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Research Article
Diatom Colonization on Cowhide Substrate in Batang Kuranji River, Padang City, for Forensic Drowning Diagnosis

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Abstract

Background and Objective: Diatoms are unicellular algae that can survive extreme conditions because of their silica-based cell walls. As an indication of the murder scene and the duration of the victim's drowning, diatoms might be utilized for forensic diagnosis. The purpose of this study is to determine the diatom species that colonize the cowhide substrate, the community and the relationship between physico-chemical factors in the Batang Kuranji River in Padang City. **Materials and Methods:** Drowning diagnosis was investigated by submerging cowhide substrates at sampled river sites to collect colonizing diatoms over 10 days. Diatom samples were collected, preserved and identified microscopically, while key water quality parameters were measured concurrently. Statistical analysis, including ANOVA and PCA, was used to correlate diatom colonization with location, submersion time and environmental factors. **Results:** It showed that diatom colonization based on the number of species on the 8th day showed colonization influenced by an increase in the number of individuals, such as the species *Fragilaria capucina* and *Synedra ulna*. In the observation, 94 species of diatoms were found, 42 species were obtained as a pointer to the crime scene of the drowning victim and 68 species as a pointer to the time of the victim's drowning. Diatoms on cowhide substrate consisted of 2 classes, 2 orders and 19 families with diatom density ranging from 3.81±0.16 to 22.80±1.34 ind/cm^2. The highest relative density was *Fragilaria capucina* (28.59%). **Conclusion:** The diatom community that was identified had no dominant species, an equally distributed individual population and diversity index requirements that were categorized as moderate to high. The Batang Kuranji River's chemical and physical factors are still largely ideal for diatom colonization on cowhide substrates, according to the PCA analysis's results, which can also serve as a guide for drowning victims.

Key words: Colonization, diatoms, forensic, cowhide substrate, drowning

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Diatoms are part of the Bacillariophyceae class of unicellular algae. Diatoms can be found in a variety of freshwater and marine aquatic ecosystems[1]. It has been estimated that there are over 100,000 species of diatoms, making them the most varied category of microalgae[2]. A distinctive silica-based cell wall known as a frustule is present in diatoms[3,4]. The majority of aquatic and humid settings are home to diatoms, a group of algae that exhibit remarkable diversity in terms of taxonomy, morphology and ecology[5]. Diatoms are primary producers in water and have a significant impact on the worldwide ecology. Furthermore, diatoms can be used as markers of environmental health, particularly water quality[6]. Diatoms are generally the earliest and most abundant primary colonizers on natural and artificial substrate surfaces. The presence of bacteria and unicellular algae in biofilms can promote further substrate colonization[7]. The condition of the aquatic environment can affect the abundance, species distribution of the diatom community. In addition, nutrient availability is important for diatoms in enhancing reproduction and colonization development[8].

Colonization is a process where a pioneer species comes and occupies a new habitat by other organisms, then adapts to the environment and can survive and reproduce so that it forms a new colony. Colonization is the process of gathering similar biota that group and form a colony. Colonization is one of the important processes in the succession of biota in a new habitat[9]. Studies on diatom colonization and development have been conducted by collecting diatoms on substrates over time[10,11].

Diatom colonization on substrates can be used in supporting forensic diagnosis, namely as a pointer to the scene of the crime and a pointer to the length of time the victim drowned. The substrate used in this study is a cowhide substrate that was submerged in the Batang Kuranji River. The Batang Kuranji River is one of the major rivers in Padang City. This river has its headwaters in the Bukit Barisan Mountains area, which is included in the Kerinci Seblat National Park. Previous research on diatoms in supporting forensic diagnostics has been conducted in West Sumatra, including[12-14]. However, there is no research information regarding diatom colonization as a forensic diagnosis of drowning victims, especially using cowhide substrates in the Batang Kuranji River. The results of this study are expected to support the process of diatom colonization on cowhide substrate drowned in Batang Kuranji River, Padang City, in assisting forensic diagnosis of drowning victims.

The aims of this study were (i) Analyze the species of diatoms colonizing the cowhide substrate as a forensic diagnosis of the crime scene and the time of the drowning victim, (ii) Analyze the composition and structure of the diatom community and (iii) Analyze the relationship between physical and chemical factors in the Batang Kuranji River, Padang City.

