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Scientific Article

Potential of liquid organic fertilizer from horse and cow urine on shoot growth of *Cattleya labiata* Lindl.

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Abstract

Liquid organic waste from animals can be fermented into liquid organic fertilizer, so it can be an alternative to increase plant growth. This study aims to determine the effect of giving horse and cow urine LOF, as well as its interaction on shoot growth in the *Cattleya labiata* Lindl orchid plant. The method used is a factorial experimental method with a randomized block design and three replications. Liquid organic fertilizer from horse and cow urine with a concentration of 0%, 10% and 20%. There were 9 treatment combinations with 27 experimental units. The results showed that the LOF application of horse and cow urine and their interactions had a very significant effect on the growth of *Cattleya labiata* shoot diameter in the 4th week. The application of horse urine LOF and its interaction with cow urine LOF had no significant effect on the growth of the number and height of shoots, but had a very significant effect on the administration of cow urine LOF. The best treatment was found in the application of 20% cow urine LOF without horse urine LOF (K0S2); obtained the highest growth of one shoot 4.8 cm with the highest diameter of 1.8 cm. LOF application of cow urine is more effective in increasing the growth of orchid shoots including: number, height, and diameter of shoots.
Keywords: fermentation, nutrients, orchid, vegetative propagation.

Resumo

Potencial de adubação orgânica líquida com urina de cavalo e vaca no crescimento da parte aérea de *Cattleya labiata* Lindl.

Resíduos orgânicos líquidos de animais podem ser fermentados em fertilizante orgânico líquido, podendo ser uma alternativa para aumentar o crescimento das plantas. Este estudo teve como objetivo determinar o efeito da administração de LOF à urina de cavalo e vaca, bem como sua interação no crescimento da parte aérea da orquídea *Cattleya labiata* Lindl. O método utilizado é um método experimental fatorial com delineamento em blocos ao acaso e três repetições. Fertilizante orgânico líquido de urina de cavalo e vaca com concentração de 0%, 10% e 20%. Houve 9 combinações de tratamento com 27 unidades experimentais. Os resultados mostraram que a aplicação de LOF de urina de cavalo e vaca e suas interações tiveram um efeito muito significativo no crescimento do diâmetro da parte aérea de *Cattleya labiata* na 4ª semana. A aplicação de LOF de urina de cavalo e sua interação com LOF de urina de vaca não teve efeito significativo sobre o crescimento do número e altura de brotos, mas teve um efeito muito significativo sobre a administração de LOF de urina de vaca. O melhor tratamento foi encontrado na aplicação de 20% LOF de urina de vaca sem LOF de urina de cavalo (K0S2); obteve o maior crescimento de um broto de 4,8 cm com o maior diâmetro de 1,8 cm. A aplicação LOF de urina de vaca é mais eficaz em aumentar o crescimento de brotos de orquídeas, incluindo: número, altura e diâmetro de brotos.
Palavras-chave: fermentação, nutrientes, orquídea, propagação vegetativa.

Introduction

The *Cattleya labiata* Lindl orchid is a type of flower that is liked by many people because of its very beautiful and attractive crown color, shape and size that is quite large. In general, vegetative cultivation of orchids is carried out in pots using artificial media from charcoal and dried coconut husks, as well as using inorganic fertilizers. Fertilizer is one of the most important needs for orchid plants, probably due to the limited growing media and space for growth.

Inorganic fertilizers are often used by orchid growers, perhaps because they are easier to find and react more quickly to plant growth. However, if used continuously with inappropriate concentrations, it will damage the growth and development of plants and can slow down the occurrence of flowering. According to Kurnianta et al. (2021), long-

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term fertilization using inorganic fertilizers without the addition of organic matter will have the potential to reduce soil fertility and cause non-optimal plant growth. While organic fertilizers can bind the ability of the soil to absorb water, increase resistance to erosion, and increase soil fertility, but will not increase residues in crop yields so it is safe for the environment and human health (Lesik et al., 2019). Such as liquid organic fertilizer from shrimp extract can significantly increase growth and productivity of chrysanthemum, can increase nutrient content and change soil functional microbes at the rhizosphere level compared to chemical fertilizer treatment (Ji et al., 2017).

