

# Bacterial Community of Klong Tub Mangrove Forest in Chonburi Province, Thailand

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## ABSTRACT

Mangrove forests are located in the transition zone of terrestrial and river/marine ecosystems, making these forests a unique environment harbouring diverse microbes. This study investigated the bacterial community of Klong Tub Mangrove Forest in Chonburi Province, Thailand. The distinct feature of this forest is its nearby location to a narrowleaf cattail wetland. Assessment of the abiotic parameters of the sediments from site#1 nearby the narrowleaf cattail wetland and site#2 in the mangrove forest revealed differences in pH and salinity values between these two sites. Biochemical identification of bacterial isolates (n=233) indicated that these species belonged to 16 families and 29 genera as follows: *Moraxellaceae* (17.60%) > *Vibrionaceae* (16.31%) > *Paenibacillaceae* (15.88%) > *Staphylococcaceae* and *Bacillaceae* (9.87% each) > *Aeromonadaceae* and *Pseudomonadaceae* (8.58% each) > *Enterobacteriaceae* (4.29%) > *Lactobacillaceae* (2.58%) > *Moraxellaceae* (2.15%) > *Comamonadaceae* (1.72%) > *Alcaligenaceae* (0.86%) > *Morganellaceae*, *Burkholderiaceae*, *Pasteurellaceae* and *Streptococcaceae* (0.43% each). Among the genera, 12 were commonly isolated from both sites. Bacterial strains from 7 and 10 genera were detected only in site#1 and site#2, respectively. Analysis of the partial 16s rRNA gene sequence of four filamentous gram-positive isolates showed their high sequence similarity to three genera, including three novel species, *Streptomyces* sp. NA03103, *Micromonospora fluminis* sp. nov. and *Bacillus velezensis* sp. nov. In conclusion, the Klong Tub Mangrove Forest possesses high microbial diversity, and the bacterial taxon in the sediments differ between the narrowleaf cattail wetland and mangrove forest. Several bacterial isolates from the forest show a high biotechnological potential.

## 1. INTRODUCTION

Mangrove forests harbour a unique environment with microbial diversity because they are located in the transition zone of terrestrial and river/marine ecosystems. Mangroves with high microbial diversity are a potential natural resource for various biotechnological fields, such as industry, agriculture, and medicine. In the past decade, mangrove ecosystems have become a hotspot in the study of natural products (Azman et al., 2015; Katili and Retnowati, 2017). Several novel species, particularly actinomycetes, have been found in marine ecosystems; this breakthrough led to the discovery of newly bioactive compounds, some of which are promising drug candidates (Pathom-

aree et al., 2006; Hong et al., 2009; Gong et al., 2018; Shi et al., 2019; Xavier et al., 2021). Other mangrove-associated bacteria with high potential biotechnological interests were also documented, including an extracellular hydrolytic enzyme producer *Vibrio alginolyticus* Jme3-20, a diverse metabolic intermediate producer *Bacillus velezensis* sp. nov. and a new lipopeptide biosurfactant and mosquitocidal agent producer *Aneurinibacillus aneurinilyticus* (Das et al., 2016; Balan et al., 2017; Abouelkheir et al., 2020; Mamangkey et al., 2021). Exploring the bacterial strains in mangroves is crucial for biotechnology applications and deep understanding of the ecosystem, such as the interaction between microorganisms and

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their environment and the dynamics of microbial population resilience in response to seasonality (Fuhrman et al., 2015). The abiotic factors of each site vary and can influence its microbial taxonomy and structure (Stottmeister et al., 2003; Oguh et al., 2021). Hence, the various aspects of a mangrove forest, including status, microbial diversity and ecosystem interaction webs, are crucial in establishing policies for its protection, rehabilitation and sustainable use (Allard et al., 2020).

