

(REVIEW ARTICLE)



The role of beneficial soil microbes for degraded-land rehabilitation

Betty Natalie Fitriatin *, Anne Nurbaity, Shantosa Yudha Siswanto and Marendra Ishak Sule

Department of Soil Science and Land Resources, Faculty of Agriculture, Universitas Padjadjaran Jl. Raya Bandung Sumedang km.21 West Java, 45363, Indonesia.

International Journal of Frontiers in Life Science Research, 2023, 05(01), 001–008

Publication history: Received on 15 May 2023; revised on 29 June 2023; accepted on 01 July 2023

Article DOI: <https://doi.org/10.53294/ijflsr.2023.5.1.0071>

Abstract

Various human activities that involve many activities such as forest clearing, logging, mining, clearing of agricultural and urban land can have a negative impact on the environment in the form of damage to forest vegetation as animal habitat and the possibility of loss of rare endemic flora/fauna species as a source of nuftah plasma. potential, damage to the water system (watershed), increase the rate of surface erosion, reduce land productivity and stability as well as the biodiversity of flora and fauna. To support the success of the revegetation and rehabilitation of degraded lands, this study discusses the role and prospects of beneficial microbes such as arbuscular mycorrhizal fungi (AMF) and root nodule-forming N-fixing bacteria (Bradyrhizobium/Rhizobium) which are alternative strategies that need to be tried. and developed. The role of mycorrhizal fungi and Bradyrhizobium/Rhizobium bacteria in accelerating the growth rate, increasing the quality and viability of forestry plant seedlings on nutrient-poor marginal lands has been widely demonstrated.

Keywords: Fixing bacteria; Land rehabilitation; Mycorrhiza; Revegetation

1. Introduction

The international world's attention and response to the status and sustainability of tropical humid forests in Indonesia is quite serious. This is reasonable considering the rate of forest destruction and land degradation every year has reached an area of between 900,000 hectares to 2,000,000 hectares [1] and is estimated largest in Southeast Asia. The damage was caused by various activities such as shifting cultivation, logging, forest fires, transmigration and agricultural development [2].

The process of desertification of forest land will occur in the not too distant future, if the pressure on forest resources in Indonesia is continued continuously [3]. Many cases, such as illegal logging, forest fires, mining activities, and the ambitious project to create one million hectares of rice fields, can speed up the process of desertification. On the other hand, the forest rehabilitation process carried out by the government, the private sector and the community cannot keep up with the rate of deforestation.

To prevent and reduce more severe environmental damage, it is necessary to look for various control efforts that lead to land rehabilitation activities [4]. Seeing this fact, the rehabilitation of critical and marginal lands in the form of Industrial Plantation Forest development, reforestation, and revegetation is an alternative problem solving program that needs to be carried out and prioritize. The success of this program will not only improve Indonesia's image in the eyes [5]. the international community regarding the "Deforestation Impact" which has recently received the spotlight [6].

* Corresponding author: Betty Natalie Fitriatin

The development of plantation forests is one of the solutions to save the remaining natural forests, but it must be anticipated that in the future the development of plantation forests will create new problems [7].

There is still enough time for us to anticipate the problems that will be caused by the development of plantation forests, while there is no longer any time for us to stop the loss of the remaining natural forests. The plantation forest should be optimized development so that pressure on natural forests can be reduced immediately [8].

Revegetation activities is one of the vegetative techniques that can be applied in an effort to reclaim damaged lands [9]. The aim is not only to improve unstable and unproductive lands and to reduce surface erosion, but also in the long term is expected to improve the microclimate, restore biodiversity and improve land conditions in a more productive direction [10].

In its implementation, the reforestation and revegetation programs often encounter serious obstacles due to unfavorable soil conditions [11]. Obstacles to the physical and chemical properties of soil that are often encountered include low soil reaction (low soil pH), nutrient deficiencies, especially phosphorus (P) and nitrogen (N), thin soil layers and poor organic matter. This condition is the main obstacle for plant growth and successful reforestation. Newly planted seedlings often grow slowly, languish and have low viability. This is mainly due to unfavorable land conditions to support plant growth. Plants are difficult to grow and have low vitality [12].

