

Exploratory study on the genus diversity of tropical paludi-microalgae as a potential source of human nutrition and other products

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SUMMARY

Population growth and limited availability of agricultural land have created immense pressure for accessible food and nutritional supplements in Indonesia. To establish new food sources that will be sustainable in the long term, various government programmes have opened up land for agriculture in the peatlands of Central Kalimantan. However, the establishment of both large-scale plantations and community-based agriculture has required drainage of the peatlands leading to a decline in the habitat along with increased greenhouse gas emissions and incidence of peatland fires. The option of rewetting and adopting wetland agriculture (paludiculture) in these areas could significantly improve the preservation of both terrestrial carbon stocks and the native flora and fauna. Cultivating microalgae as part of paludiculture could provide additional resources including food for both humans and aquatic animals, benefitting agriculture and fisheries in the future. This preliminary study aims to identify microalgae genera in the tropical peat swamp waters of Central Kalimantan, and in particular to determine which of the microalgae present may have potential as sources of human nutrition as well as for other uses. Peat water samples were obtained from three distinct locations in Palangka Raya City. Specimens were identified using techniques based on morphological characteristics observed by photomicrography, and the nutritional potential of each genus was determined by literature review. The analysis revealed paludi-microalgae belonging to a total of 17 genera, two from Class Bacillariophyceae (*Pinnularia*, *Navicula*), two from Class Chlorophyceae (*Pandorina*, *Scenedesmus*), three from Class Cyanobacteria (*Microcystis*, *Anabaena*, *Oscillatoria*), four from Class Euglenophyceae (*Euglena*, *Trachelomonas*, *Lepocinclis*, *Phacus*) and six from Class Zygnematophyceae (*Closterium*, *Cosmarium*, *Euastrum*, *Micrasterias*, *Spirogyra*, *Pleurotaenium*). The highest number of microalgae genera was found in peat swamp ponds in Palangka Raya City Forest, which contributed 95 % of all genera obtained. Literature review revealed that several of these genera such as *Spirogyra*, *Navicula* and *Euglena* have high potential as food sources with significant nutritional properties, containing carbohydrates, proteins, lipids, fats, minerals and a range of vitamins. Various other useful properties were also identified.

KEY WORDS: aquatic habitat, Central Kalimantan, food, Indonesia, paludiculture, peat swamp forest

INTRODUCTION

Peatland is classified as wetland or swamp (Latin “palus”), and is a singular terrestrial ecosystem that boasts a diverse array of flora and fauna. It is identifiable by the presence of a layer of peat, which accrues over time when the rate of organic matter production exceeds the decomposition rate. In its natural state peat contains 90 % water and 10 % decomposing plants; and is acidic (pH 3–5), anaerobic and deficient in nutrients (Joosten & Clarke 2002, Osaki *et al.* 2016, Giesen & Sari 2018). Peat water is either groundwater or surface water and typically has

a brownish-red colour due to high levels of Fe and Mn, a sour taste from low pH (pH < 7), low cations, and high turbidity with low suspended particle content due to high organic substance content.

Central Kalimantan, located on Indonesia's Kalimantan Island (Borneo), is known for its vast peatland area, which makes up 18 % of Indonesia's peatlands and 55 % of Kalimantan's peatlands (Ritung *et al.* 2011, Uda *et al.* 2017). These and other peatlands in Indonesia formed over sea mud and sand during the past 14,000 years and were initially covered by tropical rainforests (Dommain *et al.* 2011, Dohong *et al.* 2017). These tropical peatlands are

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recognised for harbouring exceptional biodiversity of flora, fauna and microorganisms, making them ecologically valuable and worthy of preservation (Yule 2010, Posa *et al.* 2011, Husson *et al.* 2018, UNEP 2022, Han *et al.* 2023).

The growing human population and the limited availability of agricultural land have created immense pressure for accessible food and nutritional supplements in Indonesia. As a result, it is crucial to consider alternative food sources and prioritise the long-term sustainability of food supplies (Henchion *et al.* 2017, Llewellyn 2022). To address this issue, the government has implemented various agricultural programmes such as the Food Estate and Mega Rice Projects, which have aimed to open up agricultural land in the peatlands of Central Kalimantan (Uda *et al.* 2020b, Page *et al.* 2022, UNEP 2022). However, this has involved drainage of the peatlands meaning they become usable (if at all) for only dryland-type agriculture such as large-scale plantations and community-based farming (Uda *et al.* 2020a, Jaya *et al.* 2022, UNEP 2022). This has led to a decline in wetland habitat along with increased greenhouse gas emissions and risk of peatland fires. The option of re-converting these areas to paludiculture (the cultivation of wetland plants as crops on rewetted peatland; Joosten *et al.* 2016, Uda *et al.* 2020a, Nielsen *et al.* 2023) could have a significant effect in preserving both terrestrial carbon stocks and the native flora and fauna (Giesen & Sari 2018, Uda *et al.* 2020a).

Microalgae are microscopic primary producers that are found in both freshwater and marine ecosystems (Hidayah *et al.* 2023). With over 25,000 species, they can be divided into eukaryotic microorganisms and prokaryotic cyanobacteria, also known as blue-green algae (Guiry 2012, Koyande *et al.* 2019). Algae play important roles in aquatic ecosystem function. The species composition of phytoplankton, especially diatoms, is widely used as an indicator of the biological integrity, chemical conditions and ecological status of wetlands (Costa & Schneck 2022, Dubey *et al.* 2022). Microalgae are also widely used in various fields to meet human needs. They can be sources of food (Koyande *et al.* 2019, Prihanto *et al.* 2022), renewable energy (Medipally *et al.* 2015, Milano *et al.* 2016, Flassy *et al.* 2020, Almomani *et al.* 2023), cosmetics (Mourelle *et al.* 2017) and bioremediation agents (Dewi & Nuravivah 2018, Viswanaathan & Sudhakar 2019, Nie *et al.* 2020, Zohoorian *et al.* 2020), as well as bioindicators of environmental quality (Omar 2010, Dell'Aglio 2017, Kadam *et al.* 2020).

