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# Inhibitory Effects of Bamboo Leaf on the Growth of *Pyricularia grisea* Fungus

**Nguyen Phu Toan [1], Tran Dang Xuan [1,*], Pham Thi Thu Ha [1,2], Truong Thi Tu Anh [1] and Tran Dang Khanh [3]**

1 Graduate School for International Development and Cooperation (IDEC), Hiroshima University, 1-5-1 Kagamiyama, Higashi-Hiroshima, Hiroshima 739-8529, Japan; nguyenphutoan1983@gmail.com (N.P.T.); phamthithuhabt@gmail.com (P.T.T.H.); tuanhbp@gmail.com (T.T.T.A.)

2 Faculty of Applied Science, Ton Duc Thang University, 19 Nguyen Huu Tho street, Tan Phong Ward, District 7, Ho Chi Minh City 700000, Vietnam

3 Agricultural Genetics Institute, Pham Van Dong, Tu Liem, Hanoi 123000, Vietnam; tdkhanh@vaas.vn

* Correspondence: tdxuan@hiroshima-u.ac.jp; Tel./Fax: +81-82-424-6927

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**Abstract:** In this study, the effects of bamboo leaf were examined on mycelial growth of *Pyricularia grisea*, a fungus of rice blast disease that causes a great loss in rice production. The hexane extract exhibited maximal reduction on growth of *P. grisea* ($IC_{50}$ = 0.62 mg/mL), followed by aqueous and ethyl acetate extracts, while the methanol extract was least effective ($IC_{50}$ = 9.71 mg/mL). At 0.5–1.0 mg/mL doses, all extracting solvents showed inhibition on the growth of *P. grisea*, but at a 0.1 mg/mL concentration, the antifungal activity was solely observed on hexane and ethyl acetate extracts. By GC-MS (gas chromatography-mass spectrometry), 25 constituents were identified, principally belonging to long-chain fatty acids, sterols, phenols, phenolic acids, volatile oils, and derivatives of terpenes. It was suggested that compounds originated from hexane and ethyl acetate extracts such as fatty acids, oils, and phenols, and their derivatives were responsible for the antifungal activity of bamboo leaf. Non-polar constituents were accountable for the antifungal activity, although water-soluble compounds may play a role. Bamboo leaf appears to be a potent natural source to manage the infestation of *P. grisea* in rice cultivation.

**Keywords:** bamboo leaf; extracting solvents; *Pyricularia grisea*; antifungal activity; mycelial growth; GC-MS; inhibition

## 1. Introduction

Fungal diseases have caused extensive yield loss in many crops and thus have become a major challenge for agricultural production [1]. Rice blast is caused by a highly variable fungal pathogen, *Pyricularia grisea.* It is a serious disease, affecting rice yield with up to 70–80% losses [2]. The use of rice varieties resistant to this fungus has been shown to be effective. However, the resistance levels governed by the resistance genes have been reported to be inherently unstable and fast-evolving [3,4]. On the other hand, the application of synthetic chemical fungicides has certain advantages, but they also apparently result in environmental and health problems. In recent years, the use of natural products as environmentally friendly agricultural chemicals to manage plant diseases, including the biological control on *P. grisea*, has been targeted to a greater extent.

It was documented that the crude extract of *Epicoccum* sp., the leaf extracts of *Prosophis juliflora*, and *Ziziphus* sp inhibited the mycelial growth of *P. grisea* [5,6]. In addition, extracts from *E. aromatica*, *P. guineense*, and *G. kola* could serve as bio-fungicides against the growth of *P. grisea* [7]. Several other

plants have been tested and showed significant results against *P. grisea* [7–9]. However, the application of plant extracts in agricultural practice is labor-intensive and costly. Therefore, the search for bioactive chemicals from plants against the fungus *P. grisea* is much desired.