MATERIALS AND METHODS

Study area: This research was conducted from February to August, 2024. Submergence of cowhide substrate and measurement of water physico-chemical factors were conducted in Batang Kuranji River, Padang City, West Sumatra. The research was conducted at five sampling locations. Diatom sample identification and data analysis were conducted at the Animal Ecology Research Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Andalas, Padang. Geographically, Batang Kuranji River is located at 100020'31.20"-100033'50.40" East Longitude and 0055'59.88"- 0047.24" South Latitude with an area of 202,000,000 m^2. South Latitude, which has an area of 202.7 km^2. Batang Kuranji River is dendritic, where the upstream part of the river is fed by two tributaries, namely the Padang Keruh and Padang Jernih Rivers. These two rivers unite at Lubuk Siarang (Patamuan) and flow to form the Sekayan River. The Sekayan River flows and joins the Limau Manis River in the Gunung Nago Area, called Batang Kuranji (Fig. 1).

Drowning of cowhide substrate: The survey method was used to conduct this research. Purposive sampling was used to identify the location based on differences in land use and ambient hue, which affect diatom life. For 10 days, each site was covered in cowhide substrate. Diatom samples were taken on the 2nd, 4th, 6th, 8th and 10th days after drowning on a cement plate covered in cowhide for this research.

Collection and identification of diatom samples: Collection of diatom samples was carried out by brushing the cowhide substrate in a bucket containing 10 L of water, after which the water was filtered using a plankton net. Water from the plankton net filter was then put into a 20 mL sample bottle and 4% formalin was added up to 1 mL and 2 drops of Lugol. The sample bottle was then labeled with the location and time of collection.

Identification of diatom samples was carried out as much as 1 mL drop by drop, then placed on a glass object and covered using a cover glass and then observed using a microscope with a magnification of 10×40 and then

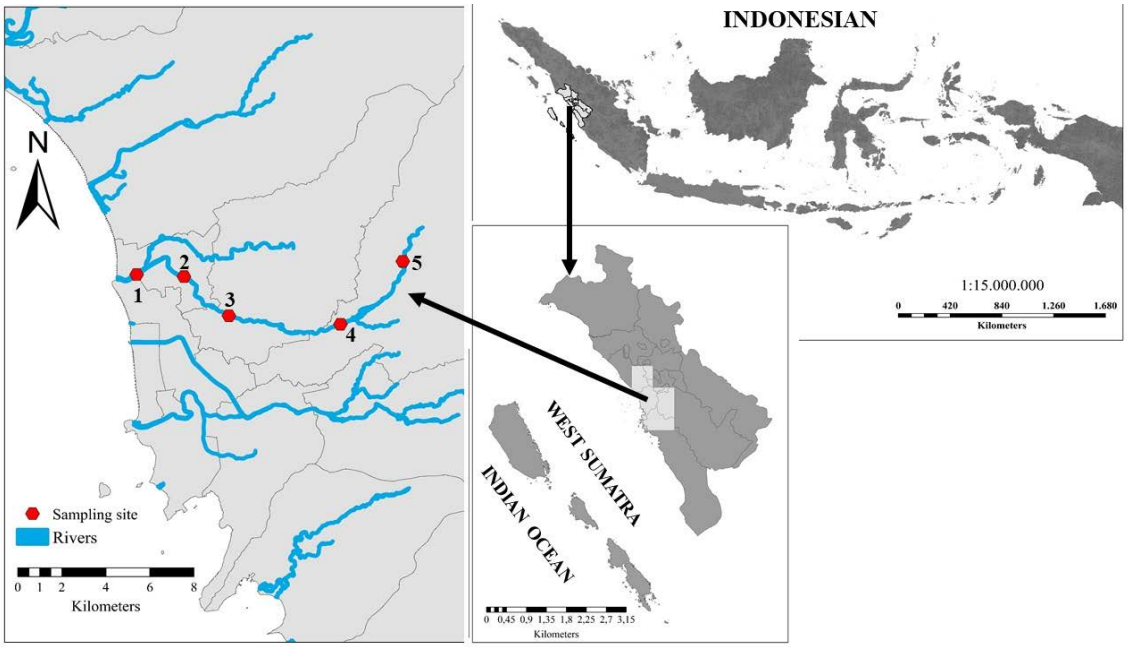

Fig. 1: Geographical map of sampling locations in Batang Kuranji River, Padang, West Sumatra, Indonesia
(1) Kuranji Batang Estuary, (2) Surau Gadang Waters, (3) Kalumbuk Waters, (4) Gunung Nago Waters and (5) Batu Busuk Waters

photographed. Observations were made of the general shape of diatom cells and diatom species using identification books, including[15-17], which refers to www.algabase.org and www.diatom.org.