The effectiveness of growing media can be increased by adding organic products. Organic fertilizers can encourage the growth of *Cattleya labiata* for the better (Hoshino et al., 2016). The application of organic fertilizers at certain concentrations can have the same potential as the application of inorganic fertilizers to optimize the growing media. Similarly, from an economic point of view, the costs and benefits of high-performance organic fertilizers will be invaluable (Jigme et al., 2015). Thus, the use of inorganic fertilizers can be reduced by utilizing organic materials that have become waste. Plant waste or livestock waste can be processed into organic fertilizer through a fermentation process. Several studies have shown that liquid organic fertilizers managed from plant waste can increase plant growth, such as water hyacinth (Apzani et al., 2017), watermelon peels (Christina et al., 2021), and banana peels (Anhar et al., 2021). Likewise, with livestock waste, such as livestock urine.

Livestock urine waste that accumulates on farms will have a negative impact on the environment if it is not managed properly. Currently, livestock urine waste can be managed through a fermentation process to produce liquid organic fertilizer (LOF). Plants fertilized with animal-based liquid fertilizer showed higher total biomass with more new organ development. Liquid organic fertilization resulted in increased absorption of macro and micro nutrients compared to plants with mineral fertilizers (Martínez-Alcántara et al., 2016). Several studies have shown that livestock urine can be used as a liquid organic fertilizer that can increase plant growth and production, such as cow urine on wheat plants (Sadhukhan et al., 2018), rabbit urine on bok choy plants (Kurnianta et al., 2021), and goat urine on Mucuna cuttings (Sitinjak and Pratomo, 2019).

Cow urine has been used as an LOF that can increase plant growth and production through foliar spraying or application to soil (Vala and Desai, 2021). Organic fertilizers increase crop yields and quality significantly, but with improper application they can cause serious environmental risks such as nitrate pollution (Li et al., 2017). In 24 hours, a cow can excrete 6 liters of urine. If a farmer can have 2 cows on his farm, he can provide 4380 liters of urine in one year which is equal to 65 kg of nitrogen, this amount of nitrogen is equal to 136 kg urea (Vala and Desai, 2021).

Cow urine industrial waste is quite a lot found in cattle farms, and has been used quite a lot as liquid organic fertilizer in several cultivated plants, such as rice plants (Sutardi et al., 2021), tomato plants (Setiati et al., 2018), and on stem cuttings of *Vitis vinevera* (Hansah et al., 2020). But no one has used it on *Cattleya* orchids yet. While horse urine waste is not only very difficult to find, no one has processed it into liquid organic fertilizer, and no one has used it for orchids. For this reason, it is necessary to conduct this study with the aim of knowing the effect of LOF in cow urine and LOF in horse urine, as well as their interaction with shoot growth in *Cettleya labiata* orchids.

Material and Methods

The main ingredients used were cow urine obtained from cattle breeders, horse urine obtained from horse breeders, Effective Microorganism (EM4), brown sugar, mother plant of *Cattleya labiata* Lindl orchid as seedlings, charcoal, coconut belt which dry and clear water.

The seedling used was taken from orchid plants grown on artificial media that were about three years old. Each seedling has two leaves, and each treatment uses two seedlings, so that in each replication there are 18 seedlings. The flowering *Cattleya labiata* orchid plant can be seen in Figure 1.

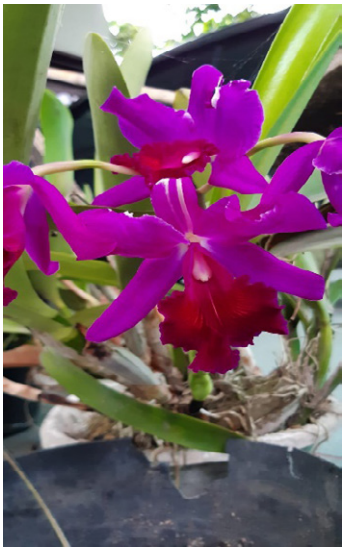

Figure 1. *Cattleya labiata* Lindl.

The experimental method used in this study was factorial design with a randomized block design and 3 replications. Obtained 9 treatments and 27 experimental units. The concentration of LOF from cow urine consists of 3 levels, namely: 0% (S0), 10% (S1), and 20% (S3), while the concentration of LOF from horse urine consists of 3 levels, namely: 0% (K0), 10% (K1), and 20% (K2). The parameters observed were the number of shoots, shoot height (cm), and shoot diameter (cm) that grew from the mother plant of the *Cattleya* orchid. Parameters were observed after 2 weeks of application or 3 weeks after planting. To measure the height of the shoot, it is measured from the base of the shoot to the tip of the shoot. To measure the diameter, measured near the base of the bud.

