

Original Research Paper

The Dynamics of Chemical Properties and Soil Microbial Types at Agroforestry System in Supporting Environmental Friendly Agriculture

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Abstract: Agroforestry is described as the land-use system producing and conserving various agricultural, livestock, and fishery commodities combined with forestry commodities. The research aims to determine the chemical properties of the soil and types of microbial (especially the presence of soil bacterial) in the cocoa farming system. The two types of analysis were conducted: (1) Soil chemical properties included pH, C-organic, N-total, P-available, and Potassium; (2) the type and number of bacterial. The research from November 2018 to July 2019 at Mamuju Regency, West Sulawesi. Soil samples used the sampling method (ring and composite). The analysis results showed the soil pH ranged from acidic to slightly acidic (between 4.73-5.76). The content of C-organic ranging from (1.39-2.56%); N-total value is very low (0.20-0.25%); P-available is low (15.6-15.91 ppm) and Potassium also low (0.13-0.21 cmol kg⁻¹). The genus of bacterial found in the research location: *Pseudomonas* in Kalonding (289 cfu), Boda Boda (238 cfu), and Sisango (220 cfu); *Alkaligenes* were found in Salukayu 2 (255 cfu) and Salumasa (220 cfu) and *Klebsiella* were found in Batupapan (265 cfu).

Keywords: Soil Chemistry, Texture, Soil Microbial, Agroforestry, Environmental Friendly

Introduction

The one alternative land-use system for production and conservation purposes is known as the agroforestry system. On the other hand of agroforestry system means the management of agricultural, livestock, or fishery commodities combined with forestry commodities in the form of trees. For the famous example, cocoa is one of the most common plantation crops grown by the community in South Sulawesi. On the other hand, cocoa (*Theobroma cacao* L.) is one of the export commodities giving many contribute to increasing the country's foreign exchange. After palm oil and rubber, cocoa placed in the third position for export in the plantation sector, contributing increase national community income. The important issue and fact, cocoa has a reasonably stable market and high prices in international trade (Suryani and Zulfebriansyah, 2007).

The suitable planting media contain good quality organic matter promise plant growth faster when compared to planting media with less organic matter content, especially in the terrestrial areas (Delgado-Baquerizo *et al.*, 2016a). Few physical conditions of the

soil determine such as plant root penetration into the soil (Eisenhauer *et al.*, 2017), water absorption, drainage, aeration, and plant nutrients. The physical properties of the soil depend on the amount, size, shape, composition of the mineral in the soil particles, type and amount of organic matter, volume and shape of the material pores, the ratio of water and air in the pores at the time formed (Singh *et al.*, 2010; Beugnon *et al.*, 2021). According to Nelvia *et al.* (2012); Bonner *et al.* (2018); Ngatimin and Nasruddin (2019); Djohan *et al.* (2020), soil fertility can improve through the application of fertilizers and the presence of beneficial microbes in the soil. The application of fertilizers added chemical fertility and improves soil's physical and biological fertility. Kizilkaya and Dengiz (2010); Lange *et al.* (2015) and Huang *et al.* (2018) found the changes in land use from forest to agricultural and livestock land significantly affect a decrease in soil organic matter, porosity, the total nitrogen content, and stability of soil aggregates. Furthermore, Rousk *et al.* (2010) and Berek (2013) stated that soils on primary forest land commonly contain better chemical properties,

characterized by a pH in the neutral position (6.59), C-organic content (5.16%), N-total (0.53%), P-available (27.05%) and higher CEC (24.80) compared to agroforestry and cocoa plantations.