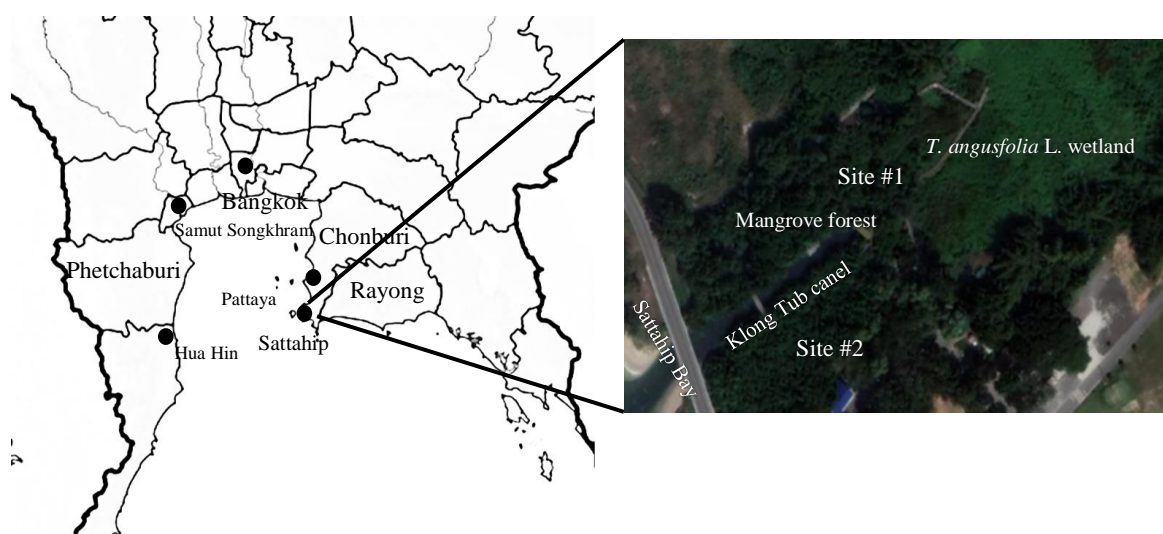
The Klong Tub Mangrove Forest is well preserved under the care of the Air and Coastal Defense Command, Royal Thai Navy. It covers a small area of 0.0112 km<sup>2</sup> but contains a high diversity of plant species from at least 15 genera, including *Rhizophora*, *Acrostichum*, *Lumnitzera*, *Ceriops*, *Clerodendrum*, *Sonneratia*, *Bruguiera*, *Avicennia*, *Hibiscus*, *Xylocarpus*, *Nypa*, *Flagellaria*, *Barringtonia*, *Wollastonia*, and *Derris* (Mangrove Forest at Air and Coastal Defense Command, 2014). This forest has distinct characteristics because it is located near a narrowleaf cattail (*Typha angustifolia* L.) wetland whose rhizopheric bacterial colonies exhibit high taxon richness (Gao and Shi, 2018). The Klong Tub Mangrove is also an undisturbed and unpolluted forest because it is isolated from the community, tourists and any other activities. Several publications have reported anthropogenic effects on the biodiversity of mangrove ecosystems, including the alternation and abundance of their bacterial communities (Ghizelini et al., 2012; Mishra et al., 2012; Oguh et al., 2021; Palit et al., 2022). Given that it is undisturbed by any human activity, contains a variety of plant species and is located close to a narrowleaf cattail wetland, the Klong

Tub Forest may possess high microbial diversity and contain bacterial strains with high biotechnological potential. This study aimed to investigate the bacterial diversity of Klong Tub Mangrove Forest and to compare the structure of bacterial community in the sediments between near the narrowleaf cattail wetland and close to the Sattahip Bay. The findings will be used in the evaluation of the forest status and the establishment of management plan for sustainable uses in biotechnological applications.

## 2. METHODOLOGY

### 2.1 Sample collection

Klong Tub Mangrove Forest, Sattahip District, Chonburi Province, Thailand is located on the coast of Sattahip Bay, the Upper Gulf of Thailand (Figure 1) and about 30 km from Pattaya City, a famous touring place in Eastern Thailand. This forest has a distinct environment because it is located in the interface of a marine ecosystem and a narrowleaf cattail (*Typha angustifolia* L.) wetland. Sampling was carried out in two sites (Figure 1): site#1 samples were collected nearby *T. angustifolia* wetland, and site#2 samples were from the mangrove area close to the Sattahip Bay. Soil samples were collected at the depth of 5 cm from the surface, placed in sterile bags, kept on ice during transportation, and maintained at 4°C until microbiological examination. Three sediment samples were collected about 2 m apart for each site, and their temperature was measured at the time of sampling using a glass thermometer. Salinity and pH were measured at the laboratory using a portable salinity/resistivity metre (SUNTEX, model SC-110, Taiwan).



