Biological characterization of geo-referenced soil samples from major ~~rice growing~~rice-growing tracts of Southern Kerala

Abstract

A Survey was accomplished to figure out the soil biological fertility index in the five ~~agro ecological~~agro-ecological, units of rice growing belts of Southern Kerala*viz.*,Onattukara sandy loam (AEU 3), Kari soil (AEU 4), Pokkali soil (AEU 5), Southern laterites (AEU 8) and Midland laterites (AEU 9). Twenty ~~geo referenced~~geo-referenced, surface soil samples @ 0- 15 cm were collected in every AEU ~~in accordance with~~by the prescribed method. The collected samples were subjected to the characterization of soil biological properties such as dehydrogenase activity, organic carbon, microbial biomass C, microbial biomass N, and soil respiratory rate following the standard protocol. The findings showed that organic carbon in AEU 5 with the highest mean value (2.73 %), followed by AEU 4 (2.19 %), AEU 9 (2.01 %), AEU 8 (1.06 %), and AEU 3 (0.60 %). Dehydrogenase activity was highest in AEU 4 (593.01µg TPFhydrolysed per gram per 24 hr), followed by AEU 5, AEU 9, and AEU 8, and lowest in AEU 3 (79.30 µg TPFhydrolysed per gram per 24 hr). AEU 5 exhibited the highest microbial biomass carbon and microbial biomass nitrogen activities, while AEU 8 showed the lowest. Soil respiration was highest in AEU 5 and lowest in AEU 3. Based on these soil microbiological indicators, soils were classified into three categories: low, medium, and high fertility. The Biological Fertility Index (BFI) scores indicated favorable conditions in AEU 4 and 5, moderate in AEU 9, and low in AEU 3 and 8.

~~Key words~~Keywords: ~~Agro ecological~~Agroecological, units, Soil biological properties ~~Biological~~biological, fertility index, Rice, Wetland soils

Introduction

Soil biological characteristics reflect both the direct and indirect impact of the organisms residing within a specific soil. These biological processes involve the cycling of carbon and nutrients, biotic interactions contributing to soil formation, microbial abundance, enzyme functions, as well as microbial biomass of C, N, and S (Azcon-Aguilar and Barea 2015).Among the array of enzymes present in soil environments, dehydrogenase holds significant importance as ~~they~~ it~~serve~~ servesas ~~a~~ anindex of overall microbial activity within the soil. Their significance lies in the fact that they are intracellular and present in all living microbial cells (Moeskops*et al.* 2010; Zhao *et al.* 2010; Yuan and Yue. 2012). Furthermore, their function is closely associated with microbial oxidoreduction processes. Soil organic matter (OM), is regarded as a key indicator of soil quality due to its role as a reservoir and contributor of nutrients that can improve soil physical and chemical attributes, while also stimulating biological processes (Salazar *et al*, 2011). Remarkably, it's not solely the quantity of OM in the soil that holds significance, but primarily its quality, as OM influences the provision of energy for microbial proliferation and the production of enzymes. Manral*et al.* (2023) revealed that the microbial biomass within soil significantly influences soil processes like nitrogen mineralization and serves as a bio-indicator

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of current climatic shifts. Soil microorganism activity in organic matter decomposition governs the mineralization and immobilization of nutrients, influencing nutrient availability in soil crucial for plant growth. Soil microbial biomass functioning as a reservoir and consumer of accessible nutrients, holds significant importance in the transformation of nutrients within the soil. Alterations in microbial biomass have the potential to impact the turnover of soil organic matter. Consequently, soil microbial activity directly influences the stability and fertility of soil. The assessment of microbial biomass serves as a valuable tool for evaluating soil quality. Soil respiration quantifies the CO_2 emitted by soil, indicating the collective CO_2 release from various living organisms such as bacteria, fungi, earthworms, protists, roots, and more. These measurements serve in carbon balance calculations and serve as an index for soil health (Gao *et al*. 2020). At the global scale, soil fertility is emerging as a significant concern because changing climatic conditions are leading to declines in soil properties attributed to alterations in soil biota, encompassing both fauna and flora (Gurjar*et al*. 2017). Kerala state delineated into twenty-three ~~agro-ecological~~agroecological units has been established considering climate, topography, and soil characteristics. Each AEU corresponds to specific soil and climatic conditions. Therefore, the present study is envisaged to investigate the biological properties of various ~~agro-ecological~~agroecological units and thematic maps were generated highlighting the biological fertility index of wetland soils of Southern Kerala.

