The International Review of Multidisciplinary Research
Volume 1, Issue 4, April 2026, pp. 858-868
https://doi.org/10.5281/zenodo.20028161

The International Review of Multidisciplinary Research
Advancing knowledge through quality research

Efficacy of Tilapia (*Oreochromis Niloticus*) Viscera and Powdered Eggshell as Biofertilizer in the Growth of Rice (*Oryza Sativa*)

[1]Trisha Mae K. Alimagno, [2]Pollyn S. Coronado, [3]Lucky Anne J. Paran, [4]Asnar L. Aloro
Laguna State Polytechnic University
[4]asnar.aloro@lspu.edu.ph

Article Details:

Received: 7 March 2026
Revised: 9 April 2026
Accepted: 19 April 2026
Published: 30 April 2026
Corresponding Email:
asnar.aloro@lspu.edu.ph

Recommended Citation:

Alimagno, T. M. K., Coronado, P. S., Paran, L. A. J., Aloro, A. L. (2026). Efficacy of Tilapia (Oreochromis Niloticus) Viscera and Powdered Eggshell as Biofertilizer in the Growth of Rice (Oryza Sativa). The International Review of Multidisciplinary Research. 1 (4), 858-868.
https://doi.org/10.5281/zenodo.20028161

Index Terms:

biofertilizer, oreochromis niloticus, oryza sativa, powdered eggshell, tilapia viscera

Abstract. Rice (*Oryza sativa*) is a primary staple food in most Asian countries particularly in the Philippines. It plays an important role in the diet and health of an individual. With this, rice consumption is relatively high, amounting to 90% all over Asia. Therefore, the production of rice is necessary to deal with the continuously increasing demand for rice. Fertilizer, specifically chemical fertilizer is one of the necessities in the production of rice crops. However, the excessive use of chemical fertilizers has a severe impact on the health of the soil over some time. As a sustainable alternative, biofertilizers composed of fish derivatives and organic matter like eggshell offers a potential solution to this problem. This study investigates the effectiveness of a biofertilizer formulated from tilapia *(Oreochromis niloticus)* viscera and powdered eggshell on the growth of rice (*Oryza sativa*). The researchers examined its impact on key growth parameters, including plant height, root length, and the number of leaves. Furthermore, researchers formulated six various setups with different concentrations of tilapia *(Oreochromis niloticus)* viscera and powdered eggshell. Based on the findings, set-up treatment with a high concentration of eggshell and low fish amino acid concentration has high effectiveness as a biofertilizer among the other treatments. The results imply that the product has the potential to be an eco-friendly and cost-effective biofertilizer that can be utilized and be helpful to the community.

Introduction

Rice (*Oryza sativa*) is an important food, particularly in the Philippines, where it is commonly consumed with fish asserted by Tacio (2021). According to the Rice Research Institute (IRRI), rice is the principal food source for over 60% of the global population. With rising global demand for rice, sustainable agricultural practices are necessary to ensure food security while minimizing environmental impact. One such practice involves the use of biofertilizer that offers an eco-friendly alternative to chemical fertilizer. Aside from biofertilizer as a provider of essential nutrients, as stated by Kumar et al. (2022) it also assists in preserving the soil fertility.

Soil fertility must be taken into consideration as it is one of the major key factors for a successful rice cultivation. Especially here in the Philippines wherein majority of the farmers utilize chemical fertilizers that gradually decreases the natural fertility of the soil leading to lower rice production over time. According to a study conducted by Calubaquib et al. (2016) most soils in Luzon have already been degraded, characterized with low levels of nitrogen, phosphorus, potassium and pH levels due to excessive usage of chemical fertilizers, pesticides and plus the unpreventable environmental factors leading to rice production constraints**.** This issue may extend to other regions of the Philippines, posing a significant concern for sustainable rice production.

Furthermore, this study maximizes the potential utilization of both Tilapia (*Oreochromis niloticus*) viscera and eggshell waste as a biofertilizer that promotes rice (*Oryza sativa)* growth and development while also enhancing soil fertility. According to Jaies et al. (2024), typically fish wastes contain the NPK (nitrogen, phosphorus, and potassium) material essential for the crop's growth. The Tilapia (*Oreochromis niloticus)* viscera in particular demonstrates its value as a source of these essential nutrients, including nitrogen (N), phosphorus (P), and potassium (K) all of which are vital constituents for rice cultivation. This is supported by the result of the fish amino acid sample tested by the College of Agriculture and Food Science at the University of the Philippines, Los Baños, Laguna.

Additionally, the use of Tilapia (*Oreochromis niloticus*) viscera can also aid in fish waste management as it remains a challenge and has emerged as a significant environmental concern worldwide asserted by Lee et al. (2022). Similarly, as discussed by Abdulrahman et al. (2014), eggshell waste, which is rich in calcium carbonate, is discarded as agricultural waste, contributing to environmental pollution. By converting these wastes into valuable agricultural inputs such as biofertilizer, it promotes both agricultural productivity and waste management.

With that, this study supports Sustainable Development Goal 12 (SDG 12), which advocates sustainable consumption and production through the responsible management of natural resources and waste. This study aimed to develop a potential biofertilizer combining fish wastes, specifically fish amino acids and eggshell derivatives, to enhance rice growth, improve soil fertility, and contribute to the reduction of environmental waste pollution.

