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Research Article

Impact of Lighting Durations on Production, Quality, and Fungal Contamination in Green Fodder

[1]Suci Wulandari, [1]Adib Norma Respati, [1]Satria Budi Kusuma, [2]Amal Bahariawan and [3]Siswanto

[1]*Department of Animal Science, Politeknik Negeri Jember, Jember, Indonesia*
[2]*Agricultural Technology Department, Politeknik Negeri Jember, Indonesia*
[3]*Biology Department, Faculty of Science and Mathematics, Jember University, Indonesia*

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Corresponding Author:
Suci Wulandari
Department of Animal Science,
Politeknik Negeri Jember, Jember,
Indonesia
Email: suci@polije.ac.id

Abstract: This study aimed to identify hydroponic fodder seeds with optimal production and chemical quality and to determine the fungal species contaminating the green fodder produced. This study used a 2´5 Factorial Completely Randomized Design pattern, with 2 replications. Factor I consisted of two lighting treatments: (1) 12 hours of natural sunlight and (2) 24 h of light (natural sunlight followed by LED light at 500-600 nm). Factor II included different plant types: Gramineae (maize and rice) and Leguminosae (mung beans). Significant differences were further analyzed using Duncan's Multiple Range Test (DMRT) for mean comparison. Data were processed using SPSS version 24. Extending the lighting duration to 24 hours did not significantly affect fresh and dry matter production, ether extract content, ash content, Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF), or the Digestibility of Dry Matter (DMD) and Organic Matter (OMD) in the three types of green fodder (maize, mung beans, and rice). The highest Dry Matter (DM) production was observed in green fodder rice, while the highest digestibility was found in green fodder maize. Green fodder mung beans had the highest protein content. The microbiological analysis identified four fungal species (A, B, C, and D) from the genus *Aspergillus* and three species (A, B, and C) from the genus *Acremonium*. Among these, *Aspergillus* B was present in maize, mung beans, and rice.

Keywords: Fungi on Green Fodder, Lighting Durations, Plant Types

Introduction

The main problem that occurs in ruminant livestock farming is the decreasing availability of green fodder land and the shortage of green fodder during the dry season. Green fodder plays a crucial role in ensuring optimal milk production and quality. A limited supply of green fodder can significantly reduce the productivity of ruminant livestock.

This issue is urgent and must be addressed promptly to enhance livestock productivity in Indonesia, particularly for ruminant livestock. Hydroponics is an agricultural system that utilizes liquid media enriched with essential nutrients for plant growth. It offers a viable alternative to overcoming green fodder shortages caused by limited land resources. Fodder refers to all parts of a plant, whether fresh or processed, that are provided to livestock as green fodder (Wulandari *et al.*, 2023). Hydroponic fodder, or green fodder, is produced by growing plants in water or a nutrient-rich solution without the use of soil (Naik *et al.*, 2012).

The advantages of this system include A higher success rate for plant growth and production, minimal water and land usage, shorter growth period, high and continuous yield, richness in essential nutrients and positive impact on livestock (highly digestible, palatable and nutritious) and it requires minimal labour (Ghorbel and Koşum, 2022). The study by Wulandari *et al.* (2024) showed that maize fodder crops achieve the best growth and biomass production with a harvest time of 10 days.

The photoperiod directly affects the time of the plant's photochemical reaction (Li *et al.*, 2023). In general, the longer that plant gets sunlight will be more intensive the photosynthesis process, so the production can be higher. It was stated by Zhang *et al.*, (2018) stated that extending the photoperiod can enhance carbohydrate production, promote growth and stimulate photosensitive pigments in lettuce, thereby inducing relative gene expression and increasing nutrient absorption for improved quality.

A limitation of green fodder is its susceptibility to fungal Growth on the roots, leading to contamination.

According to Li *et al.* (2014), *Colletotrichum* sp., *Fusarium* sp., *Phytophthora* sp., *Pythium* sp., *and Rhizoctonia* sp. are common fungal species found in hydroponic systems. Therefore, it is essential to identify the fungal contaminants that may develop during green fodder production to determine whether they are pathogenic to plants or harmful to livestock that consume them. Proper identification ensures appropriate handling and mitigation strategies.

The purpose of this study was to obtain hydroponic fodder seeds with optimal production and chemical quality and to find out the types of fungi that contaminate the green fodder produced. The novelty of this study lies in providing information on hydroponic fodder seeds that have been tested for both production and chemical quality as alternative ruminant feed, as well as information on the types of fungi that contaminate the green fodder, whether they are pathogenic to plants or harmful to livestock that consume them.