Efforts to rehabilitate critical lands and develop new forest plantations have experienced many obstacles. As explained above, the problems that are often encountered in the field include acidic soil pH (a characteristic of soils in the wet tropics), poor nutrients, very thin topsoil, and there are still many problems that must be solved. It is necessary to pay attention to the need for special handling for this land rehabilitation activity [13].

The preparation of seedlings on an operational scale is still below standard (in terms of quality and quantity), as a result many forest plant seedlings experience transplant shock and even die after being transferred to the field [14]. Another factor that is more dominant is the non-technical factor in the critical land rehabilitation program. The land rehabilitation program with a very large area and quite a lot of funds was carried out with a very short time frame, immature planning and a bidding system that involved chains of sub-contractors. long, so it is very clear success planting failed from year to year.

Based on experience in the field, to help grow and increase the viability of seedlings on these marginal lands, in addition to proper silvicultural techniques and selection of suitable species, it also requires high and expensive energy inputs such as liming, phosphate saturation, complete fertilization and organic matter (management [15]. In the context of developing forests with an environmental perspective, it is necessary to look for other alternative strategies that are not only effective but are cheaper and environmentally friendly.

To support the success of the revegetation program (greening) and rehabilitation of degraded lands, this paper will discuss the role and prospects of PSM, arbuscular mycorrhizal fungi and root nodule-forming N-fixing bacteria (*Bradyrhizobium/Rhizobium*) as an alternative strategy that needs to be considered. tried and developed.

The role of mycorrhizal fungi and *Bradyrhizobium/Rhizobium* bacteria in accelerating growth rates, increasing the quality and viability of forestry plant seedlings on marginal lands that are poor in nutrients has been extensively demonstrated [16], [17].

This paper attempts to provide an overview potential of soil beneficial microbes and their application prospects as biological tools in order to support government programs to rehabilitate marginal and critical lands in Indonesia.

2. Degraded-Land Conditions in Indonesia

2.1. Soil Physical Condition

Mineral mining activities produce waste, one of which is called tailings. Tailings are rock powder that has been finely ground after the copper, gold and silver minerals have been physically separated using grinding and flotation techniques. The most prominent general characteristics of critical lands that are heavily damaged, for example post-mining lands, are non-profile soil layers [18]. The normal soil profile has been disturbed by dredging, backfilling and compaction of heavy equipment. The activities of stockpiling and compacting soil in the reconstruction of planting land

cause damage to the structure, texture, porosity and bulk density of the soil as soil physical characteristics which are very important for plant growth.

In addition, compact soil conditions due to compaction can cause poor water management systems (water infiltration and percolation) and aeration which can directly have a negative impact on root function and development. Roots cannot develop properly and their function as a means of nutrient absorption will be disrupted. As a result, plants can develop normally, but still grow stunted and miserable.

Damage to texture and structure also causes the soil to be unable to store and absorb water during the rainy season, resulting in high surface run-off and an impact on increasing erosion rates. On the other hand, during the dry season the soil becomes dense and hard, making it very difficult to cultivate, which indirectly results in an increase in the need for labour.

2.2. Soil Chemical Conditions

In a normal soil profile, topsoil is a source of essential macro and micro nutrients for plant growth, as well as a source of organic matter to support potential soil microbial life and activity. Thin and lack of topsoil and organic matter is considered as the main cause of poor soil fertility on degraded lands. Deficiency of essential nutrients such as N and P, and acid (low pH) or alkaline (high pH) soil reactions, as well as low CEC values (cation exchange capacity) are common problems found in critical lands.

The tropics have high rainfall and have high/hot temperatures, so that it can cause a high level of soil weathering, the higher the weathering the soil has an acidic pH. The low soil pH causes poor base cations which in turn can indirectly result in easy erosion of the soil.

2.3. Soil Biological Conditions

Loss of top soil can result in the loss of the litter layer as a source of carbon (C) which is a source of energy to support the growth and survival of potential soil microbes, is one of the main causes for reducing the population and activity of soil microbes which play an important role in the provision of nutrients or the transformation of soil elements. This will indirectly greatly affect plant life, especially tree species that need to associate with Rhizobium, Bradyrhizobium and arbuscular mycorrhizal fungi (AMF) to help their growth. The existence of these potential soil microbes can play a very important role for the development and survival of plants.