Conceptual Framework

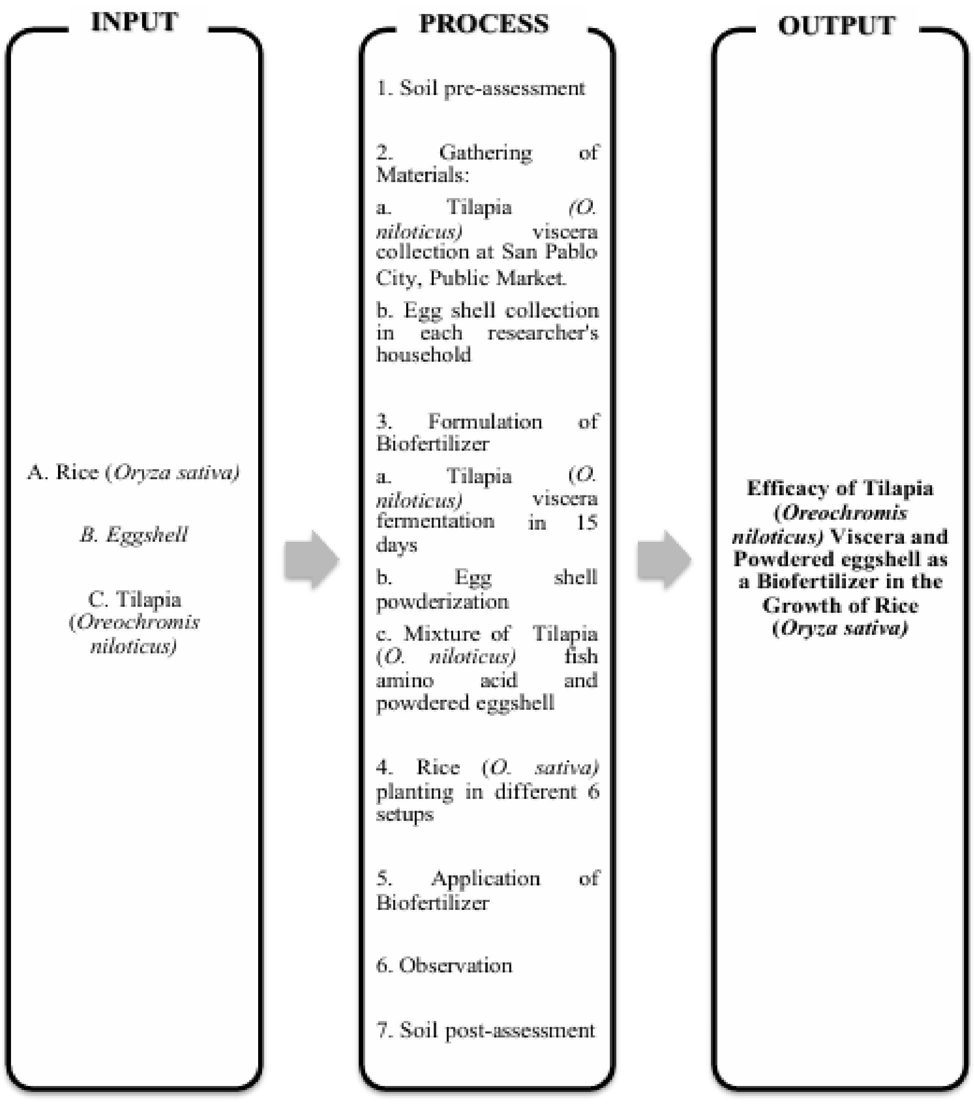

Figure 1. Research Paradigm

Figure 1 shows the research paradigm of the study, which consists of input, process, and output. The input includes rice (*Oryza sativa*), eggshells, and Tilapia (*Oreochromis niloticus*) viscera. The process involves several steps such as collection of soil samples, gathering of materials, preparation of the biofertilizer, fermentation of fish viscera, rice planting, and application of the biofertilizer. Then, the output reflects the study's results, specifically the evaluation of the efficacy of Tilapia (*O. niloticus*) viscera and powdered eggshell as biofertilizers in promoting the growth of rice (*Oryza sativa*).

Statement of the Problem

This study aimed to determine the synergistic effect of Tilapia (*O. niloticus*) viscera and eggshell as a biofertilizer to the growth of rice (*O. sativa*).
Specifically, it sought to answer the following questions:

1. What is the plant height of the Rice (*O. sativa*) with and without Biofertilizer treatment at different time spans?
2. What is the number of leaves of the Rice (*O. sativa*) with and without Biofertilizer treatment at different time spans?
3. What is the root length of the Rice (*O. sativa*) with and without Biofertilizer treatment?
4. What is the NPK (Nitrogen, Phosphorus and Potassium) and pH levels of the soil before and after planting?
5. Is there a significant difference on the plant height of the Rice (*O. sativa*) with and without Biofertilizer treatment at different time spans?
6. Is there a significant difference on the number of leaves of the Rice (*O. sativa*) with and without Biofertilizer treatment at different time spans?
7. Is there a significant difference on the root length of the Rice (*O. sativa*) with and without Biofertilizer treatment?

Hypotheses

The researchers used null hypotheses in the study to determine whether there is a significant difference among the variables. The following null hypotheses would be tested at 0.05 level of significance through Two-way Repeated Measures (ANOVA).

1. There is no significant difference on the plant height of the Rice (*O. sativa*) with and without Biofertilizer treatment at different time spans.
2. There is no significant difference on the number of leaves of the Rice (*O. sativa*) with and without Biofertilizer treatment.
3. There is no significant difference on the root length of the Rice (*O. sativa*) with and without Biofertilizer treatment.

