

ORIGINAL PAPER

The Accumulation of Microplastics by Zooplankton in Chachoengsao and Samut Songkhram Provinces, the Gulf of Thailand

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Abstract. Microplastic (MP) pollution is increasingly recognized as a pervasive environmental issue with profound ecological and socioeconomic consequences. This study examines the occurrence, abundance, and physical characteristics of microplastics ingested by zooplankton in two key estuarine environments of the Gulf of Thailand: the Bang Pakong and Mae Klong Rivers. Zooplankton samples were collected across both dry and wet seasons using a Manta net and analyzed through microscopy and Fourier Transform Infrared (FTIR) spectroscopy. Microplastics were identified in 27.4% of 420 individual zooplankton spanning eight dominant taxa. Fish larvae and *Lucifer* spp. exhibited the highest ingestion rates, indicating potential susceptibility due to feeding behavior or ecological niche. The mean abundance was 0.402 particles per individual, with fibers comprising the most common shape (88.2%), predominantly composed of cellulose (59.2%), polyester, and polypropylene. Blue and black fibers were the most frequently observed colors, suggesting sources such as synthetic textiles and fishing gear. While seasonal variation in microplastic abundance and size was not statistically significant, particle ingestion occurred consistently across sites and seasons, highlighting year-round contamination. Significant interspecific differences in microplastic size and abundance suggest the influence of taxa-specific feeding mechanisms and morphological traits. These findings highlight the risk posed by microplastics to lower trophic organisms and their potential for upward transfer through the food web. This study provides essential baseline data for microplastic contamination in Thailand's estuarine zones and reinforces the urgency for enhanced monitoring, pollution mitigation strategies, and targeted policy frameworks to address marine microplastic pollution.

Keywords: Estuarine ecosystem, Gulf of Thailand, Microplastics, Trophic transfer, Zooplankton

1. Introduction

Plastic pollution has emerged as one of the most pervasive environmental challenges of the 21st century, with microplastics (MPs)-defined as plastic particles less than 5 millimeters in diameter-being increasingly detected across marine, freshwater, and even atmospheric environments (Law & Thompson, 2014; GESAMP, 2015). Due to their small size, MPs are readily ingested by a wide range of aquatic organisms, particularly zooplankton, which occupy a critical position at the base of aquatic food webs (Turner, 2015; Niyomthai et al., 2018; Sutthacheep et al., 2018; Buathong et al., 2020). Zooplankton, comprising diverse groups such as copepods, cladocerans, euphausiids, and larvaceans, play a pivotal role in marine and freshwater ecosystems by facilitating the transfer of energy from primary producers to higher trophic levels, including commercially important fish species (Cole et al., 2013). Their small body sizes, non-selective feeding behaviors, and omnipresence in aquatic environments render them highly susceptible to ingesting MPs suspended in the water column (Setälä et al., 2014; Botterell et al., 2019; Sambolino et al., 2022; Rodríguez-Torres et al., 2024).

Microplastics in the marine environment originate from a variety of sources, broadly categorized into primary and secondary microplastics. Primary microplastics are intentionally manufactured at microscopic sizes for specific industrial or consumer applications, such as microbeads used

in personal care products, industrial abrasives, and pre-production plastic pellets known as nurdles (Fendall and Sewell, 2009; Andrade, 2011; Arat, 2024). In contrast, secondary microplastics result from the fragmentation and degradation of larger plastic debris through physical, chemical, and biological processes, entering marine systems via improper waste management, the shedding of synthetic textile fibers, tire wear, and the breakdown of fishing gear and marine coatings (Browne et al., 2011; Jambeck et al., 2015; Kole et al., 2017). These particles are transported into the ocean through riverine discharge, stormwater runoff, direct wastewater disposal, and atmospheric deposition, which can carry airborne microplastics over long distances (Lebreton et al., 2017; Allen et al., 2019).

Upon entering the marine environment, microplastics pose a multitude of threats to both individual organisms and ecosystem processes. Numerous laboratory and field studies have demonstrated that zooplankton readily ingest microplastics of varying sizes, shapes, and polymer types, sometimes mistaking them for prey (Cole et al., 2015; Desforges et al., 2015; Sun et al., 2017). Size is a critical factor influencing ingestion likelihood, with many zooplankton species targeting particles within the 2–200 micrometer range, corresponding to the typical size of their natural food items (Turner, 2015; Botterell et al., 2019). While zooplankton generally exhibit low feeding selectivity, some species may discriminate against non-nutritive particles based on texture or chemical cues; however, the formation of microbial biofilms on plastic surfaces reduces this ability, making microplastics more attractive to feeding organisms (Setälä et al., 2014; Kettner et al., 2017). The surface properties of microplastics, including roughness, hydrophobicity, and polymer composition, influence their interactions with zooplankton, with smooth, hydrophobic particles being more likely to adhere to or be ingested by aquatic organisms (Chae and An, 2017; Kooi et al., 2017). Furthermore, biofilm-coated microplastics can release chemical signals that mimic those emitted by natural prey, thereby

enhancing the risk of ingestion (Savoca et al., 2017).

