

Comparison of Soil Carbon-Nitrogen Ratio at Two Different Mangrove Ecosystem in Bali, Indonesia

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ABSTRACT

The mangrove ecosystem significantly contributes to nutrient and carbon exchange. It is primarily stored in the soil as organic matter, significantly benefiting the surrounding organisms. However, it could be changed depending on its surrounding conditions. This research aimed to determine the percentage of soil carbon-nitrogen and its ratio in two mangrove ecosystems, one with high anthropogenic impact (Tahura Ngurah Rai) and the other on a small island (Lembongan Island). We collect soil samples on 14 plots at each station at 0–30 cm depth and use carbon titration and TN-Kjeldahl methods for soil carbon-nitrogen measurement. The result shows substantial disparities in soil carbon levels between these ecosystems, but the soil nitrogen content was comparable. Two specific plots at Tahura Ngurah Rai (T8 and T11) were found at low soil carbon levels due to the damage to the mangrove forest. The C/N values vary between stations, primarily because of their different sources (Tahura Ngurah Rai: human activities, Lembongan: marine organisms). The C/N value at Tahura Ngurah Rai is higher than the Redfield ratios, while Lembongan Island is on the contrary. However, its levels at both stations are still categorized as common conditions for mangrove ecosystems compared to various sites in Indonesia. Future research will involve measuring radioisotope characteristics to verify the origin of nutrients in these ecosystems. Obtaining measurements of environmental parameters is also necessary to provide a more comprehensive explanation of the results.

Keywords: Tahura Ngurah Rai, Lembongan Island, anthropogenic, redfield ratio.

INTRODUCTION

Mangrove ecosystems play a crucial role in preserving the equilibrium of coastal ecosystems. Mangrove ecosystems have a crucial role as the primary source and reservoir of organic matter, mainly carbon (C) and nitrogen (N), in coastal and marine regions (Saavedra-Hortua et

al., 2020; Sarker et al., 2021). Most carbon and nitrogen stocks are primarily stored as biomass or accumulate in soil (Shiau and Chiu, 2020). On a global scale, the total amount of carbon stored in mangrove soil can reach 4.5 petagrams (Inoue, 2019), and the nitrogen content can reach 52.03 megagrams per hectare (Alongi, 2020). The organisms living in mangrove ecosystems and their

surroundings mostly rely on these elements for their food supply (Queiroz et al., 2020; Jiang et al., 2022; Tan et al., 2022).

Carbon (C) and nitrogen (N) are essential elements that play a crucial role in the life of species in mangrove ecosystems and their adjacent areas. The C/N ratio in mangrove soil impacts the growth and survival of microorganisms inhabiting these soils (Purahong et al., 2019; Pradisty et al., 2021). A lower carbon-to-nitrogen (C/N) ratio can suggest an abundance of nutrients, while a higher C/N ratio can imply a deficiency of nutrients (Servais et al., 2019). Nutrient pollution can disrupt the organic matter decomposition process by microbes, producing harmful chemicals, including H_2S , CH_4 , and NH_4^+ (Vincent et al., 2021). It can affect the quality of water and the overall health of mangrove ecosystems.

The content and proportion of carbon, nitrogen, and their ratio in mangrove soil varies based on the specific location, environmental conditions, and the type of mangrove species present. The stable carbon-nitrogen ratio typically adheres to the Redfield ratio of 106:16, as demonstrated by Azwa et al. (2022) and Mamidala et al. (2022). Nevertheless, mangrove ecosystems located near human settlements or impacted by human-generated garbage would exhibit distinct carbon and nitrogen levels in the soil compared to pristine mangrove ecosystems (Feng et al., 2019; Romero-Mujalli and Melendez (2023). Nguyen et al. (2022) discovered that human activities significantly impacted carbon-nitrogen ratios in mangrove ecosystems, with anthropogenically altered ecosystems reaching ratios as high as 25.9, whereas wild mangrove ecosystems only reached 16.9. Deviation from the usual carbon-nitrogen ratio values in a mangrove environment can suggest pollution or nutrient depletion, as indicated by studies conducted by Queiroz et al. (2020) and Chen and Tan (2021).