Methodology

An experimental design was utilized in this study to incorporate the employment of the treatment and control groups in order to maximize the potential outcomes. Specifically, the use of experimental design in agricultural studies, as per Janvry and Sadoulet (2021), illustrates the comparative presentation between treatments and outcomes, particularly regarding the effect of fertilizer on the physiological parameters and yield of rice (O. sativa).

To better observe these effects, the researchers employed an observational technique to assert the differences in the growth rates between the control and treatment groups. The observations focused on the significant effect of the different biofertilizer combinations including setup A with 70% amino acid (49 milliliters) and 30% eggshell (21 grams), setup B with 30% amino acid (21 milliliters) and 70% eggshell (49 grams), the setup C with 50% amino acid (30 milliliters) and 50% eggshell (40 grams), the setup D with 100% amino acid (30 mL) and the setup E with 100% eggshell (40 grams). These variations of biofertilizer allowed for a detailed examination of how different proportions influenced the rice (O. sativa) plant growth.

The data collection on the rice plants' leaf length and number of leaves involved a weekly monitoring. The Increase in these parameters was taken as indicators of plant growth. Additionally, root length was measured to serve as a hallmark of plant health stability, with longer roots suggesting a healthier rice plant according to Kitomi et al. (2020). To calculate and analyze the gathered data and determine the significant effect of various combination proportions of Biofertilizer on the final growth of rice (O. sativa) in terms of plant's leaf length (cm), number of leaves, and length of roots (cm). A one-way and two-way ANOVA (analysis of variance) was used with a 0.05 threshold of significance to ensure a 95% level of confidence in making the correct decision.

Results and Discussion

This excerpt provides a detailed analysis of the significant effects of Tilapia (*O. niloticus*) and eggshells on the growth and development of rice (*O. sativa*) in terms of various parameters, including the root and leaf length and number of leaves. This section also highlights the synergistic effects of Tilapia (*O. niloticus*) viscera and powdered eggshell on the soil and their nutrient contributions on its health, which ultimately affects rice plant productivity.

Setups	**Initial**		**1 Week**		**2 Weeks**		**3 Weeks**		**4 Weeks**	
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**
A	11.00	1.50	16.40	2.30	24.45	.05	26.40	.10	28.60	1.40
B	12.00	2.90	17.750	5.65	23.85	3.35	27.05	1.05	30.20	1.80
C	11.95	2.75	16.00	1.00	23.60	.60	25.15	.65	28.20	1.80
D	11.90	2.60	15.55	1.35	22.40	1.60	23.60	2.40	23.90	1.10
E	10.90	1.40	17.15	5.65	27.15	3.65	29.75	1.25	35.65	3.85
F	11.95	1.95	18.30	7.30	22.80	4.80	26.50	2.50	28.30	1.30

Table 1. *Mean and Standard Deviation of Plant Height of Rice (O. sativa) at Different Treatments and Duration.*

Table 1 presents the mean plant height mean and standard deviation (SD) of *O. sativa* over four weeks under different treatment conditions. Generally, the growth trend value shows that all the setups exhibited a progressive increase in height over the four-week period, with variations among treatments and controls. In terms of higher standard deviation, it is seen in setup B with 30:70 (30% amino acid and 70% eggshell) has the second highest mean, and setup F (control) has the second lowest mean. During the first two-week period, there is a high standard deviation among the samples of the setup B with 30:70 (30% amino acid and 70% eggshell), which implies the greater variability in the plant height. The setup F (control) also exhibited variability during the first, second, and third week periods.

This indicates that individual samples of *O. sativa* within these conditions exhibited a broader range of growth rates. However, setup B with 30:70 (30% amino acid and 70% eggshell) still exhibited a prominent growth mean, signifying the effectiveness of this biofertilizer mixture in increasing the height of the plant despite variability.

On the other hand, the lower standard deviation with a relatively lower mean is seen in both setups such as the setup A with 70:30 (70% amino acid and 30% eggshell) and setup D with 100 F (100% amino acid) treatments during and after a four-week period. This demonstrates that the reduced standard deviation implies greater consistency in plant heights across samples, indicating that the treatment contributed to a stabilizing effect on plant growth. In addition to that it appears that the setup 100 F (100% fish amino acid) ranked as the setup with lowest mean distribution after the four-week observation.

To reiterate that such setups with high standard deviation indicates that there is a varying extreme response of plant samples either to fertilizer or to the environment. This simply means that plant samples in each setup exhibited significant growth while others had slower growth. This may be attributed to the several factors like, natural genetic variation, natural health condition, light and water unequal access. Moreover, the low standard deviation implies the consistent and uniform growth of plant samples in each setup. This means that there is no actual promotion of increased height development.

Furthermore, it is notable that the setup E with 100 E (100% eggshell) had the highest mean and standard deviation, which reflects as well that there is a greater variability in height growth. But, the attainment of highest mean suggests that this fertilizer promoted a strong height growth development for it to be considered as the most effective biofertilizer.

The differences of standard deviation and mean values across treatments reflects that biofertilizers resulted in different influences such as growth enhancement, growth height variability and stability. Overall, the setup E with 100 E (100% eggshell) promotes the most effective growth of plant height. And the setup D with 100 F (100% amino acid) is considered the least effective biofertilizer. In line with that, it also appears that the setup F (control) exhibited a moderate growth which outperformed the setup D with 100 F (100% amino acid) indicating that the application of 100 F (100% amino acid) as biofertilizer, did not show any significant effectiveness in promoting the *O. sativa* height.