The ingestion of microplastics by zooplankton can result in multiple adverse biological effects, including digestive tract blockage, reduced feeding efficiency, energy depletion, impaired reproduction, and increased mortality (Cole et al., 2015; Botterell et al., 2019; Espincho et al., 2024; Valdez-Cibrián et al., 2024). Such impacts at the individual level may scale up to affect population dynamics and disrupt energy flow through the food web, with trophic transfer of microplastics and their associated chemical contaminants posing additional risks to higher trophic levels, including commercially important fish and human consumers (Farrell and Nelson, 2013; Cedervall et al., 2012). Moreover, microplastic contamination may interfere with the biological carbon pump by altering the sinking behavior of zooplankton fecal pellets, potentially weakening carbon sequestration processes essential for mitigating climate change (Turner, 2015; Kvale et al., 2020). In benthic habitats, the accumulation of microplastics in sediments may degrade habitat quality, reduce biodiversity, and alter ecosystem functions (Van Cauwenberghe et al., 2015)

The socioeconomic impacts of microplastic pollution are substantial. Contamination of seafood by microplastics and associated pollutants threatens food safety and public health, jeopardizing fisheries and aquaculture industries (Barboza et al., 2018). Additionally, the aesthetic degradation of marine and coastal environments by plastic debris can negatively affect tourism-dependent economies. Despite growing recognition of these risks, many aspects of microplastic interactions with marine organisms, especially zooplankton, remain poorly understood.

Although the presence of microplastics in marine environments has been widely documented, detailed studies on their ingestion by zooplankton under environmentally realistic conditions remain limited. Much of the existing knowledge is based

on laboratory experiments that use high concentrations of microplastics not reflective of natural scenarios (Botterell et al., 2019). Furthermore, the roles of particle size, shape, polymer type, and surface biofilm in determining ingestion rates and biological responses have not been fully elucidated. The lack of consistent field-based data on microplastic contamination at the base of the marine food web hampers effective risk assessments and management interventions. Without a clearer understanding of how zooplankton interact with microplastics in different environmental contexts, it is difficult to predict the long-term ecological consequences, including impacts on biodiversity, ecosystem services, and human health. Addressing these knowledge gaps is critical for informing conservation strategies and supporting global efforts to mitigate plastic pollution.

Understanding the accumulation of microplastics within marine organisms, particularly zooplankton, is essential for assessing the broader ecological consequences of plastic pollution. Given the pivotal role of zooplankton in marine food webs and carbon cycling, and their vulnerability to microplastic ingestion, research into their interactions with microplastics is urgently needed. Investigating the occurrence, ingestion patterns, and ecological effects of microplastic accumulation in zooplankton will provide critical insights necessary for preserving marine biodiversity, maintaining ecosystem function, and ensuring the sustainability of marine resources in the face of ongoing environmental change. This study aimed to assess the abundance of microplastics in zooplankton in the Gulf of Thailand, analyze their spatial and temporal variability, and characterize the types and physical properties of microplastics present in zooplankton samples.

2. Materials and Methods

2.1 Study sites

The study sites were designated at the river mouths of the Bang Pakong River, Chachoengsao Province, located at the coordinates of latitude 13°45'54.08" N and longitude 101°36'37.22" E, and at the Mae Klong River, Samut Songkhram Province, located at the coordinates of latitude 13°35'32.10" N and longitude 100°01'8.90" E (Figure 1).

2.2 Data collection and analysis

Samples for microplastic detection in zooplankton were collected during both the wet and dry seasons using a sampling method adapted from the Japan guideline. A Manta net with a known mouth area of 17 × 83 cm was used for surface tows, and a flow meter was attached at the net opening. The net was towed along the water surface for 5 minutes. Collected samples were immediately preserved in 95% ethanol.

The preserved samples were then stained with 0.5% Rose Bengal solution mixed with 10% diluted formalin and incubated at room temperature (approximately 24°C) for 24 hours to allow the Rose Bengal dye to bind to the proteinaceous tissues of zooplankton. After staining, the samples were sorted and the zooplankton were taxonomically identified. The zooplankton samples were subsequently digested to remove organic matter using 20 milliliters of 30% diluted hydrogen peroxide (H₂O₂) and incubated at room temperature for 5 minutes.

Following chemical digestion, the samples were heated in a temperature-controlled water bath at 75°C for 30 minutes, and then left to settle undisturbed for 12–24 hours to allow for sedimentation. The settled samples were filtered through a 20-micrometer filter paper. The material retained on

the filter was examined under a microscope to classify the shape, morphology, color, and size of microplastics (particles smaller than 5 millimeters).

The samples were analyzed to quantify the microplastics following the NOAA Marine Debris Program (2015) protocol. The quantities and polymer types of microplastics were determined

using stereomicroscope and Fourier Transform Infrared (FT-IR) spectroscopy, respectively. The quantities and size of microplastics found in zooplankton samples were statistically analyzed using one-way ANOVA and t-test. These analyses were performed to test for significant differences in quantities and size of microplastics among the different study sites and seasons.