Bamboo grows abundantly in the subtropics and tropics. It belongs to the family Poacaea (grass), sub-family Bambusoideae, and tribe Bambuseae. *Phyllostachys pubescens* is the major species of bamboo in Japan and is widely distributed through the country [10]. Bamboo might be made up of rich phytochemicals, which should be investigated and exploited [11]. Gong et al. [12] reported that the 1-butanol fraction of bamboo leaf possessed strong antioxidant activity and high exhibition of total phenol and flavonoid contents, and various phenolic acids. Moreover, 1-butanol, ethyl acetate, and water fractions of bamboo extracts expressed a high capacity for anti-oxidation. Caffeic acid was the principal compound that exerted the highest antioxidant capacity on DPPH (2,2-diphenyl-1-picrylhydrazyl) and FRAP (Ferric-Reducing Antioxidant Power) assays [12]. Similarly, polysaccharides extracted from Moso bamboo-shoot [13] and bamboo leaf [14] also showed strong significant antioxidant activity. In addition, the leaf and root extracts of bamboo exerted inhibitory effects on growth of groundnut and corn seedlings [15,16]. The pharmacological properties, including anti-inflammatory, anticancer, and antibacterial activities of bamboo have been described [12,17]. From this evidence, it is proposed that bamboo leaf may possess potent antifungal activity and may be successfully applied in the management of plant pathogens. This study was therefore carried out to evaluate whether bamboo leaf can be utilized for the biological control of *P. grisea*, a problematic fungus in rice production. The analysis of potent phytochemicals in bamboo leaf was also conducted.

## 2. Materials and Methods

### 2.1. Collection and Preparation of Bamboo Leaf Extract

The leaves were collected from different bamboo trees grown near Higashi Hiroshima campus of Hiroshima University, Japan in September 2016, and air-dried for two weeks. The leaf powder (200 g) was made by a grinder and immersed to methanol for three days at room temperature (23–25 °C). After filtration and vacuum evaporation, the precipitate was successively partitioned into solvents with different polarities, including hexane, ethyl acetate, 1-butanol, and water. The obtained extracts were separately evaporated and diluted to different concentrations for further tests and analyses.

### 2.2. GC-MS Analysis

Each extract, obtained as described above, was evaporated, filtered, and diluted in acetone at 10 ppm for analysis by a GC-MS instrument (JMS-T100 GCV, JEOL Ltd., Tokyo, Japan), which included a DB-5MS column (30 m, 0.25 mm, 0.25 µm) (Agilent Technologies, J & W Scientific Products, Folsom, CA, USA.). Helium was used as the carrier gas and the split ratio 5:1 was employed. The initial temperature was 50 °C without hold time; the programmed rate was 10 °C for 1 min up to final temperature of 300 °C for 20 min of hold time. The injector and detector temperatures were set to 300 °C and 320 °C, respectively. The mass spectra were scanned from 29 to 800 amu. Clarification on the mass spectrum of GC-MS on each identified compound was done using the JEOL's GC-MS Mass Center System software, version 2.65a (JEOL Ltd., Tokyo, Japan)

### 2.3. Mycelia Growth Inhibition Test

The isolate (U61-i0-k101-z05-ta102) of *P. grisea*, collected from wild rice *(Oryza rufipogon)* grown in Southern Vietnam according to a method described in Hayashi et al. [18], was used. The fungal isolate was maintained on Potato Dextrose Agar (PDA) at 28 °C ± 2 °C. Three concentrations of 1 mg/mL, 0.5 mg/mL, and 0.1 mg/mL were prepared in sterilized distilled water. An aliquot of 1 mL of each extract or fraction was added directly into 10 mL of PDA medium and poured into 9 cm diameter sterile Petri dishes, then a 2 $mm^2$ of mycelium agar disc was placed in the center of the Petri dish. The dishes were then incubated at 28 °C ± 2 °C in an incubator. For the controls, the fungal

inoculum was treated with distilled water. After incubating for three days, the colony diameters of the treatments and controls were measured diametrically. The inhibition was calculated according to the following equation: I = ((C − T)/C) × 100%, where I is the inhibition (%), C is the colony diameter of mycelium from the control, and T is the colony diameter of the treatments [19]. The inhibition of different extracting solvents was also compared to the $IC_{50}$ value, which is the concentration (mg/mL) required to inhibit 50% mycelial growth of *P. grisea*. The zone of inhibition and the correspondent levels of antifungal activity on *P. grisea* were shown in Table 1.