Materials and Methods

Initially, a survey was accomplished in the five Agro ecological units of ~~rice producing~~rice-producing zones of Southern Kerala *viz*., Onattukara sandy loam (AEU 3), – ORARS, pathiyoor, cheppadandThattarambalam is located in the kayamkulam municipality of Alappuzha district,Kari soil (AEU 4), Pokkali soil (AEU 5)- Kumbalangal,pallurthy, Kalluchira and Kuzhuppilly of Ernakulam district, Southern laterites (AEU 8)- Vellayani, Chenkal, and Kalliyoor of Thiruvanthapuram district and Midland laterites (AEU 9)-Chenapara, Karikoam, Kuttikonam and Vettikavala.Wetland soil samples were collected @ 0-15 cm as per the standard procedure. The biological properties including dehydrogenase activity, organic carbon, microbial biomass carbon, microbial biomass N, and soil respiratory rate were estimated. The method for measuring dehydrogenase activity, as outlined by Casida *et al.* (1964), involved the utilization of 3% 2, 3, 5-triphenyl tetrazolium chloride (TTC). The concentration of dehydrogenase present in the sample was determined by creating a standard graph employing triphenyl formazon (TPF) as a reference. The enzyme activity was then quantified as micrograms of TPF released per gram of soil per 24 hours. The soil organic carbon was analyzed by the protocol stated by (Walkely and Black. 1934). The microbial biomass carbon was determined by the fumigation-incubation technique described by (Jenkinson and Ladd. 1981) and is expressed as µg per gram of soil. Brookes *et al*. (1985) ~~was~~ outlined the protocol used by estimating soil microbial biomass nitrogen by chloroform fumigation and indicated as µg of nitrogen per gram of soil. Anderson (1982) outlined the method for determining ~~the~~ soil respiration activity. The respiratory activity of the soil samples was determined by collecting and quantifying the CO2 evolved from a fixed quantity of incubated soil in standard alkali. According to Brookes (1995), soils were classified

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into three categories *viz.* low, medium, and high based on indicators of soil biological properties. The biological fertility index was determined ~~in accordance with~~by the score assigned to the biological fertility index. Thematic maps, illustrating the soil of five agro-ecological units, were produced using ArcGIS software.

Creation of maps using geographic information systems

Using Geographic Information Systems (GIS), a geo-referenced thematic map of the biological fertility index for five ~~Agro Ecological Units~~agroecological units (AEUs) was created in ArcGIS. The map illustrates the spatial distribution of the relative biological fertility index across wetland soils in various AEUs of southern Kerala were presented in Fig 1.