There are several stages in this research, namely: starting from the preparation of tools and materials in the manufacture of quality LOF. The brown sugar solution that has been made is mixed with the EM4 solution in accordance with the specified ratio, stirred evenly, then closed for 14 days. During the fermentation process, the mother plant of the *Cattleya* orchid which was used as seedlings and growing media in pots was prepared. The seedlings are put into pots which have been given a substrate in the form of coconut coir and charcoal as a growing medium. The pots are arranged randomly in each repetition. After making the LOF of horse and cow urine, the LOF application was carried out according to the predetermined concentration on the *Cattleya* orchid mother plant in flower pots.

Application of treatment was done by spraying onto the roots embedded in artificial media. The treatment application was only carried out once in the experiment. Then, weekly observations were made on the shoot growth parameters of the orchid mother plant. Observations in this experiment were carried out for 4 weeks.

The datas were analyzed using the analysis of variance test and then continued by Duncan's multiple range test at a significant level of 5% using SPSS version 22.

Results and Discussion

Effect of Cow and Horse Urine LOF on Shoot Growth of *Cattleya labiata* Lindl.

Growth of the Number of Shoots

Based on the observation that one week after application of LOF cow and horse urine, shoot growth was found in orchid plants, namely in plants that were given 20% LOF of cow urine (K0S2) and in plants that were given a combination treatment of 20% LOF of horse urine and 20% of LOF urine cows (K2S2). Then in the 3rd and 4th weeks after application of treatment, more shoot growth was found in orchid plants, where the number of shoots grew on average one in each orchid mother. In the first week after application K0S2 treatment can stimulate shoot growth as high as 1 cm, and K2S2 treatment can stimulate shoot growth as high as 0.5 cm. This shows that the application of LOF cow and horse urine can accelerate the growth of *Cattleya* orchid shoots, and the effective treatment is giving cow urine at a concentration of 20%. This can also be seen in the results of data analysis in the 4th week after application. Although based on analysis of variance, the application of horse urine LOF and its interaction with cow urine LOF had no significant effect, but cow urine LOF application had a very significant effect on the growth of the number of shoots in orchid plants after 4 weeks of application.

Then the statistical test showed that the application of 20% cow urine LOF without horse urine LOF (K0S2) gave the highest number of shoots. This treatment was not significantly different from the application of the combination treatment of 20% LOF of horse urine with 20% LOF of cow urine (K2S2), combined treatment of K1S2, and even 10% LOF of cow urine (K0S1). However, based on single factor treatment, the application of 20% cow urine LOF (S2) gave a higher number of shoots than all treatments. This treatment was significantly different from the application of 10% (S1) and 0% (S0) LOF of cow urine (Table 1).

Table 1. Effect of Horse and Cow Urine LOF on growth of the number of shoots of *Cattleya labiata* after 4 weeks of application

Horse Urine LOF	Cow Urine LOF			Average
	S0	S1	S2	
K0	0.71 c	1.05 ab	1.22 a	0.993
K1	0.71 c	0.88 bc	1.05 ab	0.880
K2	0.71 c	0.71 c	1.22 a	0.880
Average	0.71 c	0.88 b	1.16 a	

Note: The data in the table is the result of the transformation, and numbers followed by the same letter show no significant difference in the Duncan's test 5%

The Figure 2 shows that the administration of cow urine LOF, without combination or combination with horse urine LOF, still has the potential to stimulate the growth of orchid plant shoots. The best concentration of cow urine LOF is 20%, producing an average number of shoots of about 1 individual shoot.