Assessment of water quality: Aquatic parameters were measured *in situ* and *ex situ* at each sinking location. Physical and chemical parameters of water are very important in supporting the growth of diatom colonization[18]. Water parameters include physics consisting of current speed, depth and temperature. Chemical parameters included pH, dissolved oxygen (DO), free Carbon Dioxide (CO_2), Biological Oxygen Demand (BOD_5), total suspended solid (TSS), nitrate, nitrite, ammonia, silica, phosphate and salinity, which were measured at the time of cowhide drowning and when collecting diatom samples on cowhide.

Statistical analysis: Diatom species colonizing the cowhide substrate were presented in graphical form based on the location and time of sampling. To obtain diatom species as a pointer to the scene of the crime and the length of time the victim drowned, it was determined based on species that were only found at certain locations and times. The composition of diatom species found in cowhide substrates was grouped by class order and family. The data were tested using parametric statistics. To determine the effect of cowhide substrate on the number of species and density of diatoms in each research location, a normality test was first conducted, after which an One-way Analysis of Variance (ANOVA) test was conducted. If significant differences were found ($p<0.05$) at the 95% confidence level, then it was continued with Duncan's New Multiple Range Test (DNMRT) to determine differences between research locations[19]. Furthermore, to analyze the relationship of physical and chemical factors in the waters of Batang Kuranji River, it was tested using the Principal Component Analysis (PCA) method with the help of the XLSTAT program.

RESULTS

Physical and chemical factors of water: During the field time at each location, the study recorded the water's chemical and physical qualities. Current velocity, depth, water temperature, air temperature, pH, DO, free CO_2, BOD_5, TSS, nitrate, nitrite, ammonia, silica, phosphate and salinity were among the physico-chemical characteristics of the waters of Batang Kuranji River, Padang City, that were measured. The findings showed that the ideal circumstances for promoting the establishment and development of diatom colonization on cowhide substrate included the physical and chemical characteristics of the waters obtained from the five sampling locations.

All measurements in this study are presents in Table 1. Water temperature during the study at Batang Kuranji Rivers ranged from 22.49±0.28 to 30.50±0.96°C with pH ranged from 7.19±0.10 to 7.84±0.06 and salinity between 0.15±0.13

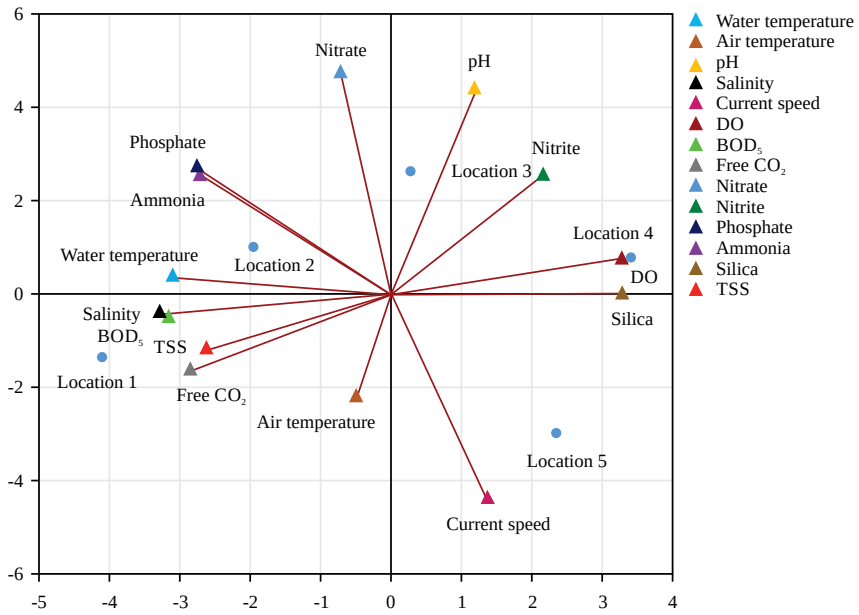

Fig. 2: Principal Component Analysis (PCA) ordination diagram for water physicochemical parameters in Batang Kuranji River, Padang City

Table 1: Environmental parameters (Mean±Standard Error) at Batang Kuranji River, Padang City

