.

Chemical Properties	Soil	Cow Manure	Chicken Manure
pH	5.62	6.50	6.40
Organic C (%)	0.18	29.55	40.51
N (%)	0.14	2.68	1.37
C/N	1.29	14.21	29.57
Available P (ppm)	10.52	0.17	0.67
CEC (me/100 g)	14.33	-	-
Water (%)	-	72.90	60.63

The manure was applied evenly according to the

However, most farmers cannot afford to buy lime and fertilizer to improve soil fertility to increase the productivity of the *Z. montanum* they cultivate. Alternatively, farmers use animal manure as a source of organic matter to improve soil fertility and production of *Z. montanum*. Several research workers demonstrated the positive effects of animal manure on the physical, chemical, and biological properties of Ulnisol, particularly in alleviating soil acidity and Al toxicity, as well as a valuable source of essential macro and micronutrients (Zhou et al., 2013; Ngo et al., 2014; Ch'ng et al., 2015; Peng et al., 2016; Shi et al., 2019; Ye et al., 2019). Malhotra et al. (2018) and Masnoudi et al. (2020) reported that manure increases plant productivity, soil organic matter and structure, water infiltration and holding capacity, and over time, the application of manure can reduce sediment loss and soil erosion, which has advantageous effects on plant growth and development. Therefore, manure application is necessary to reduce the problems of N, P, and K deficiency for plants and poor fertility in acid upland soils (Castellanos-Navarrete et al., 2015).

This study aimed to examine the impact of animal manure on the fertility of a degraded Ulnisol of East Kalimantan on the growth and secondary metabolite content of *Z. montanum*.

**Materials and Methods**

**Place and time**

The experiment was conducted at the Experimental Farm of Mulawarman University Teknik Dairi, East Kalimantan, lasting about five months from

N (%) 0.14 2.08 1.37  
 C/N 1.29 14.21 29.57  
 Available P (ppm) 10.52 0.17 0.67  
 CEC (me/100 g) 14.33 - -  
 Water (%) - 72.90 60.63

The manure was applied evenly according to the treatment and then mixed with soil at 15-20 cm depth using hand hoes. The plot had a length of 6 m and a width of 1 m. Planting one crop per planting hole resulted in a spacing of approximately 50 x 100 cm. To prevent waterlogging and seedling rot, seedlings were planted in ditches with good drainage. In order to facilitate landfiling later, planting was done in the trench. A non-factorial completely randomized design is as follows:

$$Y_{ij} = \mu + \tau_i + \epsilon_{ij}$$

where:

- $Y_{ij}$  = The observed value in the  $i$ -th treatment and  $j$ -th repetition
- $\mu$  = General mean value
- $\tau_i$  = Effect of  $i$ -th treatment
- $\epsilon_{ij}$  = Effect of error (error) in the  $i$ -th treatment experiment and  $j$ -th repetition

The parameters measured in this study were chemical properties of soil, cow manure and chicken manure, i.e., pH using the electrometric method, organic C using the Walkley and Black method, total N using the Kjeldahl method, available P using the Bray II method, and cation exchange capacity (CEC) using the leaching method with 1N ammonium acetate at

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pH 7. Plant height, number of leaves, and number of tillers were measured at 3, 6, 9, 12, 15, and 18 WAP (weeks after planting). At harvest, fresh weight, dry weight, root length, secondary metabolic, and antioxidant levels of the plants were measured.

**Laboratory measurement**

Soil analyses were carried out in the Soil Science Laboratory, Faculty of Agriculture, Mulawarman University. The initial examination of soil reaction was determined using soil to water mixture of 1:1. Organic carbon was extracted using the Walkley and Black method. Total N was extracted using the Kjeldahl method. Exchangeable aluminum was extracted using KCl 1 N. Base cations and CEC were extracted using an ammonium acetate (NH<sub>4</sub>OAc) solution pH 7.0. Clay to CEC ratio (CCR) was calculated by dividing CEC by clay percentage. Since the observed soil pH was acidic, the ECEC was applied as the CEC reference for calculating base saturation.

**Data analysis**

Soil data were analyzed descriptively, while the crop

as well as supporting the biological and chemical properties of the soil (Melasail et al., 2019). Therefore, fertilization aims to replenish lost nutrients and increase the amount of nutrients available to plants, thereby increasing plant quality and quantity by gradually releasing nutrients into the soil solution and maintaining nutrient balance for the healthy growth of crop plants. They also act as an effective energy source for soil microbes, improving soil structure and crop growth (Shaji et al., 2021).

As a function of soil chemistry, organic manure can increase soil CEC that is important to hold a given inorganic fertilizers and soil buffering capacity so that the crops can avoid the negative effect of soil acidity (Shi et al., 2019). The use of animal manures increased the availability of some nutrients and improved the efficiency of P absorption by crops because in the process of organic matters decomposition produces humic acid and fulvic acid (polyelectrolyte) that can bind Al and Fe in the soil (Al-Jubury et al., 2021). To eliminate P fixation in the soil, the active anion of organic manure forms a chelate bond with Si-Al-ODPCR (allophane). The higher the carboxyl and phenolic compounds in

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a 5% level to detect significant differences among treatments. The results of the secondary metabolic level identification and antioxidant activity tests were analyzed qualitatively using the descriptive method. The comparison of phytochemical compound levels in each treatment was based on secondary metabolism levels. Antioxidant activity was assessed by comparing the IC<sub>50</sub> values in each treatment. A spectrophotometry method was used for all of the parameters of the secondary metabolic level test.

**Results**

**Manure quality**

The results of manure analysis (Table 1) showed that the organic C content was relatively high (29.55%) for cow manure and 40.51% for chicken manure, and the water contents were characterized at a moderate level of 72.29% and 60.63%, respectively. The total N contents were 2.08% for cow manure and 1.37% for chicken manure, with C/N ratios of 14.21 and 29.57, respectively. The manure contained some micro elements required by the plant. Therefore, the quality of manure used in this study was high. Manure can improve soil chemical and physical properties effectively, and plant growth is affected by the maturity of manure (Cai et al., 2019). Animal manure contains nitrogen as well as other minerals like magnesium, potassium, and calcium. The primary benefit of manure is that it preserves the physical structure of the soil, allowing roots to grow properly.

total N and N mineralization, soluble P, exchangeable K, N uptake by plants and soil water content can increase (Kafel et al., 2023).