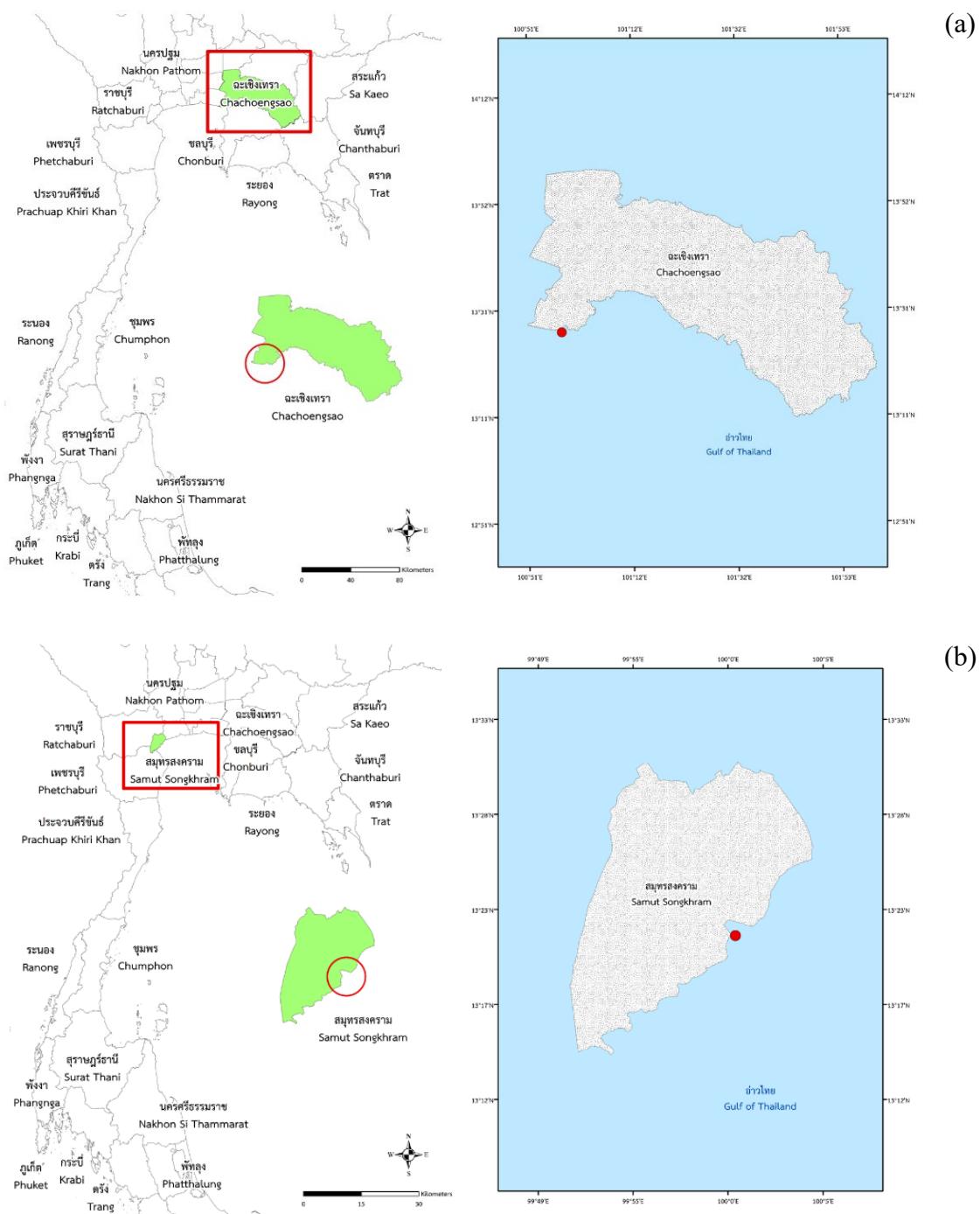


Figure 1. Map of the study sites: (a) the river mouth of the Bang Pakong River, Chachoengsao Province; (b) the river mouth of the Mae Klong River, Samut Songkhram Province.

3. Results

Eight dominant zooplankton groups were selected for microplastics analysis under a stereomicroscope, i.e., Calanoid copepod, Harpacticoid copepod,

Lucifer, Zoea, Shrimp, Jellyfish, Fish larvae and Chaetognatha. Microplastics were detected in all zooplankton taxa examined (Figure 2).



Shrimp



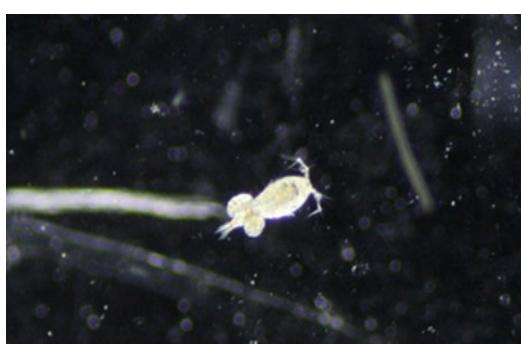
Fish Larva



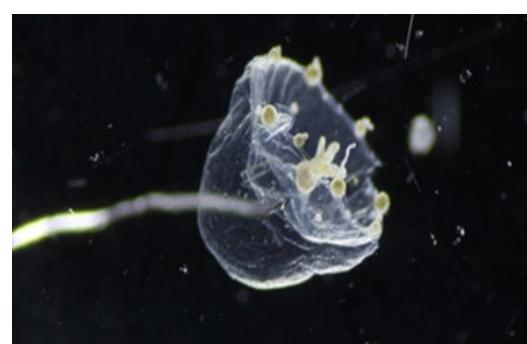
Zoea



Calanoid Copepod



Harpacticoid Copepod



Jellyfish Larva



Chaetognatha



Lucifer

Figure 2. Dominant zooplankton groups observed at the study sites

3.1 Occurrence of Microplastics in Zooplankton

Microplastics were detected in zooplankton samples from both study sites (Bang Pakong River and Mae Klong River) and across both seasons (dry and wet). Overall, microplastics were found in 27.4% of the total 420 zooplankton individuals examined. The detection rates were similar between study sites: 27.1% at Bang Pakong and 27.7% at Mae Klong. Seasonally, detection rates were slightly higher during the dry season

(27.7%) compared to the wet season (27.1%) (Figure 3).

By taxon, fish larvae exhibited the highest frequency of microplastic occurrence (38.0%), followed by Lucifer (35.5%), while Chaetognatha had the lowest occurrence (5.0%). No microplastics were detected in Chaetognatha during the dry season. These findings suggest that the likelihood of microplastic ingestion may be associated with the feeding strategies or ecological niches of different zooplankton taxa (Figure 4).