As we know, commonly soil conditions could be maintained in two ways: Physically and chemically. On the other hand, McGuire *et al.* (2010); Liu *et al.* (2018), and Djohan *et al.* (2020) found that soil organisms such as bacterial have a very important role, where soil organisms have the potential to change the organic matter in the fresh or semi-fresh or being weathered. Based the time, changes in organic matter become a form of other compounds that give more advantages to soil fertility. When knowledge is viewed from the plant aspect, there are two major soil organisms such Beneficial soil organisms and harmful or damaging other soil organisms. The beneficial groups of soil organisms include all organisms that carry out the weathering of organic matter to be inorganic and nitrogen. While the group of harmful organisms is a group of organisms that take the soil nutrients and caused nutrient competition with the primary plant. In the long processes, harmful organisms cause plants weaker (performance and adaptability), shortages, and susceptible attacking by pests and diseases.

The presence of soil and microorganisms is influenced by environmental conditions and depends on the type of soil uses and management (Saraswati *et al.*, 2007; Chodak *et al.*, 2016). The soil used in agriculture has criteria in the good environmental conditions to well maintain the microorganisms in the soil. The number and types of microorganisms found in the soil indicate the soil is fertile. The main indicators of fertile soil showed sufficient organic matter in the soil, sufficient water availability, unsupportive soil ecological conditions, and appropriate temperature and humidity (Irfan, 2014).

Microbes are known as soil organisms giving many benefits as components of natural habitats. The presence of soil microbes such as bacteria has essential roles and functions in supporting the implementation of environmentally friendly agriculture through various processes. The activities of bacteria include Decomposition of organic matter, mineralizing of organic compounds, nitrification, denitrification, nutrient fixation, and solvents. The roles of soil microbes especially bacteria activities still considered very important microorganisms and responsible for soil fertility. Also, the presence of bacteria becomes one of the main indicators in determining the soil quality index (Karlen *et al.*, 2006; Delgado-Baquerizo *et al.*, 2016b). When soil contains a higher beneficial microbes population improving the biochemical

activity and the soil quality index, and impacts plant development. The soil microbial populations are not pathogenic and are considered an indicator of being environmentally friendly in the agriculture system. Based on the statement previously, the research aims to determine the chemical properties of the soil and types of microbial (especially the presence of bacterial) in the cocoa farming system. The activities will support more environmentally friendly agriculture.

Materials and Methods

Research Location

The research was conducted from November 2018 to April 2019, in the cocoa farming system at Mamuju Regency, West Sulawesi, Indonesia (Fig. 1).

The location of research divided into six places namely: Kalonding, Boda Boda, Sisango, Salukayu 2, Salumasa and Batupapan. The soil chemical analysis was conducted at the Soil Science Laboratory, Faculty of Agriculture, Hasanuddin University. The bacteria culture analysis activities worked at the Faculty of Medicine, Hasanuddin University, Makassar Indonesia.

The Sampling Methods

Collecting soil as the samples of research was conducted randomly in the cocoa farming system. The soil samples taken were assumed to represent the soil properties of every research location. The whole soil sample used a sample ring and its composite was put in a plastic bag. The composite soil was analyzed for chemical properties used procedures: Soil sample from the field was air-dried, and sieved, then each sample (six samples) was analyzed its pH, content of N-total, Phosphorus, and Potassium available.

Analysis

The method used in analyzing the chemical, types, and amount of microorganisms (soil bacterial) contains in the soil samples from the cocoa farming system refer to Table 1.



Fig. 1: Research Location in The Cocoa Farming System, West Sulawesi, Indonesia

Table 1: Soil Samples Analysis Methods Used in The Research (Kizilkaya and Orhan, 2010; Umatermate *et al.*, 2014)

No	Parameter	Tool and Methods
A	Chemical Characteristic	
1	Soil pH (H ₂ O) and (KCl)	Extraction (1: 2.5)
2	C-organic	Walkley and Black
3	N-total	Kjeldahl
4	P ₂ O ₅	Bray and Olsen
5	K	Ammonium acetate extract (pH 7)
B	The Biological Analysis	
1	The type of soil bacteria	Culture
2	Total bacteria	Culture

The purpose of biological analysis or culture analysis is to determine the type and number of soil bacteria present in the soil sample. The activities are detailed as follows (Chaudhary *et al.*, 2019): (1) The soil sample weighed 10 g, put into BHIB (*Brain Heart Infusion Broth*) medium with a volume of 50 mL, homogenized (soil samples were finely separated), then incubated at 37°C for two hours; (2) inoculated samples that have grown microbes in BHIB on two different medium: MacConkey (MC) and Nutrient Agar (NA); (3) identification of organism colonies (especially soil bacterial) growing on MacConkey and NA medium used biochemical tests and (4) the biochemical test reactions conducted to determine bacteria species.

