International Journal of Science and Research Archive
eISSN: 2582-8185
Cross Ref DOI: 10.30574/ijsra
Journal homepage: https://ijsra.net/

(RESEARCH ARTICLE)

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Effect of arbuscular mycorrhizal fungi on the fructification of Pleurotus ostreastus

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International Journal of Science and Research Archive, 2025, 14(02), 1018-1026

Publication history: Received on 31 December 2024; revised on 07 February 2025; accepted on 10 February 2025

Article DOI: https://doi.org/10.30574/ijsra.2025.14.2.0418

Abstract

The subterranean roots of most plants contain colonies of soil microorganisms named Arbuscular Mycorrhizal fungi (AMF). Scientific research indicates that AMF plants form a symbiotic connection with the substrate they grow in the substrates including soil. Water uptake becomes better through AMF while nutrients become more accessible and the fungi aid in protecting roots from microbial attacks and regulating hormone production. The Researchers in this research investigated how AMF influences the growth rate and fruiting performance of *Pleurotus ostreatus* in this study. The current mushroom supply levels are insufficient to meet worldwide demand especially in Nigeria since mushrooms mainly come from imported sources. Therefore, yield increases become necessary. Food security may be achieved by increasing the overall mushroom mycelia yield due to the effects of AMF on plant yield. The research explored yield effects from using four (4) different types of AMF. *Glomus gigaspora*, *Glomus clarum*, *Glomus mosseae*, and *Glomus derserticola* belong to the group of AMF being studied. Mushroom producers utilized sawdust along with calcium carbonate and rice bran as their growth substrates. The researchers calculated the Biological Efficiency (B.E.) that reflected mycelia yield levels. The AMF *Glomus clarum* had the highest yield with 938.33±35.92g with B.E. of 62.55±2.9%. This is seconded by *Glomus derserticola* with the yield of 850.67±46.23g with B.E. of 56.77±3.08%. *Glomus gigaspora* had a yield of 752.00±37.47g with B.E. of 50.13±2.50% while *Glomus mosseae* is the least with the yield of 715.33±15.37g with B.E. of 47.69±1.03%. Comparing the average yield, AMF inoculated substrates had more mushroom yield with about 15% increase in mycelia yield. Cultivators of mushrooms are to be encouraged to improve on the yield of mushroom by adding AMF into the substrates after sterilization of the substrates.

Keywords: *Pleurotus Ostreatus; Glomus Clarum; Glomus Mosseae; Glomus Derserticola* and *Glomus Gigaspora*; Biological Efficiency; Mycelia Yield

1. Introduction

The soil microbes known as Arbuscular Mycorrhizal Fungi (AMF) infiltrate most plant roots, create a bond between the plant and the substrate, aid in the synthesis of plant growth hormones, improve nutrient availability, and prevent root infections [1]. AMF has significantly enhanced the plant's vegetative growth and promoted its overall health. Numerous studies demonstrated that AMF improves shoot biomass, which raises crop yield. As stated by [2], AMF inoculation raised crop yields by 23% in 2022. AMF inocula not only enhanced crop biomass of shoots and roots by 24 and 29 percent, respectively, but also increased seed number and pod/fruit. AMF has been shown to decrease the buildup of molybdenum toxicity in plants in addition to promoting plant growth and biomass [1]. By mediating a sequence of intricate communication events between the plant and the fungus, AMF helps host plants grow vigorously under

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stressful conditions, resulting in enhanced photosynthetic rate and other gas exchange-related traits [3], in addition to enhanced water absorption. .

The impact of AMF on *Pleurotus ostreatus* growth and productivity will be evaluated in this study. P. *ostreatus* or White Oyster mushrooms are a plant-like fungus that is highly sought after as a meat substitute worldwide. To satisfy the demand worldwide, the yield must be increased. Food security will be attained if the overall mycelia yield of mushrooms improves in tandem with the impact of AMF on plant yield.