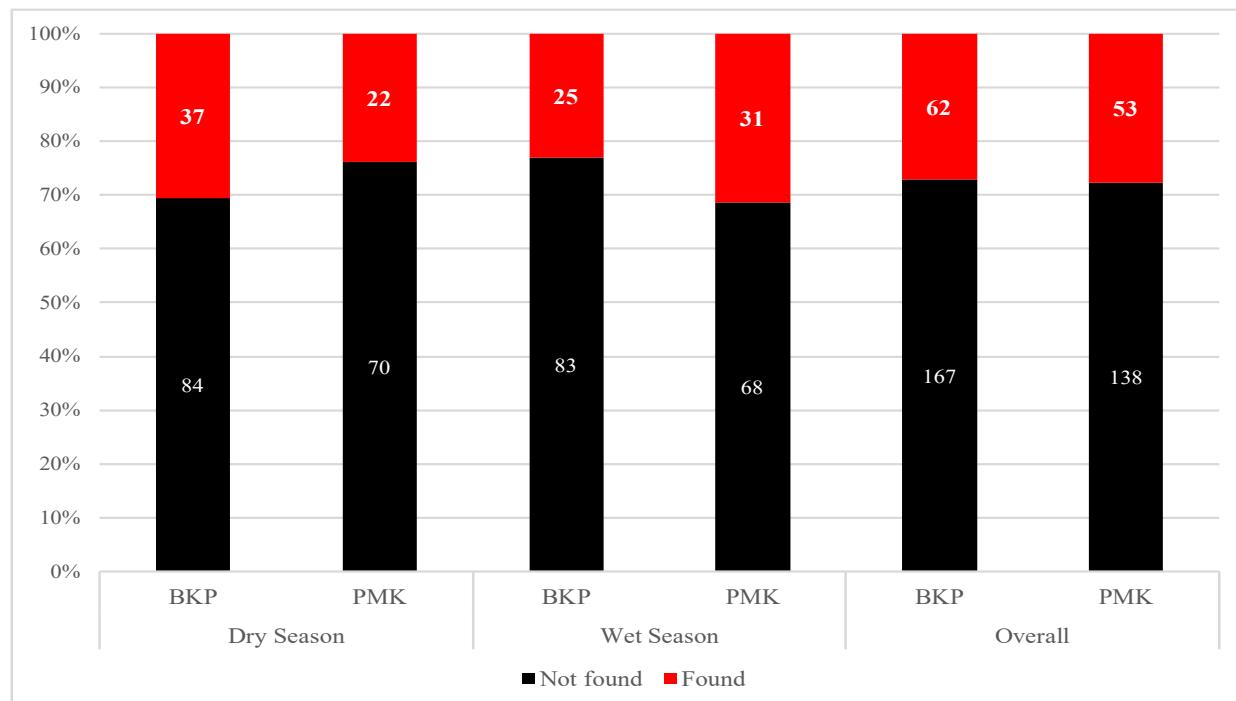


Figure 3. Occurrence of microplastics in zooplankton by seasons (BKP= Bang Pakong River; PMK= Mae Klong River)

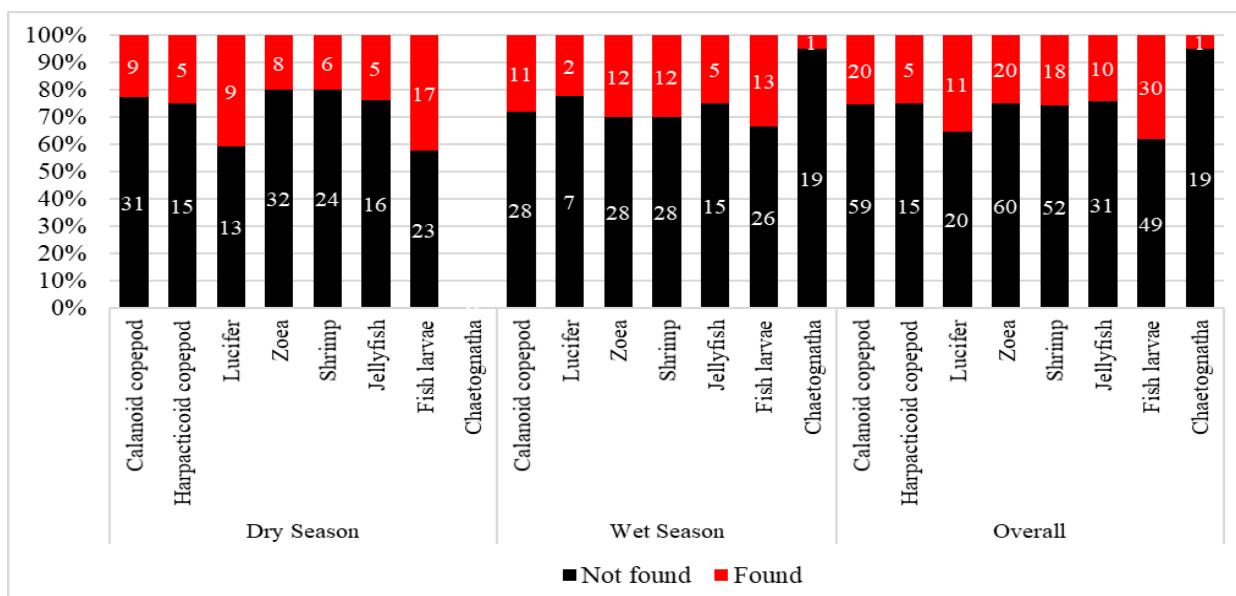


Figure 4. Occurrence of microplastics in zooplanktonic taxa and season

3.2 Quantity of Microplastics in Zooplankton

The quantity of microplastics, measured as particles per individual, ranged from 0 to 7 particles per individual, with an overall mean of 0.402 particles per individual. No significant differences in microplastic abundance were found between seasons at either study site ($p > 0.05$), as shown in Figure 5.