Several studies have been conducted on microalgae diversity in Indonesia. They have been

identified and recorded from Sumatra (Pane & Harahap 2023, Pane *et al.* 2023), Java (Arsad *et al.* 2022, Fendiyanto *et al.* 2023, Hidayah *et al.* 2023), Sulawesi (Rukminasari & Sahabuddin 2012, Tambaru *et al.* 2021), Kalimantan (Apriansyah *et al.* 2021, Adam *et al.* 2023) and Papua (Purbani *et al.* 2019, Flassy *et al.* 2020). Recent studies have reported various types of microalgae in Central Kalimantan waters, such as those belonging to the classes Chlorophyceae, Zygnematophyceae and Euglenophyceae (Adam 2022, Adam & Haryono 2022; Figure 1).

The Central Kalimantan peatland is characterised by a network of water bodies supplied by multiple rivers including freshwater and peat-water rivers (Uda *et al.* 2020a, Adam *et al.* 2023). Peat waters in this region have unique features such as a blackish-brown colour, a pH range of 3–5, a relatively high organic matter content, a depth of 30–50 cm, a temperature around 31.4 °C, and total dissolved solids (TDS) 13 ppm. The high organic substance content serves as a food source for aquatic microorganisms (Adam & Haryono 2022, Prihanto *et al.* 2022, Han *et al.* 2023), and microalgae thrive in these waters.

In this article the microalgae found in peatland waters are referred to as paludi-microalgae, meaning microalgae that come from waters in peat swamp areas. They are threatened by peatland drainage, which could ultimately cause a decrease in the presence and populations of these microorganisms. On the other hand, cultivating microalgae in peat waters as part of paludiculture could provide additional resources including food for both humans and aquatic animals, benefitting agriculture and fisheries in the future.

This exploratory study aims to provide scientific insights into the diversity of microalgae in the aquatic habitats of Indonesian tropical peat swamps. The objectives are to investigate the variety of paludi-microalgae genera in Central Kalimantan peatlands and to discover which of them have potential as sources of human nutrition, as well as for other uses.

METHODS

Study sites and sample collection

The study was conducted in shallow peat waters within Palangka Raya City, Central Kalimantan, Indonesia. Purposive sampling was employed at three distinct sites: (1) peat water ponds in Palangka Raya City Forest; (2) water-filled ditches in the UPT PPIIG forest research area on the University of Palangka Raya campus; and (3) ditches in the

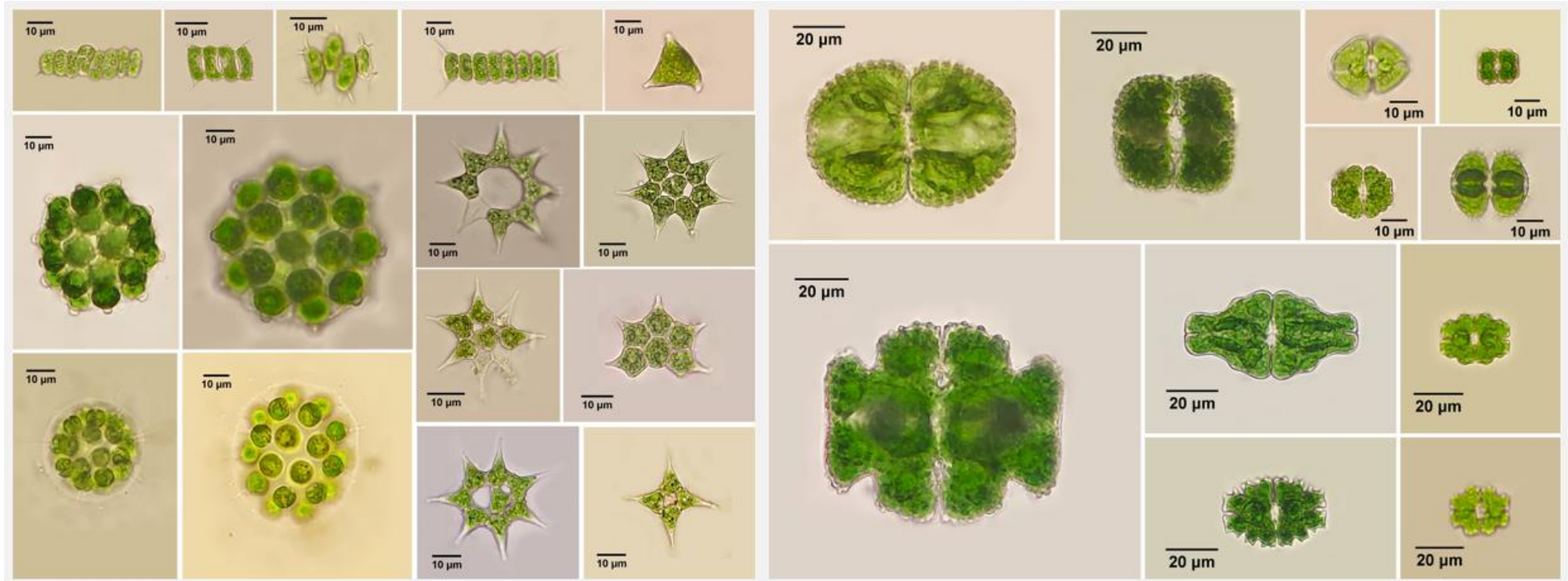


Figure 1. Microalgae previously reported from Central Kalimantan waters (adapted from Adam 2022, Adam & Haryono 2022).

residential street Jalan Mahir Mahar which form part of the 'outer ring river' of the Palangka Raya drainage network. These sites were chosen to encompass different types of wet places distributed across the city (Figure 2); the urban forest for its biodiversity, the campus forest as a forest location influenced by human activity and research, and the outer ring river for insights about the effects on microalgae of water circulation and pollution in the city's drainage system.

Samples of peat water and sediment were collected from the bottom of the water column directly into clean 300 ml plastic bottles which were labelled with information about where they were taken from. A sample from each site was collected three times between morning and evening on a single day (22 November 2022) to incorporate any diurnal variations in microalgae caused by changes in, e.g., light level and water temperature through the day. The samples were transported to the laboratory and left to settle overnight, then each bottle was sub-sampled and observed ten times, resulting in 90 observations in total.

Observation and microphotography

The microalgae species were identified using the microphotography method. The sub-samples were observed using an Olympus CX21 compound microscope. Images of specimens for further identification were captured through the microscope's eyepiece then processed using Adobe Photoshop CC for improved colour and detail. To estimate the lengths of objects in each microphotograph, a scale bar was added using ImageJ software version 1.53g (Adam 2022). The scaling method in ImageJ estimated the cell size by knowing the cell diameter in the microscope's field of view at 400× magnification (10× ocular, 40× objective) (Adam & Haryono 2022).