Microbial activity is not only limited to providing nutrients, but also plays an active role in decomposing litter and can even gradually improve the character of soil structure. Due to the low population and activity of potential soil microbes on critical lands, efforts are needed to manipulate the availability of these potential soil microbial populations.

3. Prospects and potential applications beneficial microbes in critical land rehabilitation

Plantation forest development programs and post-mining land revegetation are activities that must be carried out immediately and require serious handling. To support the success of the revegetation program and rehabilitation of degraded lands, this paper will discuss the role and prospects of arbuscular mycorrhizal fungi (AMF) and root nodule-forming N-fixing bacteria (Bradyrhizobium/Rhizobium).

3.1. Leguminous-Rhizobium/Bradyrhizobium Association

The use of Rhizobium bacteria as an inoculant has been popularly used in agricultural crops such as soybeans and other types of legumes [19]. Now this bacterium has begun to be introduced for its use also for leguminous plants which are often used for reforestation and agroforestry activities [20],[21].

Rhizobium/Bradyrhizobium bacteria have the ability to infect roots and form root nodules (nodules) in symbiosis with leguminous trees. Inside the root nodules, these bacteria are chemically capable of fixing free nitrogen (N₂) from the atmosphere and converting it into ammonia (NH₃), the last product of which can be utilized by the host plant for its growth [22]. While Rhizobium itself obtains carbohydrates as a source of energy from the host plant [23].

Nitrogen fixation by these bacteria, which are in symbiosis with leguminous plants, is actually a natural process whose level of effectiveness [24] can be manipulated and increased by inoculating superior Rhizobia strains that have been tested. In this way, not only can the growth rate of leguminosae trees be properly stimulated, but it also allows these trees to live in nitrogen-poor soil conditions. In addition, the presence of a suitable leguminosa-rhizobium association

allows for a relatively high contribution of N fixation to the soil. The system mentioned above in the long term can indirectly maintain and increase soil fertility so that it allows other plants (non-legumes) to grow, fears of a decline in forest productivity in the next rotation can be overcome [25].

The characteristics and presence of root nodules on potential leguminous tree species which are often used for Industrial Plantation Forest, reforestation and agriforestry activities, have been widely evaluated. This is now being done to see the opportunities and abilities of these plants to fix free nitrogen in the atmosphere. Of the 25 species that have been widely observed, as many as 17 species have been shown to form root nodules. Of the tree species that form nodules, the types are included in the Mimosaceae family such as: *Acacia mangium*, *Acacia auriculiformis*, *Albizia lebbbeck*, *Paraserianthes falfacataria*,

Calliandra callothyrsus, *Enterolobium cyclocarpum* and *Leucaena leucocephala*, have a high frequency and abundance of nodules. *Acacia mangium* is one of the leguminous trees that has the potential to be used to rehabilitate critical lands, besides that, its wood has bright prospects as a forestry crop commodity. To increase the productivity of this plant, on marginal land with the help of *Rhizobium/Bradyrhizobium* bacteria, it has now been carried out to isolate nodule-forming bacteria from the roots of *Acacia mangium* [20]. It was reported that this bacterial isolate could increase the height, diameter and biomass of *Acacia mangium* seedlings 2-3 times compared to the control, this is almost equivalent to giving 140-280 kg of Urea [20].

3.2. Mycorrhiza

Mycorrhiza is a root system structure that is formed as a manifestation of a mutualistic symbiotic relationship between fungi (myces) and roots (rhiza) of higher plants [26]. Based on the mode of infection of the host plant, mycorrhizae can be grouped into three groups, namely ectomycorrhizae, endomycorrhizae and ectendomicorrhizae. The ectomycorrhizal fungus is common in forest plants, especially pine. This fungus can be seen directly in the field with the naked eye. The infected part of the root will enlarge/swell and branch out into dichotomes, and the root surface will be covered with mycelia which is commonly called the mantle. The hyphae of this ectomycorrhizal fungus develop between the cell walls of the cortex tissue.