By taxa, fish larvae had the highest mean abundance (0.557 particles per individual), followed by calanoid copepods and Lucifer species. A one-

way ANOVA revealed significant differences in microplastic abundance among zooplankton taxa ($p = 0.028$), with post-hoc analysis (Fisher's LSD) indicating that Chaetognatha differed significantly from Lucifer and fish larvae ($p < 0.05$) (Figure 6). This suggests that certain taxa may accumulate microplastics more readily than others, likely due to differences in feeding behavior or microhabitat. Considering by each taxa, seasonal variation was found only in calanoid copepods (Figure 7).

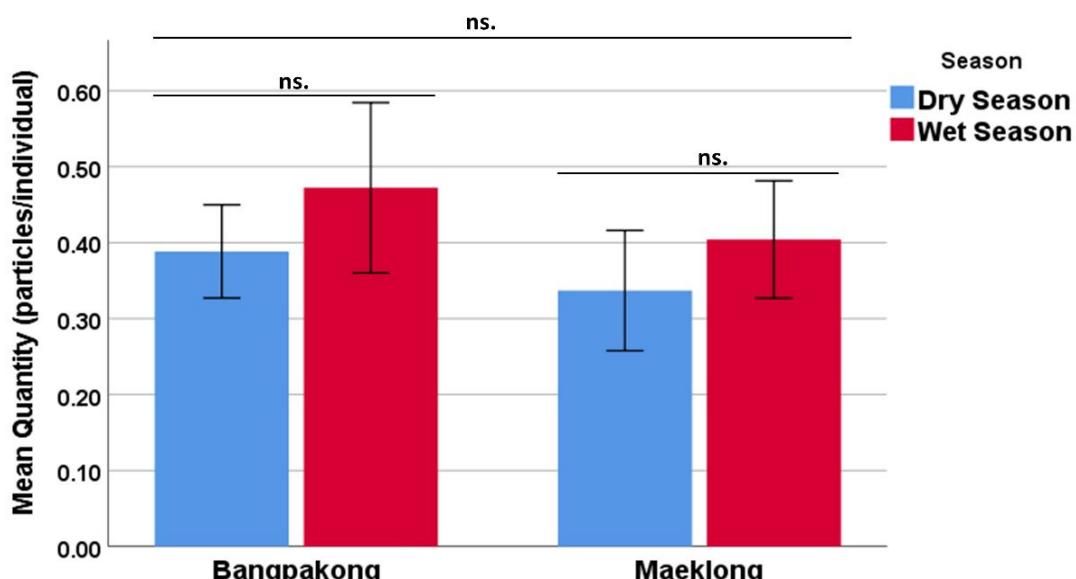


Figure 5. Differences in microplastic quantities between study sites and seasons (ns. denotes non-statistical difference)

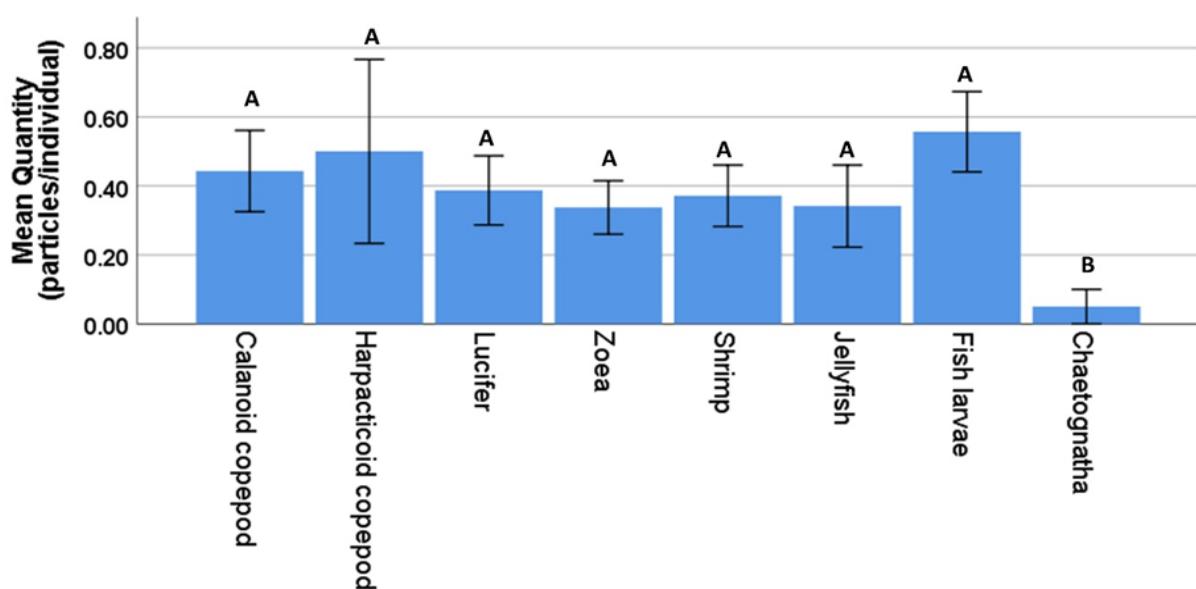


Figure 6. Variation of microplastic quantities between study sites. Means that share a letter are not statistically significant.