Morphological analysis and identification

Morphological analysis was performed on the samples by assessing the photomicrographs in terms of microalgae taxonomy (Adam & Haryono 2022). The identification of microalgae involved a detailed examination of their morphological characteristics including locomotion, colour, cell shape and size. This analysis was conducted both qualitatively and quantitatively. To accurately identify the microalgae genus, morpho-taxonomic identification was carried out by referring to key microalgae identification guides (Prescott 1964, Belcher & Swale 1976, Shayler & Siver 2006, van Vuuren *et al.* 2006, Serediak & Huynh 2011, Guiry & Guiry 2013, Jeuck & Arndt 2013).

Potential uses of the microalgae identified

Information on the nutritional properties and other potential uses of each microalgae genus identified was gathered by literature review. We used search engines such as Wikipedia, Google and Google Scholar to find relevant sources. Examples of the key words used in searches are "microalgae", "nutritional properties" and the names of individual genera.

RESULTS

In this research, the microalgae were taxonomically classified at the genus level. The results revealed the presence of five different classes of microalgae in the Central Kalimantan peat swamp waters, i.e.: Bacillariophyceae, Chlorophyceae, Cyanobacteria, Euglenophyceae and Zygnematophyceae. The study identified a total of 12 families and 17 genera of paludi-microalgae including two genera of Bacillariophyceae, two genera of Chlorophyceae, three genera of Cyanobacteria, four genera of Euglenophyceae and six genera of Zygnematophyceae (Table 1).

Bacillariophyceae

Bacillariophyceae, also known as diatoms, are a diverse group of microalgae commonly found in marine, freshwater and terrestrial environments. They exhibit a wide range of cell shapes from elongated to circular, triangular and pentagonal. The cell walls of diatoms are composed of frustules, which consist of two valves or shells connected by a thin silica band known as the girdle band (Burckle & Akiba 1978). With over 20,000 known species out of an estimated 72,500 algae species (Guiry 2012), diatoms contribute significantly to the oxygen released into the environment each year (De Tommasi 2016, Benoiston *et al.* 2017, Llewellyn 2022). They typically range in size from 3 µm to 2 mm (Svensson *et al.* 2014) and contain chlorophyll a, chlorophyll c and fucoxanthin, as well as other pigments such as xanthophylls and carotenoids that give them a golden brown appearance (Strain *et al.* 1944, Bertrand 2010). In a recent study of tropical peat waters in Central Kalimantan, two genera of diatoms (*Pinnularia* and *Navicula*) were identified. *Pinnularia* (Pinnulariaceae) (Figure 3a–d) has elongated golden brown cells. According to Mahoney (2018), *Pinnularia* valves are linear with wide parallel margins, and circular at the terminal. In our study, two variations of *Pinnularia* form were observed which were thought to be different species. *Navicula* (Naviculaceae) (Figure 3f) is termed a boat-shaped diatom because of its cell morphology.

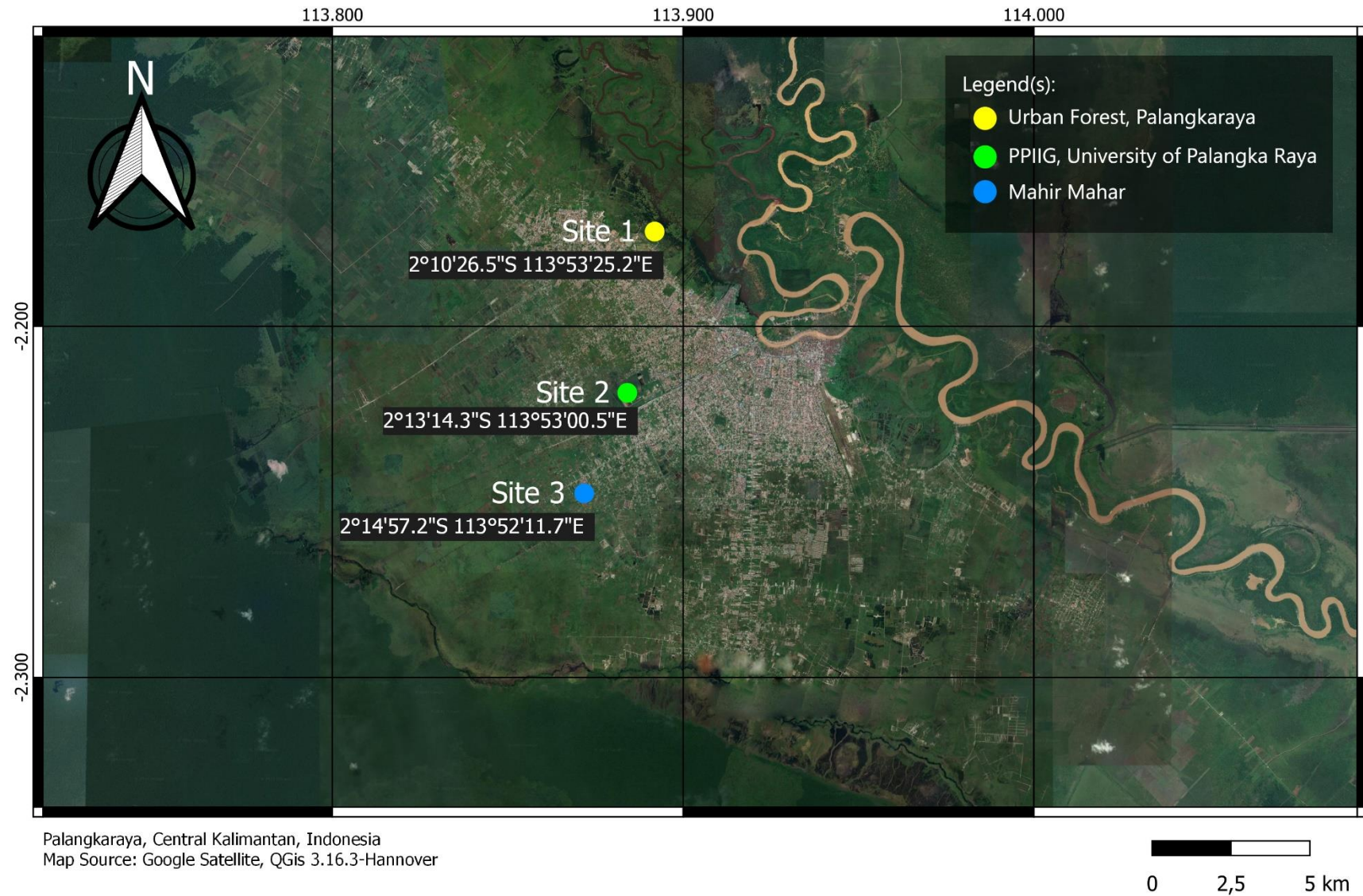


Figure 2. The locations in Palangka Raya (Central Kalimantan, Indonesia) where water samples were collected. Sources: Google Satellite, QGis 3.16.3-Hannover.