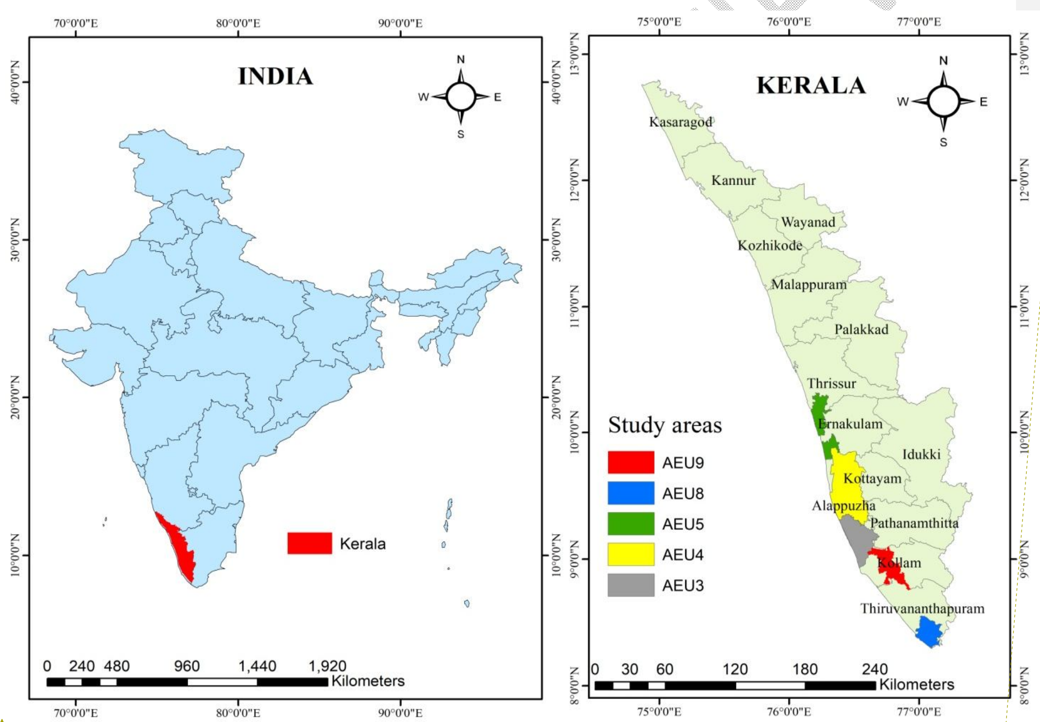

Fig 1: Location map of study area different AEUs

Results and discussion

Organic carbon

Soil samples were collected from 20 locations in different Agricultural Ecological Units (AEUs). The highest mean value of organic carbon was recorded in AEU 5 (2.73 %), followed

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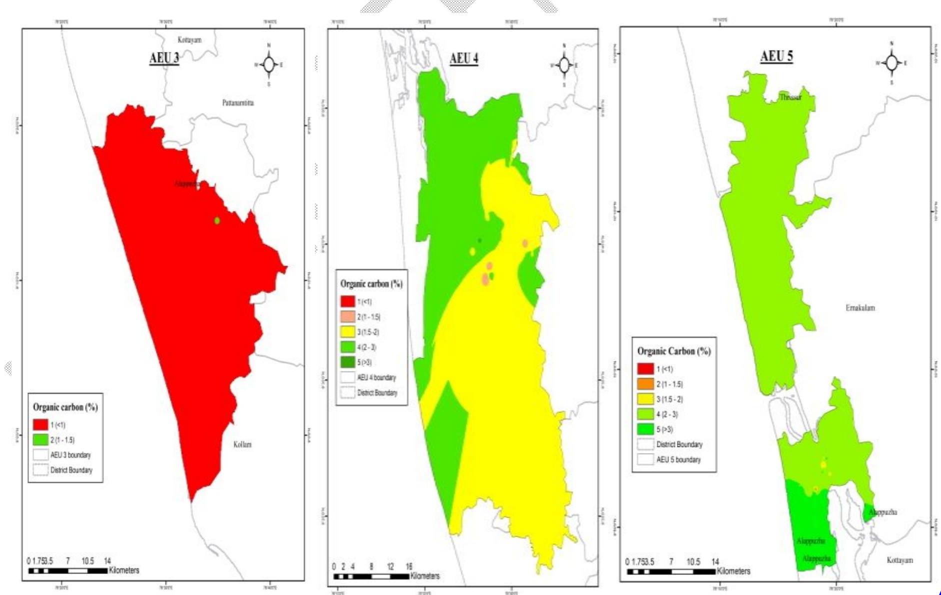
by AEU 4 (2.19 %), AEU 9 (2.01%), AEU 8 (1.06 %), and AEU 3 (0.60 %). Organic carbon content ranged from 0.02 to 1.00 in AEU 3 ~~indicated~~ indicatingthat low to medium organic carbon, 1.00 to 3.14 in AEU 4 was medium to high organic carbon content, 1.00 to 6.20 in AEU 5 showed medium to high organic carbon content, 0.52 to 3.38 in AEU 9, the organic carbon content was from medium to high range, and 0.50 to 1.86 in AEU 8 it was varied from low to medium organic carbon content Fig 2. The low organic carbon content in AEU 3 is attributed to its lateritic soil type and sandy texture. High temperatures and heavy rainfall in this region accelerate the decomposition of organic matter, thereby reducing organic carbon content. Conversely, AEU 4 predominantly features high organic carbon content (>1.5%), largely due to the abundance of partially decomposed fossil woods and roots at various stages of decomposition. The high organic carbon status is also due to the slow decomposition rate of large amounts of organic matter under flooded anaerobic conditions (Unnikrishnan and Jayashree. 2021). AEU 5, characterized by Pokkali soils, is known for its high fertility and acid saline conditions, along with high organic carbon content. This is likely due to the presence of partially decomposed roots and other organic materials (Beena *et al*. 2017). In AEU 9, the accumulation of organic matter is attributed to leaf litter, plant residues, and root biomass, which enrich the soil. The slow decomposition rates in anaerobic conditions further contribute to the high organic matter levels. AEU 8 has medium organic carbon content, with a moderate quantity of dry matter and a range of major and minor nutrients being added to the soil (Aparna *et al*. 2023; Sheeba *et al*. 2019).