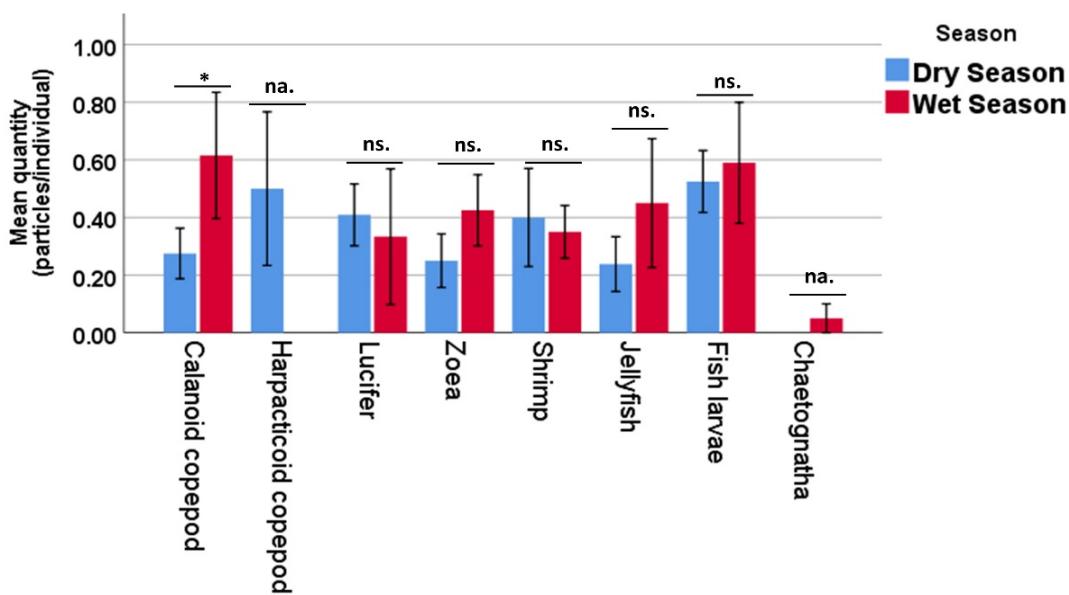


Figure 7. Difference in microplastic quantities between study sites and seasons (ns. denotes non-statistical difference)

3.3 Size of Microplastics in Zooplankton

The size of microplastic particles found in zooplankton ranged from 25.02 μm to 7304.09 μm , with a mean size of approximately 1,099 μm . No significant differences in particle size were observed between seasons at either study site ($p > 0.05$) (Figure 8). However, one-way ANOVA analysis revealed significant differences in microplastic particle sizes among zooplankton taxa ($p = 0.034$). Post-hoc test showed that

chaetognath, jellyfish, and calanoid copepods tended to contain significantly larger microplastic particles compared to other taxa such as harpacticoid copepod, lucifer, zoea, and fish larvae ($p < 0.05$) (Figure 9). This could reflect differences in ingestion capacity, particle selectivity, or habitat use. Seasonal variation can be only found in shrimp ($p < 0.05$), illustrating that the size of microplastics found in dry season is significantly higher than that in wet season (Figure 10).

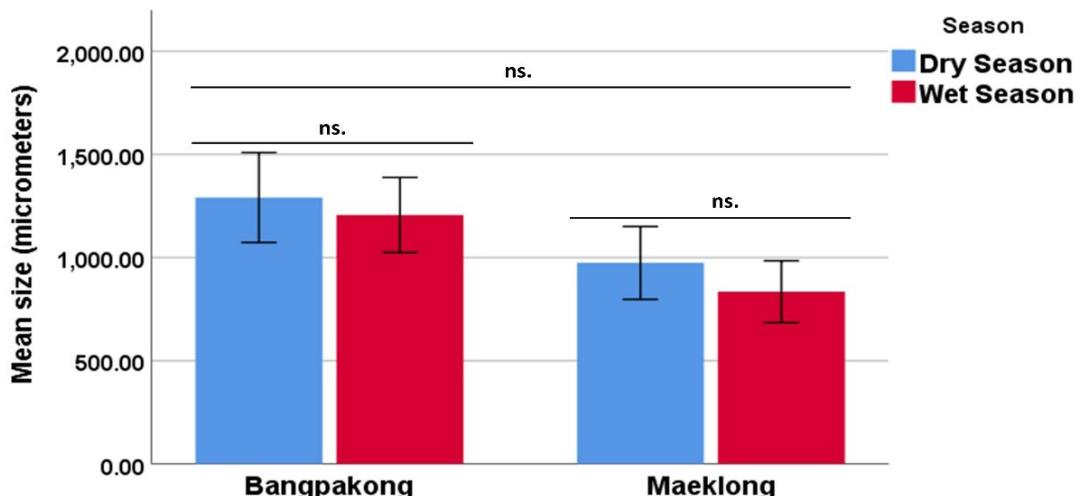


Figure 8. Differences in sizes of microplastics between study sites and seasons (ns. denotes non-statistical difference)