	Locations				
Parameters	1	2	3	4	5
Water temperature (°C)	29.32±0.34	28.78±0.55	28.15±0.32	22.49±0.28	25.78±0.78
Air temperature (°C)	29.67±1.68	30.50±0.96	28.83±0.72	30.00±1.05	28.67±0.93
pH	7.20±0.12	7.38±0.09	7.84±0.06	7.60±0.07	7.19±0.10
Salinity (%)	4.07±0.78	1.49±1.19	1.15±0.81	0.15±0.13	0.07±0.01
Current speed (m/sec)	0.41±0.13	0.24±0.05	0.25±0.09	0.39±0.14	0.72±0.09
DO (ppm)	3.25±1.09	4.42±0.36	5.46±0.35	5.65±0.28	5.69±0.75
BOD_5 (ppm)	2.14±0.54	2.07±0.57	0.96±0.27	0.76±0.25	0.91±0.72
Free CO_2 (ppm)	1.41±0.29	1.17±0.16	1.06±0.38	0.50±0.08	1.13±0.31
Nitrite (mg/L)	0.15±0.02	0.38±0.21	0.35±0.23	0.72±0.41	0.21±0.11
Nitrate (mg/L)	1.17±0.11	1.34±0.21	1.44±0.13	1.28±0.06	0.90±0.03
Phosphate (mg/L)	0.08±0.01	0.08±0.12	0.07±0.01	0.07±0.01	0.06±0.01
Ammonia (mg/L)	0.12±0.01	0.14±0.01	0.14±0.02	0.07±0.01	0.07±0.01
Silica (mg/L)	80.02±4.88	96.96±5.37	144.45±7.00	149.56±7.20	159.17±7.60
TSS (mg/L)	17.56±0.01	3.94±0.05	6,.9±0.27	4.00±0.01	4.11±0.01

DO: Dissolved oxygen, BOD_5: Biological Oxygen Demand, TSS: Total suspended solid, 1: Batang Kuranji Estuary, 2: Surau Gadang Waters, 3: Kalumbuk Waters, 4: Gunung Nago Waters and 5: Batu Busuk Waters

to 4.07±0.78%. The current speed values obtained ranged from 0.24±0.05 to 0.72±0.09 m/sec, DO value ranged from 3.25±1.09 to 5.69±0.75, BOD_5 between 0.76±0.25 to 2.14±0.54 ppm and free CO_2 content ranged from 0.50±0.08 to 1.41±0.29 mg/L. Nitrate ranged from 0.90 to 1.44 mg/L, Nitrite between 0.15±0.02 to 0.72±0.41 mg/L and TSS content obtained ranged from 3.94±0.05 to 17.56±0.01 mg/L.

The results of PCA analysis showed that the content of free CO_2, BOD_5, TSS, salinity and air temperature had a positive relationship and influence on diatom life on cowhide substrate (location 1). The content of ammonia, phosphate, nitrate and water temperature (location 2), pH, DO, nitrite and silica (locations 3 and 4) and current speed (location 5) have a relationship with diatom life and affect the diatom species found on cowhide substrates (Fig. 2).

Colonization of diatom species as a diagnosis of drowning victims: According to studies conducted in Padang City's Batang Kuranji River, 94 various types of diatoms have been identified to be colonizing the cowhide substrate. There were the most diatom species (61 species) at location 1 and the

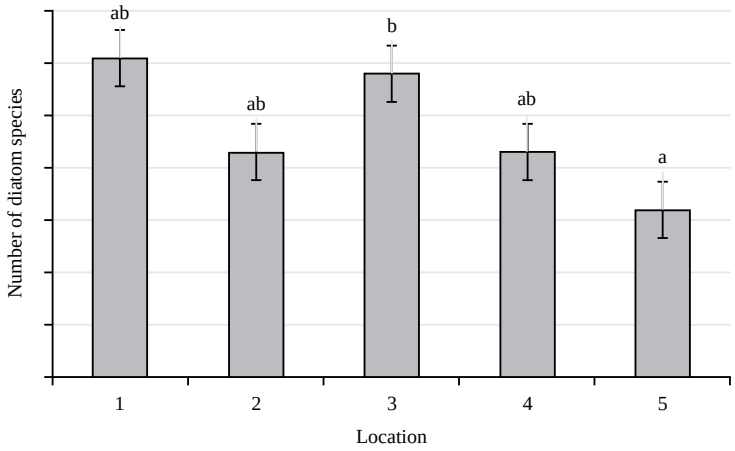

Fig. 3: Number of diatom species colonising the cowhide substrate at each sampling location

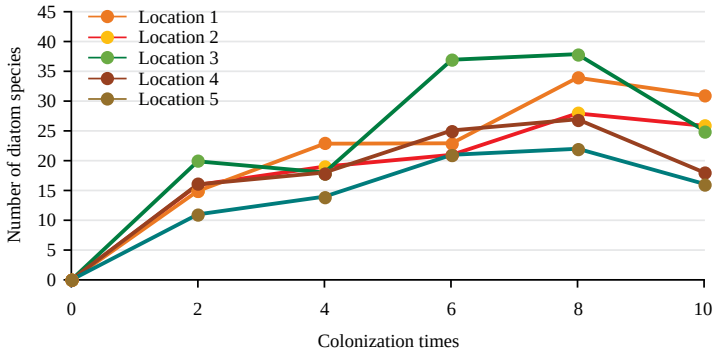

Fig. 4: Colonization dynamics of diatom species on cowhide substrate in Batang Kuranji River, Padang City

least number (32 species) at location 5 (Fig. 3). The results of the One-way Analysis of Variance (ANOVA) statistical test showed that there was no significant difference in the number of diatom species found between the study locations ($p>0.05$). However, after further testing by Duncan's New Multiple Range Test (DNMRT), it was found that the number of diatom species at location 3 was significantly different from location 5, but not different from locations 1, 2 and 4. Colonization of diatom species over time on the cowhide substrate was found to differ between locations (Fig. 4).