**Table 1.** Zone of inhibition and correspondent levels of inhibitory activity.

| Zone of Inhibition (mm) | Inhibitory Activity Level |
|---|---|
| >17 | +++, strong |
| 12–16 | ++, moderate |
| 7–11 | +, weak |
| 6–0 | −, negative |

### 2.4. Statistical Analysis

The data for mycelial growth inhibition test was analyzed by analysis of variance (ANOVA), and the mean values were compared by the Duncan's Multiple Range Test using IBM SPSS Statistics for Windows, version 20.0 ($p < 0.01$) (IBM cooperation, Armonk, New York).

## 3. Results

### 3.1. Chemical Profiles Identified by GC-MS

In this study, GC-MS was used to analyze and identify the chemical components in different extracts, as shown in Table 2 and Figures S1–S5. There were 25 constituents identified, principally belonging to long-chain fatty acids, volatile oils, sterols, phenols, derivatives of terpenes, and amines. In details, the methanol extract included 17 compounds, whilst the number of constituents identified in hexane, ethyl acetate, 1-butanol, and water were eight, eight, two, and one, respectively (Table 2). Methanol was the most effective solvent to extract phytochemicals in bamboo leaf, followed by hexane and acetate, while 1-butanol and water showed the least efficacy.

**Table 2.** Chemical profile in different extracting solvents identified by GC-MS.

| Peaks Number | Extracting Solvents | | | | | Chemical Class |
|---|---|---|---|---|---|---|
| | Methanol | Hexane | Ethyl Acetate | 1-Butanol | Water | |
| 1 | Propanoic acid, 2-oxo-, methyl ester | - | - | + | + | Long chain fatty acid |
| 2 | Glycerin | - | + | + | - | Polyol |
| 3 | Coumaran | + | + | + | - | Colorless oil |
| 4 | Phytol | - | - | - | - | Acyclic diterpene alcohol |
| 5 | *n*-Hexadecanoic acid | - | + | - | - | Long chain fatty acid |
| 6 | Phytol | + | + | - | - | Diterpene alcohol |
| 7 | cis, cis, cis-7,10,13-Hexadecatrienal | - | - | - | - | Volatile oil |
| 8 | Allene | - | - | - | - | Polyene |
| 9 | Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester | - | - | - | - | Long chain fatty acid |

**Table 2.** *Cont.*

| Peaks Number | Extracting Solvents | | | | | Chemical Class |
|---|---|---|---|---|---|---|
| | Methanol | Hexane | Ethyl Acetate | 1-Butanol | Water | |
| 10 | Cis, cis, cis-7,10,13-Hexadecatrienal | - | - | - | - | Volatile oil |
| 11 | 1-Tridecyn-4-ol | - | - | - | - | Volatile oil |
| 12 | Campesterol | - | - | - | - | Sterol |
| 13 | Stigmasterol | | - | - | - | Sterol |
| 14 | γ-Sitosterol | + | + | - | - | Sterol |
| 15 | β-Amyrin | + | + | - | - | Triterpene |
| 16 | Urs-12-en-24-oic acid, 3-oxo-, methyl ester, (+)- | + | - | - | - | Triterpene |
| 17 | Phytol, acetate | - | - | - | - | Acyclic diterpene |
| 18 | - | Hydrazinecarboxamide | | | - | Amino urea |
| 19 | - | Hexadecanoic acid, methyl ester | - | - | - | Long chain fatty acid |
| 20 | - | Methylamine | - | - | - | Amine |
| 21 | - | - | Phenol | - | - | Phenol |
| 22 | - | - | Benzaldehyde, 4-hydroxy- | - | - | Phenolic acid |
| 23 | - | - | - | Pentane, 3-methyl- | - | |
| 24 | - | - | - | 4H-Pyran-4-one, 5-hydroxy-2-methyl- | - | Phenol |
| 25 | - | - | - | - | Ketene | Simple ketene |

GC-MS: gas chromatography-mass spectrometry; +: detected; -: not detected.