Results and Discussion

Soil Chemical Properties

The soil properties are very decisive in supporting plant growth and development including physical, chemical, and biological properties of the soil. The soil's chemical properties include soil pH and nutrient content (nitrogen, phosphorus, potassium, and organic matter) (Rousk *et al.*, 2010; Sitanala, 2010). The soil chemical analysis-based sample from six locations was showed in Table 2.

Based on Table 2, an analysis of the soil's chemical properties showed the soil pH in the research location was categorized as acidic to slightly acidic. Commonly acidic soil is caused by the H⁺ content in the soil higher than OH⁻. The high number of H⁺ ions is also caused by the high anaerobic activity of microorganisms in producing humid acids in the soil (Hardjowigeno, 2007; Irfan, 2014). Commonly aerobic conditions caused most soil microbes number increase in the huge population (Biswas *et al.*, 2000). In addition, the decomposition of organic matter as soon increases the availability of nutrients in the soil. The good condition of the environment becomes a source of energy for soil microbes. It means good quality of environment increasing the number and activity of soil microbes (Meryandini *et al.*, 2009; Huang *et al.*, 2018). Soil acidity is a common phenomenon in Indonesian areas. Areas with high rainfall caused alkaline leaching from the adsorption complex and run-off through drainage water. Besides the presence of aluminum and

hydrogen, the decomposition products of organic matter and pyrite oxidation also caused acid soil reactions (Delgado-Baquerizo *et al.*, 2016b). One of the decomposition products of organic matter is H₂CO₃. These compounds play a role in dissolving alkaline from the rocks. The alkaline will be quickly washed away or absorbed by plants. The loss of alkaline compounds is one cause of the growth of acid soil reactions, while the effect of H₂CO₃ on soil pH is not much because it is a fatty acid. The formation of organic acids such as H₂SO₄ and HNO₃ are commonly formed from the mineralization of organic matter. Based the activity of microorganisms on inorganic fertilizers is also a contributor to the acid reaction in the ecosystem (Rousk *et al.*, 2010; Begnon *et al.*, 2021).

The lowest nitrogen ranges were found throughout all of the study areas. This condition assumed the nitrogen transported at harvest was greater than the amount of nitrogen in the soil. Crowther *et al.* (2019) reported in addition, that inorganic nitrogen compounds are highly soluble and easily lost in drainage water or lost to the atmosphere. Most soil nitrogen comes from free air nitrogen and a small part comes from organic matter. The nitrogen from the atmosphere enters the soil through (1) Anchorage or fixation by microorganisms, in cases symbiotic with plant and non-symbiotic organisms, (2) rainwater incidence, and (3) fertilizers (Lange *et al.*, 2015). The plant can absorb nitrogen in the ion forming of NH₄⁺ and NO₃⁻. These ions in the soil come from the transformation process of organic and fertilizer. The transformation takes place through three stages: (1) Aminization, (2) ammonification, and (3) nitrification, respectively (Suryani and Zulfebriansyah, 2007).

The phosphorus (P) element was found in the low range for the entire study area or location. This is greatly influenced by nature, characterization of the soil, and management of the land by humans. For important information, the addition of phosphorus into the soil does not occur by biochemical binding such as nitrogen but only comes from deposits or rocks and minerals containing phosphorus in the soil. The availability of phosphorus was strongly influenced by pH number. The forming of phosphorus ions in the soil also depends on the pH of the solution. Phosphorus ions will easily combine with Al, Fe, or Mn at low pH, forming insoluble compounds. Meanwhile, soluble phosphorus ions will be bound by Ca at high pH to form insoluble compounds (Tilak *et al.*, 2005; Nurida *et al.*, 2012).