Ngurah Rai Grand Forest Park (TAHURA Ngurah Rai) and Nusa Lembongan Mangrove are two distinct mangrove ecosystems that vary in size and geographical location. The mangrove ecosystem of TAHURA Ngurah Rai is situated in the southern part of Bali Island, covering an area of 1,373.5 hectares. Numerous coastal settlements surround it, and the convergence point for five significant rivers (Loloan, Buaji, Badung, Mati, and Sama Rivers) that flow into the vicinity (Central Bureau of Statistics Denpasar City, 2022). The Nusa Lembongan Mangrove is

a compact mangrove environment situated on a small island with a total area of 202 hectares. It is sparsely populated, with just 8,890 individuals residing in the vicinity, according to the Central Bureau of Statistics Nusa Penida in 2021. However, these mangrove ecosystems share similarities regarding the major mangrove species belonging to the genus *Rhizophora* (Palguna et al., 2017; Sugiana et al., 2022). The existing research primarily focuses on quantifying organic carbon levels in mangrove soil. In TAHURA Ngurah Rai, the carbon stock soil was found to be $216.17 \text{ MgCha}^{-1}$, according to Mahasani et al. (2016); however, in Nusa Lembongan Mangrove, it was only 68.10 MgCha^{-1} , as reported by Priscilla et al. (2021). Additional research is required on nitrogen stock and carbon-nitrogen ratios to address the insufficient data. This research will contribute to evaluating the health status of mangrove ecosystems, particularly in the two specified locations.

This study aims to quantify the carbon and nitrogen content in mangrove soil and determine the ratio between the two elements in two distinct mangrove ecosystems. The mangrove environment of Tahura Ngurah Rai is situated near numerous coastal communities, which possess the capacity to generate carbon and nitrogen pollution. In contrast, the Nusa Lembongan mangrove ecosystem remains comparatively pristine. Based on such characteristics, we predicted that there would be a substantial difference in the carbon and nitrogen values and ratios between the two stations. Understanding the carbon-nitrogen ratio is crucial for comprehending the ecology and well-being of mangrove ecosystems. Furthermore, it can contribute to preserving the long-term viability of mangrove ecosystems for both the environment and humans, as anomalous ratio values can serve as indicators of issues within the mangrove ecosystem.

METHOD

Study site

The research was carried out during low tide at two locations: the mangrove ecosystems in Ngurah Rai Grand Forest Park (Tahura Ngurah Rai) (-8.699762°S - 8.798416°S , 115.178133°E - 115.251148°E) and Lembongan Island (8.663148°S - 8.694237°S , 115.446668°T - 115.473524°T). The mangrove ecosystems in Tahura

Ngurah Rai and Nusa Lembongan differ in their surrounding conditions. In Tahura Ngurah Rai, the mangroves are predominantly bordered by numerous towns and river outlets, but in Nusa Lembongan, there are only a limited number of settlements. Fourteen soil samples were collected from each location (Figure 1). The sampling was conducted in December 2022, during the rainy season.

Soil collection and carbon-nitrogen laboratory analysis

Methodology for soil sampling and preparation

Soil samples were collected using a soil auger during low tide. The soil auger was submerged to a 0–30 cm depth below the surface and then carefully raised to retrieve the soil samples without damage. The initial step involved separating soil samples from mangrove trash to minimize any potential bias in the test results. Subsequently, the samples were homogenized, and up to 100 grams were obtained to examine total organic carbon and

nitrogen. The wet soil samples were subsequently subjected to drying at a temperature of 80 °C until a consistent weight was achieved, which typically took around 48 hours. Subsequently, the desiccated soil samples were pulverized using a mortar and filtered using a 2-mm sieve. To the organic carbon and total nitrogen content, three-gram samples of soil that could pass through a filter with a maximum opening size of 2 millimeters were collected. An analysis was performed at the soil laboratory of Udayana University.

Soil carbon-nitrogen measurement

The dry soil samples were filtered and subsequently subjected to extraction using the acid-base method. It involved using sulfuric acid (H₂SO₄) to separate the organic carbon. The procedure involved utilizing a reflux apparatus consisting of a flask equipped with a condenser, where soil samples and H₂SO₄ were inserted. The device was then heated to a temperature of 120 °C. The acid solution containing organic carbon was