**Figure 1.** Map of Klong Tub Mangrove Forest, Sattahip District, Chonburi Province, Thailand and sample sites in this study (modified from Google map and own work based on Nordwest). Site#1: 12°38'48.7"N 100°55'53.7"E and site#2: 12°38'46.4"N 100°55'50.9"E

## 2.2 Bacterial isolation

Bacterial strains were isolated within 12 h after sample collection using the modified protocol of [Jalal et al. \(2010\)](#). Briefly, 1 g of each soil sample was suspended in 9 mL of sterile distilled water. The mixture was shaken to thoroughly combine the suspension before centrifugation to collect the supernatant. The supernatant of each sample was diluted by 10-fold serial dilution method to obtain  $10^{-1}$ - $10^{-5}$  dilutions. For the total plate count of heterotrophic bacteria, 100  $\mu$ L of each dilution was spread on nutrient agar supplemented with 1% NaCl (NA+1% NaCl). The plates were incubated at 37°C for 24-48 h and used for bacterial identification.

Actinomycetes were isolated and characterised using ISP medium#2 (ISP-2) containing 10 g/L glucose, 5 g/L meat extract, 4 g/L malt extract, and 18 g/L agar-agar with pH 7.0 and incubated at 30°C ([Taechowisan et al., 2017](#)). Actinomycete colonies, evidenced by a filamentous growth pattern, were then subjected to amplified partial 16S rRNA gene sequencing for identification.

## 2.3 Biochemical test for the identification of bacterial isolates

For bacterial strain identification, colonies were randomly selected according to their morphological differences and streaked on NA+1% NaCl. The pure culture was observed for the presence of Gram's reaction. The strains were subsequently identified using the following morphological and standard biochemical features/tests: carbohydrate fermentation, triple-sugar iron agar, indole production, methyl red and Voges-Proskauer tests, citrate utilisation, motility, urease, oxidase, catalase and decarboxylation. Primary bacterial identification was performed using the ABIS online tool available at <https://www.tgw1916.net/> ([Sorescu and Stoica, 2021](#)). According to the guidelines of the ABIS online tool, the bacteria were identified according to the isolates' metabolic characteristics, cultural and phenotypical characteristics, ecology data and similarity percentage ( $\geq 87.00$ ).

## 2.4 Identification of bacteria by 16S rRNA PCR

Genomic DNA was extracted using the modified protocol of [Taechowisan et al. \(2017\)](#). Briefly, the isolates were cultured in ISP-2 broth by shaking at 100 rpm and 28°C for 7-14 days. After centrifugation at 6,000 rpm for 15 min, the mycelial pellet was ground to powder in liquid nitrogen using

mortar and pestle. The powder was mixed with 400  $\mu$ L of lysozyme (20 mg/mL) and then incubated at 25°C for 20 min. Subsequently, the mixture was added with 20  $\mu$ L of 25% (w/v) SDS and 10  $\mu$ L of RNase (100 mg/mL) and incubated at 37°C for 30 min, followed by protein digestion with 50  $\mu$ L of proteinase K (20 mg/mL) at 37°C for 2 h. Finally, the mixture was subjected to phenol:chloroform (1:1) extraction and subsequent DNA precipitation with 2 volume of absolute ethanol and 0.1 M NaCl. The DNA pellet was rinsed with 70% ethanol and resuspend in TE buffer. DNA concentration was measured using Nano drop (Thermo Fisher Scientific, USA), followed by PCR.

The 16S rRNA gene was amplified using primers A7-26f (5'-CCGTCGACGAGCTCAGATTTGATCCTGGCTCAG-3') and B1523-1504r (5'-CCCGGGTACCAAGCTTAAGGAGGTGATC-CAGCCGCA-3'), and the thermal cycle for PCR was described by [Taechowisan et al. \(2017\)](#). The PCR condition was started with denaturation at 95°C for 5 min, followed by 35 cycles of denaturation at 95°C for 1 min, annealing at 56°C for 1 min, extension at 73°C for 1 min, and a final extension at 72°C for 10 min. The PCR products were then subjected to sequence analysis (Illumina Miseq, Celemics, South Korea). The resulting sequences were submitted to the National Center for Biotechnology Information database using Basic Local Alignment Search Tool (BLAST) algorithm ([Zhang et al., 2000](#); [Morgulis et al., 2008](#)), Ribosomal Database Project (RDP) ([Wang et al., 2007](#)) and EzBioCloud ([Yoon et al., 2017](#)).