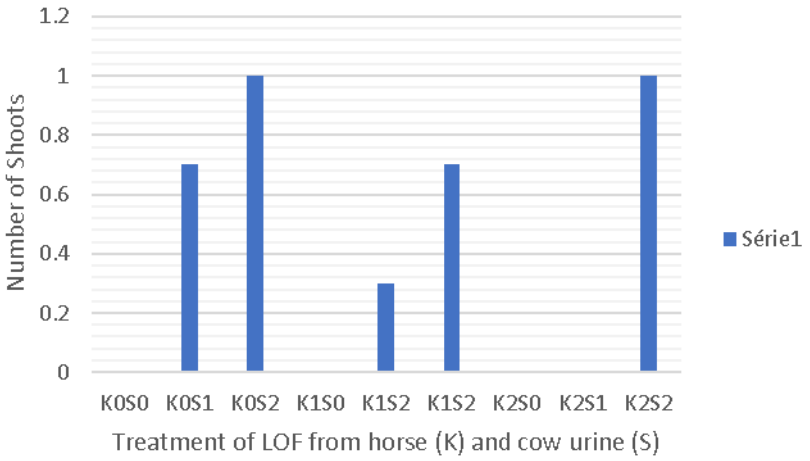

Figure 2. Effect of LOF of horse and cow urine on the number of shoots of *Cattleya labiata* orchids after 4 weeks of application

Growth of shoot height

Based on the analysis of variance, the application of LOF from horse urine and its interaction with LOF in cow urine had no significant effect, but the application of LOF from cow urine had a very significant effect on shoot height growth of *Cattleya* orchids. Then the statistical test showed that the application of a combination treatment of 10% horse urine LOF with 20% cow urine LOF (K1S2) gave the highest shoot height growth in orchid plants after 4 weeks of application. However, this treatment was not significantly different from the application of 20% cow urine LOF (K0S2). Then the single factor treatment also showed that the application of 20% cow urine LOF (S2) gave the best results compared to the 0% cow urine LOF treatment (S0) and even 10% cow urine LOF treatment (Table 2).

Table 2. Effect of Horse and Cow Urine LOF on *Cattleya labiata* shoot height growth after 4 weeks application

Horse Urine LOF	Cow Urine LOF			Average
	S0	S1	S2	
K0	0.71 c	0.997 c	2.30 ab	1.33
K1	0.71 c	1.05 c	2.82 a	1.53
K2	0.71 c	0.71 c	2.06 b	1.16
Average	0.71 c	0.92 b	2.39 a	

Note: The data in the table is the result of the transformation, and numbers followed by the same letter show no significant difference in the Duncan's test 5%

Application of LOF from horse urine and LOF from cow urine can increase shoot height growth in orchid plants. The best treatment that can increase the growth of the highest shoot height is the provision of LOF from 20% cow urine (treatment K0S2), which can reach about 4.8 cm in shoot height and has one leaf after 4 weeks of application. Then followed by a combination treatment of 20% horse urine LOF and 20% cow urine LOF (K2S2) which resulted in a shoot height of about 3.8 cm after 4 weeks of application (Figure 3).

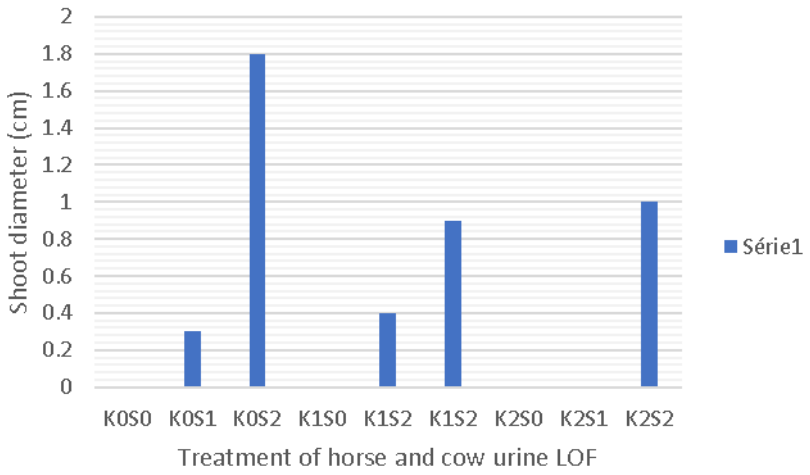

Figure 3. Effect of LOF of horse and cow urine on the height of shoots of *Cattleya labiata* orchids after 4 weeks of application

Growth of shoot diameter

Based on analysis of variance (ANOVA) that the application of horse urine LOF and cow urine LOF, and their interactions have a very significant effect on the growth of orchid shoot diameter at week 4 after application. The growth of shoot diameter has begun to be seen at the time of the growth of shoot height. The increase in the size of the shoot diameter was more evident in the K0S2 treatment when compared to the K0S1 treatment, even with other combination treatments. This can be seen in Figure 4.