Potassium (K) was found in low amounts for all of the study areas. It is indicated limited availability in soil, is also affected by losses because of leaching by rainwater. The primary source of soil potassium is the earth's crust contains more acid and the mineral potassium. As an element, potassium cannot stand alone but is always present as a compound in various rocks, minerals, and salt solutions. Potassium cannot enter directly into the soil through other

sources, the circulation process of potassium is more likely to be lost. Most potassium is in primary minerals that are poorly soluble and not available to plants. The availability of potassium was defined as material that is possibly exchanged and absorbed by plants. Then potassium availability in the soil is highly dependent on the addition of external sources, fixation by the soil, and added potassium in the fertilizers (Karlen *et al.*, 2006; Suryani and Zulfebriansyah, 2007).

Schroth and Weinhold (1986); Fanin *et al.* (2012) and Crowther *et al.* (2019) found soil organic matter is all types of organic compounds contained in the soil including Litter, light organic matter fraction, microbial biomass, organic matter dissolved in water and stable organic matter or humus. Schmidt *et al.* (2011); Simanjuntak *et al.* (2013) and Delgado-Baquerizo *et al.* (2020) reported that soil organic matter is the multiple elements and playing important role in the ecosystem property. It must be broken down into simple compounds and then utilized optimally by plants. The provision of organic matter on land with a sandy texture needs more help to improve soil properties and increase the ability of the soil to bind the water surrounding them.

As we know, organic matter is a stabilizing material for soil aggregates. About half of the Cation Exchange Capacity (CEC) comes from organic matter as the source of plant nutrients. Based on their role, organic matter is the main energy source of most soil organisms. In playing this role, organic matter is primarily determined by its source and composition, the smoothness of its decomposition, and the results of the decomposition. Litter on moist soil undergoes fragmentation faster compared the dry litter on dry soil (Schmidt *et al.*, 2011; Fanin *et al.*, 2012). Handayanto and Hairiah (2007) stated that the characterization of soil organic matter could be performed in various ways. It includes chemical analysis, total C, and N. In addition, the application of compost and *Arbuscular Mycoriza Fungi* (AMF) on land containing sandy texture will improve soil aggregates and increase soil CEC (Nurida *et al.*, 2012). Moreover, the soil contains high organic matter content and has a high population and activity of microorganisms, including beneficial bacteria.

Number and Types of Soil Bacterial

The soil bacterial is one of the famous microbes that play important roles in the decomposition process in the environment. In general, the function of microbes in nature is divided into four tasks: (1) Increasing the availability of plant nutrients in the soil; (2) remodel of organic matter in the soil and mineralization organic elements; (3) rhizosphere-endophytic bacteria have specific function stimulate plant growth by forming enzymes and protecting roots from pathogenic microbes; (4) the biological control agents of pests and plant diseases, respectively (Saraswati *et al.*, 2007). The decomposition

and mineralization of nutrients from organic matter in the soil and nitrogen fixation by *rhizobium*. The soil bacterial known as *Rhizobium* is the microbes with the ability to catch free nitrogen from the atmosphere. For the information, soil microbial activities play an essential role in increasing soil fertility. In addition, microbes as intermediaries in chemical reactions and physical, metabolic processes on the surface and the soil. This organism can reduce the negative impact of heavy metal complexity contamination that is commonly found in agricultural land. The effort to clean the soil from chemical contamination has converted complex or straightforward chemical compounds into harmless material. The list containing the number and types of bacteria found in the soil colonies at the six locations was showed in Table 3.