### 3.2. Inhibitory Activity on Mycelia Growth of *P. grisea*

The antifungal activities of bamboo leaf in different extracting solvents on *P. grisea* were examined (Table 3; Figures S6–S10). It was found that the methanol extract was inactive on growth of *P. grisea* at all concentrations. At the lowest dose (0.1 mg/mL), the extracts of 1-butanol and water also showed negative results. However, the antifungal activity of ethyl acetate was weak, whilst that of hexane exhibited a moderate level of inhibition (Table 3). The aqueous extract at 0.5–1.0 mg/mL revealed strong inhibition on mycelial growth of *P. grisea* (Table 3).

**Table 3.** Antifungal activity of bamboo leaf extracts on mycelial growth (mm) of *P. grisea.*

| Extracts | Concentrations (mg/mL) | | | | | |
|---|---|---|---|---|---|---|
| | 0.1 | Level | 0.5 | Level | 1.0 | Level |
| M | −0.967 g | − | 0.367 g | − | 0.33 g | − |
| H | 16.70 de | ++ | 19.70 d | +++ | 38.70 ab | +++ |
| E | 9.033 f | + | 38.033 abc | +++ | 31.367 c | +++ |
| B | 1.700 g | − | 23.033 d | +++ | 34.367 bc | +++ |
| W | 6.367 fg | − | 12.367 ef | ++ | 44.367 a | +++ |

+++: Strong; ++: Moderate; +: Weak; −: Negative; M: methanol; H: hexane; E: ethanol; B: 1-butanol; W: water. Means were the diameter of the mycelial growth (mm) of *P. grisea*. Means with similar letters are not significantly different at $p < 0.01$.

The antifungal activity of different extracting solvents was also compared using the $IC_{50}$ value, which was the concentration required to inhibit 50% growth of *P. grisea*. Thus, the lower $IC_{50}$ value reflected stronger antifungal activity. Table 4 showed that the level of inhibition was the maximal with hexane (0.62 mg/mL), followed by water (0.70 mg/mL), and ethyl acetate (0.72 mg/mL),

whilst methanol showed the least activity (9.71 mg/mL). Statistically, the inhibitory levels between ethyl acetate and water were not significantly different.

**Table 4.** $IC_{50}$ values of different extracts on mycelial growth of *P. grisea.*

| Extracting Solvents | $IC_{50}$ (mg/mL) |
|---|---|
| Methanol | 9.71 [a] |
| Hexane | 0.62 [d] |
| Ethyl acetate | 0.72 [c] |
| 1-butanol | 0.81 [b] |
| Water | 0.70 [c] |

Means with similar letter in a column were not significantly different ($p < 0.01$). $IC_{50}$ is the concentration (mg/mL) required to inhibit 50% mycelial growth of *P. grisea.*

## 4. Discussion

Many rice pathogens have shown resistance against a number of the fungicides currently available [20]. The use of plant extracts to control the infestation of fungi on rice was potent and environmentally friendly compared with synthetic fungicides [21]. In a previous work, Hubert et al. [8] reported that aqueous extracts of *Coffee arabica* at 10% and 25% (*v*/*v*) exhibited 81.12 and 89.40% inhibition on *P. grisea*, respectively. Suriani et al. [22] documented that the crude extract of *Piper canium* inhibited mycelial growth of *P. grisea*. Several phenolic acids including caffeic, chlorogenic, and ferulic acids were identified in bamboo shaving extracts and were found to correlate to antioxidant activity [12]. Several C-glycosyl flavonones such as orientin, isoorientin, vitexin, and isovitexin [23], and phenolic acids including caffeic, chlorogenic, and ferulic acids [12] were detected in bamboo leaf extract and correlated to antioxidant activity. Bamboo shoot exerted antibacterial (*S. aureus*, *B. subtilis*, *P. aeruginosa*, and *E. coli*) and antifungal (*Fusarium oxysporum*) properties. Chemicals including tannins, steroids, phenols, glycosides, flavonoids, carbohydrates, and proteins were identified and were responsible for the antimicrobial activity [24]. Amadioha [25] reported that the growth of rice blast fungus was significantly suppressed by water and ethanol extracts of the leaves and oil extracts of neem seeds.