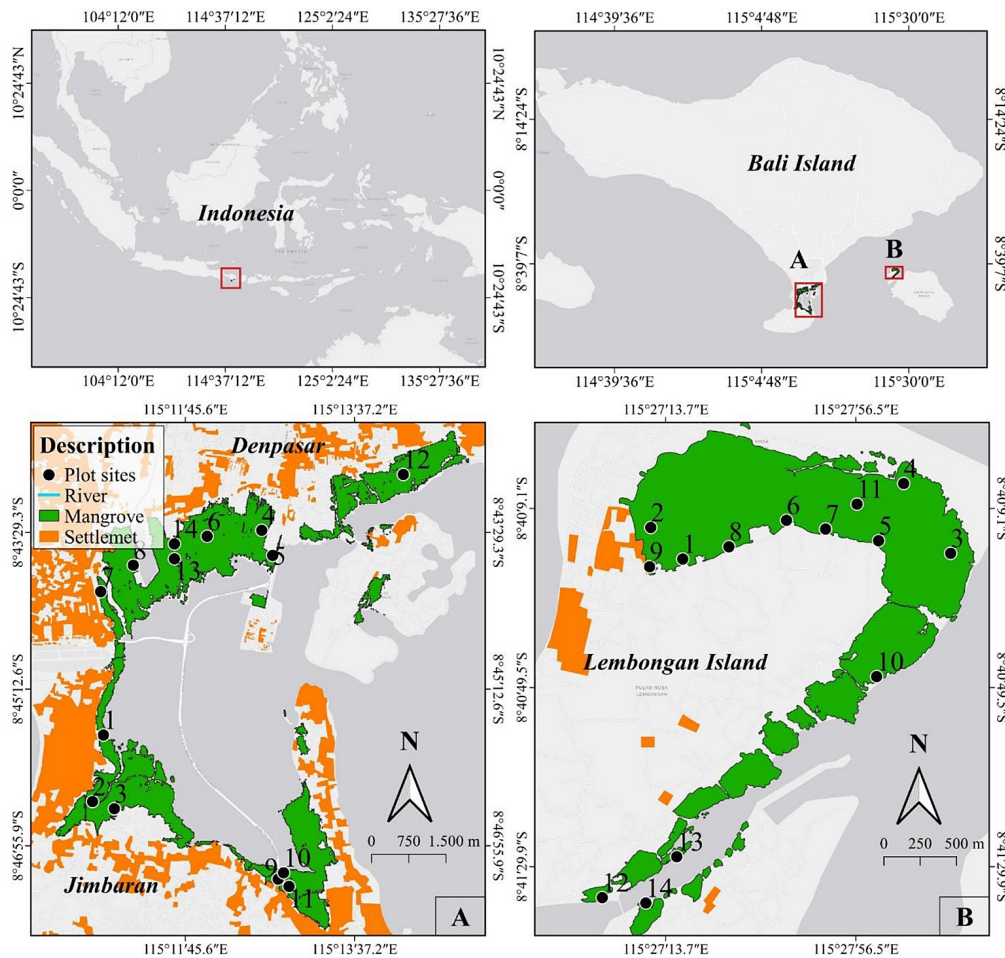


Figure 1. Distribution of sampling plots of each station

diluted with distilled water. Subsequently, the acid solution, which had been diluted, was titrated using a standardized solution of potassium dichromate ($K_2Cr_2O_7$). To ascertain the equivalence point, titration is conducted using a colour change indicator called phenolphthalein. The organic carbon concentration is determined by measuring the quantity of standard solution required to reach the equivalence point, considering the dilution factor and the volume of the soil sample.

Meanwhile, the determination of the total nitrogen content is nearly identical, involving the dissolution of the sample in sulfuric acid (H_2SO_4) with the inclusion of a catalyst, copper sulphate ($CuSO_4$). The dissolution process is conducted in a Kjeldahl flask equipped with a condenser to prevent the loss of vapor. In addition, the flask is subjected to a temperature of $350\text{ }^\circ\text{C}$ to transform organic nitrogen into ammonia. Upon heating, the chemical mixture will be introduced into the distillation system, where it will undergo a reaction with sodium hydroxide solution ($NaOH$). The end outcome is the production of vapor containing ammonia, which is then absorbed by a solution of boric acid (H_3BO_3) to create an ammonium salt and phenolphthalein indicator. The concentration of total nitrogen is determined by observing the colour change of the indicator, which is estimated using the dilution factor and the volume of the extracted soil sample.

Mapping the spatial distribution of carbon-nitrogen concentrations and ratio

The spatial distribution of organic carbon parameters, nitrogen, and their ratio is determined using the interpolation approach, specifically the IDW (inverse distance weighting) plugin in QGIS 3.2. The interpolation procedure involves inputting field measurements in latitude and longitude coordinates and concentration values for each parameter. The interpolation results are further segmented according to the size of each mangrove region to ascertain the distribution pattern.

Statistical analysis

The Shapiro-Wilk normality test was used to analyze univariate data, specifically percentages of organic carbon, nitrogen, and their ratios. The purpose was to evaluate the distribution of data among observation stations. Due to the non-normal distribution of the data ($p > 0.05$), the Kruskal-Wallis non-parametric test was employed to

assess the magnitude of difference between each parameter across stations. The study was conducted using Rstudio 4.0.2 software, following the principles outlined by Lubis (2021), with a statistical significance threshold set at 0.05.

RESULT AND DISCUSSION

Percentage and distribution of soil carbon

The soil carbon % exhibits variability throughout the collection points of each research station. The Mangrove Tahura Ngurah Rai exhibited a more significant soil carbon content percentage, ranging from 0.79% to 4.30%, with an average of $2.92 \pm 1.03\%$. The Lembongan Mangrove had a soil carbon content ranging from 0.79% to 3.90%, with an average of $1.97 \pm 1.10\%$ (Figure 2). The spatial distribution analysis revealed that the carbon content was only poor at two specific locations, namely T8 and T11, in Tahura Ngurah Rai. In contrast, over half of the data collecting stations in Lembongan exhibited low soil carbon values, namely below 2%. Plots with low soil carbon were observed near land at both stations. In particular, the Lembongan Mangrove had low soil carbon levels, with additional low soil carbon sample points discovered in mangroves near the sea (Figure 3). The Kruskal-Wallis non-parametric test revealed a statistically significant disparity in soil carbon levels between the two stations (Kruskal-Wallis; $H_{2,14} = 4.670$, $p = 0.031$).