Colonization at locations 1, 2, 4 and 5 showed an increase in the number of species from the second day to the 8th day, then decreased on the 10th day. However, at location 3, the number of diatom species decreased on the 4th day; this was due to the ongoing adaptation process of diatoms to the cowhide substrate. Furthermore, it increased again until the 8th day, then decreased on the 10th day. Table 2 shows the typical diatom species found on cowhide substrate at each study location.

Diatom community composition and structure on cowhide substrate: The results showed that 94 species of diatoms were found on cowhide substrate in Batang Kuranji River, Padang City. The species composition of diatoms found included 2 classes, namely Bacillariophyceae and Coscinodiscophyceae as well as 2 orders (pennales and centrales) and 19 families. The highest percentage of diatom species is found in the Bacillariophyceae class, which is 98.94%. The density of diatoms found on cowhide substrate ranged from 3.81±0.16 to 22.80±1.34 ind/cm^2 (Fig. 5).

The results of the One-way Analysis of Variance (ANOVA) statistical test showed that there was a significant difference in the density of diatoms found at each sampling location ($p<0.05$). The results of Duncan's New Multiple Range Test (DNMRT) further test found that the density of diatoms at location 3 was significantly different from locations 1, 2 and 5, but not significantly different from location 4. The difference in diatom density found was due to the physical and chemical factors of the water that were found to be different at each location. However, the density of diatoms in locations 3 and 4

Table 2: Typical diatom species found on cowhide substrate at each study location

	Locations				
Species	1	2	3	4	5
Achnanthes coarctata	-	-	√	-	-
Achnanthes longipes	-	-	√	-	-
Amphipleura pellucida	√	-	-	-	-
Amphora ovalis	-	-	-	-	√
Amphora sp.	√	-	-	-	-
Cymbella aspera	-	-	√	-	-
Cymbella naviculiformis	-	√	-	-	-
Cymbella sinuata	√	-	-	-	-
Cymatopleura elliptica	-	-	√	-	-
Cymatopleura sp.	√	-	-	-	-
Donkinia recta	-	-	√	-	-
Eunotia bilunaris	√	-	-	-	-
Frustulia appendiculata	√	-	-	-	-
Frustulia vulgare	-	-	-	√	-
Gomphonema augur	-	-	√	-	-
Melosira arctica	√	-	-	-	-
Navicula cari	√	-	-	-	-
Navicula laterostrata	-	-	√	-	-
Navicula pygmaea	-	-	-	√	-
Nitzschia acicularis	-	√	-	-	-
Nitzschia acuminata	-	-	√	-	-
Nitzschia capitata	-	-	-	√	-
Nitzschia delicatissima	√	-	-	-	-
Nitzschia hungarica	√	-	-	-	-
Nitzschia longissima	-	√	-	-	-
Nitzschia obtuse	√	-	-	-	-
Pinnularia borealis	-	-	-	√	-
Pinnularia dactylus	-	-	√	-	-
Pinnularia interrupta	-	√	-	-	-
Pinnularia rectangulata	-	-	-	√	-
Pleurosigma acrosphenia	√	-	-	-	-
Pleurosigma affinis	√	-	-	-	-
Pleurosigma angulatum	√	-	-	-	-
Pleurosigma normanii	√	-	-	-	-
Pleurosigma rectum	-	-	√	-	-
Pleurosigma rigidum	√	-	-	-	-
Surirella capronii	-	-	√	-	-
Surirella linearis	-	-	√	-	-
Synedra tabulata	-	√	-	-	-
Thalassiothrix frauenfeldii	√	-	-	-	-
Tryblionella coarctata	-	-	√	-	-
Tryblionella punctata	-	√	-	-	-
Number of species	17	6	13	5	1

√: Found, -: Not found, 1: Batang Kuranji Estuary, 2: Surau Gadang Waters, 3: Kalumbuk Waters, 4: Gunung Nago Waters and 5: Batu Busuk Waters

that were not different was due to the almost the same concentration of pH, nitrite, dissolved oxygen and silica in both locations.