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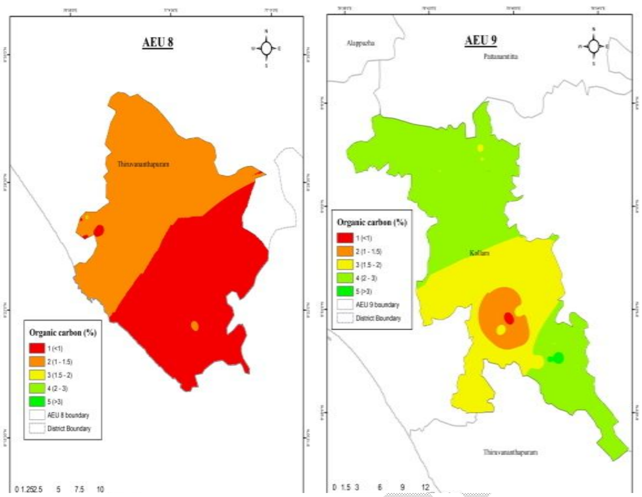

Fig 2: Spatial distribution of soil organic carbon of AEU 3, 4, 5, 8 and 9

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Dehydrogenase

Dehydrogenase ~~play~~ playsa key role in the biological oxidation of soil organic matter, and ~~their~~ itsactivity is used as an indicator of microbial activity in the soil. The range of dehydrogenase activity was 121.89 to 888.68 in AEU 4 with the highest mean value was (593.01µg TPFhydrolysed per gram per 24 hr) followed by a range 288.58 to 886.00 in AEU 5 with ~~the~~ amean value of (489.35µg TPFhydrolysed per gram per 24 hr), 177.85 to 355.48 in AEU 9 with mean value (242.93µg TPFhydrolysed per gram per 24 hr), 48.79 to 158.33 in AEU 8 with the mean value (90.74µg TPFhydrolysed per gram per 24 hr) and the lowest activity was AEU 3 with mean value (79.30µg TPFhydrolysed per gram per 24 hr) Fig 3. Unnikrishnan and Jayashree (2021) reported that dehydrogenase activity is highly correlated with the organic carbon content in soil. Higher levels of organic matter can provide ample substrate to support greater microbial biomass, which in turn leads to increased enzyme production (Yuan and Yue. 2012).

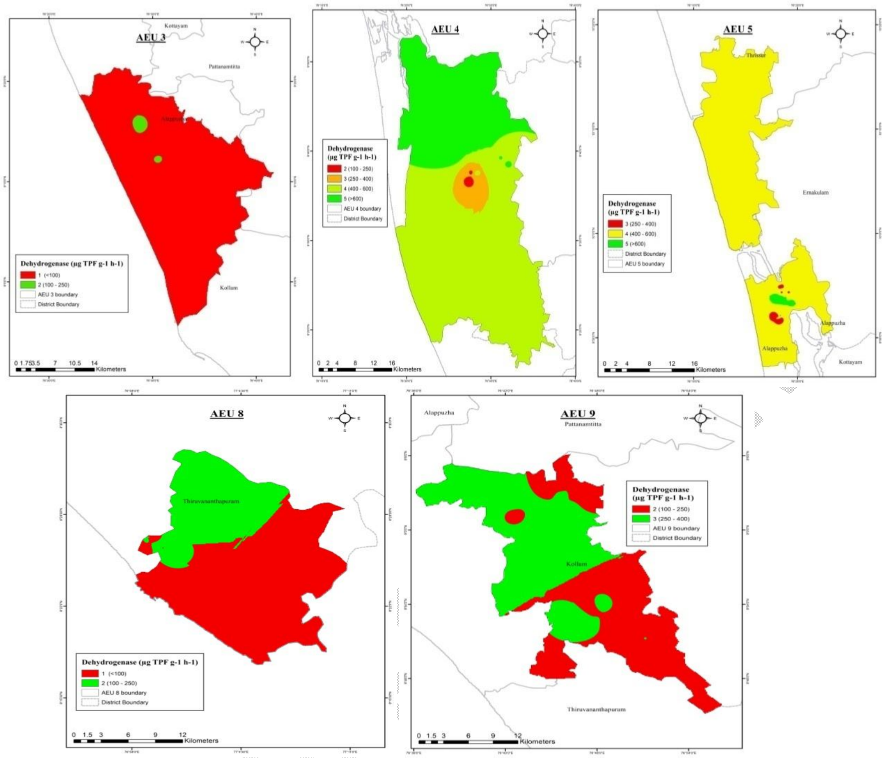

Fig 3: Spatial distribution of soil dehydrogenase ~~activityof~~ activity of AEU 3, 4, 5, 8, and 9