This situation is neither uncommon nor novel because this has already occurred in a similar study conducted by Johari et al. (2020). The study findings highlight the application of FAA has a substantial impact on the Okra plants' shoot length and height at various sampling dates. Basically, the FAA application and sample times significantly affected shoot length, indicating that high FAA concentration can alter plant height depending on application rates and timing. This significantly entails that higher concentrations of FAA would result in either a decrease of plant's height or a promotion of growth.

Likewise, Xu and Mou (2017) clarified that the application of the excessive fish amino acid concentration has a potential toxicity that decreases the growth promotion of a plant's height and root length as it weakens the structure of the root and stem components. Thus, the 100% fish amino acid fertilizer certainly has a direct effect on the *O. sativa*. The 100%

concentration value of fish amino acids that were applied can be considered as "excessive" as it resulted in the least growth of the setup D.

Concerning biofertilizer concentrations, this study highlights the mixture proportion that demonstrates the highest effectiveness. Based on the statistical result, the comparison between setup B with 30:70 (30% amino acid and 70% eggshell) and setup A with 70:30 (70% amino acid and 30% eggshell) and 50:50 (50% amino acid and 50% eggshell) indicates that the proportion of eggshell should be higher than fish amino acid to achieve better outcome.

To assert that, the researchers cited an observed finding by Kasim et al. (2023) in their study that the plant height increased when eggshells were added to the growing media; their results indicate that eggshells can be a useful soil supplement to encourage vegetative growth. Overall, this emphasizes that applying biofertilizers with a concentration of 100E (100% eggshell) and a 30:70 ratio (30% amino acid and 70% eggshell) is statistically advantageous than the absence of biofertilizer in enhancing the plant height of *O. sativa*.

Setups	Initial		1 Week		2 Weeks		3 Weeks		4 Weeks	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
A	21.00	2.00	22.00	4.00	19.00	3.00	21.00	1.00	20.00	.00
B	22.50	2.50	28.00	1.00	30.00	2.00	30.50	1.50	31.00	1.00
C	20.00	2.00	22.50	1.50	24.50	2.50	25.50	1.50	25.50	1.50
D	16.50	1.50	19.00	2.00	22.50	2.50	24.50	3.50	25.00	4.00
E	22.00	5.00	28.00	4.00	34.50	1.50	36.00	3.00	33.50	.50
F	20.50	.50	23.00	.000	19.00	1.00	17.00	1.000	17.00	1.00

Table 2. Mean and Standard Deviation of Number of Leaves of Rice (O. sativa) at Different Treatments and Duration

Table 2 presents the mean and standard deviation of the number of leaves of Rice (*O. sativa*) at different treatments over a four-week duration. The results show that the setup E with 100 F (100% eggshell) had the highest mean leaf count with 33 leaves. However, it has a high standard deviation during the first and third week of observation indicating a considerable variation in number of leaf growth, with samples having obtained significantly more or fewer leaves over such time. Despite the inconsistency in leaf number growth, the setup E with 100 E (100% eggshell) still has the highest mean that indicates its strong promotion of the leaf number development.

On the contrary, the setup B with 30:70 (30% amino acid and 70% eggshell) appears to have the second highest mean of leaf numbers accounting to 31 leaves, but it has a more consistent growth leaf number across samples during the four-week period of time as compared to the setup E with 100 E (100% eggshell). This simply justifies that the setup B with 30:70 (30% amino acid and 70% eggshell) is considered as the ideal mixture of biofertilizer, because it is able to promote an increase of leaf number while maintaining the uniform leaf number growth across the samples.

Moreover, the setup F (control) revealed the lowest number of leaf mean among all the setup that only has 17 leaves. It also has a lower standard deviation entailing that there is uniform and limited growth of each sample during the four-week observation. This result aids to validate the potential effectiveness of the biofertilizers in enhancing *O. sativa* leaf numbers.

Therefore, 100 E (100% eggshell) alone is the most effective among all the biofertilizers in increasing the leaf number. And when it comes to the mixture of both fish amino acid and eggshell, the 30:70 ratio (30% fish amino acid and 70% eggshell) shows the best results, as it ranked second in producing leaf number. This refers to the ideal optimal ratio of eggshell and fish amino acid when combined. Therefore, eggshell must be greater than the fish amino acid in a mixture.

To support that significant proportion, there is Sari and Muhsanati (2023) that states the eggshell powder application per plant had a good effect on growth, including leaf development. As to their study, adding eggshell powder to soybean plants produced a great number of leaves. Furthermore, fish amino acid concentration must be lower as Jumar et al. and Siti (2021) concluded in their study that fish amino acid application on Amaranthus plants showed that, in comparison to higher concentrations and control groups, a 1% FAA foliar spray resulted in noticeably more leaves per plant, therefore application of FAA must remain optimal.

Setups	Root Length	
	Mean	SD
A	11.4500	2.45000
B	18.5500	1.75000
C	15.4500	3.55000
D	18.1000	2.10000

E	27.8000	1.20000
F	11.4500	1.45000

Table 3. Mean and Standard Deviation of the Root Length of Rice (O. sativa) at Different Treatments and Duration.

Table 3 presents the mean and standard deviation (SD) of the root length of Rice (*O. sativa*) at different treatments over a four-week duration. The statistical results revealed that the setup E with 100 E (100% eggshell) exhibited the longest mean root length of 27.8 cm, with a low standard deviation of 1.2 cm, indicating a high level of consistency in root development among the plants. This uniformity suggests that the treatment effectively promoted stable and extensive root growth.