Based on Table 3, the analysis results found three genus of bacteria found in the soil at the research location: *Alcaligenes*, *Klebsiella*, and *Pseudomonas*. Commonly the presence of microorganisms such as bacteria in the agricultural field is strongly influenced by pH number and soil organic matter content such as the availability of nitrogen, phosphorus, potassium, and C-organic elements. The presence of microorganisms such as fungi is more adaptable at all soil acidity levels than the bacterial population. At the same time, the presence of bacteria (including fixation type) is very influenced by soil pH. Hardjowigeno (2007), Rousk *et al.* (2010), and Hanafiah (2018) state that the optimum range of pH is about 5.5 resulting in the good development of bacteria. In contrast, if the number pH is below 5.5, the bacterial growth is disrupted. Unfortunately, we are not counting the number of bacteria at the beginning of the research because we focused on the types of bacteria around the cocoa farming system.

Alcaligenes is the bacterial genus with character: The reaction is gram-negative, aerobic, and rod-shaped. The type of bacterial included in the genus of non-fermenting bacteria. Some kinds of *Alcaligenes* are capable of anaerobic respiration, if available nitrate or nitrite supports the process. In addition, *Alcaligenes* sp. is an essential bacterial species used for biotechnology development because the microbes produce a plastic-like material such as Polyhydroxybutyrate Biopolymer (PHB). Chaudhary *et al.* (2019) stated that the advantage of PHB is easier degraded by biological processes and does not leave polluting debris in the environment. In addition, the bacterial population commonly found in soil help more in metabolize, and neutralize organic and inorganic matter in the agricultural field.

Klebsiella has the opposite character to *Alcaligenes* because they have a gram-negative reaction, oxidase negative, rod-shaped completing with prominent capsule-based polysaccharides. The bacterial cells are rod-shaped,

wide about 0.3 to 1.5 μm ; long about 0.5 to 5.0 μm . The better condition for growing *Klebsiella* is between 35 and 37°C, pH of around 7.2. Commonly the species of *Klebsiella* live as facultative anaerobic. Tilak *et al.* (2015) and Delgado-Baquerizo *et al.* (2016b) found that most bacterial strains can survive on citrate and glucose as main carbon sources, including ammonia as an important source of nitrogen. *Klebsiella* as a microbe is easier found on a variety of host plants. The species of *Klebsiella* such as *Klebsiella pneumoniae* and *Klebsiella oxytoca* play tasks as fixed-atmospheric nitrogen and preparing into the form ready for the plant. On the other hand, the character of *Klebsiella* is called associative nitrogen fixed.

Besides the presence of *Alcaligenes* and *Klebsiella*, *Pseudomonas* sp. is the free-living or nitrogen-fixing bacterial in the root and tissue areas of rice plants. This genus lives around the cocoa farming ecosystem. These bacteria can fix free nitrogen from the atmosphere, both symbiotic and non-symbiotic. stated that the use of nitrogen-fixing bacteria in agriculture, whether applied through the soil or sprayed on plants, gives benefits in increasing the efficiency of nitrogen fertilization. To achieve the goal of environmentally friendly and sustainable agriculture, the use of nitrogen-fixing bacteria very flexible reduces the demand for natural nitrogen fertilizers, increasing plant productivity and farm income with good inputs cheaper than synthetic fertilizers (Sitanela, 2010). Although we are not counting several bacterial, based presence of soil bacterial in the research location, we assumed the soil bacterial give more help increasing the fertility of the soil and increasing plant health. As we know, litter content in the soil plays a role in protecting microbes from the extreme condition. This is very useful knowledge to manage sustainable agriculture and reduce pesticide application.

Table 2: Soil Chemical Analysis from Six Locations in Mamuju Regency

Profile	Location	pH Extract 1:2.5		C-Organic (%)	N (%) Kjeldahl	P (ppm) Bray	K (cmol/kg)
		H ₂ O	KCl				
1	Kalonding	4.73	3.57	1.39	0.20	15.60	0.16
2	Salukayu 2	5.76	5.00	1.87	0.23	15.76	0.19
3	Batupapan	4.91	3.36	2.10	0.22	15.70	0.16
4	Boda Boda	5.21	4.47	2.56	0.25	15.67	0.15
5	Salumasa	4.93	3.65	2.20	0.23	15.76	0.21
6	Sisango	5.07	4.23	2.06	0.22	15.91	0.13