Soil carbon variations in mangrove ecosystems are influenced mainly by internal factors, including hydrological and vegetative conditions (Hapsari et al., 2022). Differences in hydrological conditions, particularly pH, oxygen availability (DO), nutrients, water content, temperature, and salinity, can change the usefulness of carbon stored in the soil for microbial metabolism under aerobic and anaerobic conditions. Consequently, this can result in fluctuations in carbon availability in the soil (Chen et al., 2016). The amount of carbon stored in the soil is influenced by vegetation conditions, with each species of mangrove having a distinct rate of soil carbon deposition (Chen et al., 2021). Furthermore, degraded mangrove ecosystems tend to release stored soil carbon due to their diminished capacity to sequester carbon in their roots (Das et al., 2022; Romero-Urbe et al., 2022). Mangroves that have undergone degradation will exhibit less canopy cover, leading to more sunlight penetration into the soil.

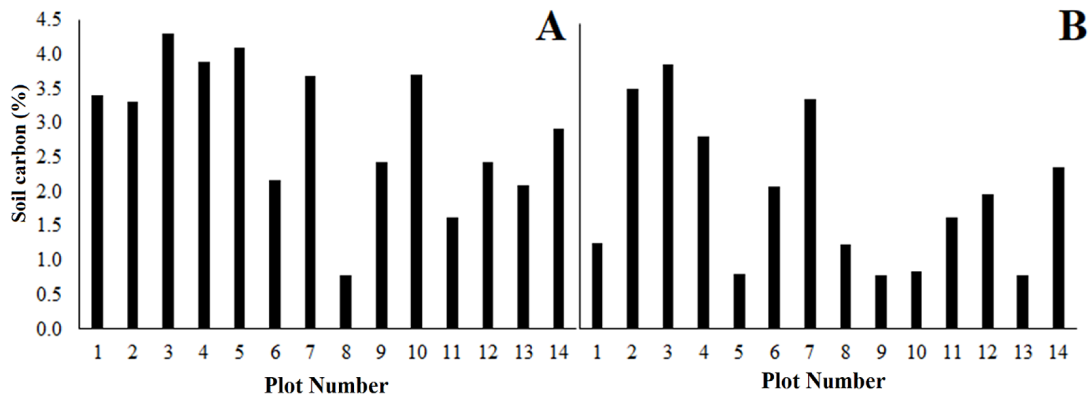


Figure 2. Percentage of soil carbon at both research stations

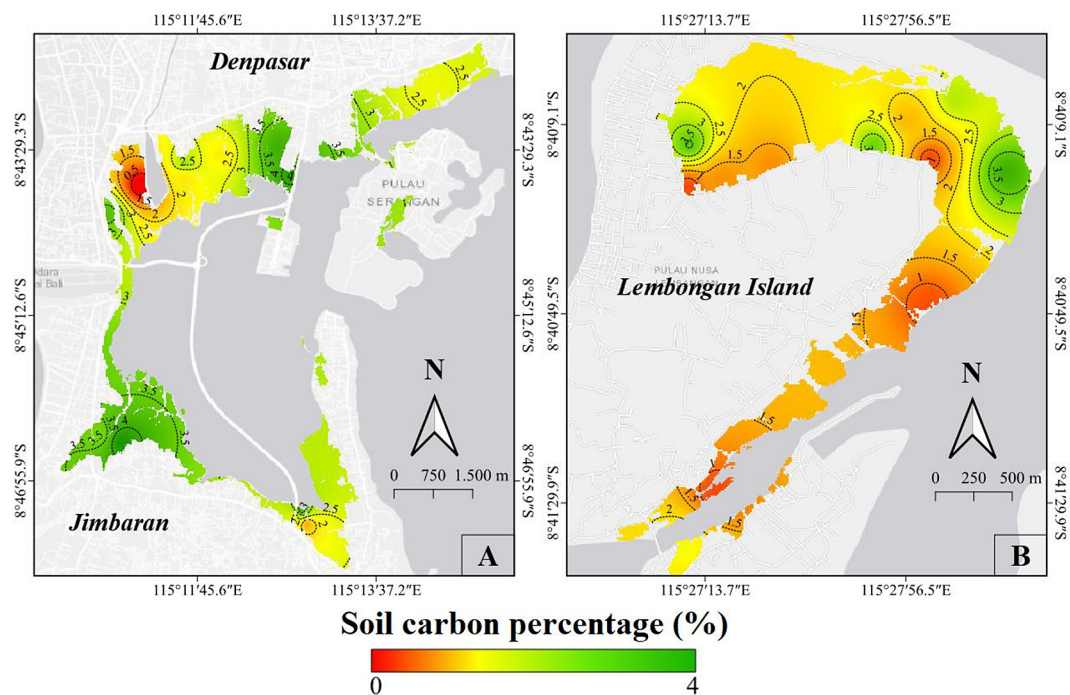


Figure 3. Spatial distribution of soil carbon percentage at both research stations