According to Mosee (1981) [27], an infection by endomycorrhizae can be characterized by the formation of vesicles and arbuscles, so it is known as MVA (vesicle-arbuscule mycorrhiza). This type of mycorrhiza infection occurs inside the cell. Vesicles are in the form of a kind of sac, usually located at the end of the internal hyphae which contain lots of fat, functioning as a storage organ for food reserves. Arbuscules (intracellular) are hyphae that enter the cortical cells of the host plant, then the hyphae branch out. It is suspected that it is through these arbuscules that translocation of nutrients (phosphates) occurs between the host plant and mycorrhiza. Arbuscules are generally formed about 2 to 3 days after the root is infected.

In addition to vesicles and arbuscules, this endomycorrhizal has other components, namely inter and intra cellular hyphae in the cortex and external mycelium around plant roots. Hyphal tissue located on the outside of the root is an extension of the root surface. The presence of these hyphae allows the roots to absorb nutrients, especially P, with a wider and farther range [28].

In contrast to ectomycorrhizae, arbuscular mycorrhizal colonization does not cause changes in root morphology, so quantification of this fungal colonization must be through observation with a microscope. In addition to helping plants absorb nutrients, AMF can make plants more tolerant of environmental stresses such as drought, extreme temperatures and soil acidity [29]. Mycorrhiza can also increase the formation of soil aggregates around plant roots so that the physical properties of the soil become better.

Mycorrhizae need P as an energy source while plants need P physiologically for their metabolism. The presence of mycorrhizae will increase the concentration of P in the root area of the plant, by releasing the fixed P bonds to become available P [26].

According to Smith and Read (1997) [30], that the amount of P absorbed from the soil and transferred to plants is the result of the following three mechanisms:

- Uptake of P by hyphae from the soil
- P translocation along the hyphae
- Transfer of P from hyphae to plants through the interface between the fungus and the plant.

The real function of AMF to plant nutrients depends on the exploration of hyphae in the soil and the expansion of the root surface [27]. The formation of AMF can have a positive effect on several physiological aspects of plants. Inoculation of AMF influences the plant growth and increases water moisture uptake, consequently improving plant tolerance against stresses like drought and salinity. Exploitation of AMF for plant growth in various biological ecosystems can contribute greatly to organic culturing for growth promotion and yield production [29].

4. Critical land rehabilitation technique

4.1. Critical Land Rehabilitation Technique

Degraded lands usually have damaged landscapes. For example, post-mining lands are usually in the form of pits which are not suitable for direct planting. To be planted, the land must be laid out first by stockpiling. In carrying out these activities, several things that need to be considered are the type and origin of the fill material, the thickness of the water flow system (drainage) that might be disturbed.

It is better if the excavated materials are returned to their original state close to their original state. The recommended thickness of soil cover (sub-soil) ranges from 70 – 120 cm. After the planting land is formed, the activity is continued with the redistribution of top soil. The existence of top soil as a potential source of nutrients, organic matter and microbes is absolutely necessary for plant growth. In order to obtain good top soil quality, it is necessary to carry out strict supervision during dredging, storage and redistribution. Re-allocation of top soil to planting fields can be done locally (per planting hole) or spread evenly with sufficient depth. This will be determined by the type of plants to be planted, the degree of slope, the quality and availability of top soil.

4.2. Revegetation of Critical Land

The main obstacle in implementing revegetation in degraded lands is the unfavorable soil conditions for plant growth as previously described. By taking into account the existing constraints, revegetation activities on critical lands need to be designed as follows:

4.2.1. Selection of tree species

Selection of the right tree species is the key to successful revegetation. The various approaches commonly used in selecting tree species are as follows: adaptability, fast growing, known silvicultural techniques, availability of plant material and potential symbiosis with microbes such as mycorrhiza and rhizobia.

4.2.2. Improvement of soil conditions

The main obstacle in implementing revegetation on degraded lands is soil conditions (chemical, physical and biological) which are marginal for plant growth. To overcome this problem, there are various strategies that need to be implemented, namely: as follows: improvement of plant growth space, provision of top soil and organic matter, basic fertilization, liming and application of humic acid.