Microbial biomass carbon

The mean value of microbial biomass carbon (MBC) was highest in AEU 5 (483.83µg g^{-1} of soil), followed by AEU 4 (441.29µg g^{-1} of soil), AEU 9 (435.78µg g^{-1} of soil), AEU 3 (160.09µg g^{-1} of soil), and lowest in AEU 8 (158.68µg g^{-1} of soil). The MBC values ranged from 68.56 to 276.20 in AEU 3, 288.66 to 699.15 in AEU 4, 199.43 to 825.10 in AEU 5, 103.14 to 675.28 in AEU 9, and 41.28 to 264.21 in AEU 8 Table 1. Joseph (2014) reported that microbial biomass carbon in Pokkali soil varied from 50.12 mg kg^{-1} to 658.52 mg kg^{-1}. Soil microbial biomass is highly sensitive to even slight changes in the organic matter content, which directly serves as an energy source. There is a positive correlation between organic carbon and microbial biomass carbon (Haripal and Sahoo. 2014)

Microbial biomass nitrogen

The microbial biomass nitrogen was found to be highest mean value at AEU 5 (193.53µg g^{-1} of soil) and varied from 79.77 to 330.04 followed by AEU 9 (173.17µg g^{-1} of soil) and the range from 58.67 to 270.11, AEU 4 (146.07µg g^{-1} of soil) varied from 79.61 to 236.08, AEU 3

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(76.09 µg $g^{-1}$of soil) from 33.55 to 138.10 and lowest MBN was noticed in AEU 8 (57.81µg $g^{-1}$of soil) varied from 35.58 to 89.22 Table1.

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~~Table.~~Table 1. Mean value of microbiological parameters in various ~~agro ecological~~agro-ecological units

	OC (%)	Range	Dehydrogenase (µg TPF hydrolysed per gram per 24 hr)	Range	Microbial biomass carbon (µg g^{-1} of soil)	Range	Microbial biomass nitrogen (µg g^{-1} of soil)	Range	Soil respiration (mg g^{-1} of soil)	Range
AEU 3	0.60	0.02 to 1.00	79.30	58.40 to 121.47	158.68	68.56 to 276.20	76.09	33.55 to 138.10	2.75	1.87 to 5.70
AEU 4	2.19	1.00 to 3.14	593.01	121.89 to 888.68	441.29	288.66 to 699.15	146.07	79.61 to 236.08	4.81	2.88 to 11.2
AEU 5	2.73	1.00 to 6.20	489.35	288.58 to 886.00	483.83	199.43 to 825.10	193.53	79.77 to 330.04	7.76	5.28 to 11.01
AEU 8	1.06	0.50 to 1.86	90.74	48.79 to 158.33	160.09	41.28 to 264.21	57.81	35.58 to 89.22	1.62	1.45 to 4.07
AEU 9	2.01	0.52 to 3.38	242.93	177.85 to 355.48	435.79	103.14 to 675.28	173.17	58.67 to 270.11	3.51	2.80 to 6.70

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Soil respiration

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Soil respiration, which refers to the CO_2 produced by the biological activity of soil organisms, is a significant component of the global carbon cycle (Phillips *et al.* 2015). The highest mean value of soil respiration was observed in AEU 5 (7.76 mg g^{-1} of soil), with a range from 5.28 to 11.01. This was followed by AEU 4 (5.52mg g^{-1} of soil) with a range from 2.88 to 11.21, AEU 9 (3.51 mg g^{-1} of soil) ranging from 2.80 to 6.70, AEU 3 (2.75 mg g^{-1} of soil) ranging from 1.87 to 5.70, and the lowest was recorded in AEU 8 (1.62 mg g^{-1} of soil) with a range from 1.45 to 4.07. Soil respiration rates typically increase with ambient temperature, as higher temperatures accelerate carbon cycling processes (Tang *et al.* 2019).