**Soil chemical properties**

The effect of manure on the soil chemical properties presented in Table 2 showed that the initial soil pH (before planting) was 5.62 and increased to 4.21 after manure applications. A significant decrease in soil acidity was obtained due to the addition of chicken manure at 60 t/ha. The addition of manure reduced soil organic C at all levels of addition of manure. A significant decrease in organic C occurred with the addition of chicken manure at 20 t/ha (Table 3). In soil ecosystems, C-organic is a critical component that influences soil properties to support plant growth, namely as a source of energy for soil organisms and a trigger for nutrient availability for plants (Qiu et al., 2018).

Application of all levels of manure increased the N content in the soil (Table 4). The highest increase in N content in the soil was obtained due to the addition of chicken manure at 60 t/ha. The C/N ratio of organic matter is the ratio between the elemental carbon (C) and elemental nitrogen (N) presented in an organic matter. Good manure must have a C/N ratio of <20 (Macias-Corral et al., 2019). This study showed that the C/N ranged from 2.00 to 11.91 and decreased to 2.05 after 18 weeks of incubation (Table 5). Manure input at all application levels increased the P content in the soil along with

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the incubation time. The highest increase in P in the soil was obtained due to the addition of chicken manure at 60 t/ha (Table 6). The results of this study indicated that the addition of all levels of manure provided an increase in cation exchange capacity. The highest increase in cation exchange capacity was obtained due to the addition of cow manure at 60 t/ha, as shown in Table 7.

Table 2. Soil pH after manure treatment.

Manure Treatments	pH						
	Week of Incubation						
	0	3	6	9	12	15	18
Control	5.62	5.62	5.62	5.61	5.62	5.62	5.62
Cow manure 20 t/ha	6.40	6.20	6.00	5.80	5.70	5.70	5.60
Cow manure 40 t/ha	6.50	6.40	6.30	5.90	5.70	5.50	5.40
Cow manure 60 t/ha	6.80	6.60	6.40	6.60	6.20	6.00	5.80
Chicken manure 20 t/ha	5.83	5.60	5.40	5.00	4.83	4.70	4.51
Chicken manure 40 t/ha	6.04	6.00	5.80	5.04	4.74	4.90	4.80
Chicken manure 60 t/ha	6.08	5.43	5.08	4.76	4.77	4.08	4.21

Table 3. Soil organic C after manure treatment.

Manure Treatments	Organic C (%)						
	Week of Incubation						
	0	3	6	9	12	15	18
Control	0.18	0.18	0.18	0.18	0.18	0.18	0.17
Cow manure 20 t/ha	0.83	0.80	0.78	0.64	0.56	0.46	0.40
Cow manure 40 t/ha	1.31	1.27	1.20	1.19	1.08	1.07	1.01
Cow manure 60 t/ha	1.40	1.37	1.35	1.32	1.31	1.31	1.30
Chicken manure 20 t/ha	0.36	0.33	0.32	0.30	0.27	0.21	0.18
Chicken manure 40 t/ha	0.42	0.42	0.41	0.38	0.35	0.31	0.26
Chicken manure 60 t/ha	0.98	0.98	0.95	0.92	0.87	0.80	0.78

Table 4. Soil N after manure treatment.

Manure Treatments	N (%)						
	Week of Incubation						
	0	3	6	9	12	15	18
Control	0.14	0.14	0.14	0.14	0.14	0.15	0.14
Cow manure 20 t/ha	0.08	0.10	0.15	0.16	0.18	0.19	0.19
Cow manure 40 t/ha	0.11	0.11	0.12	0.17	0.19	0.19	0.21
Cow manure 60 t/ha	0.13	0.15	0.16	0.16	0.17	0.19	0.23
Chicken manure 20 t/ha	0.18	0.18	0.19	0.19	0.20	0.21	0.21
Chicken manure 40 t/ha	0.20	0.21	0.22	0.30	0.32	0.33	0.35
Chicken manure 60 t/ha	0.20	0.22	0.32	0.35	0.36	0.37	0.38

Table 4. Soil N after manure treatment.

Manure Treatments	N (%)						
	Week of Incubation						
	0	3	6	9	12	15	18
Control	0.14	0.14	0.14	0.14	0.14	0.15	0.14
Cow manure 20 t/ha	0.08	0.10	0.15	0.16	0.18	0.19	0.19
Cow manure 40 t/ha	0.11	0.11	0.12	0.17	0.19	0.19	0.21
Cow manure 60 t/ha	0.13	0.15	0.16	0.16	0.17	0.19	0.23
Chicken manure 20 t/ha	0.18	0.18	0.19	0.19	0.20	0.21	0.21
Chicken manure 40 t/ha	0.20	0.21	0.22	0.30	0.32	0.33	0.35
Chicken manure 60 t/ha	0.20	0.22	0.32	0.35	0.36	0.37	0.38

Table 5. Soil C/N ratio after manure treatment.

Manure Treatments	C/N ratio						
	Week of Incubation						
	0	3	6	9	12	15	18
Control	1.29	1.29	1.29	1.29	1.29	1.20	1.21
Cow manure 20 t/ha	10.38	8.00	5.20	4.00	3.11	2.42	2.11
Cow manure 40 t/ha	11.91	11.55	10.00	7.00	5.68	5.63	4.81
Cow manure 60 t/ha	10.77	9.13	8.44	8.25	7.71	6.89	5.65
Chicken manure 20 t/ha	2.00	1.83	1.68	1.58	1.35	1.00	0.86
Chicken manure 40 t/ha	2.10	2.00	1.86	1.27	1.09	0.94	0.74
Chicken manure 60 t/ha	4.90	4.45	2.97	2.63	2.42	2.16	2.05

Cation exchange in the soil occurs due to a negative charge from the soil colloid, which adsorbs cations in an exchangeable form (Mecetei et al., 2020). Soils with high CEC can absorb and provide nutrients