Among the compounds identified by GC-MS (Table 2), phytol has been reported to have antimicrobial [26,27] and anti-inflammatory activities [28]. Shukla and Dwivedi [29] described the inhibitory effect of phenol against growth of *Fusarium oxysporum.* Salehan et al. [30] noted that an ethyl acetate extract of *Cosmos caudatus* suppressed the spore formation of *Phytophthora palmivora*, which caused black pod disease of cocoa. In this study, hydrazinecarboxamide, a compound exhibiting antibacterial [31] and antimycobacterial [32] activities, was detected in the hexane extract. Recently, some novel chemicals have been synthesized such as azaloes and *N*-(substituted)-2-isonicotionoylhydrazinocarbothioaide which showed promising antimicrobial activities [33,34]. However, many plant extracts were effective against pathogenic fungi of cucumber [35], and some other economically important crops including rice, soybean, tomato, sesame, potato, and watermelon [30,36], suggesting that natural products were more favorable. In this study, among the different extracting solvents, hexane was the most active, followed by ethyl acetate and water. 1-Butanol showed negligible inhibition as compared with ethyl acetate and water, whereas methanol did not show any antifungal activity (Tables 3 and 4; Figures S6–S10).

Although the chemicals identified by GC-MS were most abundant in the methanol extract (17 compounds), the other extracting solvents included fewer constituents (eight, eight, two, and one for hexane, ethyl acetate, 1-butanol, and water, respectively) (Table 2). It was concluded that the number of chemicals identified was not proportional to the level of antifungal activity observed on *P. grisea* (Tables 2–4). The polarities of hexane, ethyl acetate, 1-butanol, methanol, and water were 0.009, 0.228, 0.586, 0.762, and 1.000, respectively. Both hexane and ethyl acetate showed strong inhibition on the growth of the harmful fungus *P. grisea*, indicating that non-polar compounds in bamboo leaf

might be responsible for the antifungal activity. Based on the GC-MS results, the presence of the identified constituents was not water-soluble (Table 2). It appears that water-soluble compounds have not yet been detected by the use of GC-MS in this study. Because the ethyl acetate and water extracts showed similar antifungal activity (Table 4), it was proposed that water-soluble constituents may also be involved in the inhibition of bamboo leaf on *P. grisea*. Further analytical instruments such as LC-MS (liquid chromatography-mass spectrometry), and CC (column chromatography) should be applied to isolate and identify potent water-soluble chemicals in bamboo leaf. In addition, the inhibition of each chemical identified by this study, as well as their mixture, on growth of *P. grisea* should be investigated.

Although hexane, ethyl acetate, and butanol were effective in the search for potent phytochemicals, the aqueous extract of bamboo leaf showed strong inhibition on the growth of the harmful fungus (Table 4). This evidence revealed that bamboo leaf was a promising natural source for biological control of *P. grisea*. Rice farmers, especially in developing countries, may extract bamboo leaf with water to utilize as a cheap, convenient, and environmentally friendly material to reduce the yield loss caused by this rice blast disease.

## 5. Conclusions

The findings of this study highlighted that bamboo leaf appeared to be a promising source to manage the infestation of *P. grisea* in rice production. The analysis by GC-MS showed that non-polar constituents in bamboo leaf may be responsible for the antifungal activity, although water-soluble compounds may also play a role. Purification and isolation of potent phytochemicals should be conducted to further understand the possibility of using bamboo leaf for the biological control of *P. grisea*.