4.3. Production of Mycorrhizal Inoculants

Exploration of AMF species in various intact and disturbed ecosystems is an initial study that has been carried out in stages in several regions in Indonesia. From this activity, it has been possible to identify and map the dominant AMF species that are specific to an area. This activity is very important to do because in addition to knowing the distribution patterns of AMF species and their ecosystems, it can also identify potential AMF species that can be used as a source of material in the manufacture of biological fertilizers and have adapted to local conditions.

Screening in order to find superior isolates that can spur growth and improve the quality of forestry plant seeds, which are planted on critical and marginal lands is the main object of research that must be continuously carried out. Some research results show that not all types of plants always respond positively to the application of AMF, for example the seeds of Balsa (*Ochroma bicolor*) and Matoa (*Pometia pinnata*) trees, although both can be infected intensively but do not show any growth response. This is not only determined by the level of effectiveness of the isolate and the nutritional status of the substrate used, it is also determined by the level of dependence of the plant on mycorrhizal (mycorrhizal dependency) [31].

Provision of AMF inoculants on a large scale, for application in the field is still under control. One of the reasons is that this type of fungus is obligate, so to live and reproduce it always requires a host plant. Until now, AMF has not been able

to make pure culture (arsenic). One common technique used to propagate AMF is pot culture. In this way, AMF is allowed to grow and reproduce in the host's root system which is grown in pots containing certain nutrient-dense substrates. For the supply of inoculants on a large scale, this technique is still considered inefficient, because it is not only voluminous and heavy, but also heavy requires a large enough space in its manufacture. One way to overcome this problem has been to develop high propagule density inoculant [20]. The inoculant was prepared by first studying the characteristics of the AMF culture which were related to the suitability of the host, the suitability of the substrate, the pH range and the optimum nutritional requirements of each isolate which had been tested to be effective. With this knowledge, the density of active propagules (apora, mycorrhizal and mycorrhizal roots) as a potential inoculum source can be manipulated and their number increased several tens of fold. The use of high propagule density inoculants in combination with the pre-inoculation technique (inoculation in the nursery) can overcome the constraints in providing AMF inoculants for field applications. This technique is not only practical and effective but can also reduce the weight (weight) of the inoculant needed by up to 60 %.

Production of mycorrhizal propagules, as conducted in this experiment, permits the inoculation of these organisms in plants growing in soils where AMF inoculum levels are reduced [30]. Different type of soils will have different responses to the AMF inoculum and soil type influenced spore density as well as the percentage of mycorrhizal colonization of roots during cultivation period [32].

5. Conclusion

Mycorrhizal fungi (ecto and endo) and Rhizobium/Bradyrhizobium are potential biological natural resources found in nature. The level of effectiveness in terms of spurring growth and improving the quality of forestry plant seeds can be manipulated and increased through a series of laboratory studies and field tests. In this way, it can be produced and packaged in the form of inoculums and used as a biological tool to stimulate plant growth.

Based on its ability to increase plant growth on unproductive land conditions, the application of inoculants is very suitable to be directed at assisting government programs in rehabilitating critical and marginal lands such as open post-mining lands, and deforested and main forest lands. is the grassland area as the largest reforestation zone in Indonesia. Apart from that, it is also possible to try the application of biological fertilizers as an alternative strategy to increase the productivity of transmigration and shifting cultivators.

The importance of awareness of the importance of ectomycorrhizal inoculation to produce quality forest plant seeds is needed so that the success of the critical land rehabilitation program in Indonesia can be realized in practice. Of course, evidence in terms of practicality and use and the low cost of inoculum production and ease of inoculation techniques in nurseries are urgently needed to be produced through intensive research and development.

The inoculant application is actually an ecological integrity because in principle it utilizes potential biological natural resources with simple and inexpensive technology, is safe to use (non-pathogenic, does not cause environmental pollution), plays an active role in nutrient cycles and can even improve soil fertility status. It is hoped that its application on a broad scale will not only help increase the growth and quality of seedlings, increase the productivity of marginal lands but also improve biodiversity and ecosystem stability.

Compliance with ethical standards

Acknowledgments

The authors are immensely grateful to the Academic Leadership Grant (ALG) of Universitas Padjadjaran for providing funding of this work.