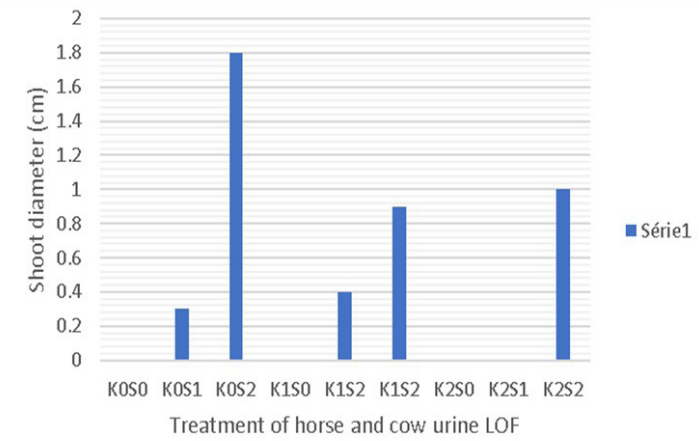

Figure 4. Growth of shoot diameter after 4 weeks of application

Statistical test analysis showed that the best treatment was the application of cow urine LOF with a concentration of 20% without a combination with horse urine LOF (K0S2) can increase the growth of the highest shoot diameter of about 1.8 cm at the 4th week after application. This treatment was significantly different from the other treatments (K0S0, K0S1, K1S0, K1S1, K1S2, K2S0, K2S1, and K2S2) (Table 3).

Table 3. Effect of Horse and Cow Urine LOF on growth of *Cattleya labiata* shoot diameter after 4 weeks of application

Horse Urine LOF	Cow Urine LOF			Average
	S0	S1	S2	
K0	0.71 d	0.99 c	1.52 a	1.074 a
K1	0.71 d	0.87 cd	1.18 b	0.918 b
K2	0.71 d	0.71 d	1.21 b	0.876 b
Average	0.710 c	0.858 b	1.30 a	

Note: The data in the table is the result of the transformation, and numbers followed by the same letter show no significant difference in the Duncan's test 5%

The application of the combination treatment of horse urine and cow urine LOF with the highest concentration (20% horse urine LOF and 20% cow urine LOF = K2S2) only reached a shoot diameter of 1.0 cm at the 4th week. This combination treatment (K2S2) was not significantly different from the combination treatment K1S2 (10% horse urine LOF and 20% cow urine LOF) which can increase the growth of orchid shoot diameter by about 0.9 cm in the 4th week after application. So it can be said that although horse urine LOF and cow urine LOF, and their interactions can significantly increase shoot diameter growth, to obtain the highest shoot diameter growth in this orchid plant only cow urine LOF with a concentration of 20% (K0S2) is needed. This shows that cow urine LOF can be considered as an effective alternative fertilizer in stimulating the speed of cell division, especially in meristem tissue to form plumules and radicles to form orchid plant shoots. The growth of shoots resulting from the application of K0S2 treatment can be seen in Figure 5.

Figure 5. Growth of shoots from mother plant orchids after 4 weeks of application of 20% cow urine LOF.

Liquid organic fertilizer from livestock urine can stimulate plant growth. Because orchid plants in general live and require food from the remains of rotting or weathered plant material, and the growth media is very limited, the cow urine LOF that was applied may have contained sufficient available nutrients needed by the mother plant of the *Cattleya* orchid in the process of formation and growth of shoots when experiencing propagation. So that, cow urine LOF was more effective in increasing shoot growth (number of shoots, shoot height, and shoot diameter) of orchid plants compared to horse urine LOF. According to Gensch et al. (2011), that fertilization increases crop yields only if each plant nutrient supplied is one of the limiting growth factors. If factors other than nutrients are limiting, for example: water, light, pH, salinity, light or temperature, adding more nutrients will not increase yield. Like Liebig's law of minimums: plant growth is controlled not by the total available resource but by the most limited resource. Nutrients can be divided into two categories; macronutrients and micronutrients. The most important major macronutrients are Nitrogen, Phosphorus, and Potassium. Nitrogen is often the most limiting nutrient for plant growth, and the use of N is usually higher than the use of total macronutrients and other micronutrients together. However, the N requirements of the grown crops can be very different.