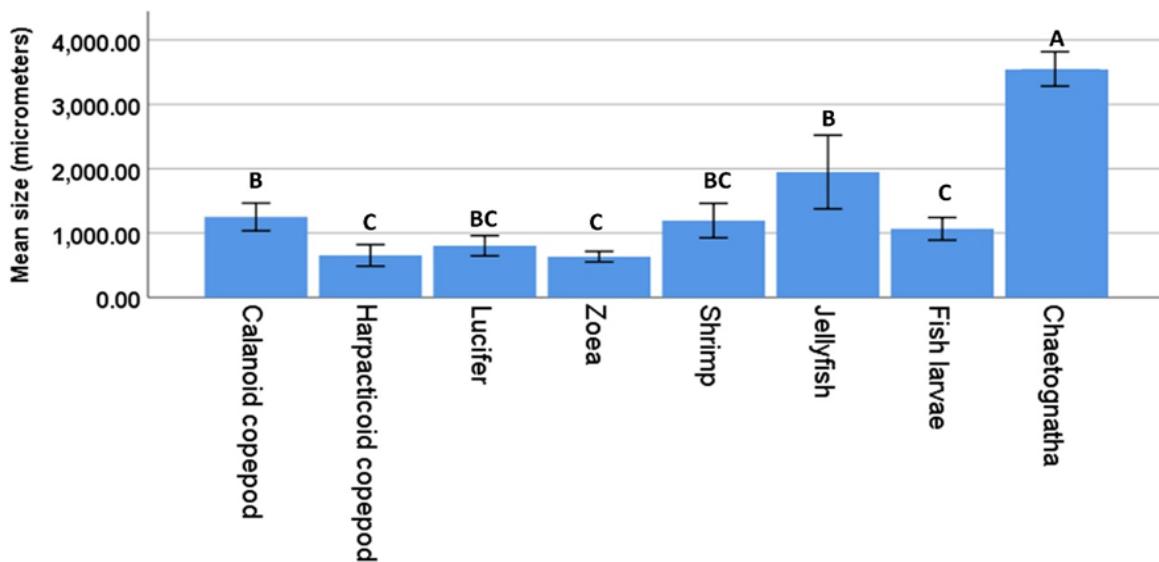


Figure 9. Variation of sizes of microplastics between study sites. Means that share a letter are not statistically significant.

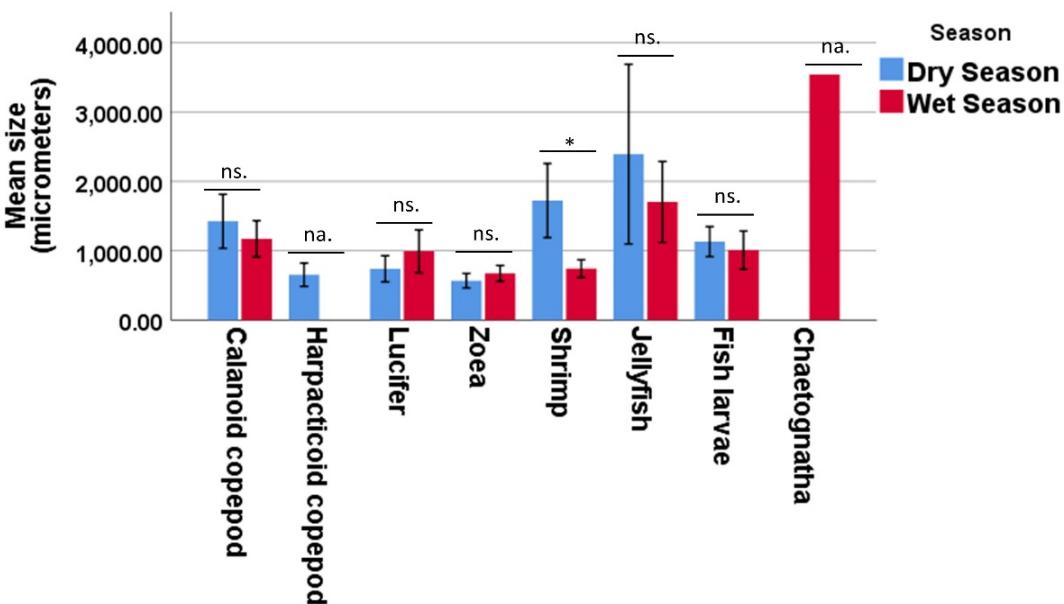


Figure 10. Difference in sizes of microplastics between study sites and seasons (ns. denotes non-statistical difference)

3.4 Shape, Color, and Polymer Type of Microplastics

Regarding shape, fibers were the dominant form of microplastics found (88.2% of particles), followed by films (8.3%) and fragments (3.6%). This predominance of fibers aligns with previous studies suggesting that synthetic fibers, primarily from textiles, are a major source of marine microplastic pollution (Figure 11).

The distribution of microplastic types (fragments, films, and fibers) across various marine species in the Gulf of Thailand during the dry and wet seasons, as well as overall, is showed in Figure 12. Calanoid copepods consistently show the highest microplastic counts, particularly in the dry season with 10 microplastics, and wet season with 20 microplastics. The dominant microplastic type across all species and seasons is fragments, with a minimal presence of fibers and films. Harpacticoid copepods also show

significant contamination, especially in the wet season (18). Other species, such as shrimp and Lucifer, show lower microplastic counts, but fragments still dominate. Overall, calanoid copepods and harpacticoid copepods have the most microplastic contamination, and the

primary microplastic type is fragments, while films and fibers are relatively rare. The wet season generally shows a higher level of microplastic contamination than the dry season, particularly for copepods (Figure 12).