The relative density of diatom species found at each location varies. Figure 6 shows the diatom species that belong to the dominant group (relative density >5%). Species that have the highest relative density are *Navicula cryptocephala* at location 1, *Synedra ulna* at location 2, *Fragilaria capucina* at locations 3, 4 and 5 (Fig. 6). The diversity index obtained in this study ranged from 2.36±0.12 to 3.41±0.09, which is classified into the medium to high category. The equitability index of diatoms on cowhide substrate ranged from 0.58±0.02 to 0.83±0.02, with the highest value found in location 1 and the lowest in location 3. The dominance index of diatoms on cowhide substrate ranged from 0.06±0.01 to 0.15±0.02 (Fig. 7).

DISCUSSION

The results showed that the water and air temperature at five sampling locations had satisfied the water quality standards in supporting diatom life. According to

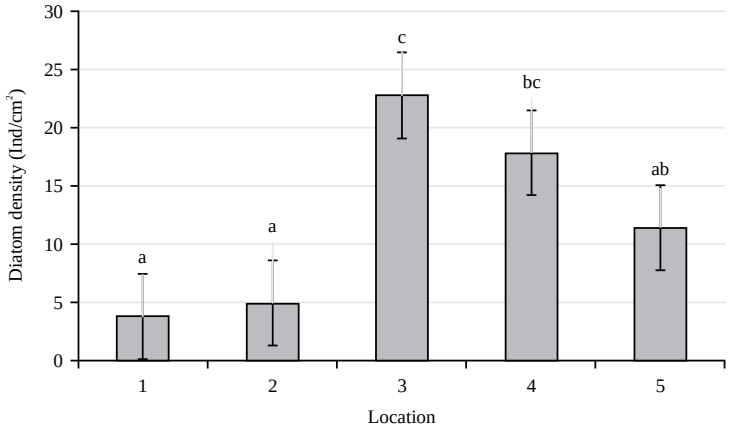

Fig. 5: Diatom density on cowhide substrate in Batang Kuranji River

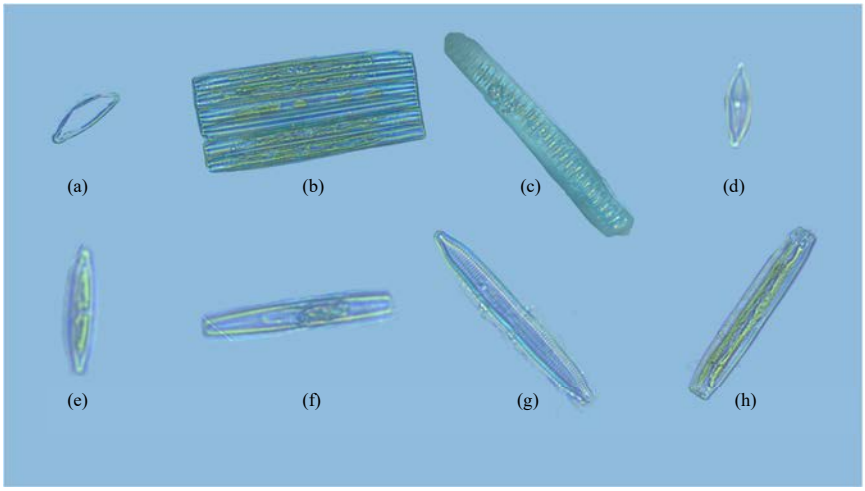

Fig. 6(a-h): Dominant diatom species with relative density >5% on cowhide substrate at each location (40×10 magnification), (a) *Cymbella tumida*, (b) *Fragilaria capucina*, (c) *Fragilaria construens*, (d) *Navicula anglica*, (e) *Navicula cryptocephala*, (f) *Nitzschia amphibia*, (g) *Synedra acus* and (h) *Synedra ulna*

Virta and Hedberg[20], changes in temperature in a body of water can affect the life and diversity of diatoms; the optimum growth of diatoms requires a temperature in the range between 25-30°C.

There was no discernible variation across the five sampling locations (Table 1). In this study, the pH value remains within the ideal range for sustaining diatom life in colonizing cowhide substrates. According to Baharuddin *et al.*[21], pH is one of the environmental factors that may encourage the growth and survival of life in the water. Location 1 is the highest salinity compared to other locations, because is situated in an estuary or the meeting place of freshwater and the sea, it has the most salinity of any location. According to Frost *et al.*[22], changes in salinity cause phytoplankton to maintain osmosis pressure between protoplasm and water, so that salinity in waters can affect the abundance and distribution of phytoplankton.