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better than soils with low CEC. Because these elements are in the soil adsorption complex, water does not quickly lose or wash away these nutrients. Mineralization of soil organic fractions provides limited supplies of N, S, and micronutrients; during mineral dissolution and surface exchange reactions, re-supply P, K, Ca, Mg, and micronutrients. Nutrient mobility in soil influences ion transport to plant roots, evaluation of nutrient availability to plants, and ultimately nutrient management decisions (Havlin, 2020). The nutrient added to the soil with low CEC can not be held and is easily lost. This condition was reflected in the increasing soil organic C contents in the treatment with manure application. It did not increase significantly in the treatment without manure application, and vice versa. This study showed that the treatments of giving organic fertilizers in the form of chicken and cow manure gave significant differences in plant height, number of tillers, number of leaves, root length, fresh weight of rhizomes, and dry weight of rhizomes. Only root length was not affected significantly by the treatments. For growing zingiber, N, P, and K play as essential nutrients.

**Table 6. Soil available P after manure treatment.**

Manure Treatments	P (ppm)							
	Week of incubation							
	0	3	6	9	12	15	18	
Control	10.52	10.52	10.52	10.52	10.51	10.52	10.52	
Cow manure 20 t/ha	0.35	0.36	0.36	0.37	0.37	0.38	0.38	
Cow manure 40 t/ha	0.40	0.40	0.42	0.43	0.44	0.45	0.45	
Cow manure 60 t/ha	0.44	0.44	0.45	0.47	0.48	0.48	0.49	
Chicken manure 20 t/ha	12.78	12.78	12.8	12.81	12.90	12.92	12.93	
Chicken manure 40 t/ha	13.42	13.44	13.47	13.48	13.49	13.49	13.5	
Chicken manure 60 t/ha	15.68	15.68	15.68	15.69	15.70	15.72	15.73	

**Table 7. Soil CEC after manure treatment.**

Manure Treatments	CEC (me/100g)							
	Week of incubation							
	0	3	6	9	12	15	18	
Control	14.33	14.33	14.32	14.33	14.34	14.33	14.33	
Cow manure 20 t/ha	19.15	19.15	19.17	19.17	19.18	19.18	19.19	
Cow manure 40 t/ha	19.15	19.15	19.16	19.17	19.17	19.18	19.19	
Cow manure 60 t/ha	19.15	19.16	19.16	19.17	19.17	19.18	19.18	

**Table 8. Plant height**

Table 8 shows that when the plant was about six weeks old, the application of cow manure at a dose of 60 t/ha produced the best plant growth (26.16 cm), followed by chicken manure at a dose of 60 t/ha, chicken manure of 40 t/ha, and cow manure of 40 t/ha. Compared to the previous week, the plant age of nine weeks after planting showed a rapid increase in plant height. At nine weeks after planting, the best dose was 40 t/ha of chicken manure with an average plant height addition of about 32.45 cm. The best results were obtained at week 12, with cow manure at a dose of 60 t/ha that yielded an average plant height of 37.24 cm. The application of cow manure at a dose of 20 t/ha and chicken manure at a dose of 20 t/ha did not produce significantly different plant heights from the control. The best dose at 15 weeks after planting was 60 t cow manure/ha, with an average plant height of 40.47 cm and 38.38 cm of chicken manure. The highest dose of cow manure yielded an average plant height of 42.78 cm at 18 weeks after planting.

**Table 9. Number of tillers**

Table 9 shows that the highest number of tillers was found at a dose of 60 t/ha of chicken manure, with an average of 1.65 tillers at 6 weeks after planting. At 9 weeks after planting, a control with an average of about 2.55 tillers produced tillers that were not significantly different from chicken manure of 20 t/ha. The best application of cow manure was 60 t/ha, which yielded an average number of 4.15 tillers. At 12 weeks after planting, the effect of manure treatment revealed that the best dose was 40 t/ha chicken manure, with an average of 6.65 tillers. This best dose did not differ significantly between chicken and cow manure at 60 t/ha or chicken manure at 20 t/ha. Cow and chicken manure at a dose of 60 t/ha at 15 weeks after planting produced the highest yields, with cow manure yielding 9.50 tillers and chicken yielding 9.00 tillers, respectively. At 18 weeks, the

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control did not differ significantly from the cow manure doses of 20 and 40 t/ha. The highest number of tillers was observed for the treatment of cow manure at 60 t/ha, with an average of 14.45 tillers, and chicken manure at 60 t/ha, with an average of 13.37 tillers.

**Table 8. Effect of cow and chicken manure on plant height (cm).**

Manure Treatments	Week After Planting (WAP)			
	6	9	12	18
Control	11.38 a	18.61 a	24.68 a	20.41 a
Cow manure 20 t/ha	17.46 b	20.26 ab	25.44 a	24.02 ab
Cow manure 40 t/ha	20.51 c	27.05 b	29.81 ab	35.57 cd
Cow manure 60 t/ha	26.16 e	31.18 b	37.24 c	40.47 d
Chicken manure 20 t/ha	17.36 b	27.21 b	30.16 abc	29.15 bc
Chicken manure 40 t/ha	22.84 cd	32.45 b	34.83 bc	37.61 cd
Chicken manure 60 t/ha	25.68 de	32.17 b	36.84 bc	38.38 d

Notes: Based on Duncan's Multiple Range Test at a 5% significance level, values in the same columns followed by the same letter do not differ significantly.

**Table 9. Effect of cow and chicken manure on the number of rhizome tillers.**

Manure Treatments	Week After Planting (WAP)			
	6	9	12	18
Control	0.80 a	2.55 a	3.97 a	5.80 a
Cow manure 20 t/ha	0.70 b	3.45 b	5.20 b	6.60 a
Cow manure 40 t/ha	1.70 ac	3.40 b	5.05 ab	7.35 ab
Cow manure 60 t/ha	1.60 c	4.15 b	6.10 bc	9.50 c
Chicken manure 20 t/ha	1.30 bc	3.15 ab	5.70 bc	7.40 ab
Chicken manure 40 t/ha	1.50 c	4.00 b	6.65 c	8.45 bc
Chicken manure 60 t/ha	1.65 c	3.75 b	6.05 bc	9.00 c

Notes: Based on Duncan's Multiple Range Test at a 5% significance level, values in the same columns followed by the same letter do not differ significantly.