## 3. RESULTS AND DISCUSSION

### 3.1 Abiotic parameters of the sediments from Klong Tub Mangrove Forest

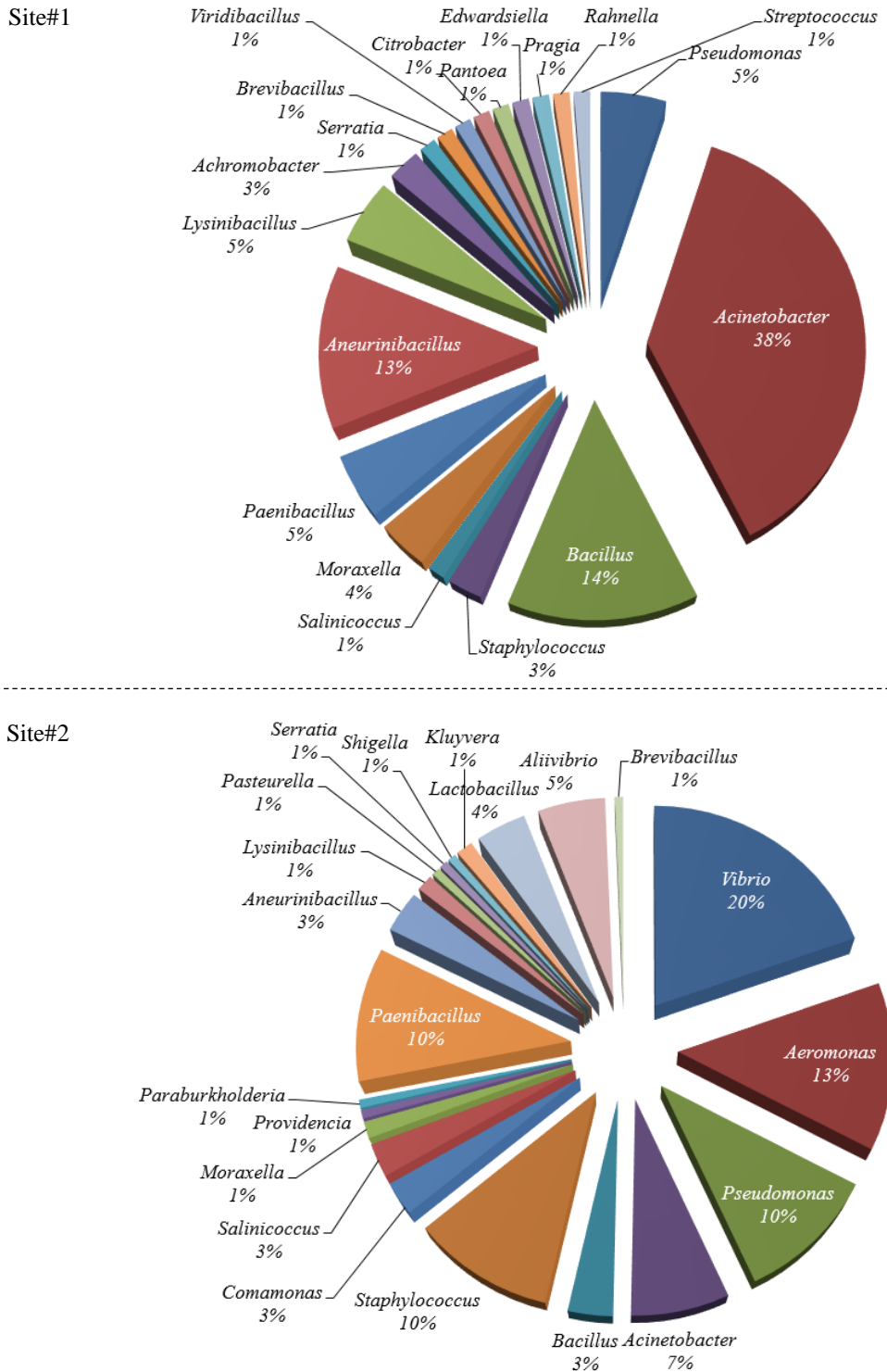
Three abiotic parameters influencing micro-organism growth were measured in the sediment samples from the Klong Tub Forest. The pH of site#1 sediments was higher than that of site#2 sediments with ranges of 7.55 to 8.12 and 7.11 to 7.53, respectively. Meanwhile, the salinity of site#1 sediments ranged from 0.54 to 1.20 ppt and was lower than that of site#2 sediments at 18.40 to 23.42 ppt. The temperature of site#1 sediments was between 28°C and 30°C, and that of site#2 sediments was between 26°C and 28°C.