As versatile fungi, mushrooms provide numerous nutritional and health advantages to people from all walks of life [4]. In the 18th century, the first mushrooms were produced [5]. In addition to meeting dietary needs, the full essential amino acid profile of mushroom proteins may offer some financial benefits over those found in animal and plant sources. Few locals or experts are aware of how to identify and enjoy edible mushrooms because they have grown in the wild over the years and have been classified as either poisonous or edible [6]. According to [7], AMF enhances plant nutrition by increasing the availability and translocation of different nutrients. To increase the mycelial yield, it is imperative to utilize AMF activity in mushroom cultivation.

Many people all over the world eat mushrooms, which are the fruit bodies of macrofungi [8]. Like many other fungi, mushrooms reproduce by fusing two hyphae that are sexually compatible to create a large number of spores. When these spores land in a favorable environment, such as moist or damp soil or surfaces made of decaying wood, they grow into mushrooms. China alone has identified roughly 966 edible mushrooms in Asia. 576 of these are therapeutic in nature and are employed to treat various illnesses [9]. While eating mushrooms has many health and nutritional advantages [4], eating mushrooms serves more purposes than just nutrition. Because they are rich in phytochemicals, nutrients, and minerals, mushrooms support healing and the advancement of health of the consumers [10].

The production of mushrooms is thought to have begun in the 18th century and has recently become increasingly popular throughout the world. Currently, it is estimated that over 34 million tons of edible and medicinal mushrooms are produced worldwide. China produces more than 30 million tons of mushrooms annually, making it the world's largest producer [5]. Approximately 87% of the total production was accounted for by this. Due to their nutritional value and therapeutic qualities, mushrooms have been consumed for more than 2000 years and are a widely distributed food on Earth. The nutrients found in mushrooms, such as digestible protein, carbohydrates, fiber, vitamins, and antioxidants, have improved human health in addition to their delicious flavor and taste [10]. Polysaccharides, lectines, lactones, terpeniods, and alkaloids are among the many bioactive compounds from medicinal mushrooms that have been the subject of extensive research and are used extensively in western Asia [11] examined the composition, synthesis, and function of bioactive mushroom polysaccharides. In addition to their pharmacological characteristics, mushrooms are becoming more significant in our diet because of their high protein content, low fat/energy content, and nutritional value [4]. All nine of the essential amino acids needed by humans are present in the protein found in mushrooms.

The main medicinal uses of mushrooms and fungi are antioxidant, anticancer, antidiabetic, antiallergic, immunomodulating, cardiovascular protector, anticholesterolemic, antiviral, antibacterial, antiparasitic, antifungal, detoxification, and hepatoprotective effects; they also aid in preventing the growth of tumors and inflammation [12, 13].

Evaluating the impact of AMF on mushroom yield and exploring the possibilities for large-scale mushroom production with AMF are the goals of this study. It is anticipated that the study will offer agribusiness solutions for food scarcity and unemployment in the study area through mushroom farming. *Glomus Clarum*, *Glomus Mosseae*, *Glomus Derserticola*, and *Glomus gigaspora* are among the chosen AMF.

2. Materials and methods

Substrate: Sawdust is the substrate utilized in this study. It has been demonstrated that sawdust is a productive substrate for commercial mushroom cultivation. $CaCO_3$ and rice bran were added to the sawdust. After thoroughly mixing them with water, the substrates passed the "moisture squeeze test," indicating that there was a sufficient amount of moisture in the bagged substrate mixture inside the nylon bags [14]. The laboratory autoclave was used to sterilize the substrates after they were placed in nylon bags. After adding the AMF to sterile substrates, the mushroom mother spawn was injected into the substrates. *Glomus clarum*, *Glomus mosseae*, *Glomus derserticola*, and *Glomus gigaspora* are among the arbuscular mycorrhizal inoculum that was acquired from the Department of Soil Science at the University of Ibadan in Oyo State, Nigeria.

The term "mother spawn" describes the mycelium that has been meticulously multiplied on grains or agars. The mother spawn of *Pleurotus ostreatus* was acquired from MycoFarms Ltd. in Benin City, Edo State, Nigeria.

For four to five weeks, the inoculated substrates must be kept in a dark room. The substrates were moved from the dark room to the growth house to provide greater light intensity, ventilation, and humidity after the mycelium (spawn run) had completely colonized them. Water was sprayed often (morning and evening) until it reached maturity and fruit. A cropping room would be constructed at the Federal Polytechnic's Science Laboratory Technology Department in Orogun, Delta State, using materials that would provide light, ventilation, and a suitable temperature of 27±2^{0}C.