*Number of leaves* cow and chicken manure of 20 and 40 t/ha. Cow manure of 60 t/ha with an average of 52.80 leaves.

Notes: Based on Duncan's Multiple Range Test at a 5% significance level, values in the same columns followed by the same letter do not differ significantly.

*Number of leaves* cow and chicken manure of 20 and 40 t/ha. Cow manure of 60 t/ha with an average of 52.80 leaves provided the best dose at 12 weeks after planting. The highest number of leaves (84.55) was found in cow manure treatment at a rate of 60 t/ha at week 15. The application of 60 t/ha cow manure produced the largest number of leaves, 116.65 in the 18<sup>th</sup> week. The application of chicken manure of 60 t/ha increased the number of leaves by 105.80.

**Table 10. Effect of cow and chicken manure on the number of rhizome leaves.**

Manure Treatments	Week After Planting (WAP)			
	6	9	12	18
Control	4.70 a	19.60 a	35.80 a	52.25 a
Cow manure 20 t/ha	5.55 a	25.00 ab	41.05 ab	55.65 a
Cow manure 40 t/ha	9.65 b	28.25 abc	42.70 abc	66.00 ab
Cow manure 60 t/ha	10.05 b	37.95 c	52.80 c	84.55 c
Chicken manure 20 t/ha	9.20 b	26.00 ab	40.25 ab	56.85 a
Chicken manure 40 t/ha	9.65 b	30.90 bc	45.25 abc	67.05 bc
Chicken manure 60 t/ha	11.05 b	30.45 abc	50.75 bc	79.55 bc

Notes: Based on Duncan's Multiple Range Test at a 5% significance level, values in the same columns followed by the same letter do not differ significantly.

**Rhizome root length, fresh weight and dry weight**

Table 11 shows that the lowest root length of 24.36 cm was obtained by the application of 20 t/ha cow manure. The control yielded root length that was not statistically different from the application of 20 t/ha cow and chicken manure. This result indicates that the application of different doses of manure did not

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affect the root length of the bangle plant. The chicken manure dosage of 20 t/ha yielded the lowest rhizome fresh weight of 392.35 g. In comparison, the control yielded fresh weight that was not significantly different from 20 t/ha and 40 t/ha cow and chicken manure. The fresh weight of the bangle rhizome yielded from the application of 40 t/ha chicken and cow manure also did not differ significantly from those yielded from the application of 60 t/ha chicken manure. The treatment of 60 t/ha cow manure yielded the highest rhizome fresh weight of 822.00 g, which was significant for all treatments.

The lowest dry weight of bangle rhizome was shown by the cow and chicken manure treatment at a dose of 20 t/ha, respectively 86.00 g for cow manure treatment and 78.25 g for chicken manure treatment. These values were lower than that of the control, with an average of 93.75 g. The control yielded insignificantly different rhizome dry weight with chicken manure at 20 and 40 t/ha, cow manure at 20 and 40 t/ha, and chicken manure at 60 t/ha but yielded significantly different rhizome dry weight with the best dose of 60 t/ha cow manure with an average rhizome dry weight of 179.75 g, followed by

60 t/ha chicken manure t/ha with an average rhizome dry weight of 135.75 g.

**Secondary metabolic level**

The results of the secondary metabolism test showed that the bangle rhizome contained phenolic, flavonoid, and tannin-containing active substances, and also steroids, alkaloids, and terpenoids. Different levels of the compounds in this secondary metabolism presented in Table 12 show that, despite using the same amount of fertilizer, the active compound content of the bangle rhizomes was highest when the rhizome was dry (low moisture content) as opposed to when the rhizome was still fresh. The amount of chicken manure that produced the highest phenolic content in the rhizome under fresh and dry conditions, correspondingly around 178.56 mg/L and 202.79 mg/L, was 60 t/ha. A dose of 60 t/ha of chicken manure also yielded higher levels of active flavonoids and tannins than other doses. In comparison with the fertilizer treatment, the control had the lowest active compound content. The plant yields of the cow manure treatments were lower than those of the chicken manure treatments.

Table 11. Effect of cow and chicken manure on root length and rhizome weight.

Manure Treatments	Root length (cm)	Rhizome fresh weight (g)	Rhizome dry weight (g)
Control	31.90 ab	415.25 ab	93.75 ab
Cow manure 20 t/ha	24.36 a	444.75 ab	86.00 a
Cow manure 40 t/ha	35.36 b	530.00 bc	119.00 ab
Cow manure 60 t/ha	36.50 b	822.00 d	179.75 c
Chicken manure 20 t/ha	33.20 ab	393.25 a	78.25 a
Chicken manure 40 t/ha	38.95 b	487.50 abc	99.75 ab

Manure Treatments	Root length (cm)	Rhizome fresh weight (g)	Rhizome dry weight (g)
Control	31.90 ab	415.25 ab	93.75 ab
Cow manure 20 t/ha	24.36 a	444.75 ab	86.00 a
Cow manure 40 t/ha	35.36 b	530.00 bc	119.00 ab
Cow manure 60 t/ha	36.50 b	822.00 d	179.75 c
Chicken manure 20 t/ha	33.20 ab	393.25 a	78.25 a
Chicken manure 40 t/ha	38.95 b	487.50 abc	99.75 ab
Chicken manure 60 t/ha	41.03 b	618.75 c	134.75 bc

Notes: Based on Duncan's Multiple Range Test at a 5% significance level, values in the same columns followed by the same letter do not differ significantly.

Table 12. Secondary metabolic levels in bangle rhizome in each treatment (mg/L).