Consequently, this will elevate the metabolic activity of bacteria that rely on carbon deposited in the soil (Liu et al., 2020). The presence of low soil carbon at plots number 8 and 11 in Tahura Ngurah Rai and plots number 1, 5, 8, 9, 10, and 13 in Lembongan (Figure 4) can be attributed to this factor.

External factors such as position and soilation rate can influence measured soil carbon values. Coastal areas with mangrove vegetation typically have lower soil carbon levels due to the regular tidal flushing they experience, resulting in the accumulation of less carbon in these regions (Chen et al., 2021; Yin et al., 2023). It explains the rationale behind the proximity of plots number 10 and 13 in Lembongan to the sea,

where the soil carbon content is relatively low. Additionally, changes in soil accretion rate have an impact on soil carbon levels. Specifically, smaller soil particles such as silt and clay have a higher capacity to retain organic matter, mainly carbon (Matsui et al., 2015). According to Yin et al. (2023), soil containing smaller particles will have higher levels of carbon. However, the notable disparity in the proportion of carbon stocks at the two stations might also be attributed to a small river estuary in Tahura Ngurah Rai, whereas Lembongan lacks a flowing river. Therefore, the carbon stocks in Tahura Ngurah Rai have a more significant total percentage than those in Lembongan. Comparing the average soil carbon stores at both sites to several studies conducted in Indonesia,



Figure 4. Mangrove damage in Tahura Ngurah Rai (left) and Nusa Lembongan (right)

the figure appears to be relatively lower. According to Marbun et al. (2020), the study revealed that soil carbon stocks in mangrove habitats in Bolaang Mongondow Regency, North Sulawesi, tended to be more significant, with an average of $10.57\% \pm 4.87\%$. Similarly, the Kawal and Lagoi areas in Bintan Island, Riau, have an average mangrove coverage of 10.35% and 4.84%, respectively, according to Hapsari et al. (2022). The Dukuh Setapak Mangrove Area in Semarang has a mangrove coverage of 4.4%, as reported by Hakim et al. (2016). Nevertheless, the carbon stock percentage at both stations remains greater compared to the converted mangrove lands in Indramayu, West Java ($1.52 \pm 0.24\%$) and Pati, Central Java ($1.96 \pm 0.18\%$), as reported by Kepel et al. in 2018. The variation in soil carbon percentage can be attributed to the mangrove ecosystem's sampling position and geomorphology type. Large flowing rivers characterize the locations with higher soil carbon percentages, whereas the research station represents a mangrove ecosystem primarily influenced by tides.

Percentage and distribution of soil nitrogen

Contrary to the soil carbon percentage, Lembongan mangrove soil nitrogen percentage is generally elevated compared to TAHURA Ngurah Rai. In Lembongan, the average percentage of soil nitrogen is $0.19 \pm 0.14\%$ (range: 0.06%–0.58%). In TAHURA Ngurah Rai, the average percentage of soil nitrogen is $0.14 \pm 0.054\%$ with a range of 0.07%–0.24% (Figure 5). The second point in Lembongan, located near the settlement,

exhibits the highest levels of soil nitrogen, whilst nearly all places near the sea display the lowest values. Concurrently, the TAHURA Ngurah Rai station exhibited a relatively uniform distribution of soil nitrogen, as depicted in Figure 6. Nevertheless, the two stations exhibit comparable values, as indicated by the Kruskal-Wallis values: $H_{2,14} = 0.511$, $p = 0.475$.

Variations in soil nitrogen levels in mangrove ecosystems can arise from external inputs, such as rivers, or biological influences. Mangrove areas near the river estuary typically exhibit elevated nitrogen levels. Mangrove zones more secluded from external sources generally exhibit reduced nitrogen content due to limited fertilizer availability (Palufi et al., 2019). Biological processes also influence soil nitrogen fluctuations. Biological activities, such as denitrification processes, also impact nutrient cycling and nitrogen utilization in mangrove soils (Queiroz et al., 2019). Mangrove areas with dense vegetation and more biodiversity exhibit heightened microbial activity and decomposition processes. This action enhances the nitrogen supply in soil by accelerating the breakdown of organic materials. Conversely, mangrove zones that are more open and have less dense vegetation generally exhibit reduced biological activity, leading to decreased nitrogen content in soil (Kanti et al., 2019).