**Supplementary Materials:** The following are available online at http://www.mdpi.com/2077-0472/8/7/92/s1, Figure S1: Gas Chromatography-Mass Spectrometry (GC-MS) chromatogram of methanol extract, Figure S2: GC-MS chromatogram of hexane extract, Figure S3: GC-MS chromatogram of ethyl acetate extract, Figure S4: GC-MS chromatogram of butanol extract, Figure S5: GC-MS chromatogram of aqueous extract, Figure S6: Growth of *P. grisea* on Potato Dextrose Agar (PDA) medium with different concentrations as control (C), 0.1 mg/ml (M1), 0.5 mg/ml (M2), and 1 mg/ml (M3) of methanol fraction, Figure S7: Growth of *P. griseas* on PDA medium with different concentrations as control (C), 0.1 mg/ml (H1), 0.5 mg/ml (H2), and 1 mg/ml (H3) of hexane extract, Figure S8: Growth of *P. grisea* on PDA medium with different concentrations as control (C), 0.1 mg/ml (E1), 0.5 mg/ml (E2), and 1 mg/ml (E3) of ethyl acetate extract, Figure S9: Growth of *P. grisea* on PDA medium different concentrations as control (C), 0.1 (B1), 0.5 (B2), and 1 mg/ml (B3) of butanol extract, Figure S10: Growth of *P. grisea* on PDA medium with different concentrations as control (C), 0.1 mg/ml (W1), 0.5 mg/ml (W2), and 1 mg/ml (W3) of aqueous extract.

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## References

1. Darma, R.; Purnamasari, I.M.; Agustina, D.; Pramudito, T.E.; Sugiharti, M.; Suwanto, A. A strong antifungal—Producing bacteria from bamboo powder for biocontrol of *Sclerotium rolfsii* in melon (*Cucumis melo* var. amanta). *J. Plant Pathol. Microbiol.* **2016**, *7*, 334. [CrossRef]
2. Khush, G.S.; Jena, K. Current status and future prospects for research on blast resistance in rice (*Oryza sativa* L.). In *Advances in Genetics, Genomics and Control of Rice Blast Disease*; Wang, G.L., Valent, B., Eds.; Springer: Dordrecht, The Netherlands, 2009; pp. 1–10.
3. Michelmore, R.W.; Meyers, B.C. Cluster of resistance genes in plants evolve by divergent selection and a birth- and -death process. *Genome Res.* **1998**, *8*, 1113–1130. [CrossRef] [PubMed]
4. Mundt, C.C. Durable resistance: A key to sustainable management of pathogens and pests. *Infect. Genet. Evol.* **2014**, *27*, 446–455. [CrossRef] [PubMed]