Condition	Manure Treatments	Level mg/L		
		Phenolic	Flavonoid	Tannin
Fresh	Control	31.64	23.08	2,034.83
	Cow manure 20 t/ha	43.18	41.55	2,991.50
	Cow manure 40 t/ha	77.03	64.30	3,151.50
	Cow manure 60 t/ha	86.90	71.14	3,198.17
	Chicken manure 20 t/ha	97.67	74.79	4,066.50
	Chicken manure 40 t/ha	107.54	78.71	4,144.83
Chicken manure 60 t/ha	178.56	104.39	3,861.33	
Dry	Control	124.97	38.44	1,982.33
	Cow manure 20 t/ha	133.56	67.23	2,504.00
	Cow manure 40 t/ha	148.44	68.76	3,034.83
	Cow manure 60 t/ha	181.38	99.43	4,734.67
	Chicken manure 20 t/ha	175.23	120.06	4,903.00
	Chicken manure 40 t/ha	198.09	156.10	5,088.00
Chicken manure 60 t/ha	202.79	181.91	5,406.33	

Note: The number followed by yellow denotes the best outcome in each observation variable for the flavonoid, tannin, and phenolic contents.

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### Antioxidant

Results of antioxidant analysis of bangle rhizomes based on fresh and dry rhizomes are shown in Table 13. IC<sub>50</sub> value obtained described how well the sample captured free radicals. The dose of chicken manure of 60 t/ha was found to give the lowest IC<sub>50</sub> value of 9.52 ppm in the fresh sample, while the control gave the highest IC<sub>50</sub> value of 53.58 ppm in the fresh rhizomes. The same quality was obtained for the dry rhizomes. The application of chicken manure of 60 t/ha produced the lowest IC<sub>50</sub> value, and the largest was in control. The results of an analysis of variance (ANOVA) at a 5% level showed that the treatments of giving organic fertilizers in the form of chicken and cow manure gave significant differences in plant height, number of tillers, number of leaves, root length, fresh weight of rhizomes, and dry weight of rhizomes. The treatments did not significantly affect root length.

**Table 13. Antioxidant based on IC<sub>50</sub> value.**

Antioxidant (IC <sub>50</sub> ) (mg/L)	Manure Treatments (t/ha)						
	Control	Cow Manure			Chicken Manure		
		20	40	60	20	40	60
Fresh rhizomes	53.58	48.55	47.36	47.14	32.73	27.42	9.52
Dry rhizomes	40.91	38.30	34.78	34.46	24.41	14.05	8.06

Note: The smaller the IC<sub>50</sub> value, the stronger the antioxidant.

The increment in incubation time seemingly exhibited an inconsistent effect on the soil. Adedkiya et al. (2020) reported that utilizing organic manure to meet crop nutrient requirements would be an unavoidable practice to enhance sustainable agriculture. This is

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Note: The smaller the IC<sub>50</sub> value, the stronger the antioxidant.

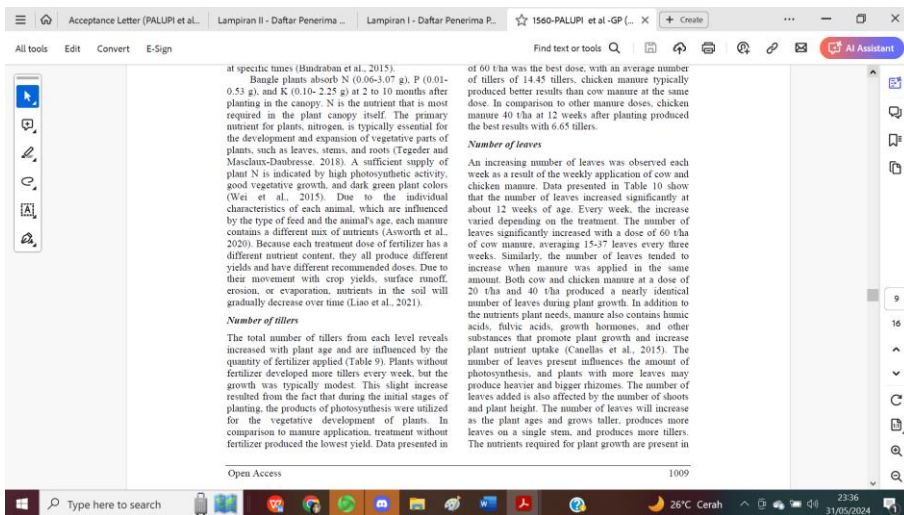
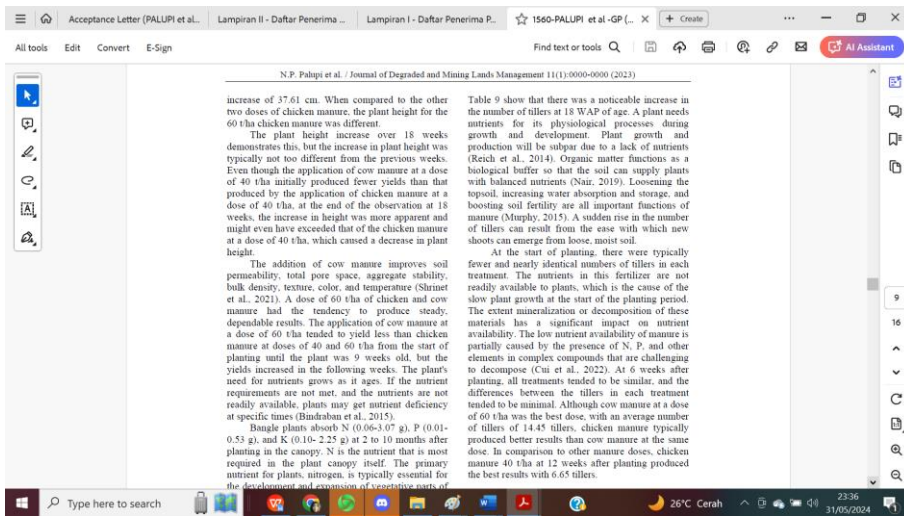
The increment in incubation time seemingly exhibited an inconsistent effect on the soil. Adedkiya et al. (2020) reported that utilizing organic manure to meet crop nutrient requirements would be an unavoidable practice to enhance sustainable agriculture. This is because the physical, chemical and biological properties of soil are generally improved by the addition of organic manure, which in turn enhances crop productivity and maintains the quality of crop production. Poultry litter treatments were positively correlated with greater soil fertility levels, as well as higher crop yield and soil biodiversity. These results underscore linkages between manure additions and cropping sequences within the nutrient cycling, soil health, and crop production continuum (Asworth et al., 2016).