Materials and Methods

Materials

The materials used in this experiment included seeds from the Gramineae family—maize (*Zea mays*) and rice (*Oryza sativa*) and seeds from the Leguminosae family, namely mung beans (*Vigna radiata*). Additional materials included AB Mix liquid mineral fertilizer (Hydroponics Surabaya, Indonesia), a 10% sodium hypochlorite solution (Bayclin brand, SC Johnson, Surabaya, Indonesia), a fungicide (PT. Bayer, Surabaya, Indonesia) and LED lights (λ = 500–600 nm).

Procedures

Preparation of Liquid Fertilizer Solution

The liquid mineral fertilizer solution (AB mix) was prepared by mixing 5 mL of fertilizer solution A and 5 mL of fertilizer solution B into one litre of water.

Seed Preparation

The seeds were first soaked overnight, and any floating seeds were discarded. The remaining seeds were then treated by soaking them in a fungicide solution for 20 min. For storage, plastic trays with perforated bottoms were used to prevent water stagnation. These trays were then placed on each planting rack.

Planting Seeds

Seeds that were ready for transfer to the hydroponic system were first prepared by sanitizing the gutters. The gutters were treated with a 10% sodium hypochlorite solution for 15 min to minimize fungal growth, then rinsed with clean water and aired for 10 min to remove any residual odor.

The seeds were then spread onto planting trays at a density of 0.36 g/cm². The trays were covered with large black plastic and sprayed with water three times a day to maintain humidity for two days until sprouting began. After sprouting, the plastic covering was removed, and the plants were grown openly until harvest.

Fertilization was carried out through spraying, while watering was done daily. The trays were drained three times a day to maintain optimal growing conditions. AB Mix nutrients were applied as fertilizer on days 3, 5 and 7. Additionally, a fungicide was applied at a rate of 2 grams per square meter on day 7.

Harvesting

Harvesting was conducted at the age of 10 days (Wulandari *et al.*, 2023). Furthermore, the harvest results are analyzed according to observation parameters.

Experimental Design

This experiment employed a 2×5 factorial completely randomized design with two replications. Factor I consisted of two lighting durations: 12 h of sunlight and 24 h (sunlight followed by LED light at λ = 500–600 nm) (Matysiak *et al.*, 2021)). Factor II included different plant types: Gramineae (*Zea mays* (maize) and *Oryza sativa* (rice)) and Leguminosae (*Vigna radiata* (mung beans)). Significant differences were analyzed using Duncan's Multiple Range Test (DMRT) for mean comparison. Data processing was conducted using SPSS version 24 software.

Observed Parameters

The following parameters were analyzed in the study: Fodder Production; Proximate analysis of fodder (Latimer, 2023); Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF) (Latimer, 2023); Digestibility: Dry Matter Digestibility (DMD), Organic Matter Digestibility (OMD); Identification of fungi to genus (macroscopic and microscopic) (Arumugam *et al.*, 2024).

Microscopic and Microscopic Observation

Fungal isolation was performed by taking parts of the fodder plant that were attacked by fungi, namely the roots and lower stems (Figure 1B). The fodder sample was then chopped into smaller pieces. Next, 10 grams of fodder sample was taken and put into an Erlenmeyer flask containing 90 mL of distilled water. The Erlenmeyer flask was then tightly closed, and the suspension was shaken for 15 min. The suspension solution obtained was then diluted.

The dilution series was carried out in stages. One millilitre of the suspension solution was pipetted and added to a test tube containing 9 mL of physiological solution, maintaining a 1:9 ratio. The staged dilution used for fungal isolation was a dilution of 10^{-2}-10^{-8}. The isolation method used was the Pour Plate method, where each diluted suspension was poured into a petri dish and incubated for four days. The fungal isolate was ready for macroscopic and microscopic observation.

An ose needle was used to inoculate by scratching the fungus on the surface of Potato Dextrose Agar (PDA). After that, the petri dish was then sealed with an insulating tape to prevent contamination of other microbes. The streaking was performed in a sterile cabinet. The plates were incubated for four days and the streaking process was repeated until a pure culture (single colony) was obtained (Salvamani and Nawawi, 2014). The pure culture obtained was used for macroscopic and microscopic observations.