Nitrogen-deficient plants can be associated with changes in color and shape: the color of the leaves changes to yellow-green, the veins turn white, and the shape is irregularly distorted. When the chlorophyll content in the palisade is depleted, the spongy tissue becomes loose, and the cell gap increases. The absence of phosphorus is associated with upward curling of the leaves and the appearance of white spots at the edges which develop gradually into necrotic spots, while the center remains dark green. The chlorophyll content remains high in the leaves. Without potassium, the new leaves are softer and more yellow, and the tops show a significant change in distortion reaction. Nitrogen supply affected the development of main roots, phosphorus in lateral roots, and potassium on the number of fibrous roots. Especially when nitrogen and potassium elements are lacking at the same time, root development is significantly inhibited, and biomass accumulation is significantly decreased (Yue et al., 2022).

In this study, all shoots that grew from the mother plant of the *Cattleya labiate* orchid physically grew normal and healthy, especially the shoots that grew the best from the application of 20% cow urine LOF treatment without a combination with horse urine LOF (K0S2). The growth of these shoots was the best in terms of morphology compared to all shoots that grew. It is possible that the nutrients contained in the LOF of cow urine are more available and in accordance with the needs for the growth of the orchid plant shoots compared to the LOF of horse urine. Kgasudi and Modiri (2020) also argue that cow urine, apart from being the most effective substance derived from animals, has innumerable therapeutic values. Also useful in agriculture for the manufacture of bio-fertilizers and bio-pesticides. It plays an important role in effective and inexpensive pest control and plant growth/production enhancer (Gottimukkala et al., 2019; Devasena and Sangeetha, 2022).

Cow urine also has antifungal activity so it can be used in fungal control (Jandaik et al., 2015). Devasena and Sangeetha (2022) also stated that nutrient-rich cow urine with nitrogen, potassium, and phosphorus is very beneficial

for soil for dilution and direct application or by formulation and indirect application. In addition to macronutrients, the presence of sulphur, sodium, manganese, iron, enzymes and chlorine make cow urine an indispensable natural pest repellent that requires low external inputs for sustainable agriculture. In a sense, besides being useful in providing nutrients for the growth of shoots of the *Cattleya labiata* orchid plant (where the nutrients N, P, K and pH are more likely to meet the needs of this orchid plant), LOF from cow urine also plays a role in the plant disease control system. as a biological pesticide so as to accelerate the process of shoot growth for the mother plant of the *Cattleya* orchid. So with the application of 20% cow urine LOF without a combination with horse urine LOF (K0S2) this treatment has been very effective in increasing the growth of the number, shoot height of *Cattleya* orchids, especially shoot diameter.

The effectiveness of the LOF of cow and horse urine may also be influenced by various factors, including the type and raw materials used in the manufacture of LOF (such as cow and horse urine), the concentration of EM4, and the fermentation time. According to Hartini et al. (2018), the addition of EM4 can affect the effectiveness of the quality of liquid organic fertilizer. Then Vala and Desai (2021) argue that only 20% of the nitrogenous material consumed by cattle is absorbed and 80% is excreted in the urine and feces. 52% Nitrogen returns in the form of urine while 28% returns in the form of feces. 61-87% phosphorus and 82-92% potassium are also obtained from cow urine. So in this study the application of horse and cow urine LOF can increase shoot growth on the orchid plant *Cettleya labiata*, but the application of cow urine LOF is even more effective.

Conclusions

The application of liquid organic fertilizer (LOF) derived from horse urine and cow urine, and their interactions greatly affect the diameter growth of *Cattleya labiata* Lindl. shoots. The LOF application of cow urine greatly affected the growth in height and number of shoots of the *Cattleya labiata* Lindl., but had no effect on the LOF application of horse urine and its interactions after 4 weeks of application. The best shoot growth was found in the application of 20% cow urine LOF treatment without a combination with horse urine LOF, until the 4th week after application it could reach a shoot height of about 4.8 cm, and a diameter of about 1.8 cm with one leaf. The application of horse and cow urine LOF can increase the growth of orchid shoots, especially the application of cow urine LOF can be part of an alternative technique that is very effective in increasing the growth of the number of shoots, shoot height, and shoot diameter of orchid plants. Horse and cow urine liquid waste can be managed into quality liquid organic fertilizer.

Author Contribution

R.R.S.: Conceptualization, Supervision, Project administration, Writing – review & editing

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