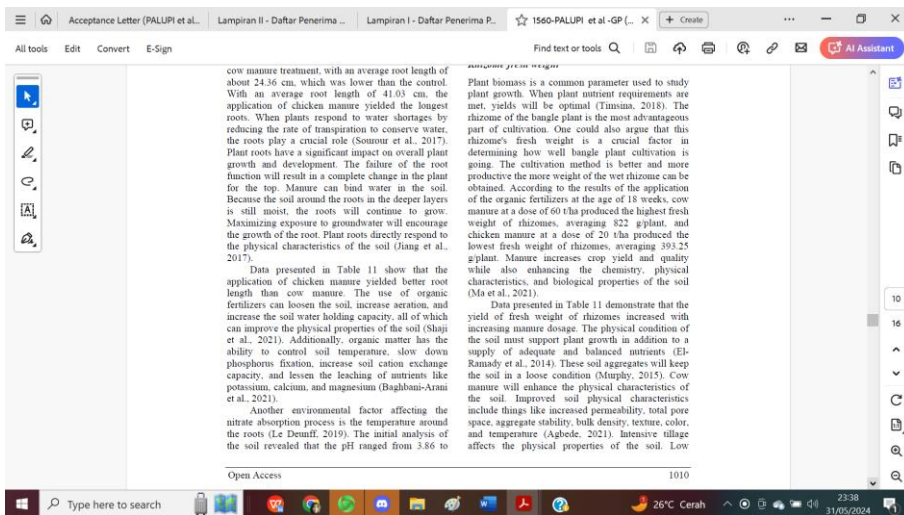
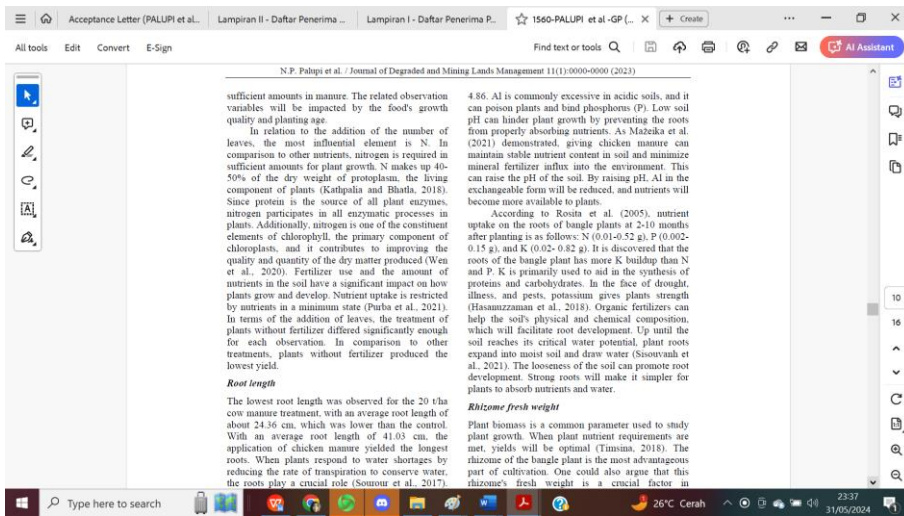
### Plant height

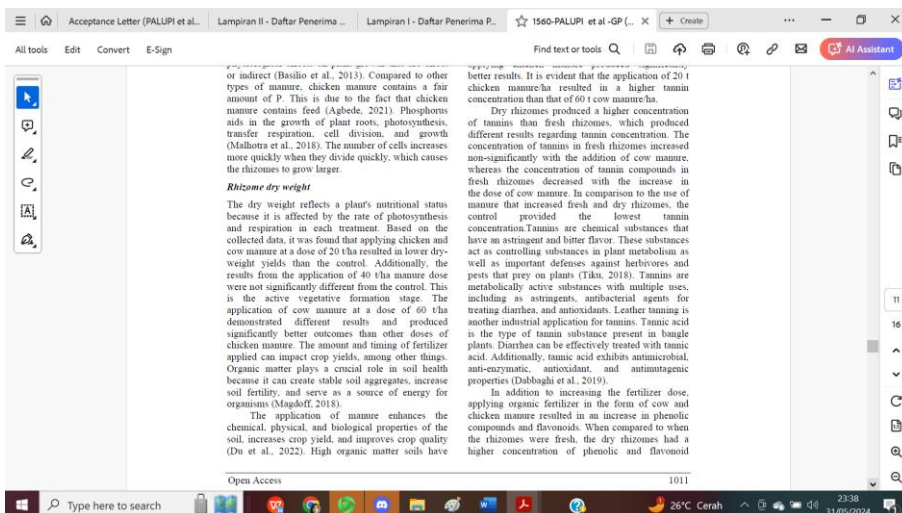
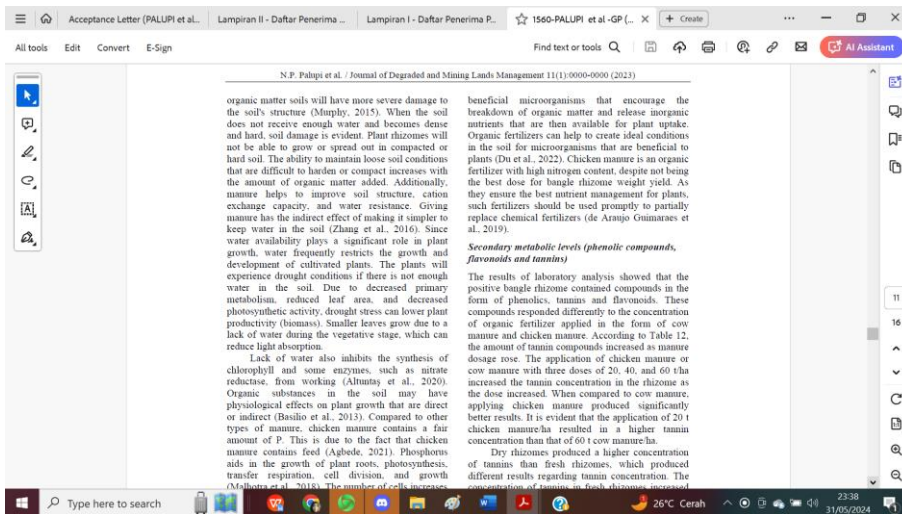
According to Table 8, the average increase in plant height starting at 6.9, 12.15, and 18 WAP tended to fluctuate. At the age of 6 weeks, the treatment without fertilizer (control) continued to grow until week 12 when it reached its peak with an average height increase of about 24.68 cm before the declining height in the following weeks. In order for a plant to grow, nutrients must be obtained from the soil by the roots through their root hairs (Wang et al., 2021). Organic matter affects plant growth by influencing the physical, chemical, and biological properties of the soil (Delgado and Gómez, 2016). The more organic matter is provided, the faster the plant will grow. Compared to chicken manure, cow manure typically produced better plant growth. The application of chicken manure at all levels significantly increased plant height at 9 weeks after planting, but at 12 weeks, the plant height slightly decreased. In the 20 t/ha chicken manure treatment, the plant height was an average of 30.16 cm at 12 weeks after planting and decreased in the following weeks. A high nitrogen content was found in chicken manure (0.38%). Although the amount of nitrogen required by plants is always higher than other nutrients, a deficiency or excess of nitrogen can hinder and disrupt plant growth (Jiang et al., 2017). After planting, the growth rate of the bangle plant accelerated between 2 and 5 months. As plants get older, their growth rate for height starts to slow down (Rademacher, 2015).