Table 1. Types of microalgae identified in peat waters from ponds in the Palangka Raya City Forest.(Site 1), ditches in the PPIIG research area on the University of Palangka Raya campus (Site 2) and ditches in Jalan Mahir Mahar, a residential street on the outskirts of Palangka Raya City. + = present; - = absent.

No.	Class	Family	Genus	Sampling Sites		
				Site 1	Site 2	Site 3
1	Bacillariophyceae	Naviculaceae	<i>Navicula</i>	-	+	-
		Pinnulariaceae	<i>Pinnularia</i>	+	+	+
2	Chlorophyceae	Scenedesmaceae	<i>Scenedesmus</i>	-	-	+
		Volvocaceae	<i>Pandorina</i>	-	-	+
3	Cyanobacteria	Microcystaceae	<i>Microcystis</i>	-	-	+
		Nostocaceae	<i>Anabaena</i>	-	-	+
		Oscillatoriaceae	<i>Oscillatoria</i>	+	-	+
4	Euglenophyceae	Euglenaceae	<i>Euglena</i>	-	-	+
			<i>Trachelomonas</i>	-	-	+
		Phacaceae	<i>Lepocinclis</i>	-	+	+
			<i>Phacus</i>			+
5	Zygnematophyceae	Closteriaceae	<i>Closterium</i>	+	+	+
			<i>Cosmarium</i>	-	-	+
			<i>Euastrum</i>	-	-	+
			<i>Micrasterias</i>	-	-	+
			<i>Pleurotaenium</i>	-	-	+
		Zygnemataceae	<i>Spirogyra</i>	-	-	+

Chlorophyceae

Chlorophyceae, a type of green algae, can exist in various forms such as unicellular, solitary, colonial and filamentous. The green hue is due to chlorophyll a and chlorophyll b, which are the dominant pigments (van Vuuren *et al.* 2006). We classified the Chlorophyceae into two types: *Pandorina* (Volvocaceae) and *Scenedesmus* (Scenedesmaceae). *Pandorina* (Figure 3f), a genus of green algae belonging to the Volvocaceae family, is found in small colonies called coenobia consisting of 8–32 cells (Nozaki 2003, Kita *et al.* 2010). These cells form circular coenobia. *Scenedesmus* (Figure 3g), on the other hand, is a genus of green algae from the Scenedesmaceae family that forms coenobia consisting of 4–32 cells (van Vuuren *et al.* 2006, Gopalakrishnan *et al.* 2014). *Scenedesmus* identified in this study forms coenobia consisting of four elongated cells that narrow at the terminal. Species of *Scenedesmus* can be determined on the basis of the number, shape and arrangement of cells in coenobia, as well as the ornamentation on the cell walls (Hegewald 1997, van Vuuren *et al.* 2006).

Cyanobacteria

Cyanobacteria are a type of bacteria that closely resemble algae, and are known as ‘algae-like bacteria’. Like microalgae, they are important members of the phytoplankton community in aquatic ecosystems. They use chlorophyll a and other pigments such as phycocyanin and allophycocyanin to perform photosynthesis. These pigments give cyanobacteria their characteristic blue-green colouration. Cyanobacteria can be found in various habitats, from aquatic to terrestrial. In our analysis we recognised three cyanobacteria genera, namely: *Microcystis*, *Anabaena* and *Oscillatoria* (Figures 3h–j). *Microcystis* (Figure 3h) is a cyanobacteria genus from the Microcystaceae family that forms large colonies consisting of spherical cells. *Anabaena* (Figure 3i) and *Oscillatoria* (Figure 3j) are non-branching filamentous cyanobacteria. The main difference is that *Anabaena* contains heterocysts and akinetes, which are derivatives of vegetative cells (Shayler & Siver 2006).