The current speed at this research location is included in the slow-to-moderate criteria. According to Rozon *et al.*[23], current speed is divided into 5 criteria, namely very fast (>1.25 m/sec), fast (1-1.25 m/sec), moderate (0.5-1 m/sec), slow (0.25-0.5 m/sec) and very slow (0.1-0.25 m/sec). According to Moulidia *et al.*[24], a relatively slow current speed is found in calm waters and shows a relatively high abundance of diatoms.

The DO values were still optimal for the growth and development of diatom colonization. This is in accordance with the quality standards of PP number 21 of 2021 that the

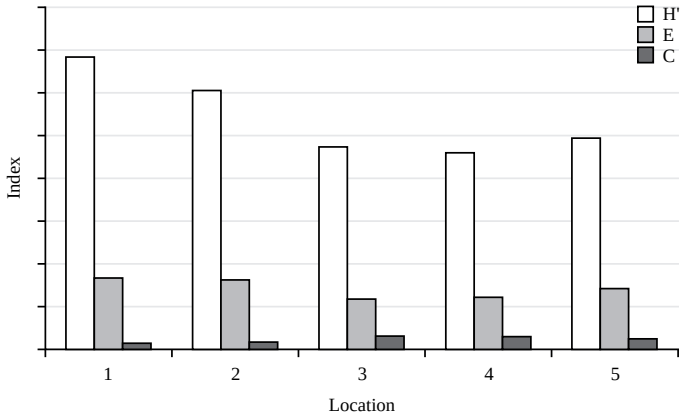

Fig. 7: Diversity index, equitability index and dominance index of diatoms on cowhide substrate at each sampling location

optimal dissolved oxygen to support the life of aquatic biota is more than 4 ppm. According to Shibabaw *et al.*[25], the higher the dissolved oxygen level, the abundance of diatoms will increase. This means that dissolved oxygen content affects the growth and development of diatom colonization in the Batang Kuranji River.

The results showed that location 1 has the highest BOD_5 content compared to other locations. This is because community activities that release waste upstream will accumulate in the estuary, causing high BOD_5 content (Fig. 2). According to Zhu *et al.*[26], BOD_5 is the total dissolved oxygen consumed by microorganisms to degrade organic matter such as food waste and other living things. The free CO_2 content in Batang Kuranji River is influenced by the activities of the surrounding community, such as direct waste disposal. According to Yang *et al.*[27], CO_2 concentration is closely related to dissolved oxygen in a water body and can affect the life of diatoms. The nitrate content obtained in this study was still within the normal range for diatom growth and development. According to Bona *et al.*[28], nitrate plays an important role in the process of protein synthesis for diatom growth. According to Wisha *et al.*[29], nitrite content in water is associated with low dissolved oxygen and nitrate content.

Table 1 shows the value of phosphate content found in the Batang Kuranji River, which ranges from 0.06±0.01 to 0.08±0.01 mg/L. According to Nurdin *et al.*[30], variations in diatom community abundance are influenced by chlorophyll in waters and an environment rich in nutrients such as nitrate, phosphate and silica. Silica content ranges from 80.02±4.88 to 159.17±7.60 mg/L. According to Arumugham *et al.*[31], silicate content is important for diatom development and growth, where silica has a special role in diatom growth. The results showed ammonia content ranged from 0.07±0.01 to 0.14±0.01 mg/L. According to the West Sumatra Governor's regulation on class II river water quality standards, the optimal ammonia content for aquatic biota life should not be more than 0.5 mg/L.

The highest TSS content is found at the mouth of the Batang Kuranji River due to the accumulation of all community activities originating from upstream and slow currents so that all particles in the waters take a long time to settle (Fig. 2). According to PP number 21 of 2021 where the quality standard threshold for TSS is 50 mg/L. Based on the measurement results, the physical and chemical parameters of the waters observed at all locations meet the quality standards and can support the growth and development of diatom colonization on cowhide substrates. According to Zhang *et al.*[32], phytoplankton growth can be influenced by water quality conditions in an environment.

The results showed that the pioneer species on the second day found on cowhide substrate were characterised by a large number of diatom individuals, mainly dominated by species of *Cymbella tumida*, *Cymbella turgida*, *Fragilaria capucina*, *Fragilaria construens*, *Navicula cryptocephala*, *Synedra acus* and *Synedra ulna* at all research locations. Diatom species such as *Fragilaria capucina*, *Fragilaria construens*, *Synedra acus* and *Synedra ulna* are found every day of the study (Fig. 6). According to Rahman *et al.*[33], the genus *Fragilaria* and *Synedra* are diatoms that are found abundantly in aquatic ecosystems and have good adaptability to changing environmental conditions. The peak colonization of diatom species occurred on the 8th day of observation in all study locations.