The application of chicken manure always resulted in the best plant response in the first growing season. This is because chicken manure decomposes relatively quickly and has enough nutrients compared to other manure of the same weight. Table 1 shows that the application of chicken manure tended to increase plant height rapidly at 6, 9, and 12 weeks of age, then declined as plant age increased. Large-scale application of chicken manure is thought to be less effective because the nutrients will exhaust quickly. The same result was also shown by the application of chicken manure at a dose of 40 t/ha, which decreased the plant height; the maximum height increase at this dose was at 15 weeks, with an average height

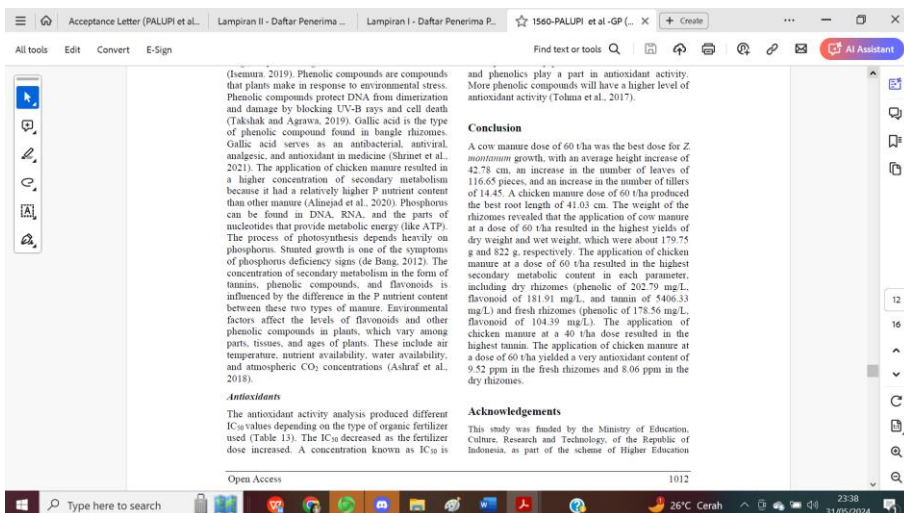
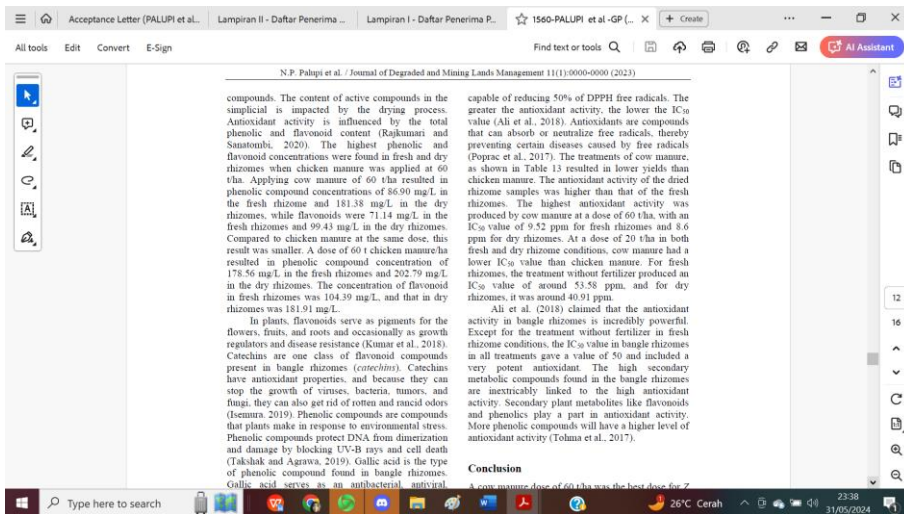
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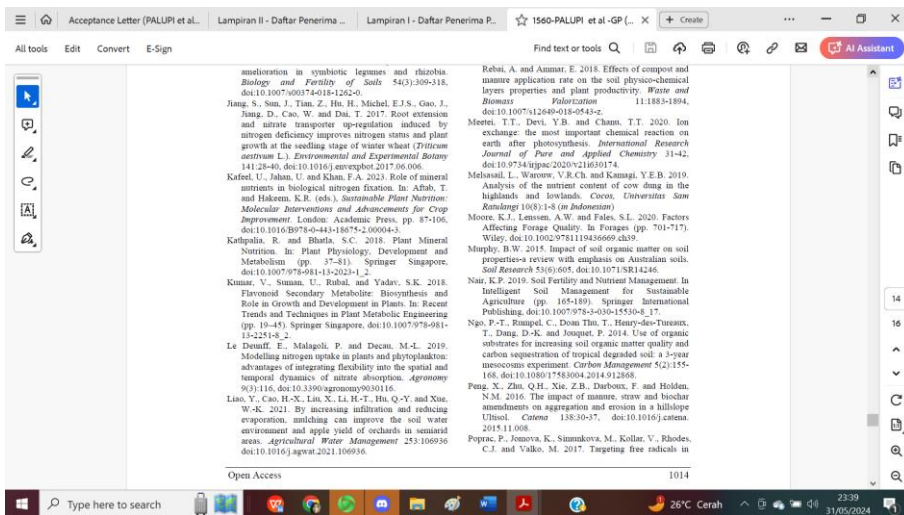