Final Stage/Harvesting: Mycelium colonization of the substrate was checked by looking for the growth of mushroom clusters after four to five weeks in the dark room. In the growth room (production house), where there is adequate light and ventilation, these were moved to shelves. Using my fingers to gently hold the base of the stalk, I carefully twisted and broke them off the substrate as they grew and matured with the cap tight on the stalk [6]. Growth Measurement: After harvesting, the quantity and size of fruiting bodies from each species-substrate component were noted, labeled appropriately, and their mean values were computed using the following methods: Counting the fruiting bodies from each specie bunch after it was harvested from its respective nylon bags.

Yield Determination: To identify the fruit bodies of the harvested mushrooms, their yields were measured in a unique way and documented appropriately [15]. In order to ascertain the mycelia yield and Biological Efficiency (B.E.), the fresh and dry fruit bodies were weighed on a scale using a calibrated pan. The proportion of the mushroom yield to the substrate weight was measured (in gramme) and the B.E. was calculated using the formula below [16].

$$B.E.v = \frac{\text{Fresh weight of mushroom}}{\text{Weight of substarte}} \times \frac{100}{1}$$

Once the substrate was bagged with nylon, its weight was 1,500g (1.5% kg).

The impact of AMF substrates on the quantity of mushroom fruit bodies was ascertained by harvesting the mushrooms, weighing the fruiting bodies, and documenting the values for each substrate to calculate the yield (Table 1).

Analysis of Variance (ANOVA) will be used to statistically analyze the experiment's data.

2.1. Study Area

The Federal Polytechnic in Orogun is the site of the study. Orogun is located in Delta State, Nigeria's Ugheli North Local Government Area. The Department of Science Laboratory Technology (SLT) of the Polytechnic established a mushroom farm in its Biological Garden. The farm's coordinates are 5° 41' 0" North, 6° 11' 0" East [17]. Orogun is bordered to the east by Abbi and Amai, to the north by Abraka, to the west by Eku, Kokori, and Agbara, and to the south by Emevor and Owhelogbo.