In contrast to the study conducted by Ardhani et al. (2020), the soil nitrogen content in Mangrove Kabupaten Demak, Central Java, is relatively elevated, ranging from 0.65% to 0.85%

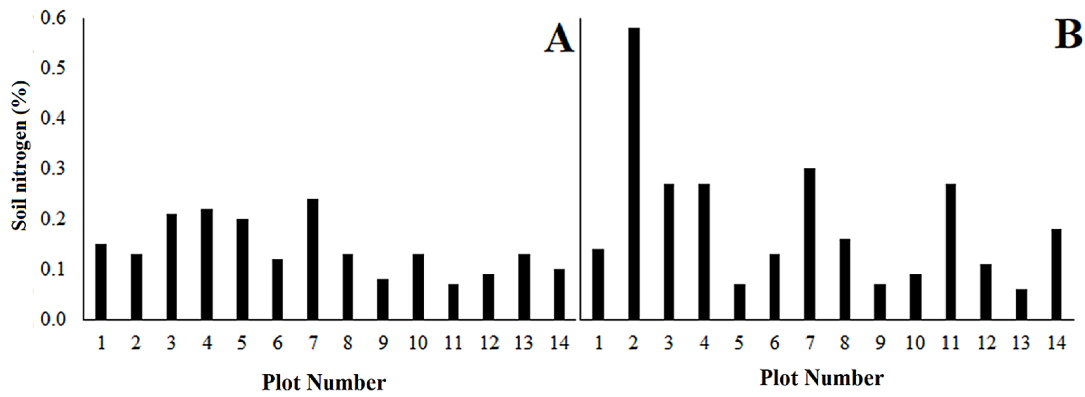


Figure 5. Percentage of soil nitrogen at both research stations

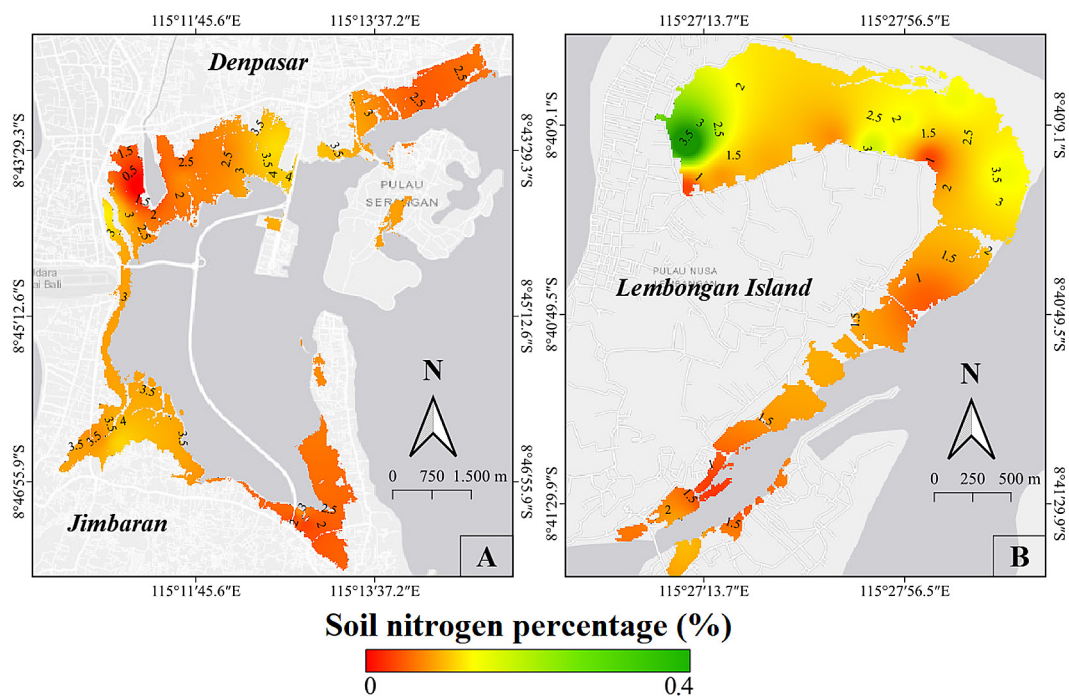


Figure 6. Spatial distribution of soil nitrogen percentage at both research stations

throughout both research stations. Nevertheless, the levels of soil nitrogen in Kepulauan Seribu, Jakarta, and Pangkal Balam Harbour, Bangka, are relatively lower, ranging from 0.02% to 0.04% and 0.01% to 0.06% correspondingly (Palufi et al., 2019; Sadewi et al., 2022). The disparity can be elucidated by the origin of nitrogen, where the mangroves in Demak Regency originate from former ponds and are situated near a river mouth. Conversely, most nitrogen is derived from the mangrove vegetation in Kepulauan Seribu, Pangkal Balam Harbour, and at the research stations (TAHURA Ngurah Rai and Lembongan). It is inclined to be influenced by hydrological processes.