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Biological fertility index

The Biological Fertility Index (BFI) is a metric used to evaluate the potential fertility of soil. It takes into account various biological factors that contribute to soil fertility, including the presence and activity of soil microorganisms, organic matter content, and the soil's ability to support plant growth (Francaviglia*et* al. 2017). The Biological Fertility Index (BFI) is an evaluation system that measures soil health and fertility by considering several key parameters: soil organic carbon, soil respiration, dehydrogenase, ~~and~~ soil microbial biomass carbon, and soil microbial biomass nitrogen. Each parameter is divided into five intervals, with each interval assigned a score from 1 to 5. These scores are based on findings from previous research (Brookes 1995; Vance *et al*. 1987). The overall biological fertility is determined by summing the scores of all parameters, which then classifies the soil into different levels of biological fertility, as illustrated in ~~Table~~ Tables2 and 3.

Based on the Biological Fertility Index (BFI) scores, the selected ~~Agro Ecological Zones~~agroecological zones (AEUs) in the study were classified as follows: AEU 3 had a BFI score of 10, placing it in the Low BFI class. AEU 4 had a BFI score of 19, classifying it as Good biological fertility. AEU 5 had a BFI score of 21, indicating Very Good biological fertility. AEU 8, with a BFI score of 8, was categorized as Low. AEU 9, with a BFI score of 17, placed into the Medium fertility class Fig 4 and Fig 5. The highest BFI scores were attributed to the high organic matter content and microbial activity in the soil.

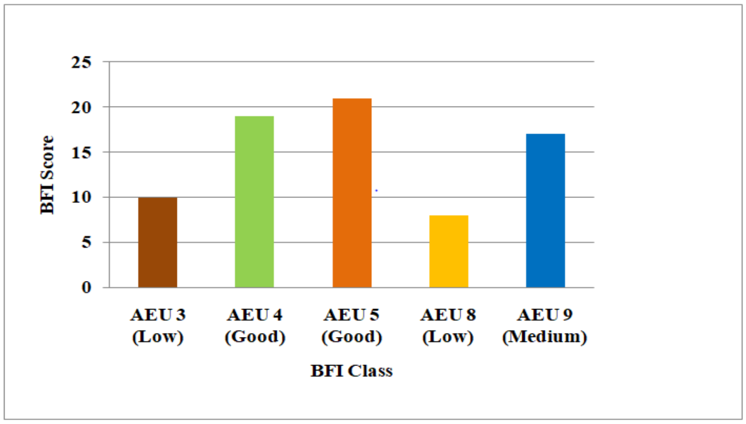
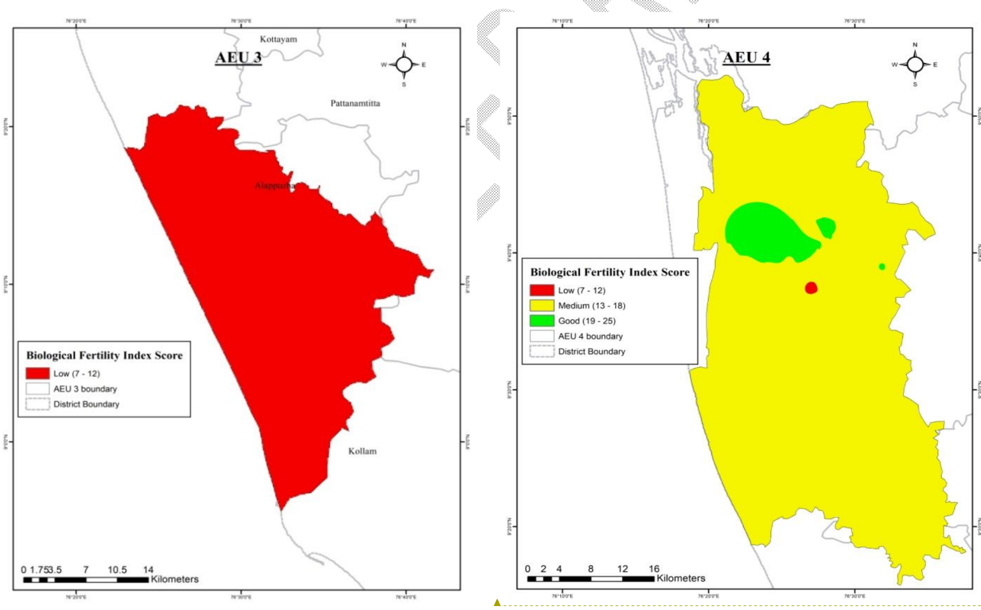