Conversely, both setup A with 70:30 (70% amino acid and 30% eggshell) and setup F (control) recorded the shortest mean root length at 11.45 cm. The setup A (70% amino acid and 30% eggshell) mean is accompanied with high standard deviation, while the setup F (control) has lower standard deviation. This means that setup F (control), has a more growth uniformity after the four-week period observation. But this result also amplifies the infectivity of the setup A with 70:30 (70% amino acid and 30% eggshell) as it retains a root length mean similar to the setup that does not receive any treatment.

Apart from that, there are setups with higher mean and standard deviation, which is the setup B with 30:70 (30% amino acid and 70% eggshell) followed by setup D with 100 F (100% amino acid). There is 0.4500 difference in their mean distribution. But both of these two setups are considered as the second for having the highest mean, next to setup E with 100 E (100% eggshell). The variability seen in this two setup pertains to the inconsistency and different responses of each sample to attain root length growth.

However, despite that, the higher mean they possess which is not seen in other setup points into one thing, these two setups are a potential effective biofertilizer for root length promotion. Therefore, it is the setup E with 100 E (100% eggshell) setup that significantly promotes the increase of root length with a uniform growth. The stability in root length growth across samples suggests that this treatment created optimal conditions for nutrient absorption and overall plant health which was stressed by Li et al. (2024).

On the other hand, although the setup B with 30:70 (30% amino acid and 70% eggshell) and setup D with 100 F (100% amino acid) contributed to root growth, it exhibited greater variability, making it a less dependable option for achieving consistent root development. However, its root mean length that is greater than other setup suggests it as the second most effective type of biofertilizer.

Set-ups	pH	Nitrogen	Phosphorus	Potassium
Pre-assessment	6.4	0.26	64.27	0.92
A	6.7	0.26	33.49	0.87
B	6.6	0.28	32.29	0.89
C	6.8	0.29	26.97	0.78
D	6.5	0.26	29.50	1.08
E	6.9	0.28	29.76	0.98
F	6.6	0.30	25.05	0.92

Table 4. The NPK (Nitrogen, Phosphorus and Potassium) and pH levels of the Soil Before and After Planting

The researchers also derived the findings from soil analysis conducted during the pre-experiment and post-experiment. It was executed to ascertain the practical influence of different setups of biofertilizers on the soil's nutritional components such as pH, nitrogen, phosphorus, and potassium.

Regarding the pH, the pre-experiment soil has 6.4 while the setup F (control) of the post-experiment has a 6.6 pH value. It appears that even though there was no biofertilizer application, the crop rice (*O. sativa*) planting can contribute to a pH increase. Nevertheless, according to a statement from Kenney (2022), the pH increase provided by the planting is limited, which means that it cannot sustain optimal pH conditions over time. The inability to control the optimal pH condition of the soil necessary for the rice (*O. sativa*) might lead to a decrease in the number of organic compounds essential to it. This highlights the importance of the biofertilizer influence to retain the optimal pH condition required by the rice (*O. sativa*).

The biofertilizer with a higher proportion of powdered eggshell implies its ability as a soil neutralizer mitigating the consequence of fish amino acid by decreasing soil pH. In addition to that, Huang et al. (2017) stated that the resulting pH values of the soil samples that range from 6.5 to 6.9 are evaluated as still conducive pH levels for the growth of *O. sativa*. Since the 6.5 is considered slightly acidic and the 6.9 is slightly neutral. Therefore, all the pH values listed above are still considered optimal pH values for rice (*O. sativa*).

Next to pH is nitrogen, the values of nitrogen levels on each setup B, C, and E in contrast to the pre-assessment soil sample demonstrate the ability of the biofertilizer to induce the nitrogen level of the soil. However, the soil samples from setup F (control) appeared to have higher nitrogen levels than those from the setups with biofertilizer application. This can be explained by the error system that occurred while the researchers were preparing the soil for soil analysis submission. The researchers performed a sun-drying method that significantly reduced the activity of all essential microorganisms that generate nitrogen. Gao et al. (2020). It also happened that the soil samples from both the control (F) and the pre-experiment setup were not exposed to high temperatures like the others which explains the setup F gaining the highest nitrogen level after all.

Another organic compound that was analyzed in this study was the phosphorus (P) content. The results implicated a drastic decrease in the phosphorus level of the soil after planting Rice (*O. sativa*). According to Julia et al. (2016), *O. sativa* is one of the plants with high phosphorus demand. In line with that, the soil analysis showed that the phosphorus content of the pre-experiment soil sample declined from 64.27% to 25.05% during the post-experiment. Therefore, this demonstrates how *Oryza sativa* maximized its phosphorus uptake. The data further highlight that the setups with biofertilizer application mitigated the adverse effects associated with rice *(O. sativa's)* enhanced phosphorus uptake from the soil. Generally, the setups with biofertilizer application were higher by 0.92% to 8.44% compared to the 25.05% of the setup F (control). This exhibited that the mixture of Tilapia (*O. niloticus*) and powdered eggshell diminished the impact of *O. sativa* on the soil, especially concerning the sustainability of the phosphorus level.

Lastly, potassium (K) was analyzed, and the results indicated that Setup D (100% Fish Amino Acid, FAA) and Setup E (100% Eggshell, ES) exhibited higher potassium levels compared to the other setups. Meanwhile, the setups combining fish amino acid and powdered eggshell showed lower potassium levels than both Setup F and the pre-assessment soil samples. This suggests that the combination of fish amino acid and powdered eggshell may have triggered a reaction that reduced the soil's potassium levels. However, Setup B, composed of 30% FAA and 70% ES, recorded the highest potassium level among the three setups, with the lowest readings. This finding highlight that a 30:70 ratio (30% FAA to 70% ES), with a lower concentration of Tilapia (*Oreochromis niloticus*) viscera fish amino acid and a high concentration of powdered eggshell, was more favorable to use. Nonetheless, further adjustments to the mixture's ratio are necessary to achieve even higher potassium levels.