Table 3: Number and types of bacteria from Six Locations in Mamuju Regency

Profile	Location	Growing Colony/ Colony Forming Unit (cfu)		Bacterial Genus
		MacConkey (MC)	Nutrient Agar (NA)	
1	Kalonding	287	289	<i>Pseudomonas</i> sp.
2	Salukayu 2	250	255	<i>Alcaligenes</i> sp.
3	Batupapan	265	265	<i>Klebsiella</i> sp.
4	Boda Boda	235	238	<i>Pseudomonas</i> sp.
5	Salumasa	215	220	<i>Alcaligenes</i> sp.
6	Sisango	295	298	<i>Pseudomonas</i> sp.

Besides beneficial bacterial application in supporting environmentally friendly agriculture, giving compost treatment to the soil increases the decomposition of organic matter by microorganisms. Glaser *et al.* (2002); Handayanto and Hairiah (2007) stated that the addition of organic matter and phosphate solubilizing microorganisms could increase the availability of phosphorus and indirectly increase the soil microelements. The example other genera of bacteria such as *Pseudomonas fluorescens* and *Bacillus thuringiensis* are phosphate solubilizing bacteria. They have the function of increasing phosphorus available in the soil and plants can utilize it for their development.

The kind of bacteria found in the research location was classified as essential microbial. They are not pathogenic in supporting the implementation of environmentally friendly agriculture through various processes, such as decomposition of organic matter, mineralization of organic compounds, nutrient fixation, nutrient solvents, nitrification, and denitrification. The activity of soil microorganisms is directly proportional to the total number of microorganisms in the soil. If the total number of microorganisms is higher, the movement of microorganisms increases. The results of the microorganism analysis showed that the Kalonding area had the highest number of microbes (289 cfu), Batupapan (265 cfu), Salukayu 2 (255 cfu), Boda Boda (238 cfu), Sisango and Salumasa (220 cfu), respectively. The higher number of microorganisms in the soil means the soil is more fertile and suitable for plant growth.

The use of soil fertilizing microbes (contain beneficial bacterial) can provide various benefits: (1) providing known source nutrients for plant growth; (2) protecting roots from attacking pests and diseases; (3) stimulating the root system to develop and extend root fully growth; (4) stimulating tissue mitosis meristem at the point of growth of shoots, flower buds, and stolon; (5) playing task as a safe antidote to some land covered heavy metals and contamination; (6) the growth regulator metabolite and (7) bio activator for organic fertilizer (Chaudhary *et al.*, 2019). Overall, the result showed the research location was classified as generally less fertile. The assumed a high rainfall in the research location and the top layer of soil enriched by organic matter as a source of nutrients. The material is easily washed away by soil erosion. To avoid and keep soil healthy by applying compost or natural organic fertilizer from a large number of rotting plant residues around the plant, then improve activities of drainage. When used chemical fertilizers, must be balanced amount according to the needs of each commodity on the plantation.

Conclusion

The chemical properties of the soil samples from six research locations indicate the soil pH ranges from acidic to slightly acidic, with a pH between 4.73-5.76. All of the chemical components range in the low status (the C-organic level, N content, Phosphorus, and Potassium criteria).

The genus of bacteria found in the research location: *Pseudomonas* in Kalonding (289 cfu), Boda Boda (238 cfu), and Sisango (220 cfu). The another bacterial *Alkaligenes* were found in Salukayu 2 (255 cfu) and Salumasa (220 cfu). Especially *Klebsiella* was found only in Batupapan (265 cfu).

Author's Contributions

Ida Suryani: Designed the field and laboratory work, identified the soil bacterial, collected the data and writing of the manuscript.

Sulfiana and Maimuna Nontji: Participated in the data analysis and wrote the manuscript.

Marliana: collected soil microbial, participated in the field and laboratory work and wrote the manuscript.

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 that no ethical issue is involved.

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