### 3.2 Determination of microbial diversity in the sediments from Klong Tub Mangrove Forest

According to the experiment using spread plate technique on NA+1%NaCl, the total plate count of

heterotrophic bacteria in site#1 and site#2 sediments ranged from 2.39 log CFU/mL to 4.39 log CFU/mL and from 2.47 log CFU/mL to 3.07 log CFU/mL, respectively. The bacterial colonies on NA+1% NaCl were categorised on the basis of morphology, such as shape, size and colour, and then randomly selected for identification. A total of 233 bacterial isolates were

subsequently identified using biochemical tests and were found to belong to 16 families and 29 genera (Figure 2). The first-, second-, and third-most identified isolates were in families *Moraxellaceae* (41 isolates; 17.60%), *Vibrionaceae* (38 isolates; 16.31%), and *Paenibacillaceae* (37 isolates; 15.88%), respectively.

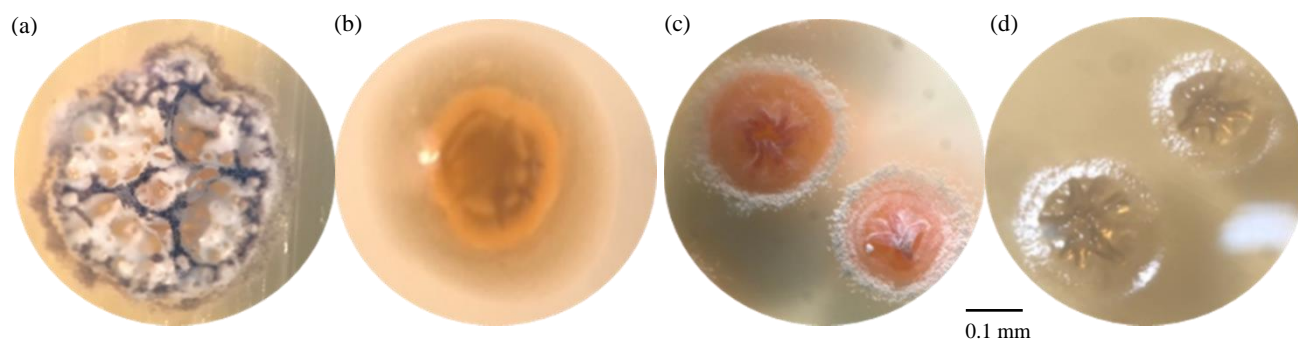


**Figure 2.** Taxonomy of bacterial strains isolated from Klong Tub Mangrove Forest at sites#1 (nearby the narrowleaf cattail wetland) and site#2 (in the mangrove)

Comparison of bacterial isolates in site#1 and site#2 sediments revealed differences in bacterial structure and composition (Figure 2). Identification of isolates from site#1 sediments (n=80) revealed that the bacteria belonged to nine families and 19 genera. The first-, second-, and third-most frequently identified isolates were in genera *Acinetobacter* (38%), *Bacillus* (14%), and *Aneurinibacillus* (13%), respectively. One isolate each from six genera including *Citrobacter*, *Panoea*, *Edwardsiella*, *Pragia*, *Rahnella*, and *Streptococcus* and two isolates of *Achromobacter* spp. were identified in site#1 sediments but not in site#2 samples. For site#2 sediments, 153 of the isolates belonged to 14 families and 21 genera. The first- and second-most frequently identified isolates were in genera *Vibrio* (20%) and *Aeromonas* (13%), and the third-most frequently identified isolates were bacterial strains from three genera *Pseudomonas*, *Staphylococcus*, and *Paenibacillus* (10% each). The following bacterial strains from nine genera were detected only in site#2 sediments: *Vibrio* spp. (30 isolates), *Aliivibrio* spp. (8 isolates), *Lactobacillus* spp. (6 isolates), *Commamonas* spp. (4 isolates), *Kluyvera* spp. (2 isolates), *Shigella* sp. (1 isolate), *Providencia*

sp. (1 isolate), *Paraburkholderia* sp. (1 isolate), and *Pasteurella* sp. (1 isolate).