5. Kamalakannan, A.; Shanmugan, V.; Suhendran, S.; Srinivasan, R. Antifungal properties of plant extracts against *Pyriclaria oryzae*, rice blast pathogen. *Indian Phytopathol.* **2001**, *54*, 490–492.
6. Sena, A.P.A.; Chaibub, A.A.; Côrtes, M.V.C.B.; Silva, G.B.; Silva-Lobo, V.L.; Prabhu, A.S.; Filippi, M.C.C.; Araújo, L.G. Increased enzymatic activity in rice leaf blast suppression by crude extract of *Epicoccum* sp. *Trop. Plant Pathol.* **2013**, *38*, 387–397. [CrossRef]
7. Olufolaji, D.B.; Adeosun, B.O.; Onasanya, R.O. In vitro investigation on antifungal activity of some plant extracts against *Pyricularia oryzae*. *Niger. J. Biotechnol.* **2015**, *29*, 38–43. [CrossRef]
8. Hubert, J.; Mabagala, R.B.; Mamiro, D.P. Efficacy of selected plant extracts against *Pyricularia grisea*, causal agent of rice blast disease. *Am. J. Plant Sci.* **2015**, *6*, 602–611. [CrossRef]
9. Netam, R.S.; Bahadur, N.S.; Tiwari, U.; Tiwari, R.K.S. Efficacy of plant extracts for the control of (*Pyricularia grisea*) blast of rice under field condition of Bastar, Chhattisgarh. *Res. J. Agric. Sci.* **2011**, *2*, 269–271.
10. Tanaka, A.; Zhu, Q.; Tan, H.; Horiba, H.; Ohnuki, K.; Mori, Y.; Yamauchi, R.; Ishikawa, H.; Iwamoto, A.; Kawahara, H.; et al. Biological activities and phytochemical profiles of extracts from different parts of bamboo (*Phyllostachys pubescens*). *Molecules* **2014**, *19*, 8238–8260. [CrossRef] [PubMed]
11. Han, J.; Xia, D.; Li, L.; Sun, L.; Yang, K. Diversity of culturable bacteria isolated from root domains of moso bamboo (*Phyllostachys edulis*). *Microb. Ecol.* **2009**, *58*, 363–373. [CrossRef] [PubMed]
12. Gong, J.; Huang, J.; Xiao, G.; Chen, F.; Lee, B.; Ge, Q.; You, Y.; Liu, S.; Zhang, Y. Antioxidant Capacities of Fractions of Bamboo Shaving Extract and Their Antioxidant Components. *Molecules* **2016**, *21*, 996. [CrossRef] [PubMed]
13. Kato, Y.; Shiozawa, R.; Takeda, S.; Ito, S.; Matsuda, K. Structural investigation of a β-D-glucan and a xyloglucan from bamboo-shoot cell-walls. *Carbohydr. Res.* **1982**, *109*, 233–248. [CrossRef]
14. Mao, J.W.; Yin, J.; Ge, Q.; Jiang, Z.L.; Gong, J.Y. In vitro antioxidant activities of polysaccharides extracted from Moso bamboo-leaf. *Int. J. Biol. Macromol.* **2013**, *55*, 1–5. [CrossRef] [PubMed]
15. Eyini, M.; Jayakumar, M.; Pannirselvam, S. Allelopathic effect of bamboo leaf extract on the seedling of groundnut. *Trop. Ecol.* **1989**, *30*, 138–141.
16. Jayakumar, M.; Eyini, M.; Pannirselvam, S. Allelopathic effect of bamboo root extract on the seedling of groundnut and corn. *Geobios* **1987**, *14*, 221–224.
17. Ying, C.; Mao, Y.; Chen, L.; Wang, S.; Ling, H.; Li, W.; Zhou, X. Bamboo leaf extract ameliorates diabetic nephropathy through activating the AKT signaling pathway in rats. *Int. J. Biol. Macromol.* **2017**, *105*, 1587–1594. [CrossRef] [PubMed]
18. Hayashi, N.; Kobayashi, N.; Cruz, C.M.V.; Fukuta, Y. Protocols for the sampling of diseased specimens and evaluation of blast disease in rice. *JIRCAS Work. Rep.* **2009**, *63*, 17–33.
19. Kartal, S.N.; Terzi, E.; Kose, C.; Hofmeyr, J.; Imamura, Y. Efficacy of tar oil recovered during slow pyrolysis of macadamia nut shells. *Int. Biodeterior. Biodegrad.* **2011**, *65*, 369–373. [CrossRef]
20. Ishii, H. Rice pathogens in Japan. In *Fungicide Resistance in Plant Pathogens*; Ishii, H., Hollomon, D., Eds.; Springer: Tokyo, Japan, 2015; pp. 341–354.
21. Gangawane, L.V. Fungicide resistance in plant pathogens in India. *Indian Phytopathol.* **1990**, *40*, 551–553.
22. Kumar, R.S.; Sivakumar, T.; Sunderam, R.S.; Gupta, M.; Mazumdar, V.K.; Gomathi, P.; Rajeshwar, Y.; Sarawanan, S.; Kumar, M.S.; Murugesh, K.; et al. Antioxidant ad antimicrobial activity of *Bauhinia racemosa* L. stem bark. *Braz. J. Med. Biol. Res.* **2005**, *38*, 1015–1024. [CrossRef] [PubMed]
23. Suriani, N.L.; Suprapta, D.N.; Sudana, I.M.; Temaja, R.M. Antifungal activity of *Piper canium* against *Pyricularia oryzae* Cav. the cause of rice blast disease on rice. *J. Biol. Agric. Healthc.* **2015**, *5*, 72–78.
24. Wu, D.; Chen, J.; Lu, B.; Xiong, L.; He, Y.; Zhang, Y. Application of near infrared spectroscopy for the rapid determination of antioxidant activity of bamboo leaf extract. *Food Chem.* **2012**, *135*, 2147–2156. [CrossRef] [PubMed]
25. Singh, S.A.; Bora, T.C.; Singh, N.R. Preliminary phytochemical analysis and antimicrobial potential of fermented *Bambusa balcooa* shoots. *Bioscan* **2012**, *7*, 391–394.
26. Gohel, N.M.; Chauhan, H.L.; Mehta, A.N. Bio-efficacy of fungicides against *Pyricularia oryzae* the incitant of rice blast. *J. Plant Dis. Sci.* **2008**, *3*, 18–192.
27. Ghanerian, M.T.; Ehrampoush, M.H.; Jebali, A.; Hekmati Moghadam, S.H.; Mahmoudi, M. Antimicrobial activity, toxicity and stability of phytol as novel surface disinfectant. *Environ. Health Eng. Manag. J.* **2015**, *2*, 13–16.