Fig.4. Biological fertility index (BFI) scores and classifications for various agro-ecological units



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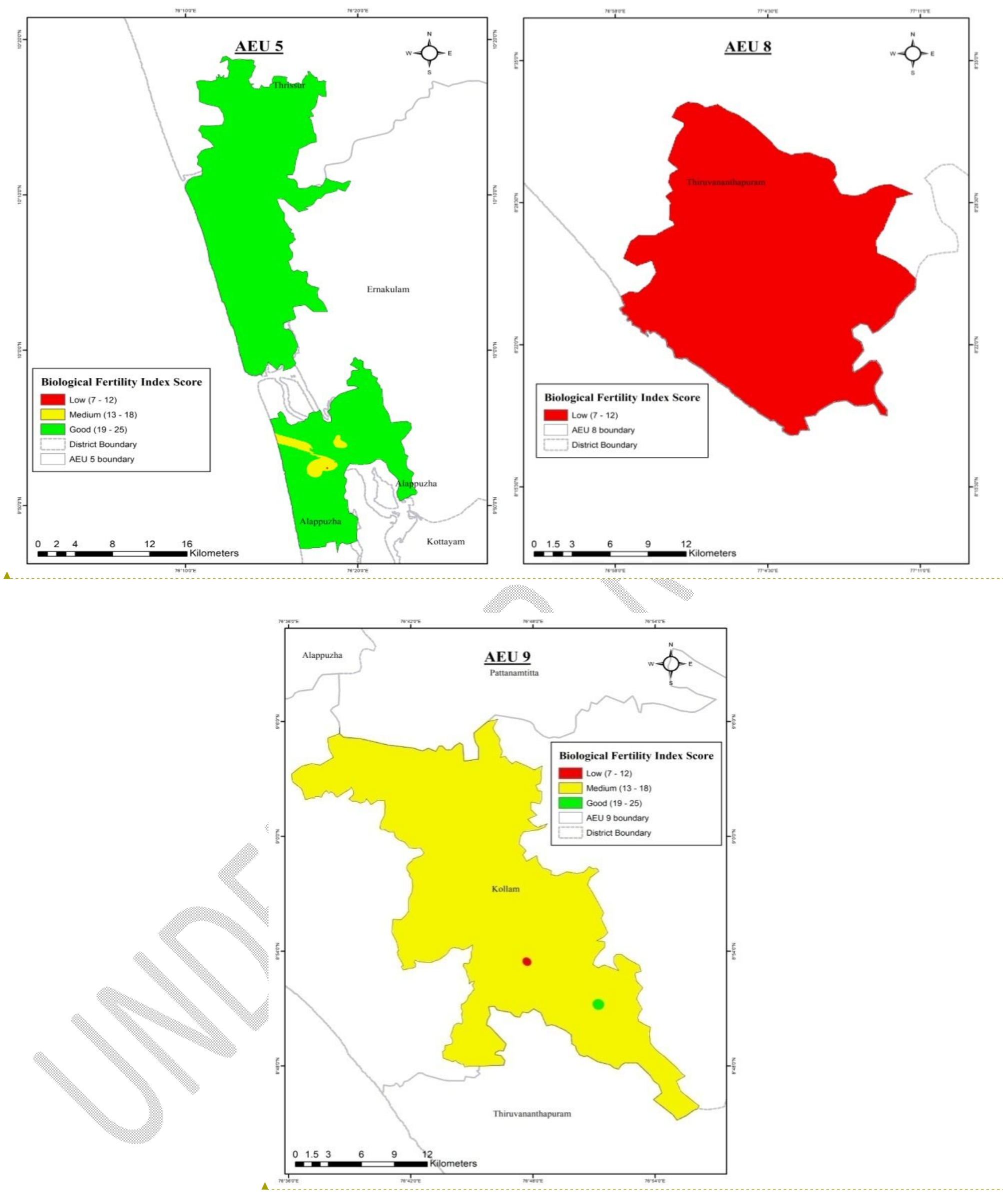

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Fig.5. Spatial distribution of soil dehydrogenase of AEU 3, 4, 5, 8 and 9

~~Table.~~Table 2. Biological fertility index score for the interval value of different parameters