Figure 3 shows the colony morphology of four bacterial isolates (A.1, B.1, C.1, and D.1) identified using 16s rRNA sequence analysis. Isolate A.1 showed a mucous, transparent spread out, edged shape with chalk-like colour. For isolate B.1, the morphology was power-like colony with pink colour in the centre. Isolate C.1 was a small, orange-brown-coloured colony covered with thick slime. Finally, isolate D.1 appeared as a powder-like colony with whitish to greyish aerial mycelium. Results from the alignment of partial 16s rRNA sequences using three different tools, BLAST, EzBioCloud, and RDP classifier, are shown in Table 1. The alignment of all sequences gave high similarity values ( $\geq 99.65\%$ ). Isolates A.1 and C.1 were closely related to *Streptomyces platensis* strain ATCC 23948 and *Streptomyces* sp. NA03103, respectively. The other two isolates, B.1 and D.1, shared sequence similarity to *Micromonospora* sp. A38 (100.00%) and *Bacillus velezensis* strain KKLW ( $\geq 99.86\%$ ), respectively. Accordingly, isolates A.1 and C.1 were identified as *Streptomyces* spp. and isolates B.1 and D.1 were *Micromonospora* sp. and *Bacillus* sp., respectively.



**Figure 3.** Colonial appearance of bacterial isolates on ISP-2 agar (a) isolate A.1, (b) isolate B.1, (c) isolate C.1, and (d) isolate D.1

**Table 1.** Closely related species shared similarity to the partial 16s rRNA sequence of isolates A.1, B.1, C.1, and D.1 as analysed by three different tools

Isolate	Best blast match with known taxa by BLAST (% identity)	Top-hit taxon by EzBioCloud (% similarity)	Genus predicted by RDP classifier at 95% confidence threshold (% probability)
A.1	<i>Streptomyces platensis</i> strain ATCC 23948 (100.00%)	<i>Streptomyces platensis</i> JCM 4662 (99.93%)	<i>Streptomyces</i> (100%)
B.1	<i>Micromonospora</i> sp. A38 (100.00%)	<i>Micromonospora fluminis</i> A38 (100.00%)	<i>Micromonospora</i> (100%)
C.1	<i>Streptomyces</i> sp. NA03103 (100.00%)	<i>Streptomyces ardesiacus</i> (99.65%)	<i>Streptomyces</i> (100%)
D.1	<i>Bacillus velezensis</i> strain KKLW (100.00%)	<i>Bacillus velezensis</i> CR-502 (99.86%)	<i>Bacillus</i> (100%)

#### 4. DISCUSSION

This study investigated the microbial diversity of Klong Tub Mangrove Forest. Observation of heterotrophic bacteria in aerobic condition was conducted in 5 cm-deep sediments because the oxygen level in the sediments at this level is higher than that of the sediments in low depths, leading to high microbial diversity (Dias et al., 2009; Somerfield et al., 1998). A total of 233 bacterial isolates were isolated and identified using biochemical approaches and ABIS online tools (Bhagobaty, 2020; Kayode et al., 2020; Dumitru et al., 2021; Ndukwu et al., 2021; Vansia et al., 2021). The results showed that the bacteria belonged to 29 genera and 16 families. The dominant strains in the microbial community were in descending ranked order as follows: *Moraxellaceae* (17.60%), *Vibrionaceae* (16.31%), *Paenibacillaceae* (15.88%), *Staphylococcaceae* (9.87%), *Bacillaceae* (9.87%), *Aeromonadaceae* (8.58%), and *Pseudomonadaceae* (8.58%). Moreover, phosphate-solubilizing bacteria that were commonly found in mangroves, namely, *Bacillus*, *Paenibacillus*, *Kluyvera*, *Pseudomonas*, *Burkholderia*, *Serratia*, and *Acinetobacter* species, were predominant in this forest (Thatoi et al., 2013; Teymouri et al., 2016). The Klong Tub Mangrove Forest is located in the transition zone between a narrowleaf cattail wetland and Sattahip Bay (Figure 1). Assessment of abiotic parameters revealed differences in abiotic values between the two site samples. Compared with site#2 samples, site#1 samples had higher pH and lower salinity. Differences in taxonomic structures were also detected between the bacterial communities. The first-, second-, and third-most frequently identified isolates from site#1 samples were from genera *Acinetobacter* (38%), *Bacillus* (14%), and *Aneurinibacillus* (13%), respectively. *Acinetobacter* spp. and *Bacillus* spp. have been reported as two of the top 26 genera associated with narrowleaf cattail rhizospheric samples (Gao and Shi, 2018). For site#2 samples, the predominant bacterial strains were *Vibrio* (20%), *Aeromonas* (13%), *Pseudomonas* (10%), *Staphylococcus* (10%), and *Paenibacillus* (10%). These bacterial groups are commonly found in marine and estuarine environments (Thompson et al., 2004; Sousa et al., 2006; Stevens et al., 2007). This study provided evidence on the influence of environmental conditions, including abiotic parameters and types of plants, on determining the compositions and diversity patterns of bacterial communities (Stottmeister et al., 2003; Oguh et al., 2021).