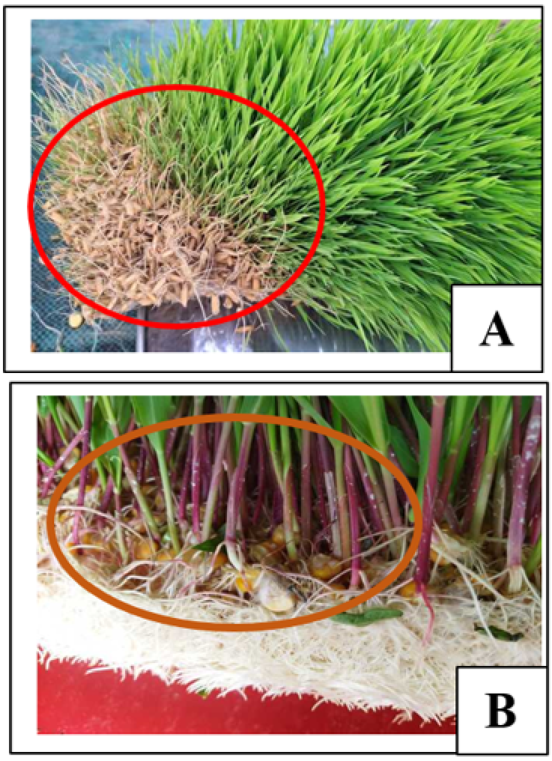


Fig. 1: A. the skin on the roots of rice green fodder; B. The roots and stems of maize green fodder are contaminated with fungi

In this experiment, macroscopic observation was conducted using PDA media. Fungal samples were incubated at room temperature for four days in a single-culture setup. Macroscopic morphological characteristics, including colony colour and culture texture, were the primary parameters observed (Salvamani and Nawawi, 2014).

Microscopic observation was conducted by preparing the object glass, then using the tip of the ose needle smeared with PDA media, and then using the ose needle inoculated to take a little fungus and mix it well with the PDA media. The object glass was then covered with a cover glass and examined under a bright field microscope. The colour, shape, appearance and arrangement of the fungal structure were documented (Salvamani and Nawawi, 2014). Photographs of the structures were taken using a digital camera (Olympus SP-350) for identification purposes.

Results and Discussion

Production of Fresh and Dry Matter

The results showed that there was no interaction between the differences in fodder plant types and lighting duration on fresh and Dry Matter (DM) production. In this case, lighting duration (12 and 24 h) had no significant effect on green fodder production. The thing that affected the difference in production ($p<0.01$) was the difference in fodder plant types (Table 1 and Figure 6).

The results of the study indicated that the highest fresh production was green fodder mung beans, as much as 18.84±1.45 kg/m^2, followed by maize (11.23±1.18 kg/m^2), and the least was rice (36.8±30.0 kg/m^2). However, this condition did not align with the Dry Matter (DM) production, where the highest DM production was actually in green fodder rice (1.89±0.16 kg/m^2), followed by maize (1.58±0.25 kg/m^2), and the smallest was mung beans (1.18±0.17 kg/m^2). This difference is due to mung beans having the highest fresh production, but their water content is also quite high, so low dry matter will be obtained. Dry matter is calculated as fresh weight minus water content (Latimer, 2023; Telling et al., 2023).

Table 1: Fresh and Dry Matter (DM) production of green fodder on different plant types and different lighting durations

Parameter	Hours	Maize	Rice	Mung Beans	Mean
Production Fresh (kg/m^2)	12	11.77±1.23	7.15±0.16	18.87±1.99	12.59±5.14
	24	10.69±0.95	6.80±0.77	18.81±0.89	12.10±5.24
Mean		11.23±1.18^b	36.8±30.0^c	18.84±1.45^a	
DM Production (kg/m^2)	12	1.60±0.15	1.89±0.18	1.23±0.12	1.57±0.31
	24	1.56±0.35	1.89±0.16	1.13±0.20	1.53±0.40
Mean		1.58±0.25^b	1.89±0.16^a	1.18±0.17^c	

Note: Means in the same row with different superscripts differ significantly (P<0.01), DM = Dry Matter

The duration of lighting did not influence fresh or dry material production. This shows that the optimal light provision for the production of green fodder maize, rice and mung beans is 12 h; extending light exposure beyond 12 h proves ineffective. According to Li *et al.* (2023) that excessive light can interfere with photosynthetic tissue and cell membranes, while insufficient light will also limit photosynthesis in the leaf mesophyll (Wang *et al.*, 2024).

In this case, the excess light energy harvested by the plant may lead to photo damage due to the formation of reactive oxygen species. To minimize damage caused by excessive light exposure, green fodder can rapidly dissipate the excess energy (Schiphorst *et al.*, 2023). This condition is a form of adaptation in green fodder, allowing it to regulate irradiation control during a 24-h light period (with sunlight followed by LED light). This adaptation is evident in the study results, which show that fresh and dry matter production under a 24-h light period was relatively maintained and did not differ significantly from green fodder exposed to 12 h of sunlight.