Diatom colonization can be used as a support for pinpointing the scene of the crime and the length of time the victim drowned. Based on observations of diatom

colonization, diatom species were found that can help indicate the crime scene of drowning victims in a body of water, where each sampling location has a unique diatom species. This is in accordance with the statement of Smol and Stoermer[34], typical diatoms in a body of water can be used as an indicator of the crime scene of drowning victims. Typical species are species that are only found in one location and not found in other locations, so they can be used to help diagnose the crime scene of drowning victims (Table 2). The results showed that the number of typical species found at each research location included 17 species (location 1), 6 species (location 2), 13 species (location 3), 5 species (location 4) and 1 species (location 5). These diatom species can be used as a pointer to the scene of drowning victims in the Batang Kuranji River and the number of typical species in this study was found to be 42 species (Fig. 4).

In addition to indicating the crime scene, diatoms can be used to indicate the time of drowning. Species that can be used are only found on one day of observation and not found on other days. The number of diatom species that can be used to indicate the time of drowning in all research locations is 68 species. Diatom species found on cowhide substrate in Batang Kuranji River showed a variety of numbers and species (Fig. 3). Diatom species that are always found in all research locations and every day of sampling are *Navicula cryptocephala*.

According to Arsad *et al*.[35], the Bacillariophyceae class is a group of organisms that have a cytoplasm that adapts to the influence of the current with the strength of the attachment tool on the substrate in the form of a gelatinous stalk. The highest density was found in location 3, due to the number of individuals obtained being more than in other locations. It was also caused by the species *Fragilaria capucina*, *Fragilaria construens* and *Synedra ulna*, which were found in the highest number.

The relative density of diatom is in line with the opinion of Mirzahasanlou *et al*.[36], that Navicula is a pennales diatom found almost evenly in aquatic ecosystems and has a high level of tolerance to pollution. According to Harmoko and Krisnawati[37], *Fragilaria* can respond quickly to increased phosphorus and is often found in abundance in fresh and marine waters. *Fragilaria* tends to occupy optimal aquatic environmental conditions.

The diversity index is thought to be due to the physical factors of water chemistry and nutrient content, such as phosphate (0.08 mg/L) at this location, which supports the growth and development of diatoms (Fig. 5). So that the number of diatom individuals increases and is almost evenly distributed for each species. The diversity index in this study is higher than Zhao *et al*.[13], who used artificial substrates in the form of cement plates. According to Vidakovic *et al*.[38], diatom diversity is influenced by changes in physical and chemical factors of waters and the availability of substrate types and nutrient absorption.

According to Nurbaya[39], the equitability value ranges from 0-1; the smaller the equitability value or the closer to 0, the more uneven the distribution of an organism in the community. The equitability index value in the Batang Kuranji River falls into the relatively even category. The distribution of individuals and species in each location is still relatively evenly distributed because the physical and chemical factors of the waters obtained are still in the optimal range for diatom growth (Fig. 7). The dominance index of diatoms in this study shows that there is no dominant diatom species on the substrate of cowhide that is submerged in the Batang Kuranji River.

CONCLUSION

There are 94 kinds of diatoms that colonize the cowhide substrate; 42 of these species can be used to determine the location of the drowning and 68 of these species can be used to determine the duration that the drowning occurred. There are 94 species, 2 classes, 2 orders and 19 families among the diatoms that are found on cowhide substrate. Diatom density ranged from 3.81±0.16 - 22.80±1.34 ind/cm^2, the dominant species was *Fragilaria capucina*. The diversity index criteria were classified as moderate to high with a relatively even distribution of individuals and no dominant species. The results of PCA analysis of the relationship between physico-chemical factors of waters in the Batang Kuranji River show different influences on diatom life including location 1 (free CO_2, BOD_5, TSS, air temperature and salinity), location 2 (ammonia, phosphate, nitrate and water temperature), locations 3 and 4 (pH, DO, nitrite and silica) and location 5 (current speed). The water condition of Batang Kuranji River is still relatively optimal to support the growth and development of diatom colonization on cowhide substrate.

SIGNIFICANCE STATEMENT

This study discovered the specific diatom species colonizing cowhide substrates in a river ecosystem, which can be beneficial for forensic science by providing a reliable biological marker for diagnosing drowning and determining the crime scene and submersion time. The established relationship between diatom colonization dynamics and physico-chemical factors offers a novel bio-monitoring tool.

This study will help researchers to uncover the critical areas of diatom succession and its forensic application that many researchers were not able to explore. Thus, a new theory on using diatom community dynamics for precise post-mortem interval estimation in drowning cases may be arrived at.

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