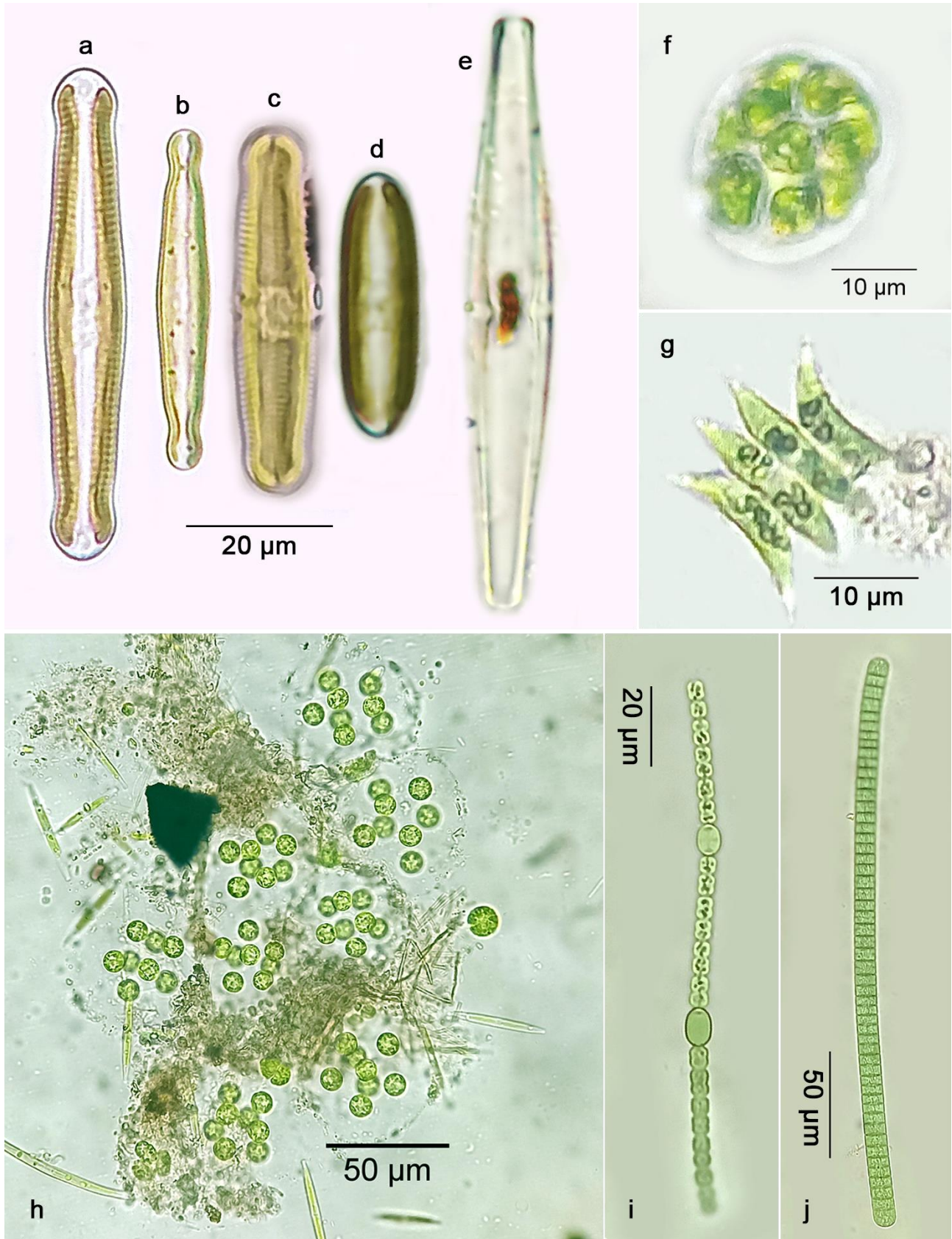


Figure 3. Bacillariophyceae: (a–d) *Pinnularia*; (e) *Navicula*. Chlorophyceae: (f) *Pandorina*; (g) *Scenedesmus*. Cyanobacteria: (h) *Microcystis*; (i) *Anabaena*; (j) *Oscillatoria*.

Euglenophyceae

Euglenophyceae is a class of green algae that uses flagella to move around in aquatic environments, from fresh to marine waters (Alves-da-Silva & Bicudo 2009, Bicudo & Menezes 2016). One of the defining features of Euglenophyceae is the presence of red eyespots, which are part of their photoreceptive organelles known as the eyespot apparatus (Belcher & Swale 1976). The eyespot apparatus consists of a paraflagellar body that connects the eyespot to the flagellum (Kivic & Vesik 1972, Kreimer 2009, Häder & Iseki 2017). In this study, four genera of Euglenophyceae were identified including *Euglena*, *Trachelomonas*, *Lepocinclis* and *Phacus* (Figure 4). The Euglenaceae family includes *Euglena* and *Trachelomonas*. *Euglena* is spindle-shaped with an oval or cylindrical body and a single anterior flagellum. It has a red eyespot that can be seen in the anterior part of the cell (Figure 4a). *Trachelomonas* is oval-shaped with a single anterior flagellum and a red eyespot visible in the anterior part of the cell (Figure 4b–c). The Phacaceae family includes *Lepocinclis* and *Phacus*. *Lepocinclis* is elliptical or spherical in shape with a clear red eyespot and a short

caudal process (Figure 4d–e). It can be described as citriform, fusiform, sub-globose, oval or elliptical (Wołowski *et al.* 2013). *Phacus* has a leaf-like shape and a red eyespot that is easily visible (Figures 4f–h). It has a single flagellum and narrows down at the posterior part of the cell (Prescott 1964, Belcher & Swale 1976, van Vuuren *et al.* 2006).

Zygnematophyceae

The class Zygnematophyceae is also known as Conjugatophyceae, which means ‘conjugation algae’ (Guiry & Guiry 2013). At the research location we identified six genera belonging to the class Zygnematophyceae, from three families: Closteriaceae, (*Closterium*), Desmidiaceae (*Cosmarium*, *Euastrum*, *Micrasterias*, *Pleurotaenium*) and Zygnemataceae (*Spirogyra*). *Closterium* (Figure 5a–f) has elongated cells shaped like crescent moons and can be curved at the terminal, straight, or have spines at both ends of the semi-cells (Prescott 1964, Oliveira *et al.* 2013). The degree of curvature varies between species and the semi-cells are separated by an indistinct median constriction. Some species can grow to 1 mm in length (van Vuuren *et al.* 2006).