Disclosure of conflict of interest

None of the authors has any conflict of interest including financial involvement in the institution or company.

References

- [1] Sitorus, S.R.P and Pravitasari, A.E. 2017. Land Degradation and Landslide in Indonesia. Sumatra Journal of Disaster, Geography and Geography Education. 1 (2) : 61-71

- [2] FAO. 2022. CHAPTER 2 FORESTS And Trees Provide Vital Goods And Ecosystem Services But Are Undervalued In Economic Systems. <https://www.fao.org/3/cb9360en/online/src/html/deforestation-land-degradation.html>
- [3] Sunkar. A., 2008. Deforestation And Rocky Desertification Processes In Gunung Sewu Karst Landscap. Media Konservasi Vol. 13, No. 3 Desember 2008 : 1 – 7
- [4] Gomiero, T. 2016. Soil Degradation, Land Scarcity and Food Security: Reviewing a Complex Challenge. Sustainability 2016, 8, 281: 1-41
- [5] Indrajaya, Y., Yuwati, T.W., Lestari, S., Winarno, B., Narendra, B.H., Nugroho, H.Y.S.H., Rachmanadi, D., Pratiwi, Turjaman, M., Adi, R.N.; et al. 2022. Tropical Forest Landscape Restoration in Indonesia: A Review. Land, 11, 328. <https://doi.org/10.3390/land11030328>
- [6] Kasaro, D., Phiri, E. and Nyambe, I. 2019. Deforestation impact on ecosystem services in Kamfinsa sub-catchment of Kafue River Basin in Zambia. African Journal of Environmental Science and Technology. Vol. 11(4), pp. 33-45
- [7] Elias, P., and Doug Boucher, D. 2014. Planting for the Future. How Demand for Wood Products Could Be Friendly to Tropical Forests. The Tropical Forest and Climate Initiative (TFCI) of the Union of Concerned Scientists (UCS).
- [8] Farooq, T.H., Shakoor, A., Wu, X., Li, Y., Rashid, M.H., Xiang Zhang, X., Gilani, M.M., Kumar, U., Chen, X., Yan, W. 2021. Perspectives of plantation forests in the sustainable forest development of China. Forest - Biogeosciences and Forestry, Volume 14, Issue 2, Pages 166-174. doi: <https://doi.org/10.3832/ifer3551-014>
- [9] Pratiwi; Narendra, B.H.; Siregar, C.A.; Turjaman, M.; Hidayat, A.; Rachmat, H.H.; Mulyanto, B.; Suwardi; Iskandar; Maharani, R.; et al. Managing and Reforesting Degraded Post-Mining Landscape in Indonesia: A Review. Land 2021, 10, 658. <https://10.3390/land10060658>
- [10] Tomar, J.M.S., Ahmed, A., Bhat, J.A., Kaushal, R., Shukla, G. and Kumar, R. 2021. Potential and Opportunities of Agroforestry Practices in Combating Land Degradation. In Agroforestry. Edited by Gopal Shukla, Sumit Chakravarty, Pankaj Panwar and Jahangeer A. Bhat. DOI: 10.5772/intechopen.97843
- [11] Dumroese RK, Landis TD, Pinto JR, Haase DL, Wilkinson KW, Davis AS. 2016. Meeting forest restoration challenges: Using the target plant concept. Reforesta 1: 37-52.
- [12] Neina, D. 2019. The Role of Soil pH in Plant Nutrition and Soil Remediation. Applied and Environmental Soil Science Volume 2019, Article ID 5794869, 9 pages <https://doi.org/10.1155/2019/5794869>
- [13] Mentis. M. 2020. Environmental rehabilitation of damaged Land. Forest Ecosystems. 7:19. <https://doi.org/10.1186/s40663-020-00233-4>
- [14] Koeser, A.K., Stewart, F.R., Bollero, G.A., Bullock, D.G., Struve, D.K. 2009. Impacts of Handling and Transport on the Growth and Survival of Balled-and-burlapped Trees. HORTSCIENCE 44(1):53–58.
- [15] Smethurst, P.J. 2010. Forest fertilization: Trends in knowledge and practice compared to agriculture. Plant and Soil 335(1):83-100. DOI:10.1007/s11104-010-0316-3
- [16] De La Cruz, R.E., Manalo, M.Q., Anggangan, N.S., & Tambalo, J.D. 1988. Growth of three legume trees inoculated with VA mycorrhizal fungi and Rhizobium. Plant Soil 108 : 111-115
- [17] Soumare, A., Diop, T., Manga, A., and Ndoy, I. 2015. Role of arbuscular mycorrhizal fungi and nitrogen fixing bacteria on legume growth under various environmental stresses. International Journal of Biosciences. 7 (4) : 31-46
- [18] Ma, K., Zhang, Y., Ruan, M., Guo, J. and Chai, T. 2019. Land Subsidence in a Coal Mining Area Reduced Soil Fertility and Led to Soil Degradation in Arid and Semi-Arid Regions. Int.J.Environ.Res.PublicHealth. 16,3929. doi: 10.3390/ijerph16203929
- [19] Nakei MD, Venkataramana PB and Ndakidemi PA. 2022. Soybean-Nodulating Rhizobia: Ecology, Characterization, Diversity, and Growth Promoting Functions. Front. Sustain. Food Syst. 6:824444.. doi: 10.3389/fsufs.2022.824444
- [20] Setiadi, Y. 2002. Mycorrhizal inoculum production technique for land rehabilitation. Jurnal Manajemen Hutan Tropika Vol. VIII No. 1 : 51-64
- [21] Shelton, H.M. 2015. Tropical forage tree legumes in agroforestry systems. Unasylva 200, Vol. 51, 2000
- [22] Lindstrom, K. and Mousavi, S.A. 2019. Effectiveness of nitrogen fixation in rhizobia. Microbial Biotechnology, 13, 1314–1335