28. Rajab, M.S.; Cantell, C.L.; Franzblau, S.G.; Fischer, N.H. Antimycobacterial activity of (E)-phytol and derivatives: A preliminary structure-activity study. *Planta Med.* **1998**, *64*, 2–4. [CrossRef] [PubMed]
29. Ryu, K.R.; Choi, J.Y.; Chung, S.; Kim, D.K. Anti-scratching behavioural effect of the essential oil and phytol isolated from *Artemisia princeps* Pamp. in mice. *Planta Med.* **2011**, *77*, 22–26. [CrossRef] [PubMed]
30. Shukla, A.; Qwivedi, S.K. Antifungal approach of phenolic compounds against *Fusarium udum* and *Fusarium oxysporum* f.*sp.*ciceri. *Afr. J. Agric. Res.* **2013**, *8*, 596–600.
31. Salehan, N.M.; Meon, S.; Ismail, I.S. Antifungal activity plant pathogens. *Int. J. Agric. Biol.* **2013**, *15*, 864–870.
32. Amadioha, A.C. Controlling rice blast in vitro and in vivo with extracts of *Azadirachta indica*. *Crop Prot.* **2000**, *19*, 287–290. [CrossRef]
33. Shelke, S.; Mhaske, G.; Gadakh, S.; Gill, C. Green synthesis and biological evaluation of some novel azoles as antimicrobial agents. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 7200–7204. [CrossRef] [PubMed]
34. Sriram, D.; Yogeeswari, P.; Priya, D.Y. Antimycobacterial activity of novel *N*-(substituted)-2-isonicotionoylhydrazinocarbothioaide endowed with high activity towards isoniazid resistant tuberculosis. *Biomed. Pharmacother.* **2009**, *63*, 36–39. [CrossRef] [PubMed]
35. Ghasemi, S.; Abbasi, S.; Bahraminejad, S.; Harighi, B. Inhibitory effect of some plant crude extracts against cucumber damping-off agents. *Aust. Plant Pathol.* **2012**, *41*, 331–338. [CrossRef]
36. Jantason, A.; Moungsrimuangdee, B.; Dethoup, T. In vitro antifungal activity evaluation of five plant extracts against five plant pathogenic fungi causing rice and economic crop disease. *J. Biopestic.* **2016**, *9*, 1–7.