Green Fodder Nutrient Content

The results of the proximate analysis (Table 2) showed a significant interaction ($p<0.01$) between plant type and lighting duration on the Crude Protein (CP) content of the green fodder. The highest CP content was observed in mung bean green fodder under both 12 and 24-h lighting conditions, while the lowest CP content was found in rice green fodder after 24 h of lighting. Notably, in rice, the CP content under 24h lighting was lower than that under 12h lighting. According to Barmudoi and Bharali (2016), white LED light or sunlight (λ = 400-700 nm) with a lighting duration of 12 h is most suitable for rice plants.

The response of plants to long photoperiods is highly genetically dependent; however, exceeding tolerable photoperiod limits can be associated with photooxidative damage to plant leaves (Fayezizadeh *et al.*, 2024). Excessive light exposure can negatively impact amino acid biosynthesis in plants by causing photoinhibition and oxidative stress, which disrupt nitrogen assimilation and redirect resources from amino acid synthesis to stress response mechanisms (Yang *et al.*, 2024). The decrease in Crude Protein (CP) content in rice fodder under 24h lighting reduces its suitability as a green fodder source. As a reference, introduced tropical grasses such as Napier grass contain 15.8–16.8% crude protein (Kamruzali *et al.*, 2021), while *Pennisetum purpureum* cv. Mott has a crude protein content of 15.69–17.51% (Widodo *et al.*, 2019).

In this study, mung beans green fodder produced a fairly high CP of 47.51±4.04% compared to maize 13.29±1.11% and rice 2.50±0.19%. Rayani *et al.* (2021) said that the highest content of green fodder in mung beans is due to mung beans being classified as high-protein grains and being a source of Protein in feed.

Table 2: Green fodder nutrient content in different types of plants and different lighting durations

Parameter	Hours	Maize	Rice	Mung Beans	Mean
Crude Protein (%)	12	12.78±0.45^{b}	8.96±2.26^{c}	46.23±5.20^{a}	22.66±17.60
	24	13.80±1.38^{b}	4.75±0.88^{d}	48.80±2.28^{a}	22.45±19.71
Mean		13.29±1.11	6.86±2.75	47.51±4.04	
Ether Extract (%)	12	4.08±0.19	2.50±0.19	1.45±0.12	2.67±1.13
	24	3.91±0.16	2.34±0.17	1.85±0.11	2.70±0.92
Mean		3.99±0.19^{b}	2.42±0.19^{a}	1.65±0.24^{c}	
Ash (%)	12	3.15±0.86	7.25±0.33	7.75±0.59	6.05±2.21
	24	2.95±0.36	7.42±1.00	7.13±0.16	5.83±2.19
Mean		3.05±0.63^{b}	7.34±0.71^{a}	7.44±0.52^{c}	

Note: Different superscripts in the same column/row indicate significant differences

Maize is the primary energy source in animal feed, followed by rice. This is also reflected in the Ether Extract (EE) content, where maize green fodder had the highest EE at 3.99±0.19%, followed by rice at 2.42±0.19% and mung beans at 1.65±0.24%.

There was no interaction in the Ether Extract (EE), but differences in plant types affected the EE of the green fodder produced, likely because both 12 h of sunlight and 24 h of lighting (sunlight followed by LED light) provided sufficient photosynthetic energy. In cases of energy deficiency, plants compensate by utilizing Ether Extract (EE) or carbohydrates, which serve as energy reserves essential for protein synthesis and Growth (Herchi *et al.*, 2015).

In general, the Ether Extract (EE) content of green fodder across all treatments is still adequate for ruminant livestock. The general recommendation is that total dietary fat should not exceed 7% of dietary dry matter, as high-fat levels in ruminant diets can negatively impact microbial fermentation (Cho *et al.*, 2023).

Maize green fodder has the lowest ash content because it does not retain any skin on the roots of the green fodder produced, unlike rice and mung beans, which still leave the skin on the roots (Figure 1A). According to Nnadiukwu *et al.* (2023), rice husks are rich in minerals such as calcium, magnesium, potassium, sodium, iron, selenium, zinc cobalt, manganese, phosphorus and copper. The results of the proximate analysis showed that mung bean skins in 100% dry matter contain 2.40% ash (Rukmini *et al.*, 2023). Additionally, Nagrale *et al.* (2018) note that mung beans are not only a source of Protein but also rich in the minerals Ca, Mg, Fe, P and K.

Differences in plant types affected ($p<0.01$) the Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF) content of the green fodder produced, but the effect of lighting duration did not have a significant effect (Table 3).