Parameter	Scores				
	1	2	3	4	5
Organic carbon	<1.0	>1.0 to <1.5	<1.5 to >2.0	>2.0 to <3.0	>3.0
Dehydrogenase	<100	>100 to <200	>250 to <400	>400 to <600	>600
Microbial biomass carbon	<100	>100 to <200	>200 to <300	>300 to <400	>400
~~MIcrobial~~ Microbialbiomass nitrogen	<50	>50 to <100	>100 to <150	>150 to <200	>200
Soil respiration	<5	>5 to <10	>10 to <15	>15 to <20	>20

~~Table.~~Table 3. Categories of Biological ~~fertility index~~Fertility Index

Biological fertility class	Low	Medium	Good	Very good
Biological fertility scores	7-12	13-18	19-25	>25

Correlation analysis

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Pearson correlation analysis across ~~Agro-ecological~~Agroecological units shows a strong positive correlation between dehydrogenase activity, microbial biomass carbon, microbial biomass nitrogen, and soil respiration with organic carbon content. As organic carbon increases, these microbiological parameters also rise, emphasizing its role in soil health and microbial activity. Blue circles represent positive correlations, brown circles indicate negative ones, and circle size reflects correlation strength (r), with significant correlations at ($p \leq 0.5$). Fig. 6.

Fig.6.Pearson correlation correlogram of various microbiological parameters across Agro-Ecological Units

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The study on the biological characterization of geo-referenced soil samples from Southern Kerala provides crucial insights into the soil microbiological health across various ~~agro-ecological~~agroecological units (AEUs) in a rice-growing region. By measuring parameters like dehydrogenase activity, organic carbon content, microbial biomass, and soil respiration, the research offers a detailed understanding of soil fertility. Notably, AEU 5 (Pokkali soil) exhibited the highest biological fertility, followed by AEU 4 (Kari soil), underscoring the significance of organic carbon and microbial activities in enhancing soil productivity. These findings are essential for improving rice cultivation practices and tailoring soil management strategies specific to each AEU, aiming to sustain productivity in ecologically varied zones (Lobo et al. 2023).

When comparing this research with studies on soil quality (Araya-Alman et al. 2020; Calero et al. 2022; Campos, 2023) and agro-environmental factors in Latin America (Olivares et al. 2022; Rey et al. 2022; Olivares, 2022, Rodriguez et al. 2023) similar methodologies often focus on soil microbiological health as a determinant of fertility and crop yield (Hernandez and Olivares, 2019)~~;~~. However, the biological fertility indexes (BFI) employed in the Kerala study offer a structured framework that could complement soil studies in Latin America, where extensive variability exists due to tropical (Hernandez et al. 2018a; Hernandez and Olivares, 2020), subtropical, and Andean agro-ecological zones (Hernandez et al. 2019b; 2018c). In Latin American studies, soil organic carbon and microbial activity are also key indicators of soil health, as seen in research conducted in regions such as the Amazon Basin and Andean highlands (Hernandez et al. 2017; Hernandez et al. 2020). These studies often emphasize the interplay between soil microorganisms, carbon cycling, and sustainable agricultural practices (López et al. 2019; López and Olivares, 2018).

The Kerala study's use of geo-referenced sampling provides a valuable model for Latin American contexts, where spatial variability in soil properties can significantly impact crop productivity (Rodriguez et al. 2016a; Olivares et al. 2015b). Both regions share challenges related to land degradation and the need for site-specific management practices to enhance soil fertility (Olivares, 2016; Olivares et al. 2015a; Olivares et al. 2011). While Kerala's rice-growing tracts and Latin America's diverse cropping systems differ, the comparative use of soil microbiological indicators in assessing agro-environmental factors reveals converging themes (Rodriguez et al. 2016b; Rodriguez et al. 2013). Such cross-regional studies help shape more effective agricultural management policies, emphasizing the critical role of soil biology in both tropical and subtropical farming systems (Montenegro et al. 2021a; 2021b).

Conclusion

The present study concluded that Agro-ecological Zones AEU 5 was the highest biological fertility index, followed by AEU 4, AEU 9, AEU 3, and AEU 8. Soil biological properties can be improved through various practices that enhance the activity and diversity of soil microorganisms, promote healthy plant growth, and maintain soil structure. Some of the effective strategies like using organic amendments like manure application, crop management practices like crop rotation, and cover cropping, nutrient management like balanced fertilization ~~and~~ green manuring, etc.

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