Factor	F value	p value
Setups	2.922	.147
Duration	156.016**	<.001
Interaction	1.422	.158

*Legend: * - significant at a = .05; ** - significant at a = 0.01*

Table 5. F value and P value between the Plant Height of Rice (O. sativa) at Different Treatments and Duration.

Table 5 presents the results of a two-way repeated measures ANOVA between the plant height of *O. sativa* at different treatments and durations. The results indicate that the duration had a highly significant effect on plant height with an F value of 156.016 and p-value of <.001. This demonstrates that the plants had substantial growth over time. On the other hand, the setups showed an F value of 2.922 and a p value of .147, which indicates that there is no significant effect on the plant height of *O. sativa*.

Additionally, the F value and p value of the interaction between the setups and duration is not significant with a value of 1.422 and .158, respectively. This supports the results on setups with an F value of 2.922 and a p value of .147; illustrating that there is no significant difference on the plant height of the rice *(O. sativa)* with and without Biofertilizer treatment at different time spans. Therefore, the researchers do not reject the null hypothesis. The P-values for each factor such as setup and interaction, showed a value that is greater than the significance level 0.01 and 0.05. And the value of the factor duration is less than 0.001, which generally suggests that plant height naturally increased over time, regardless of the application of the biofertilizer. Therefore, the observed differences for these factors are possibly caused by random variation and not by experimental treatments.

To support findings of not rejecting the null hypotheses, Ballabio et al. (2019) stated that naturally, the *O. sativa* can grow and gain an increase of height regardless of if there is an application of the biofertilizer. For instance, an organic material such as NPK is already present in the soil. Therefore, it is evident that a plant can grow even in the absence of biofertilizer. Basically, the *O. sativa* has the potential to grow on its own over time. Similar to what has occurred in this study, the setup F (control) grew independently. However, even though a soil has the nutrient provision, a biofertilizer is indeed important to increase the exponential growth of a plant, especially the height. This is because organic materials such as NPK found in a soil is limited as discussed by Cui et al. (2023). Therefore, the use of fertilizer serves as a soil amendment provider to maintain its nutritional components.

Factor	F value	p value
Setups	19.346**	<.001
Duration	33.608**	<.001
Interaction	10.687**	<.001

*Legend: * - significant at a = .05; ** - significant at a = 0.01*

Table 6. F value and P value between the No. of Leaves of Rice (O. sativa) at Different Treatments and Duration.

Table 6 shows the f value and p value of the factors such as setups, duration, and interaction. A two-way ANOVA is utilized to determine the effects of different treatments and duration to the no. of leaves of rice (*O. sativa*). The duration of applying the treatment lasts for a month with a once-a-week application. And the interaction encompasses the effectiveness of the different treatments and duration to the number of leaves grown in rice (*O. sativa*).

As shown in the table, the setup has an f-value of 19.346 with p-value of <0.001. Indicates that there is significant difference between the set ups and the number of leaves grown in rice (*O. sativa*). Particularly, the setup E with 100 E (100% eggshell) gained the greatest number of leaves. Followed by setup B with 30:70 (30% amino acid and 70% eggshell)

In terms of duration, f-value shows 33.608 and its significant value (p-value) is <0.001 which is less than the significance level of 0.05. This implies that duration significantly affects the number of grown leaves of rice (*O. sativa*). Besides, the plant naturally produced more leaves over time. With the interaction effect of the different setups and time span, the p-value of <0.001 indicates that the difference is statistically significant. This suggests that the effect of different setups of the biofertilizer as time progressed has significant results to the number of leaves in rice (*O. sativa*).

This data findings shows that some setups of the treatment specifically with 100% ES has highly notable effects on the number of grown leaves of rice (*O. sativa*). Furthermore, naturally as time progresses the plant keeps growing. With all the factors at a p-value of <0.001 which is less than the significance level of 0.05, this indicates that there is significant difference in setups, duration, and interaction between no. of leaves in rice (*O. sativa*). Therefore, the null hypothesis is rejected. The significant differences in statistical results that resulted in rejection of this null hypothesis did not occur by chance through random variation but rather through experimental treatment. This highly provides confidence in proposing the potential effectiveness of the biofertilizers with eggshell and fish amino acid.

It is also concluded by Anugrah et al. (2023) that their various number of setups with six treatments and five replications of applying eggshell fertilizer resulted to statistical differences in number of leaves of a cayenne pepper (*Capsicum frutescens L.*) that is statistically resulted to improved increase number of leaves compared to the control. As for the fish amino acid, Luna (2022) confirms that different treatment of fish amino acid application to Radish (*Raphanus sativus*) significantly increases leaf number that is better than control.

Factor	F value	p value
Setups	22.382**	<.001

*Legend: * - significant at a = .05; ** - significant at a = 0.01*

Table 7. The F and P value of the Root length of Rice (O. sativa) at different treatments.

Table 7 shows the statistical f-value and significance F (p-value) of setups using a one-way ANOVA to find out the significant difference of the root length of rice (*O. sativa*) at different treatments. The lower p value with < .001 and relatively high F value for this result, suggests that the various setups have significant differences in root length of *O. sativa*, and this reflects the rejection of the null hypothesis. Therefore, this data also emphasizes that outcome of different setups did not occur by chance but rather through an experimental treatment. This signifies that there is a potential effectiveness of the biofertilizer towards the root length of *O. sativa*.