Table 3: Content of Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF) of green fodder on different plant species and different lighting durations

Parameter	Hours	Maize	Rice	Mung Beans	Mean
NDF (%)	12	32.22±2.47	39.90±1.53	32.63±1.69	34.92±4.07
	24	35.53±2.54	41.20±3.95	32.77±3.17	36.50±4.73
Mean		33.88±2.93^{a}	40.55±2.91^{b}	32.70±2.40^{a}	
ADF (%)	12	16.22±1.52	26.80±1.94	32.76±1.21	25.26±7.23
	24	17.44±2.06	28.76±4.34	28.78±2.93	24.99±6.29
Mean		16.83±1.82^{c}	27.78±3.33^{b}	30.77±2.98^{a}	

Note: Means in the same row with different superscripts differ significantly (P<0.01), NDF = Content of Neutral Detergent Fiber, ADF = Acid Detergent Fiber

In general, the Neutral Detergent Fiber (NDF) content of green fodder for maize, rice and mung beans is generally considered good quality for feed; this is because the highest NDF value is 40.55±2.91% (rice green fodder). According to Bulcha *et al.* (2022), feed containing an NDF value of less than 45% is classified as high quality, while feed with a value of 45-65% is classified as medium quality and feed with a value above 65% is classified as low quality.

The interesting finding from this study is that the NDF content of mung bean green fodder is still lower than that of maize, but the Acid Detergent Fiber (ADF) content of mung beans is actually the highest (30.77±2.98%), compared to rice (27.78±3.33%) and maize (16.83±1.82%). This is possible due to the structural wall component (cellulose) formed being the most compared to the others. This is supported by the results showing the most lush mung beans, green fodder, and the highest fresh production (Table 1). The main component of ADF is cellulose. The main components that form cell walls are structural carbohydrates, namely cellulose and hemicellulose (Wulandari *et al.*, 2014).

Providing light can increase carbohydrate production, enhance growth and stimulate pigment photosynthesis (Zhang *et al.*, 2018). The increase in green fodder growth is associated with the formation of structural carbohydrates, which are components of crude fibre. However, in this study, it appears that green fodder harvested at 10 days and exposed to continuous light for up to 24 h did not show an increase in crude fibre compared to green fodder with 12 h of light exposure.

This suggests that a 12 h light period is the most stable for the growth and production of structural carbohydrates in maize, rice and mung bean green fodder. Exposure to light for more than 12 h appears to inhibit structural carbohydrate production. Similarly, Li *et al.* (2023) found that a 12-h light period had the most significant inhibitory effect on cellulose production in the green fodder of arugula plants. These findings indicate that light treatment with certain intensities and photoperiods can inhibit cellulose production.

Differences in plant types significantly affected (P<0.01) Dry Matter Digestibility (DMD) and Organic Matter Digestibility (OMD) under both 12 and 24 h lighting conditions (Table 4).

The nutrient utilization of hydroponic fodder in the rumen can be assessed using *in vitro* method. The *in vitro* feed evaluation technique is commonly used to measure and predict feed ingredients' digestibility, their effect on rumen fermentability and their impact on rumen microbial growth (Ndaru *et al.*, 2021). This study shows that the lighting duration of 12 and 24 h generally does not affect the nutrient content of green fodder, especially its crude fibre content, so it does not affect Dry Matter Digestibility (DMD) and Organic Matter Digestibility (OMD). As stated by Harwanto *et al.* (2022), nutrient digestibility is influenced by nutrient content. Increasing crude fibre will decrease feed digestibility because crude fibre here consists of cellulose and hemicellulose, which are sometimes accompanied by lignin, so they are difficult for the enzyme system to digest. It is further stated that nutrient digestibility is also influenced by the type of plant fodder. This is in accordance with the results of the study, which show significant differences between fodder types in terms of DMD and OMD.

Table 4: Dry Matter Digestibility (DMD) and Organic Matter Digestibility (OMD) of green fodder on different plant species and different lighting durations

Parameter	Hours	Maize	Rice	Mung Beans	Mean
DMD (%)	12	79.40±3.81	63.89±2.31	73.44±1.73	72.24±7.09
	24	79.53±2.89	63.10±3.94	73.00±1.64	71.88±7.51
Mean		79.46±3.19^{a}	63.50±3.07^{c}	73.22±1.61^{b}	
OMD (%)	12	82.94±3.71	65.60±2.49	75.39±1.80	74.64±7.79
	24	82.36±2.77	65.03±4.42	75.04±1.38	74.15±7.90
Mean		82.65±3.10^{a}	65.31±3.40^{c}	75.22±1.52^{b}	

Note: Means in the same row with different superscripts differ significantly (P<0.01), DMD = Dry Matter Digestibility, OMD = Organic Matter Digestibility

Among the three types of green fodder, maize has the highest DMD and OMD values. This is because maize green fodder does not retain skin on its roots, resulting in a lower Acid Detergent Fiber (ADF) content. ADF consists of cellulose and lignin, which are difficult for rumen microbes in ruminant livestock to digest (Wulandari *et al.*, 2014).