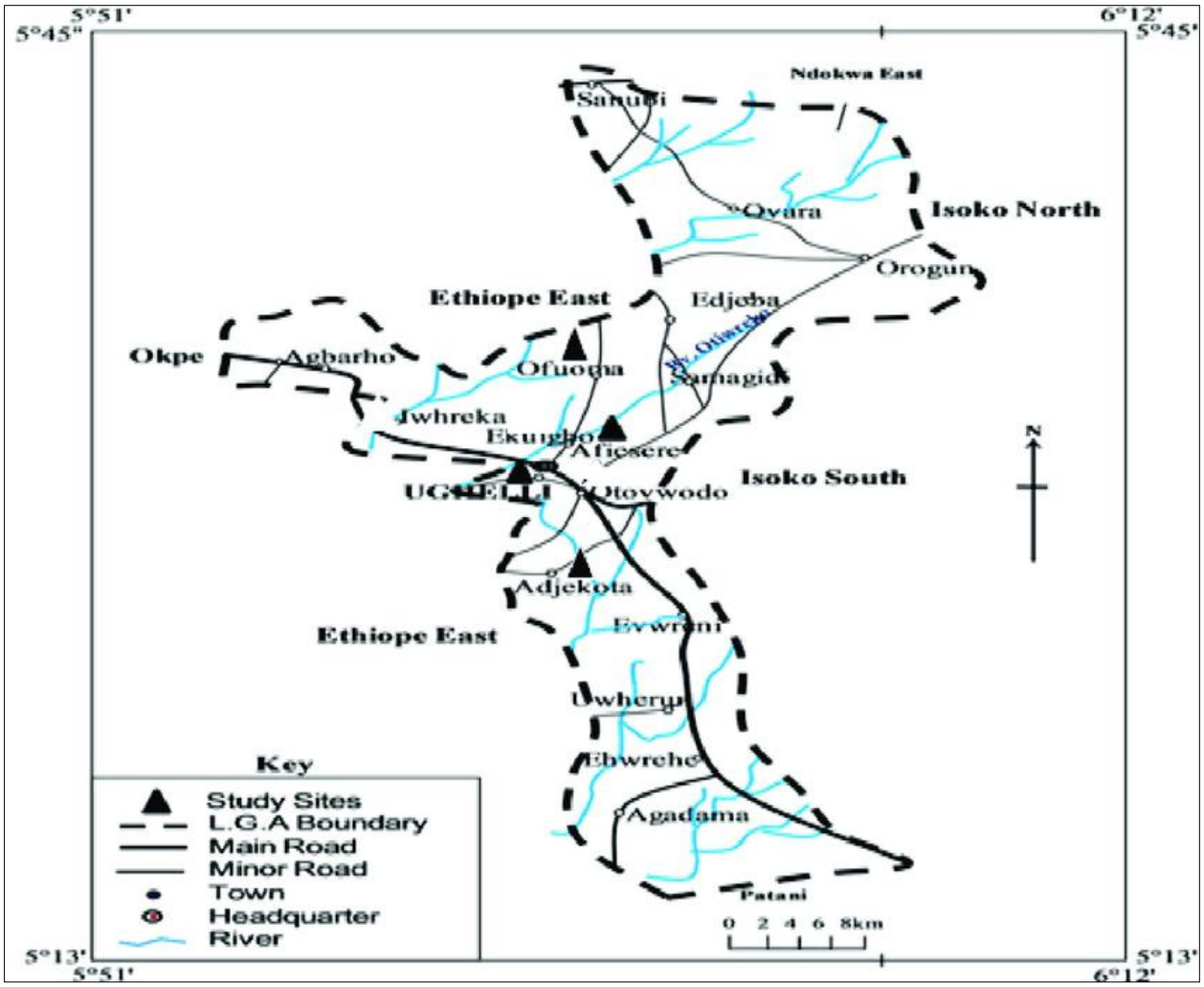

Figure 1 Map of Ughelli North showing the study area extracted from [17]

Experimental Design: The biological yield of the mushrooms was measured using a five (5) by ten (10) replicate treatment. This is equivalent to 50 fruiting nylon bags filled with White Oyster mushroom substrates that are set on wooden shelves.

3. Result and discussion

The results indicate that the AMF has a favorable impact on mushroom yield. Out of the 1,500g total weight of the substrate, the yield varies between 938g and 709g. The following tables and graphs display the detailed results. The following Table 1 displays the mycelia yields of substrates containing AMF.

Table 1 Composition of AMF in Substrates

AMF in Substrates	Mean±Std Value Yield(gram)
Substrate+*Glomus gigaspora*	752.00±37.47^{c}
Substrate+*Glomus derserticola*	850.67±46.23^{b}
Substrate+*Glomus clarum*	938.33±35.92^{a}
Substrate+*Glomus mosseae*	715.33±15.37^{c}

The values in the column with the various superscripts are significant at ($P<0.05$) and are means ± standard deviation of the triplicate determination values.

The average yield across the four (4) AMF from the Table 1 above is 814g.