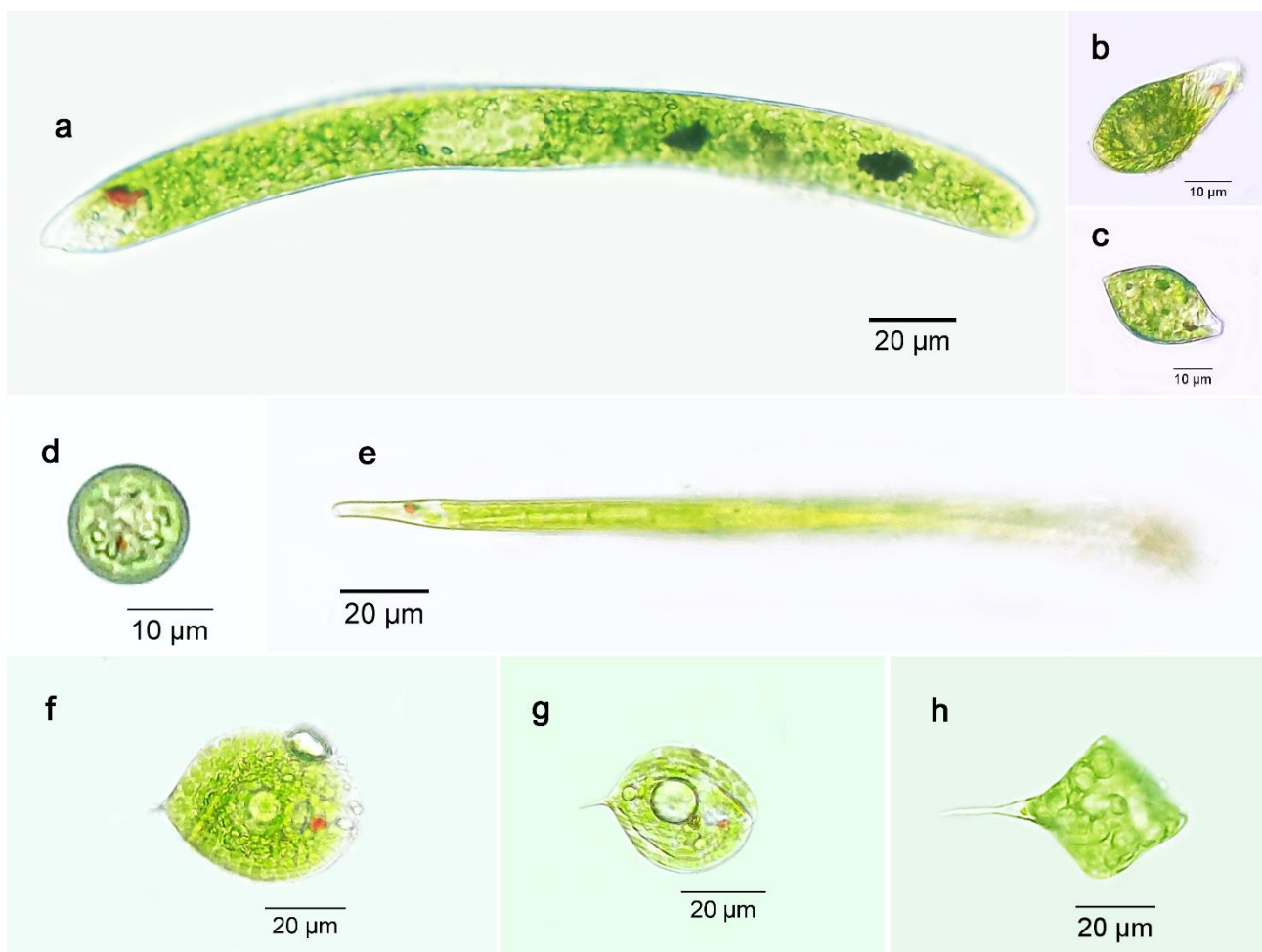


Figure 4. Euglenophyceae: (a) *Euglena*; (b–c) *Trachelomonas*; (d–e) *Lepocinclis*; (f–h) *Phacus*.

Cosmarium species are generally solitary, free-living unicellular microalgae. They have two symmetrical semi-cells divided by an isthmus (Figure 5g). The semi-cells of *cosmariums* are spherical, hemispherical or pyramidal in shape, with the nucleus visible on the isthmus (Belcher & Swale 1976, van Vuuren *et al.* 2006, Oliveira *et al.* 2013). *Euastrum* (Figures 5h–i) has semi-cells of different shapes including oval, spherical, trapezoidal and pyramidal, separated by a median (sinus) constriction. The cell walls appear flat or may have ornamentations like spines, depending on the species (Taft 1945, Prescott 1964, Belcher & Swale 1976, van Vuuren *et al.* 2006, Anissimova & Staer 2018).

Micrasterias (Figure 5j) is well-known for its elaborate star-shaped cell morphology and symmetrical semi-cells. (Lütz-Meindl 2016). *Spirogyra* (Figure 5k), a genus from the Zygnemataceae family, possesses long filaments and chloroplasts that are spiral-shaped (Coesel & Meesters 2007, Wehr & Sheath 2015). *Pleurotaenium* (Figure 5l) has semi-cells that are elongated and cylindrical, separated by a median constriction (Claassen 1985, Domozych *et al.* 2007).

Potential uses of Kalimantan paludi-microalgae

The results of our comprehensive literature review are summarised in Table 2. The review showed that