Identification of Fungi (Genus)

The results of macroscopic and microscopic fungal identification showed that four types of fungi were growing on maize green fodder infected with the disease, specifically on the roots and stems at a harvest age of 10 days. A total of four types of fungi were identified, belonging to the genera *Aspergillus* sp. and *Penicillium* sp. (Fig. 2).

The *Aspergillus* genus is commonly found infecting grains such as maize, aligning with findings by Zulkifli and Zakaria (2017), who identified seven types of *Aspergillus*, including *Aspergillus flavus*, which closely resembles the findings of this study. *Aspergillus* sp. is also frequently found in rice grains (Mawarni *et al.*, 2021) and peanuts (Atallah *et al.*, 2022) and is characterized by its distinctive stipe and conidia structures (Figure 5).

The *Acremonium* genus is widely present in soil, as demonstrated by Park *et al.* (2017), who successfully isolated it from soil samples. Additionally, this genus has also been found in aquatic environments (Lee *et al.*, 2023). *Acremonium* is characterized by its light brown colonies and distinctive spores, similar to those identified in this study (Fig. 5).

Although the seeds used for planting green fodder were soaked in a fungicide solution and sprayed with fungicide on the seventh day, fungi were still detected on the roots and lower stems in amounts of less than 1%. There were three types of fungi from the genus

Acremonium (A, B, C) found in 10-day-old green fodder and four types of fungi from the genus *Aspergillus* (A, B, C, D). The warm, humid, windless and rain-free conditions typical of hydroponic systems may contribute to a favourable environment for fungal Growth (Sudiartini *et al.*, 2021).

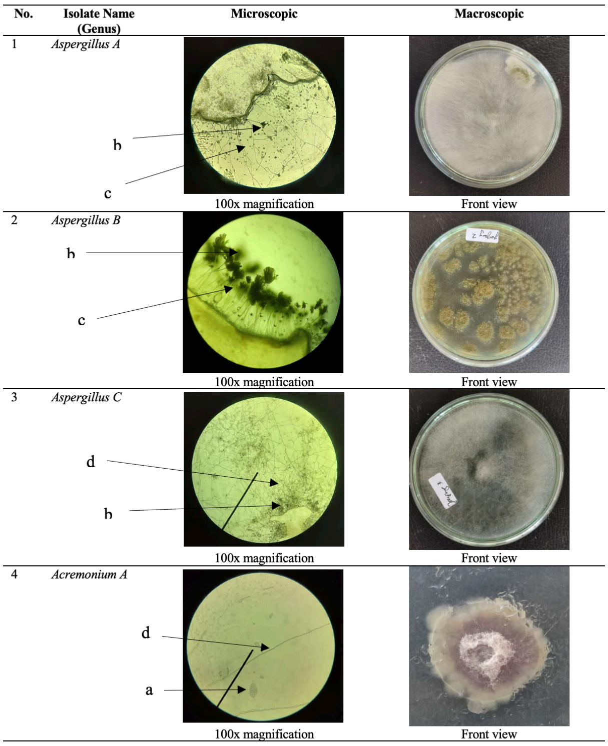


Fig. 2: Fungi growing on maize green fodder; a: Spore; b: Conidia; c: Stipe; d: Hyphae

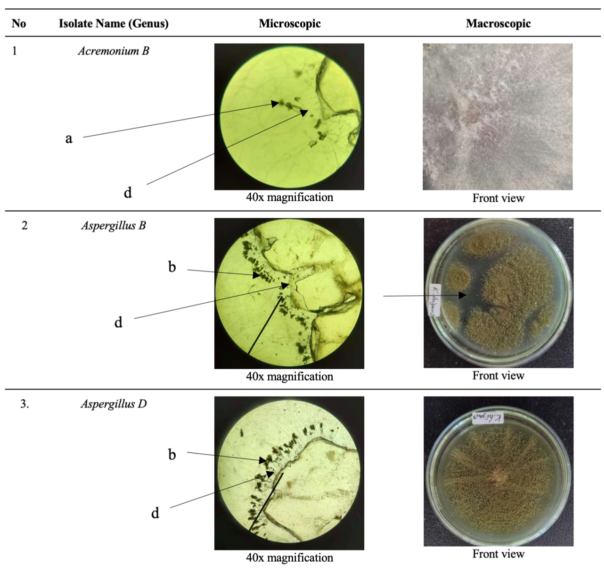


Fig. 3: Fungi growing on mung beans green fodder; a: Spore; b: Conidia; c: Stipe; d: Hyphae

The identification results of fungi contaminating green fodder on mung beans are presented in Figure (3). They showed that there were three types of fungi isolated from the roots and stems of diseased green fodder. Of these, one fungus belongs to the genus *Acremonium sp.*, while the other two belong to the genus *Aspergillus sp.*

The identification results of contaminated rice green fodder showed that there were two types of fungi that could be isolated and identified at the genus level. One fungus belongs to the genus *Acremonium sp.* and the other to the genus *Aspergillus sp.* (Fig. 4).