- [23] Yang, J., Lan, L., Jin, Y., Yu, N., Wang, D., and Wang E. (2022). Mechanisms underlying legume–rhizobium symbioses. *J. Integr. Plant Biol.* 64: 244–267.
- [24] Kawaka, F. 2022. Characterization of symbiotic and nitrogen fixing bacteria. *AMB Express.* 12:99. <https://doi.org/10.1186/s13568-022-01441-7>
- [25] Degefu T, Wolde-meskel E, Liu B, Cleenwerck I, Willems A, Frostegård Å. 2013. *Mesorhizobium shonense* sp. nov., *Mesorhizobium hawassense* sp. nov. and *Mesorhizobium abyssinicae* sp. nov., isolated from root nodules of different agroforestry legume trees. *Int J Syst Evol Microbiol* 63:1746–1753
- [26] Sieverding, E. 1991. Vesicular-arbuscular Mycorrhiza Management in Tropical Agrosystems. Technical Cooperation, Federal Republik of Germany.
- [27] Mosse, B. 1981. Vesicular-arbuscular mycorrhiza research for tropical agriculture. *Res.Bull.* 194. Hawai Institute for tropical Agriculture.
- [28] Bobbu, H. 2016. Host specificity, mycorrhizal compatibility and genetic variability of *Pisolithus tinctorius*. *International Journal of Advanced Engineering, Management and Science.* 2 (11) : 1836-1846.
- [29] Begum N, Qin C, Ahanger MA, Raza S, Khan MI, Ashraf M, Ahmed N and Zhang L. 2019. Role of Arbuscular Mycorrhizal Fungi in Plant Growth Regulation: Implications in Abiotic Stress Tolerance. *Front. Plant Sci.* 10:1068. doi: 10.3389/fpls.2019.01068
- [30] Smith S.E, and D.J. Read. 1997. *Mycorrhizal Symbiosis*. Second Edition. Academic Press. New York. Pp. 450 –110.
- [31] Janos, D.P. 2007. Plant responsiveness to mycorrhizas differs from dependence upon mycorrhizas. *Mycorrhiza* 17(2):75-91. DOI:10.1007/s00572-006-0094-1
- [32] Land S., and Schönbeck F. 1991 Influence of Different Soil Types on Abundance and Seasonal Dynamics of Vesicular Arbuscular Mycorrhizal Fungi in Arable Soils of North Germany. *Mycorrhiza* 1(1) 39-44