The microbial community in site#1 sediments (19 genera, n=80) showed higher diversity than those in site#2 sediments (21 genera, n=153). Gao and Shi (2018) investigated the taxonomic structure of seed, root and rhizospheric bacterial microbiota of narrowleaf cattail *T. angustifolia* using Illumina-based sequencing. They found that the rhizospheric bacterial community exhibited higher taxon richness compared with the seed and root endophytes. Similarly, a study of ammonium/ammonia-oxidizing prokaryote (AOP) communities in Hong Kong revealed more complex AOP community structures in man-made freshwater wetland than in natural coastal marine wetland (Wang and Gu, 2013). The authors concluded that the high diversity in freshwater AOP community was affected by narrowleaf cattail *T. angustifolia* which enhanced AOP abundances in the rhizosphere area. This finding suggested that presence of narrowleaf cattail nearby the Klong Tub Mangrove Forest may influence the high microbial diversity in site#1 sediments.

Four filamentous Gram-positive isolates were isolated from the sediment in the studied mangrove forest. Partial 16s rRNA sequence analysis using three different similarity-based search tools, BLAST, EzBioCloud, and RPD Classifier, revealed three presumptive genera with a similarity value of  $\geq 99.65\%$ . Two isolates, A.1 and C.1, shared high 16s rRNA gene sequence similarity with *Streptomyces platensis* and *Streptomyces* sp. NA03103, respectively. *Streptomyces platensis* produces a variety of antibiotics, including oxytetracycline, platensimycin, migrastatin, isomigrastatin, platencin, dorrigocin A and B and terramycine (Ju et al., 2005; Smanski et al., 2012). Genome mining of *Streptomyces* sp. NA03103 revealed the presence of an orphan nonribosomal peptide synthetase (NRPS) gene cluster (*asm*) encoding for two novel cyclopeptides ashimides A and B (Shi et al., 2019). Ashimide B has been classified as an organic compound, namely, benzoxazine. Several studies have reported the biological activities of benzoxazine derivatives in various aspects, including anti-inflammatory, antimicrobial, anticancer and anti-malaria (Akhter et al., 2011; Foto et al., 2014; Sharma et al., 2018). This finding is consistent with the study of Xavier et al. (2021) who analysed the genomic DNA of *Streptomyces* sp. NA03103 using anti-SMASH v5.2.0 and found a number of predictive BCGs which may implicate diverse secondary metabolic biosynthesis, such as polyketides, nonribosomal peptides, terpenes, and lanthipeptides

(Xavier et al., 2021). Further experiments are required to investigate the function of this potential bacterial strain on the synthesis of marine natural products.

In addition to *Streptomyces*, one rare actinomycete strain was isolated from the sediments in Klong Tub mangrove forest. Analysis of partial 16S rRNA sequence showed 100.00% similarity to *Micromonospora* sp. A38 (Table 1). Strain A38 has been classified as a novel species named *Micromonospora fluminis* sp. nov. (Poza et al., 2020). *M. fluminis* strain was isolated from freshwater, the Carpintero River in Cuba. Meanwhile, the isolated strain in the present study was from a marine environment, implying its ability to adopt to a wide range of environmental conditions. To our knowledge, the secondary metabolites of this novel strain has never been reported.