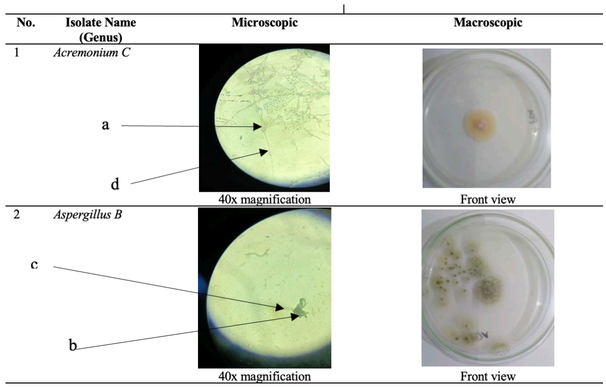


Fig. 4: Fungi growing on rice green fodder; a: Spore; b: Conidia; c: Stipe; d: Hyphae

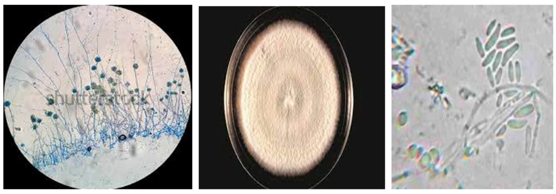
Aspergillus B was found in maize, mung beans and rice, as indicated by macroscopic observations showing consistent color and colony type. For *Acremonium* found in maize, mung beans and rice. However the shape of the colonies was different, so it was suspected that these *Acremonium* belong to different species.

Although the seeds used for planting green fodder were soaked in a fungicide solution and sprayed with fungicide on the seventh day, fungi were still detected on the roots and lower stems in amounts of less than 1%. There were three types of fungi from the genus *Acremonium* (A, B, C) found in 10-day-old green fodder and four types of fungi from the genus *Aspergillus* (A, B, C, D). The observations showed that the fungi growing on rice green fodder belonged to two genera: *Acremonium* C and *Aspergillus* B. Meanwhile, the highest fungal diversity was found on maize green fodder, with four types: *Aspergillus* A, *Aspergillus* B, *Aspergillus* C and *Acremonium* A (Table 5). The warm, humid, windless and rain-free conditions typical of hydroponic systems may contribute to a favourable environment for fungal Growth (Sudiartini *et al.*, 2021).

The results of isolating the roots and stems of green fodder maize, rice and mung beans that were affected by disease revealed the characteristics of the genus *Acremonium* sp. and *Aspergillus* sp. The genus *Acremonium* sp. found in the roots and stems of green fodder (maize, mung beans and rice) appeared to have different colonies for each plant type, suggesting that the *Acremonium* sp. from each plant belongs to different types. Generally, the genus *Acremonium* sp. has a brownish-yellow colony colour and a round colony shape like cotton. According to Irvanto and Listyowati

(2024), the morphology of *Acremonium* sp. is white, with the edges becoming increasingly dark orange with age. The results of microscopic observations show that the hyphae are hyaline, not septate, and the conidia and stipe form are rather long and branched. This mould is generally hyaline in appearance. Pangesti *et al.* (2022) describe its microscopic structure as consisting of branched and non-septate hyphae, hyaline with a smooth, single-celled, elliptical conidia surface. This genus grows optimally at temperatures between 28-30°C.

Acremonium sp. fungi are partially phytopathogenic (pathogenic to plants) and can grow on plant tissue (endophytes) at optimum temperatures (Firmansyah *et al.*, 2022). *Acremonium* sp. fungi that were successfully isolated from the roots of oil palm seedlings are pathogens of white mustard plants (Irvanto and Listyowati, 2024). In general, fungi produce organic acids, such as oxalic acid, acetic acid and lactic acid (Pangesti *et al.*, 2022). This will cause a sour odour and taste in green fodder if fungal growth is not properly controlled.



a. *Aspergillus* b. *Acremonium*

Fig. 5: Isolated *Aspergillus* and *Acremonium* according to reference (www.google.com) (Lee *et al.*, 2023)