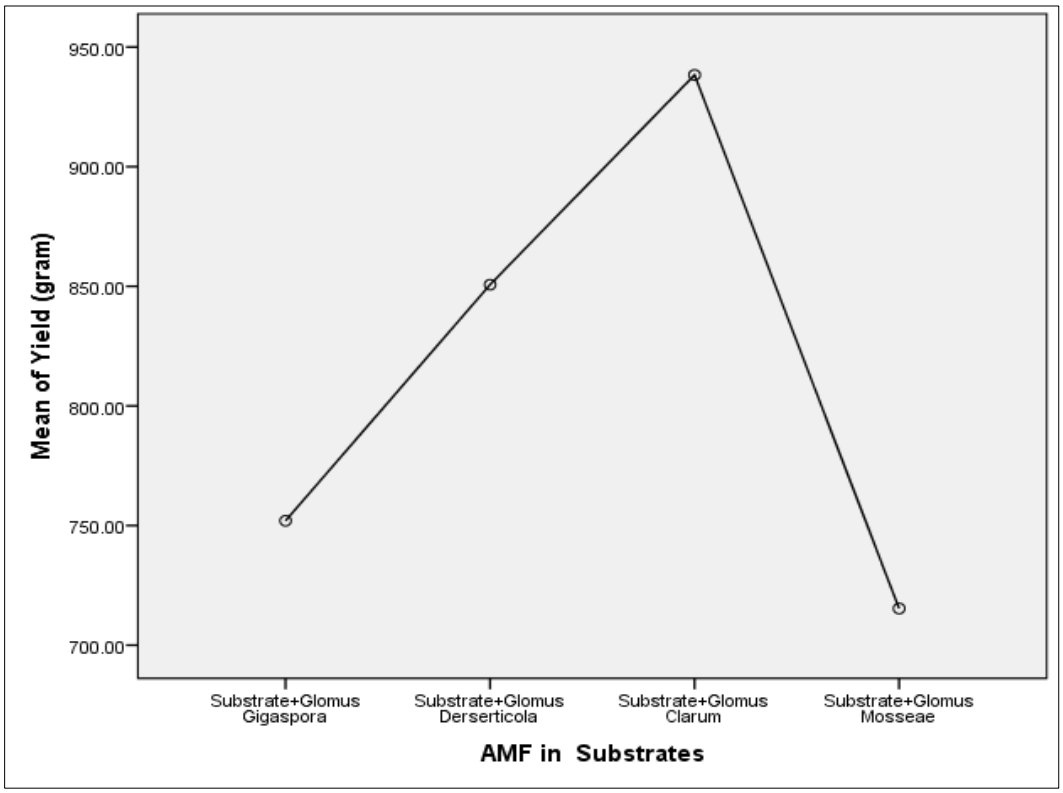

Figure 2 shows the mycelia yield of AMF-inoculated mushroom substrates graphically. The highest yield of *Pleurotus ostreatus* was found in substrates containing *Glomus clarum*, as shown in the above graph. The lowest amount of *Pleurotus ostreatus* was produced by *Glomus mosseae*.

In terms of mushroom mycelia yield, the rate of yield based on their biological efficiency performed well overall. The aforementioned formula was used to determine the biological efficiency. Below, in Table 2, the outcome is displayed.

Table 2 Composition of AMF in Substrates

AMF in Substrates	Mean±Std Value Biological efficiency (%)
Substrate+*Glomus gigaspora*	50.13±2.50^{c}
Substrate+*Glomus derserticola*	56.72±3.08^{b}
Substrate+*Glomus clarum*	62.55±2.39^{a}
Substrate+*Glomus mosseae*	47.69±1.03^{c}

P<0.05 indicates that the values with the various superscripts in the column are significant. The values are means ± standard deviation of triplicate determination values.

The biological efficiency of the substrates inoculated with *Glomus mosseae* was the lowest at 47–69 percent, whereas the highest biological efficiency was 62–55 percent for the substrates inoculated with *Glomus clarum*.