Moreover, based on the data, it is revealed that the setup E with 100 ES (100% eggshell) has the longest root with 27.8 cm, suggesting that the biofertilizer treatment improved the root growth. While setup B with 30:70 (30% amino acid and 70% eggshell) and setup D with 100 F (100% amino acid), with roots gained 18cm long thus, grew moderately. And yet, the setup F (control) and setup A with 70:30 (70% amino acid and 30% eggshell) roots grew shortly with both having a 11.45 cm length. This just indicates that setup does not show a promising efficiency for the root growth of *O. sativa* mainly because it has 100% fish amino acid concentration. To expound this, the high concentration of FAA (Fish Amino Acid) can cause overfertilization, leading to rapid plant growth without a sufficiently developed root system. According to Smith (2013), overfertilization can result in sudden plant growth, where the root system is unable to supply adequate water and nutrients to support the plant.

Overall, the 100 ES (100% eggshell) biofertilizer is the most effective and setup B and setup D is the second. The eggshell promising result was also reiterated by Anugrah et al. (2021), with their study on cayenne pepper (*Capsicum frutescens L.*) demonstrating that applying eggshell organic fertilizer significantly enhanced the root length. In further study of Sulegaon (2025), calcium carbonate present in eggshell promotes root elongation and overall root health. Applying eggshell powder to the soil can stimulate root development, resulting in increased root length.

Conclusion and Recommendations

Conclusion

This study demonstrates the effect of the various biofertilizers on rice (O. sativa) development in terms of leaf and root length and number of leaves. As well as the impact of biofertilizer application on the soil's nutrient content such as pH, nitrogen, phosphorus, and potassium. The result showcased that the most effective biofertilizer is composed of 100% powdered eggshell and the second one is composed of a combined 30% fish amino acid and 70% powdered eggshell.

The 100% fish amino acid provided inconsistent results, where at times, it was even outperformed by the control setup. This is expounded by Xu and Mou (2018) that the application of excessive fish amino acid concentration has potential toxicity that decreases the growth promotion of a plant's height and root length as it weakens the structure of the root and stem components. Thus, the 100% fish amino acid fertilizer certainly has a direct effect on the O. sativa's height.

Therefore, in conclusion, it is necessary to note that the percentage of concentration gradient plays a crucial role in determining its effectiveness. The study found that Tilapia (O. niloticus) viscera and powdered eggshells as biofertilizers had a significant effect on plant growth, specifically in terms of the number of leaves and root length. As a result, the null hypothesis was rejected for these mentioned parameters. However, the biofertilizer did not significantly impact the plant height of rice (O. sativa). Therefore, the findings support the null hypothesis, indicating no significant difference between biofertilizer and plant height.

Recommendation

Based on the findings of this study regarding the efficacy of Tilapia (O. niloticus) viscera and powdered eggshell as biofertilizer in the growth of rice (O. sativa), the researchers recommend the following to enhance the formulation of the biofertilizer as well as its utilization in the agricultural field:

Determination of the Optimal Fertilizer Concentration following the ideal proportion set by this study which is the 30% fish amino acid and 70% powdered eggshell should be implied in order to achieve the high amount of potassium level.
Utilization of other calcium carbonate sources such as seashells and mussel shells, since these types of shells have a higher calcium carbonate content than eggshell.

Exploitation of other type of fish other than Tilapia (O. niloticus) like Bangus (Chanos chanos), Galunggong (Decapterus macrosoma), Tulingan (Auxis thazard) and other fish species that can be found in the Philippines.

Acknowledgement

The authors would like to thank the colleagues and institutions who provided guidance, feedback, and support throughout the conduct of this research and the preparation of this manuscript. Any remaining errors or omissions are the sole responsibility of the authors.

Funding

This research received no external funding from any public, commercial, or not-for-profit funding agency, and no organization provided financial support for the conduct of the study, authorship, or publication of this article.

Competing Interests Statement

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this article.

Data Availability Statement

Data sharing is not applicable to this article as no new data were created or analyzed in this study; all data used were obtained from previously published sources as cited in the reference list.

References

Abdulrahman, I., Tijani, H. I., Mohammed, B. A., Saidu, H., Yusuf, H., Jibrin, M. N., & Mohammed, S. (2014). From Garbage To Biomaterials: An Overview On Egg Shell Based Hydroxyapatite. Journal Of Materials, 2014, 1–6. Https://Doi.Org/10.1155/2014/802467

Anugrah, R. D., Rafvenia, R., Meitiyani, M., & Safahi, L. (2021). The Effect Of Eggshell Organic Fertilizer On Vegetative Growth Of Cayenne Pepper (Capsicum Frutescens L). Iop Conference Series: Earth And Environmental Science, 755(1), 012001. Https://Doi.Org/10.1088/1755-1315/755/1/012001

Ballabio, C., Lugato, E., Fernández-Ugalde, O., Orgiazzi, A., Jones, A., Borrelli, P., Montanarella, L., & Panagos, P. (2019). Mapping Lucas Topsoil Chemical Properties At European Scale Using Gaussian Process Regression. Geoderma, 355, 113912. Https://Doi.Org/10.1016/J.Geoderma.2019.113912

Calubaquib, M. A. M., Navarrete, I. A., & Sanchez, P. B. (2016). Properties and nutrient status of degraded soils in Luzon, Philippines. Philippine Journal of Science, 145(3), 249–258. https://philjournalsci.dost.gov.ph/home-1/48-vol-145-no-3-september-2016/603-properties-and-nutrient-status-of-degraded-soils-in-luzon-philippines