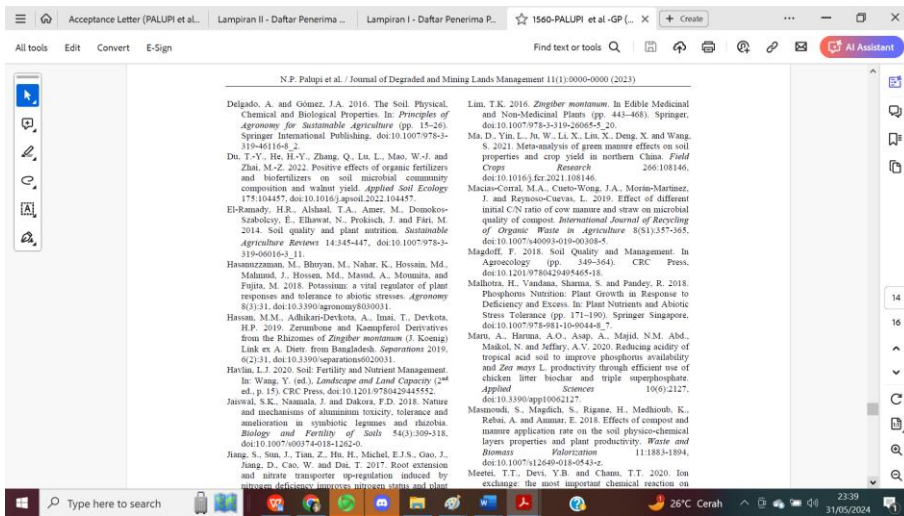
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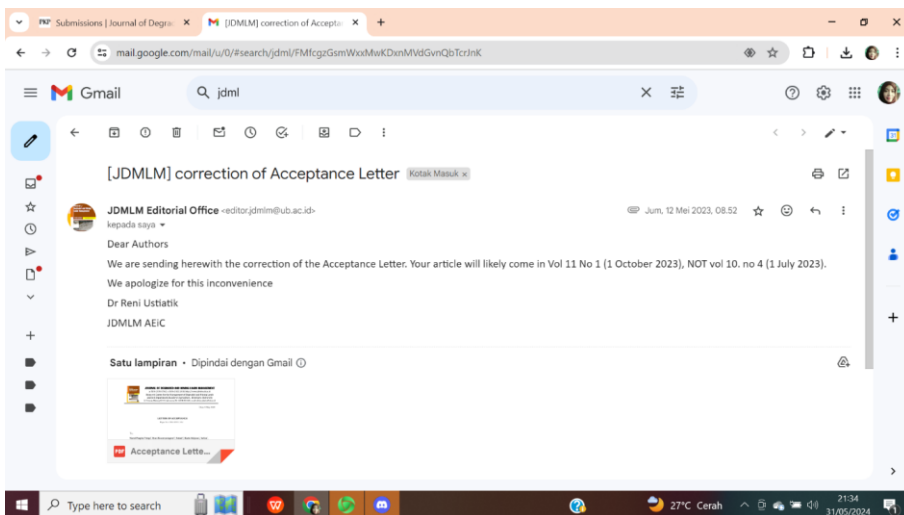
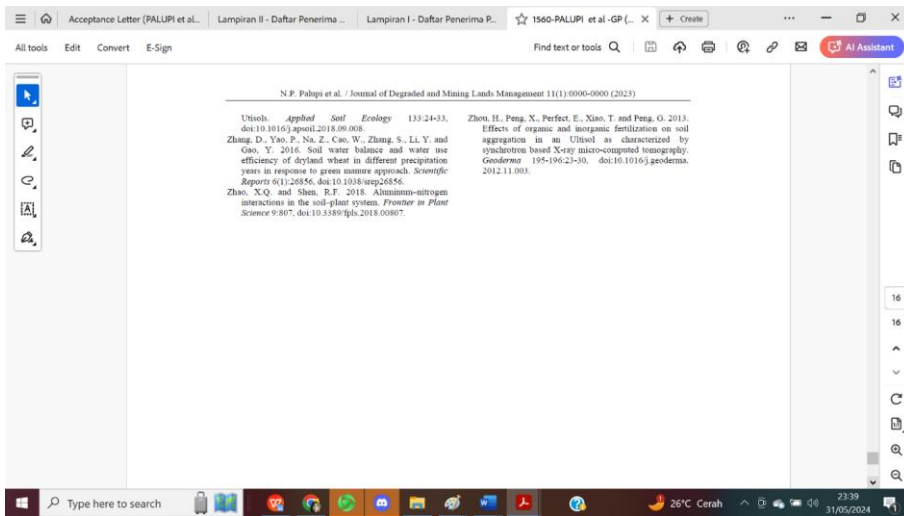
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**LETTER OF ACCEPTANCE**  
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To:  
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Dear Authors,

We are pleased to inform you that your article entitled "The use of animal manure for improving chemical properties of degraded Ultisol, yield and secondary metabolite of *Zingiber officinale*" has been accepted for publication in the Journal of Degraded and Mining Lands Management (p-ISSN: 2539-076X, e-ISSN: 2502-2458). The article will likely come in Vol. 11, No. 1 (1 October 2023).

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