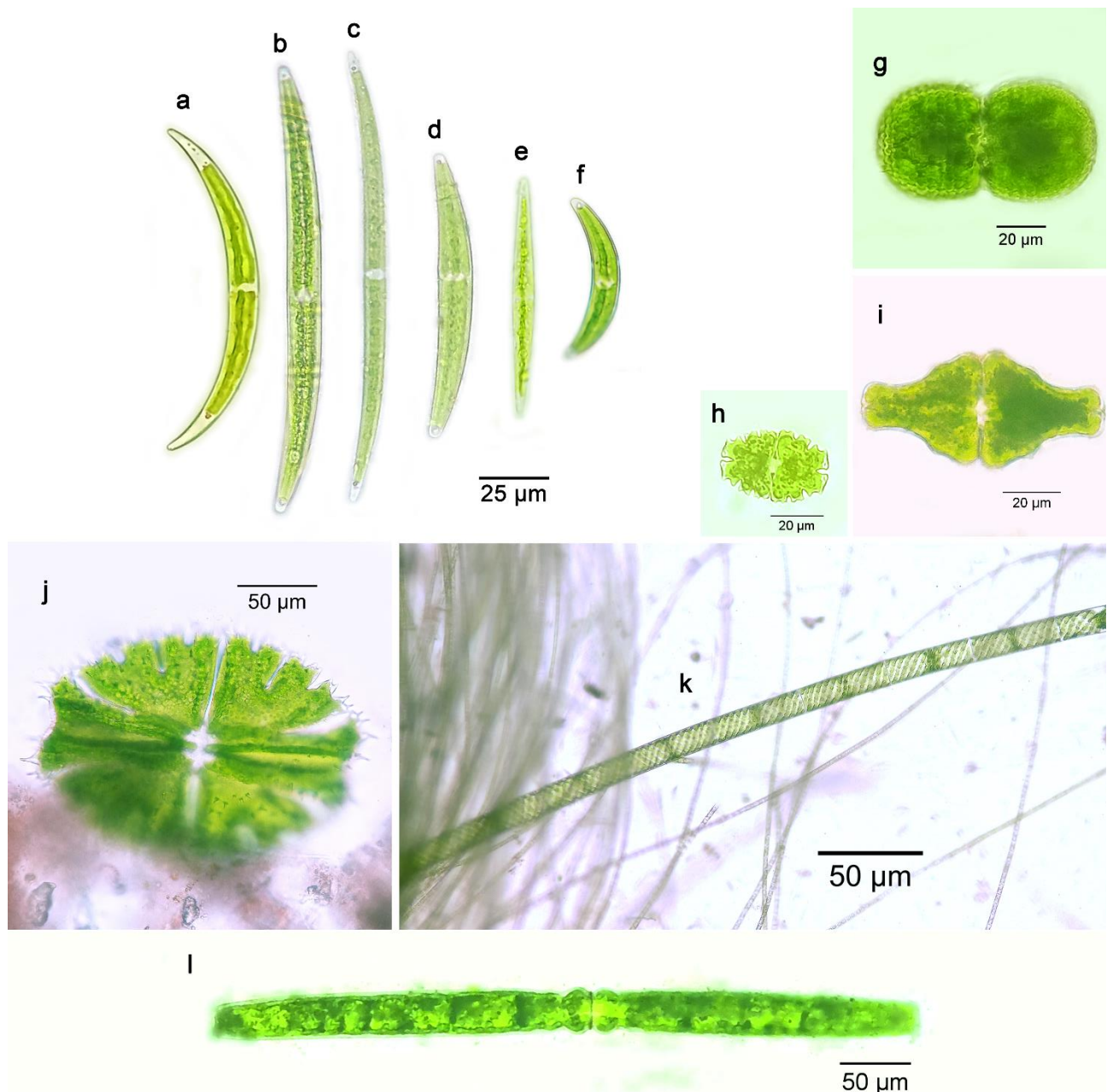


Figure 5. Zygnematophyceae: (a–f) *Closterium*; (g) *Cosmarium*; (h–i) *Euastrum*; (j) *Micrasterias*; (k) *Spirogyra*; (l) *Pleurotaenium*.

Table 2. The potentially useful properties of microalgae found in the tropical peat waters of Central Kalimantan, Indonesia.

No.	Genus	Potentially useful properties	References
1	<i>Navicula</i>	Nutritional components: carbohydrates; proteins; lipids; fatty acids such as myristic acid, pentadecanoic acid, palmitic acid, stearic acid, palmitoleic acid and eicosapentaenoic acid (EPA); the pigments chlorophyll a, chlorophyll c and fucoxanthin. Other functional properties: oxygen producer; biodiesel base material; bioactivity antibacterial; antioxidant; methanol; navicular antiviral polysaccharide; strong against HSV-1 and HSV-2 and influenza viruses.	Bertrand (2010) Mahoney (2018) Kim <i>et al.</i> (2019) Prihanto <i>et al.</i> (2022) Pane <i>et al.</i> (2023)
2	<i>Pinnularia</i>	Nutritional components: lipids; fatty acids; the pigments chlorophyll a, chlorophyll c, β -carotene and fucoxanthin. Other functional properties: biofuel base material.	Bertrand (2010) Mahoney (2018) Prihanto <i>et al.</i> (2022)
3	<i>Scenedesmus</i>	Nutritional properties: protein; carbohydrates; oil/lipids; fatty acids; the pigments astaxanthin, chlorophyll, lutein, zeaxanthin, β -carotene and echinenone. Other functional properties: biofuels; biodiesel; bio-ethanol; bioindicator; bioremediation agent for heavy metals such as Cu, Co, Pb, Zn and Hg; ammonia and phosphorus removal.	Hegewald (1997) Inthorn <i>et al.</i> (2002) Ajayan <i>et al.</i> (2011) Dell'Aglio <i>et al.</i> (2017) Wang <i>et al.</i> (2017) Arguelles (2018) Khatoon <i>et al.</i> (2019) Prihanto <i>et al.</i> (2022) Fendiyanto <i>et al.</i> (2023)
4	<i>Pandorina</i>	Nutritional properties: lipids; fatty acids; the pigments chlorophyll a and chlorophyll b. Other functional properties: bioindicator.	Claassen (1985) O'Neill <i>et al.</i> (2022) Prihanto <i>et al.</i> (2022) Chandel <i>et al.</i> (2023)
5	<i>Microcystis</i>	Nutritional properties: crude protein; oil/lipids; carbohydrates; calcium; phosphorus; fatty acids; allophycocyanin; phycoerythrocyanin. Other functional properties: antioxidant; antibacterial; biofuels; bioindicator of environmental quality of neurotoxins and hepatotoxins produced.	Lemasson <i>et al.</i> (1973) Vincent (2009) Basheva <i>et al.</i> (2018) Prihanto <i>et al.</i> (2022) Fendiyanto <i>et al.</i> (2023)
6	<i>Anabaena</i>	Nutritional properties: crude protein; carbohydrates; phosphorus. Other functional properties: Nitrogen fixation; antiviral; bioindicator of environmental quality of neurotoxins produced.	Prihanto <i>et al.</i> (2022) Wikipedia (2023a)
7	<i>Oscillatoria</i>	Nutritional properties: oil/lipids; fatty acids; phycoerythrin. Other functional properties: butylated hydroxytoluene (BHT) as an antioxidant; antiviral; food additive; industrial chemicals; biofuels.	Irmak & Arzu (2020) Prihanto <i>et al.</i> (2022) Fendiyanto <i>et al.</i> (2023)