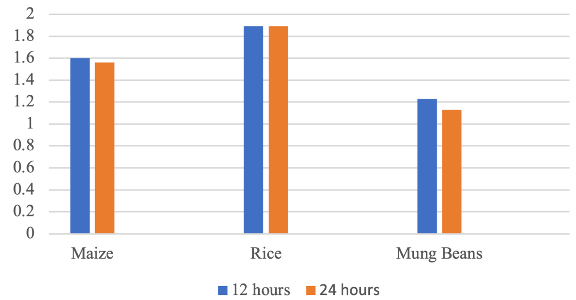


Fig. 6: Graphical impacts photoperiod on dry matter production

Aspergillus type B is found in green fodder maize, mung beans and rice. The fungi that grow on fodder maize are dominated by *Aspergillus* sp. *Aspergillus* A and C fungi macroscopically appear brownish white, while *Aspergillus* B and D are yellowish green. Microscopic observations revealed non-septate hyphae, round conidia (spores), unbranched conidiophores and hyaline colouration. According to Arumugam et al. (2024), *Aspergillus* sp. colonies initially appear as white filaments and change colour depending on the species. Microscopically, they exhibit round conidia and hyaline stipe with septate hyphae (Mawarni *et al.*, 2021). Arafat *et al.* (2022) reported that macroscopically *Aspergillus flavus* colonies in the PDA agar were observed as green, light green and dark yellowish green. *Aspergillus fumigatus* showed a green powdery surface with white to yellow, and the *Aspergillus terreus* colonies in the PDA agar were observed to be dark brown. According to Sudipa *et al.* (2023), *Aspergillus* sp colonies typically grow rapidly and range in colour from white, yellow and yellowish-brown to brown, black, or green.

Table 5: Types of isolate genus fungi in green fodder

Type plants	Isolate Genus Fungi
Maize	*Aspergillus A* *Aspergillus B* *Aspergillus C* *Acremonium A*
Rice	*Aspergillus B* *Acremonium C*
Mung Beans	*Aspergillus B* *Aspergillus D* *Acremonium B*

Aspergillus sp. has the potential to secrete organic acids such as formic, acetic, propionic, lactic, glycolic, fumarate and succinic acids. These organic acids function to dissolve phosphate elements from fertilizers or soil so that they are easily absorbed (Mawarni et al., 2021). The impact is that fodder contaminated with *Aspergillus* sp. disease will smell and taste sour, which can reduce palatability to livestock. Ruminant livestock naturally have the highest level of palatability to the smell and taste of fresh greens (Din *et al.*, 2022).

Aspergillus sp. in seeds can reduce germination and accumulation of mycotoxins in the material (Hausufa and Rusae, 2018). Feed materials contaminated by fungi can affect the health of livestock that consume them. Consequently, if humans consume products derived from these animals, their health may also be impacted by the accumulated mycotoxins (Puspitasari et al., 2015). *Aspergillus fumigatus* is the most common cause of diseases in humans with an immunity deficiency (Sousa Terada-Nascimento *et al.*, 2023).

Conclusion

Extending the lighting period to 24 h did not affect the fresh and dry matter production, ether extract content, ash, Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF), as well as Dry Matter Digestibility (DMD) and Organic Matter Digestibility (OMD) of the three types of green fodder (maize, mung beans and rice). Among the fodder types, rice green fodder had the highest Dry Matter (DM) production, while maize green fodder exhibited the highest digestibility. Mung bean green fodder was notable for having the highest protein content. The microbial analysis revealed four types of fungi (A, B, C and D) from the genus *Aspergillus* and three types (A, B and C) from the genus *Acremonium* growing in the three types of green fodder. Notably, *Aspergillus* type B is consistently found in all three fodder types. Rice green fodder demonstrated the highest dry matter production and the greatest resistance to fungal contamination, with only two fungal types detected (*Acremonium* C and *Aspergillus* B), whereas maize green fodder had the highest fungal diversity, with four types identified (*Aspergillus* A, *Aspergillus* B, *Aspergillus* C and *Acremonium* A).

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Author's Contributions

Suci Wulandari: Coordinated the implementation of research work, conducted research, contributed manuscript preparation, compiled the literary review, analyzed and interpreted the study findings, and drew conclusions.

Adib Norma Respati: Conducted research, designed the research plan, organized the study and contributed to result analysis and the writing of the manuscript.

Satria Budi Kusuma: Conducted research, analyzed and interpreted the results and contributed manuscript revisions.

Amal Bahariawan: Conducted research and contributed manuscript revisions.

Siswanto: Identification and analysis of fungi.

Ethics

This article is original and contains unpublished material. The corresponding author confirms that all of the other authors have read and approved the manuscript and there are no ethical issues involved.

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