Soil C/N ratio

There was variability in the carbon-nitrogen (C/N) ratio between the different locations. The Ngurah Rai Mangrove TAHURA exhibited the most significant ratio values, ranging from 6.1 to 30.4 (mean: 21.5 ± 6.6 or C:N = 117:6). In contrast, the Lembongan Mangrove had a C/N ratio of only 6.1 to 18.1, with an average of 11.3 ± 3.5 or C:N = 59:6 (Figure 7). The C/N distribution pattern in TAHURA Ngurah Rai is comparable to the distribution of soil carbon. Plot number 8 exhibits the lowest value, while plot number 12 in Lembongan, near the sea, demonstrates the highest C/N ratio (Figure 8).

The observed outcome can be attributed to the diminished soil carbon concentration in TAHURA Ngurah Rai at plot number 8. Conversely, in Lembongan, although plot number 12 does not exhibit the maximum soil carbon levels, the soil nitrogen content is comparatively lower. Overall, the two stations exhibited markedly distinct C/N values (Kruskall-Wallis; $H_{2,14} = 13.176, \rho = 0.0003$).

The variations in carbon-nitrogen (C/N) ratios in mangrove soil are expected to be constrained by the presence and attachment of carbon and nitrogen in the soil. Variances in C/N ratios are induced by distinct sources of these two elements (Hossain et al., 2016; Hu et al., 2022). Tahura Ngurah Rai, for instance,

has small rivers that provide more soil organic carbon than Nusa Lembongan, where the carbon input solely originates from the mangrove vegetation. Tidal movements also impact the distribution of carbon and nitrogen. The mangroves of Nusa Lembongan are small, exposed islands influenced by the sea. In contrast, Tahura Ngurah Rai has a land effect through the river. It can induce fluctuations in environmental factors such as salinity, dissolved oxygen, nutrients, and pH in the soil, hence leading to variations in the process of carbon-nitrogen binding and the microbial activity that depends on it (MacKenzie et al., 2021).

The C/N values serve as indicators of both the origin of the carbon-nitrogen deposits and