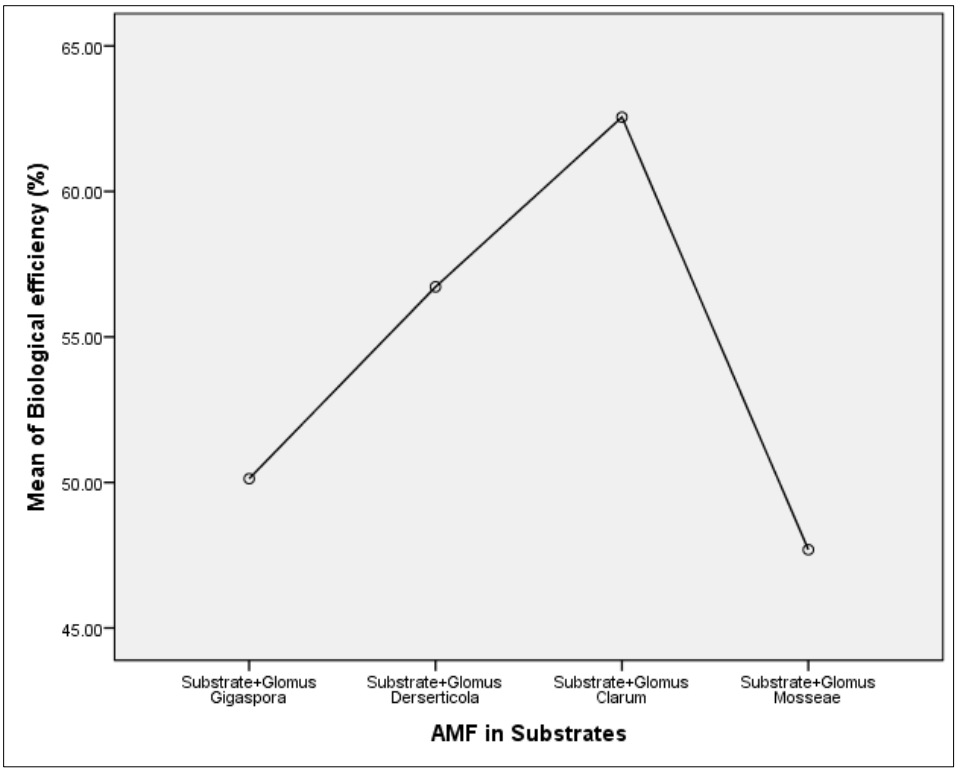

Figure 3 the biological efficiency of substrates inoculated with various AMFs, represented graphically.

AMF inoculation-free substrates make up the control treatment. Without AMF, this substrate's average yield is 709g, and its biological efficiency is 47%. The outcome is shown in Table 3 below.

Table 3 Composition of Substrates without AMF (Control) of Yield and Biological efficiency

Parameters	**Mean±Std Value**	**P-value**
Yield (gram)	709.67±13.87	0.000
Biological efficiency (%)	47.32±3.090	

Values are means ± Standard deviation of triplicate determination values are significant at (P<0.05).

A culture of *Pleurotus ostreatus* mushroom species developed with sawdust substrates combined with rice bran and calcium carbonate reached good mycelia yield totals of 709.67±87g while achieving a Biological Efficiency of 47.32±3.09%. The paper by [18] indicates that mushroom substrate selection for commercial use must achieve Biological Efficiency values greater than 40%. Fungal biomass reached its maximum levels when any type of sawdust received AMF treatment. The examined Arbuscular Mycorrhizal Fungi consisted of *Glomus clarum*, *Glomus mosseae*, *Glomus derserticola* and *Glomus gigaspora*. The mycelia yield was measured as Biological Efficiency (B.E.) where *Glomus clarum* demonstrated the highest result with 938.33±35.92g and 62.55±2.9% B.E. The mushroom yield experiments indicated that *Glomus derserticola* achieved 850.67±46.23g with B.E. of 56.77±3.08%. *Glomus mosseae* obtained the minimum yield of 715.33±15.37g with Biological Efficiency (B.E.) of 47.69±1.03% and *Glomus gigaspora* achieved 752.00±37.47g yield with B.E. of 50.13±2.50%. The substrates containing AMF showed increased yield levels which amounted to about 15% better performance than non-AMF treated substrates. The study conducted by [19] found that field-based AMF inoculation resulted in a 16% rise in cereal yield levels that were comparable to our research findings.

Mushroom producers should be motivated to get increased yields by adding AMF to their substrate material following substrate sterilization processes.

Figure 4 Mushroom fruiting bodies growing from nylon bags

Figure 5 The research team leader weighing freshly harvested mushrooms

4. Conclusion

The yield production of mushroom species *Pleurotus ostreatus* receives beneficial effects from AMF inoculation. The inoculation of AMF should be considered for mushroom growers in commercial mushroom production to achieve higher yields. The Nigerian government should create Mushroom development institutes or centers across all six geo-political regions under ministry of agriculture direction to conduct AMF harvesting and purification activities. These centers or institutes have the responsibility to provide training services along with mother spawn distribution and they must purchase mushroom from entrepreneurs who can deliver their products directly to retailers and consumers or exporters.

Compliance with ethical standards

Acknowledgments

Tertiary Education Trust Fund (TETFund) provided financial assistance under the Institution Based Research (IBR) to Federal Polytechnic, Orogun, Delta State, Nigeria for this research project.

Disclosure of conflict of interest

The researchers at no point maintain conflicts of interest throughout the research process or paper publication.

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