Cui, H., Zhu, H., Shutes, B., Rousseau, A. N., Feng, W., Hou, S., Ou, Y., & Yan, B. (2023). Soil Aggregate-Driven Changes In Nutrient Redistribution And Microbial Communities After 10-Year Organic Fertilization. Journal Of Environmental Management, 348, 119306. Https://Doi.Org/10.1016/J.Jenvman.2023.119306

Jaies, I., Qayoom, I., Saba, F., & Khan, S. (2024). Fish Wastes As Source Of Fertilizers And Manures (Pp. 329–338). Https://Doi.Org/10.1007/978-981-99-8593-7_15

Janvry, A., & Sadoulet, E. (2020b). How Experimental Research In Agriculture Has Gone From Lab To Field. World Development, 127, 104782. Https://Doi.Org/10.1016/J.Worlddev.2019.104782

Johari, N. S., Mutalib, A. A., Ismail, Z., Ismail, F., Latif, Z. A., Man, S. '. I. C., & Tang, J. R. (2020). Effects Of Fish Amino Acid (Faa) Application On Growth And Development Of Okra (Abelmoschus Esculentus) At Different Sampling Times. Journal Of Vocational Education Studies, 3(2), 35. Https://Doi.Org/10.12928/Joves.V3i2.2932

Jumar, J., Saputra, R. A., & Jannah, S. R. (2021). Effect Of Fish Amino Acid Application On Growth And N-Uptake In Plant Rice Using The System Of Rice Intensification Method. Tropical Wetland Journal, 7(1), 25–30. Https://Doi.Org/10.20527/Twj.V7i1.91

Kasim, N. N., Abidin, S. Z. Z., Ali, S. N., Omar, N. F., & Aziz, A. A. (2023). The Effect Of Eggshells As A Growing Medium Treatment On Plant Growth Performance Of Red Spinach (Amaranthus Dubius). International Journal Of Innovation And Industrial Revolution, 5(14), 22–43. Https://Doi.Org/10.35631/Ijirev.514002

Kitomi, Y., Hanzawa, E., Kuya, N., Inoue, H., Hara, N., Kawai, S., Kanno, N., Endo, M., Sugimoto, K., Yamazaki, T., Sakamoto, S., Sentoku, N., Wu, J., Kanno, H., Mitsuda, N., Toriyama, K., Sato, T., & Uga, Y. (2020). Root Angle Modifications By The Dro1 Homolog Improve Rice Yields In Saline Paddy Fields. Proceedings Of The National Academy Of Sciences, 117(35), 21242–21250. Https://Doi.Org/10.1073/Pnas.2005911117

Kumar, S., Diksha, Sindhu, S. S., & Kumar, R. (2022). Biofertilizers: An Ecofriendly Technology For Nutrient Recycling And Environmental Sustainability. Current Research In Microbial Sciences, 3, 100094. Https://Doi.Org/10.1016/J.Crmicr.2021.100094

Lee, T., Pu'ad, N. M., Alipal, J., Muhamad, Basri, H., Idris, M., & Abdullah, H. (2022). Tilapia Wastes Valuable Materials: A Brief Review Of Biomedical, Wastewater Treatment, And Biofuel Applications. Materials Today: Proceedings, 57, 1389–1395. Https://Doi.Org/10.1016/J.Matpr.2022.03.174

Li, C., Li, Y., Yang, J., Lian, B., Wang, J., & Zou, G. (2024). Regulating Root Structure Of Potted Lettuce To Magnify Absorption From App And Uan Fertilizers. Frontiers In Plant Science, 15. Https://Doi.Org/10.3389/Fpls.2024.1407984

Luna A. Growth And Yield Performance Of Radish (Raphanus Sativus L.) As Affected By Fish Amino Acid As Soil Drench Application | Thesis Repository. (N.D.-B). Https://Thesis.Msugensan.Edu.Ph/Node/1135

Sari, D. P., Warnita, & Muhsanati. (2023a, May 12). Impact Of Eggshells On Growth And Production Of Soybean (Glycine Max(L) Merril) In Ultisol. Https://Ijeab.Com/Detail/Effect-Of-Extract-Euphorbia-Hirta-Linn-Against-Leaf-And-Fruit-Pests-On-Plants-Cucumber-Cucumis-Sativus-Linn-2/

Smith, K. (2013). The Perils Of Over-Fertilizing Plants And Trees. University Of California Agriculture And Natural Resources. Retrieved From Https://Ucanr.Edu/Sites/Default/Files/2013-07/170168.Pdf

Sulegaon, R. (2025). Eggshells As Fertilizer In Horticulture Practices. International Journal Of Horticulture And Food Science, 7(3), 32–34. Https://Doi.Org/10.33545/26631067.2025.V7.I3a.277

Tacio, H. D. (2021, September 22). Rice: The Staple Food Of Filipinos The Staple Food Of Filipinos. Edge Davao. Https://Edgedavao.Net/Agri-Trends/2021/09/Rice-The-Staple-Food-Of-Filipinos-The-Staple-Food-Of-Filipinos/

Xu, C., & Mou, B. (2017). Drench Application Of Fish-Derived Protein Hydrolysates Affects Lettuce Growth, Chlorophyll Content, And Gas Exchange. Horttechnology, 27(4), 539–543. Https://Doi.Org/10.21273/Horttech03723-17

Appendices

No appendices are attached to this study.