A number of bacterial strains isolated from the Klong Tub Mangrove Forest have been documented for their biotechnological potentials. One of which was *Bacillus*, one of the abundant genera detected in the rhizosphere communities of mangroves (Sebastianes et al., 2017; Jeyanny et al., 2020). In the present study, a rhizosphere bacterium named *Bacillus velezensis*, a novel species of *Bacillus*, was isolated. *B. velezensis* produces diverse metabolic intermediates with wide applications, such as in surfactant production, antagonistic activities against phytopathogens, promotion of seed development and plant growth, antibiotics, iron chelators, antioxidants, and anticancer agents (Ruiz-García et al., 2005; Meena et al., 2018; Aloo et al., 2019; Rabbee et al., 2019). *B. velezensis* SMR isolated from seawater can produce nanocellulose, a promising material for various industries, such as medicine, food and agriculture (Abouelkheir et al., 2020). In Thailand, a number of *Bacillus* spp., such as *Bacillus aquimaris* strain TF-12 and *B. aryabhatai* strain B8W22, have been isolated from mangrove forest sediments (Foophow and Tangjitjaroenkun, 2014). These isolates can produce proteases, enzymes that are widely used in various industries (Razzaq et al., 2019). In addition to *Bacillus* spp., *Aeromonas* spp. were documented for their biotechnological potentials, particularly extracellular proteases that are involved in the degradation of different proteinaceous compounds, such as albumin, fibrin and gelatine (Liu, 2014). For the genus *Acinetobacter*, the predominant bacterial strains isolated from site#1 sediments have the abilities of hydrocarbon degradation, ester production and biosurfactant production (Jung and Park, 2015). The

other genus was *Comamonas* spp. which can be found in soil and water. The recent isolation and investigation of *Comamonas aquatica* from mangrove swamps in Rayong Province, Thailand revealed its ability to degrade polycyclic aromatic hydrocarbons (PAHs) (Chantarasiri, 2021). *C. aquatica* can also promote plant growth via the production of indole-3-acetic acid as their metabolites (Al-Mamari et al., 2020). Moreover, the *C. aquatica* strain isolated from the root of freshwater plant *Pistia stratiotes* exhibited the efficient removal of heavy metal, Ni and Cu in the environment, implying the potential of this bacterial genera in phytoremediation technology (Ghosh et al., 2020). *Aneurinibacillus aneurinilyticus* is another bacterial strain displaying various biotechnological interests. It was recognised as a plant growth-promoting rhizobacteria and also exhibited antifungal activity (Chauhan et al., 2014). Its marine strain has been recently evaluated for a new lipopeptide biosurfactant (Balan et al., 2017). Interestingly, the secondary metabolite of this bacterium has mosquitocidal potential to control mosquito-borne diseases (Das et al., 2016).

The Klong Tub Mangrove Forest is located near Pattaya City, Chonburi Province, Thailand, a famous coastal city on the Upper Gulf of Thailand. Bacterial identification revealed that none of the isolates were enterococci and *Escherichia coli*, bacterial indicators of faecal contamination (U.S. EPA, 2012). The absence of faecal contamination was confirmed by spread plate technique on eosin methylene blue agar (Abbu and Lyimo, 2009). On the basis of these results were, the Klong Tub Mangrove Forest in Sattahip, Chonburi Province, Thailand possesses high microbial diversity with minimal anthropogenic effects, making it a valuable natural resource in terms of industrial enzymes and secondary metabolite producers. Particularly, the three Actinomycete isolates require further investigation to determine their potential in biotechnological applications.

## 5. CONCLUSION

This study investigated the microbial community in sediments from Klong Tub Mangrove Forest. The results indicated that the forest possesses a number of bacterial strains. Comparison of the structure of bacterial microbiota between site#1 (nearby the narrowleaf cattail wetland) and site#2 (the mangrove forest close to the sea) revealed differences in their predominant bacterial species and compositions. This finding implied the significance of

environmental conditions on determining bacterial community compositions and diversity patterns. Moreover, two actinomycete strains (*Streptomyces* sp. and *Streptomyces* sp. NA03103) and a rare actinomycete *Micromonospora* sp. were isolated. The production of bioactive compounds from these isolates are currently under investigation. Several bacterial isolates from the mangrove forest have been documented for their biotechnological potentials, including plant growth promotion, antibiotics, antifungal and PAH degradation. According to our observation, the Klong Tub Mangrove Forest possesses high microbial diversity with minimal anthropogenic effects and high potentials for biotechnological applications.

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