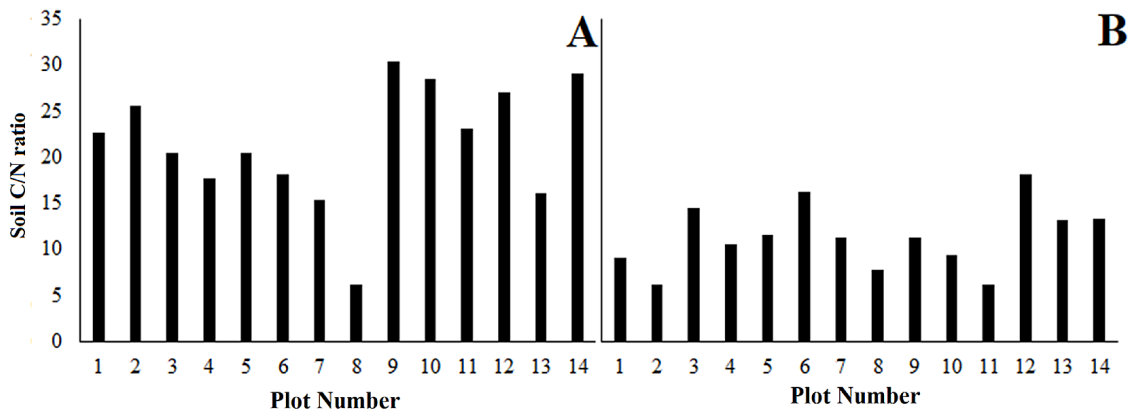


Figure 7. Soil C/N values at both research stations

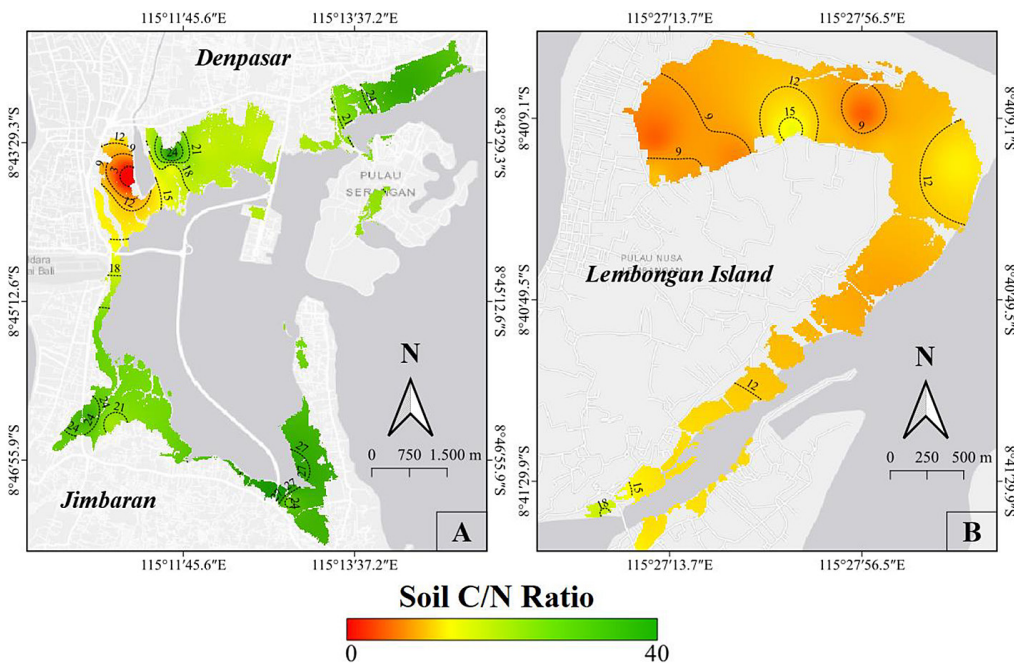


Figure 8. Spatial distribution of soil C/N at both research stations

the presence of nutrient contamination in mangrove soil (Xia et al., 2021). The C/N value conforms to the Redfield ratio of 106:16 or 6.625, indicating that the findings recorded at both stations are comparatively elevated compared to typical conditions. The Nusa Lembongan mangrove is categorized as a natural ecosystem, but Tahura Ngurah Rai has nearly half its territory designated as a rehabilitation area for former ponds (JICA, 1999). Nguyen et al. (2022) found that the average C/N ratio in undisturbed mangrove soil is 16.9, but soil impacted by human activities had a higher C/N ratio of 25.9. It indicates that the soil's carbon-to-nitrogen ratio (C/N) in Nusa Lembongan Mangrove is still lower than that of natural settings.

On the other hand, the C/N ratio in TAHURA Ngurah Rai is like the typical conditions observed in regions impacted by human activity. The decreased carbon-to-nitrogen ratio (C/N) of Mangrove Nusa Lembongan can be attributed to carbon and nitrogen supplies originating from marine biota. The C/N ratio of marine biota is often lower than terrestrial biota due to their higher nitrogen content (McGroddy et al., 2004; Li et al., 2016; Liu et al., 2020). Therefore, it may be inferred that the nutrient sources in the soil of Nusa Lembongan mangroves primarily originate from the mangrove vegetation and other marine organisms. However, Mangrove Tahura Ngurah Rai primarily derives from vegetation and inputs from the surrounding land. In comparison to research conducted on other mangrove ecosystems in Indonesia, the C/N values observed at the two research stations were generally higher than those found in the mangroves of Kepulauan Seribu, Jakarta (7.5–8, Palufi et al., 2019), and Mangrove Demak Regency, Central Java (2.4–3.4, Ardhani et al., 2020), both of which experience more frequent tidal inundation. In the same geographical area of Nusa Lembongan, Priscillia et al. (2021) conducted measurements of C/N under constrained conditions, specifically using just five samples taken at a depth of 30 cm. The results obtained were comparatively higher, ranging from 16.8 to 20.5. Meanwhile, in the mangrove community of Pangkal Balam Harbour, Bangka, the carbon-to-nitrogen ratio (C/N) in soil was discovered to be exceptionally elevated, reaching a value of 244 (Sadewi et al., 2022). It demonstrates that each mangrove habitat, characterized by distinct geomorphological, hydrological, and vegetation health conditions, can have various C/N values influenced by their respective sources.

CONCLUSIONS

The findings revealed a discrepancy in the soil carbon-nitrogen levels between Mangrove Tahura Ngurah Rai and Mangrove Nusa Lembongan. Tahura Ngurah Rai exhibits a more significant soil carbon percentage than Nusa Lembongan, whereas both areas have a similar soil nitrogen percentage. The C/N data have confirmed that the two observed mangrove ecosystems exhibit considerably distinct values. This variation is attributed to the geographical location of each mangrove ecosystem. Mangrove Nusa Lembongan is more susceptible to tidal influence, which can result in the redistribution of nutrients in the soil. As a result, the nutrient levels in this area are relatively low. In contrast, Tahura Ngurah Rai is minimally affected by tides and benefits from the presence of rivers that transport nutrients to the mangrove ecosystem. Furthermore, based on the C/N values, it can be inferred that the two stations have distinct carbon-nitrogen sources. Tahura Ngurah Rai receives contributions from human activities, whilst Lembongan benefits from additional nutrients derived from marine organisms.

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