No.	Genus	Potentially useful properties	References
8	<i>Euglena</i>	Nutritional properties: high level of carbohydrates; protein; 20 proteinogenic amino acids; lipids; polyunsaturated fatty acids; omega-3 acid; paramylon; provitamin A; β -carotene; vitamin A (retinol, retinal, retinoic acids, retinyl esters); vitamin C (ascorbate); vitamin E, docosahexaenoic. Other functional properties: source of bioproducts; pyrenoids; antioxidants; bioindicators; biodiesel; jet fuel.	Isegawa <i>et al.</i> (1993) Rodríguez-Zavala <i>et al.</i> (2010) Wołowski <i>et al.</i> (2013) Yadavalli <i>et al.</i> (2014) Häder & Iseki (2017) Henchion <i>et al.</i> (2017) Hasan <i>et al.</i> (2017) Ritala <i>et al.</i> (2017) Gissibl <i>et al.</i> (2019) Prihanto <i>et al.</i> (2022)
9	<i>Trachelomonas</i>	Nutritional properties: manganese; iron. Other functional properties: bioindicator; biofilm.	Wong & Heera (2022) Wikipedia (2023b)
10	<i>Lepocinclis</i>	Nutritional properties: oil/lipids; carotenoids; fatty acids (EFAs). Other functional properties: biofuels.	Wołowski <i>et al.</i> (2013) Fendiyanto <i>et al.</i> (2023)
11	<i>Phacus</i>	Nutritional properties: carbohydrate. Other functional properties: bioindicator.	Wołowski <i>et al.</i> (2013) Wikipedia (2023c)
12	<i>Closterium</i>	Nutritional properties: oil/lipids; rhamnose; fucose; xylose; mannose; galactose; glucose; uronic acids; iron; manganese. Other functional properties: biofuels; biofilm; bioenergy.	Oliveira <i>et al.</i> (2013) Wong & Heera (2022) Fendiyanto <i>et al.</i> (2023)
13	<i>Cosmarium</i>	Nutritional properties: glutamic acid; aspartic acid. Other functional properties: bioindicators of water environmental quality, commonly found in acidic and oligotrophic freshwater environments.	van Vuuren <i>et al.</i> (2006) Santiago-Díaz <i>et al.</i> (2022)
14	<i>Euastrum</i>	Nutritional properties: oil/lipids; triacylglyceride. Other functional properties: biofuels; antioxidant; bioindicators of water environmental quality.	Anissimova & Staer (2018) Adam (2022) Fendiyanto <i>et al.</i> (2023)
15	<i>Micrasterias</i>	Nutritional properties: protein; lipid. Other functional properties: antioxidant; bioindicator; the pigments chlorophyll a and chlorophyll b; model organisms for research on plant cell development because of their close genetic kinship with higher plants.	Sangeetha <i>et al.</i> (2009) Lütz-Meindl (2016)
16	<i>Pleurotaenium</i>	Nutritional properties: oil/lipids; protein. Other functional properties: biofuels; bioindicators; biodiesel.	Claassen (1985) Domozych <i>et al.</i> (2007) Fendiyanto <i>et al.</i> (2023)
17	<i>Spirogyra</i>	Nutritional properties: protein; peptides; amino acids; vitamin B6; vitamin B12; iron; potassium; calcium; magnesium; phosphorus; carbohydrates (starches and sugars). Other functional properties: bioethanol; biogas; biodiesel; bioplastic; antioxidant; antibacterial.	Pimpimol <i>et al.</i> (2020) Prihanto <i>et al.</i> (2022) Yongkhamcha & Buddhakala (2023)

the microalgae identified in our samples from the peat swamp forest waters of Palangka Raya have various potential applications in fields including food, environment and renewable energy (Isegawa *et al.* 1993, Inthorn *et al.* 2002, Rodríguez-Zavala *et al.* 2010, Ajayan *et al.* 2011, Hasan *et al.* 2017, Henchion *et al.* 2017, Ritala *et al.* 2017, Wang *et al.* 2017, Gissibl *et al.* 2019). Our research indicates that *Spirogyra*, *Navicula* and *Euglena* are the most extensively studied genera for their potential as food sources rich in carbohydrates, proteins, lipids, minerals and vitamins (Matos *et al.* 2017, Prihanto *et al.* 2022); while *Spirogyra* is known for its high protein and carbohydrate content (Pimpimol *et al.* 2020), *Navicula* is a good source of lipids (Kim *et al.* 2019) and *Euglena* is rich in carbohydrates (Prihanto *et al.* 2022). The species commonly linked to harmful algal blooms present specific challenges when considering their use for nutritional purposes. The presence of toxins, particularly in *Microcystis* and *Anabaena*, makes direct consumption unsafe. However, the issue of toxicity may be solved through toxin removal methods and other safety measures (De Oliveira *et al.* 2022).

DISCUSSION

Genus diversity of paludi-microalgae in tropical peat swamp waters

Biota populations that inhabit tropical peat swamp waters are typically small and unique (Joosten & Clarke 2002, Osaki *et al.* 2016, UNEP 2022), and the quality of the water plays a crucial role in determining the endemic species of different groups of organisms including microalgae (Omar 2010, Uda *et al.* 2017, Pane & Harahap 2023). The majority of microalgae genera identified in our study were collected from peat-water ponds in the Palangka Raya City Forest (Site 3). This can be attributed to the mixotrophic environmental conditions, which support a variety of microalgae genera and can be associated with the variety and density of plants in the area (Padil *et al.* 2022, Han *et al.* 2023).

Class Zygnematophyceae boasts the highest number of genera compared to other microalgae classes. Typically found in freshwater environments, this class is known for its impressive diversity, with approximately 4,000 species spanning 60 genera (Gontcharov 2008). Most of these species are found in tropical periodic waters with an acidic pH (Št'astný 2008). The genus *Spirogyra* is a member of the Zygnematophyceae and is notable for its high species diversity, with over 500 species distributed worldwide (Guiry 2012, Guiry & Guiry 2013).

Challenges and development prospects of paludi-microalgae

Despite the promising findings summarised in Table 2, research on the role of microorganisms in tropical peat swamps is still limited and information on the potential benefits of conserving microalgae in peat swamp water ecosystems remains focused on species that are relevant to the fisheries sector (e.g. *Chorella*, *Spirulina*; Haoujar *et al.* 2022).

Identification of microalgae in the current research is based primarily on genus. More specific identification methods utilising species and molecular analysis (DNA) should be pursued for greater accuracy in results (Pavan-Kumar *et al.* 2015, Seymour 2022). Genetic variation analysis of microalgae species found in peat swamp waters can reveal specific characteristics and phylogenetic data, allowing for comparison with the same species found in other aquatic habitats such as freshwater or marine waters (O'Neill *et al.* 2022, Han *et al.* 2023). Thus, further research and molecular (DNA) analysis is needed to accurately identify species of paludi-microalgae that have potential for cultivation as sources of human food, nutrients and other products.

The information we have gathered so far strongly supports the case that microalgae represent a promising food source that could be plentiful and sustainable in conjunction with other paludicultures and thus have potential to help satisfy the demands of a growing human population for food whilst reducing the pressure on marginal agricultural land and mitigating water pollution in Indonesia (Llewellyn 2022, Prihanto *et al.* 2022). To explore the potential further, there are well established protocols and methods for sampling and measuring both structural and functional properties of algae including habitat preferences, biomass, chemical composition, productivity and other metabolic functions. This presents an opportunity to develop innovative food products incorporating paludi-microalgae, as well as other products - considering that many paludiculture plants are not currently cultivated for human consumption, but for other uses.

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AUTHOR CONTRIBUTIONS

SKU and AJ are joint first authors. SKU led the study, developed the study design, performed the experimental work and data analysis, and wrote the manuscript. AJ contributed significantly to improvement of the manuscript. CA and FA undertook data collection and analysis. All authors contributed to the study design, to the writing and proofreading of the manuscript, and approved the final version.

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