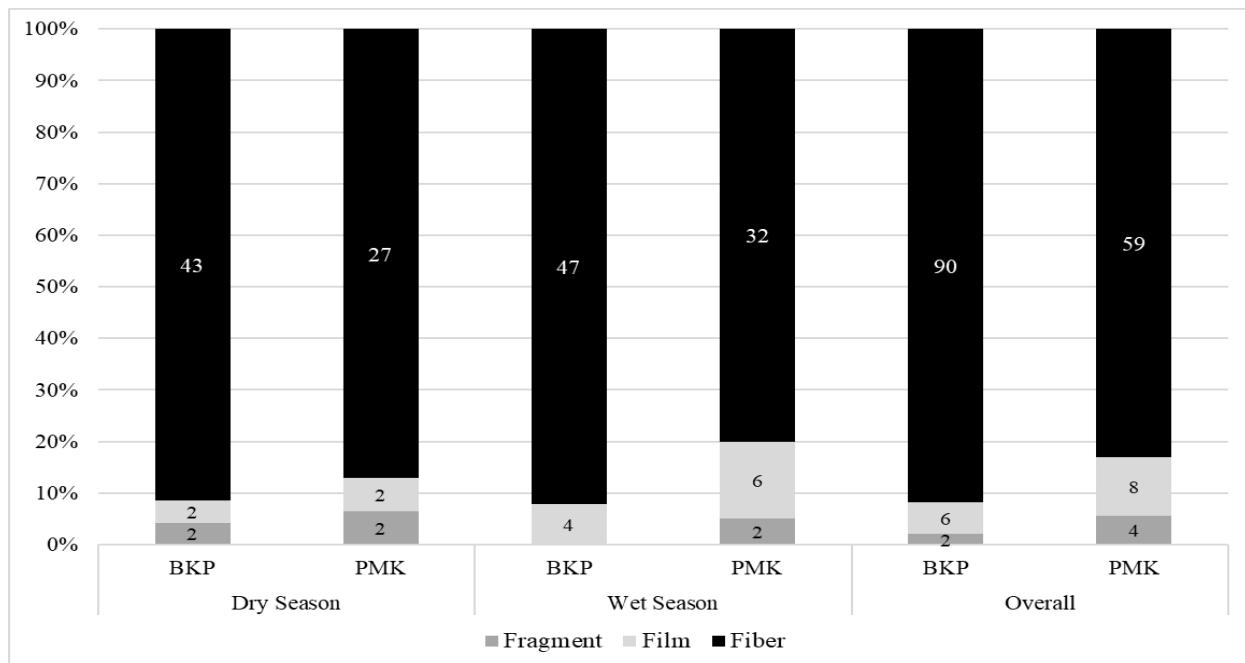


Figure 11. Proportion of characteristics of microplastics found at the study sites; Bang Pakong River (BKP) and Mae Klong River. (PMK)

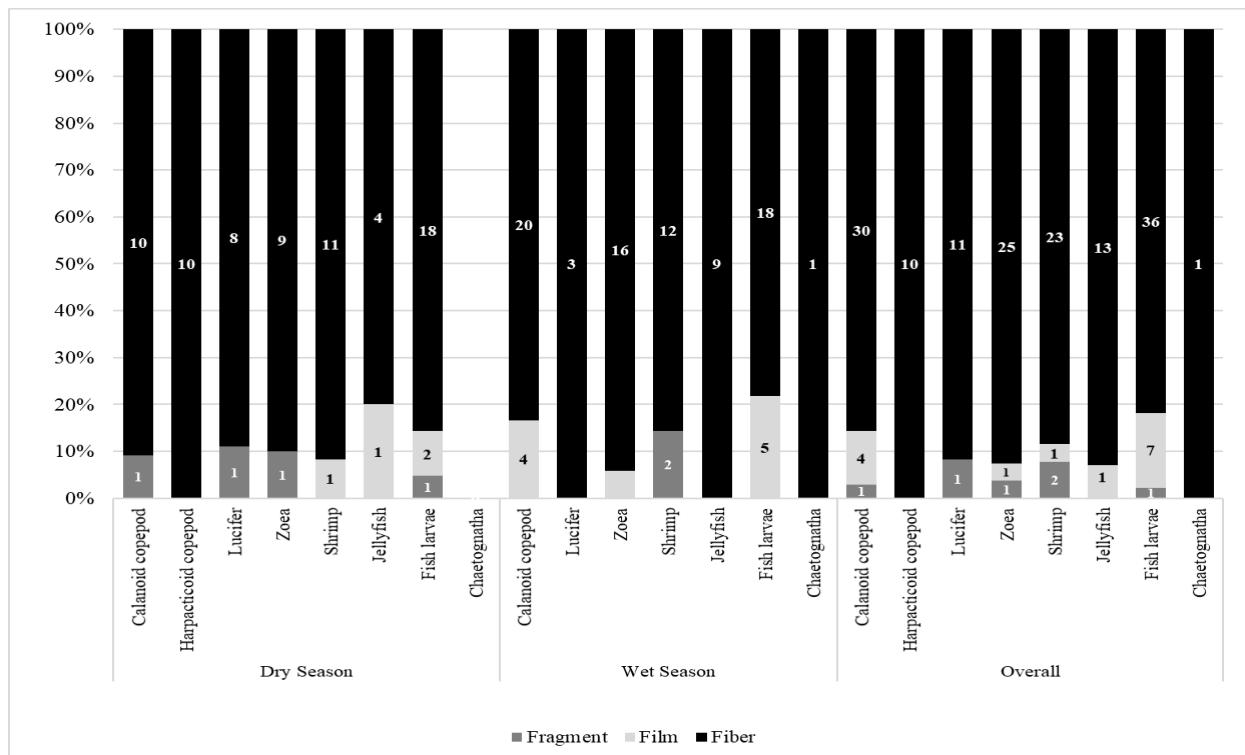


Figure 12. Proportion of characteristics of microplastics in zooplanktonic taxa

In terms of color, blue microplastics were most common (66.9%), followed by black (23.1%), red (7.1%), green (1.8%), and white (1.2%). The high occurrence of blue fibers may be related to widespread use of blue synthetic textiles and fishing gear materials (Figure 13).

The distribution of microplastics by color type (white, green, black, red, blue) across various marine species in the Gulf of Thailand during the dry and wet seasons, and overall is showed in Figure 14. Calanoid copepods consistently show the highest microplastic counts, particularly in the blue (dominantly blue microplastics) and red categories, especially during the wet season with 26 counts. The presence of red microplastics is

prominent in species like Chaetognatha, especially in the wet season (18), and shrimp (10). Lucifer and Zoea have minimal microplastic contamination, mostly blue with a few red instances. The overall trend shows that blue microplastics dominate most species, followed by red, with green microplastics being the least common. The data suggests that blue microplastics, which are the most dominant, are primarily found in copepods, while red microplastics show more variation across species, particularly in fish larvae and Chaetognatha. The wet season generally sees higher contamination, especially for calanoid copepods and Chaetognatha (Figure 14).

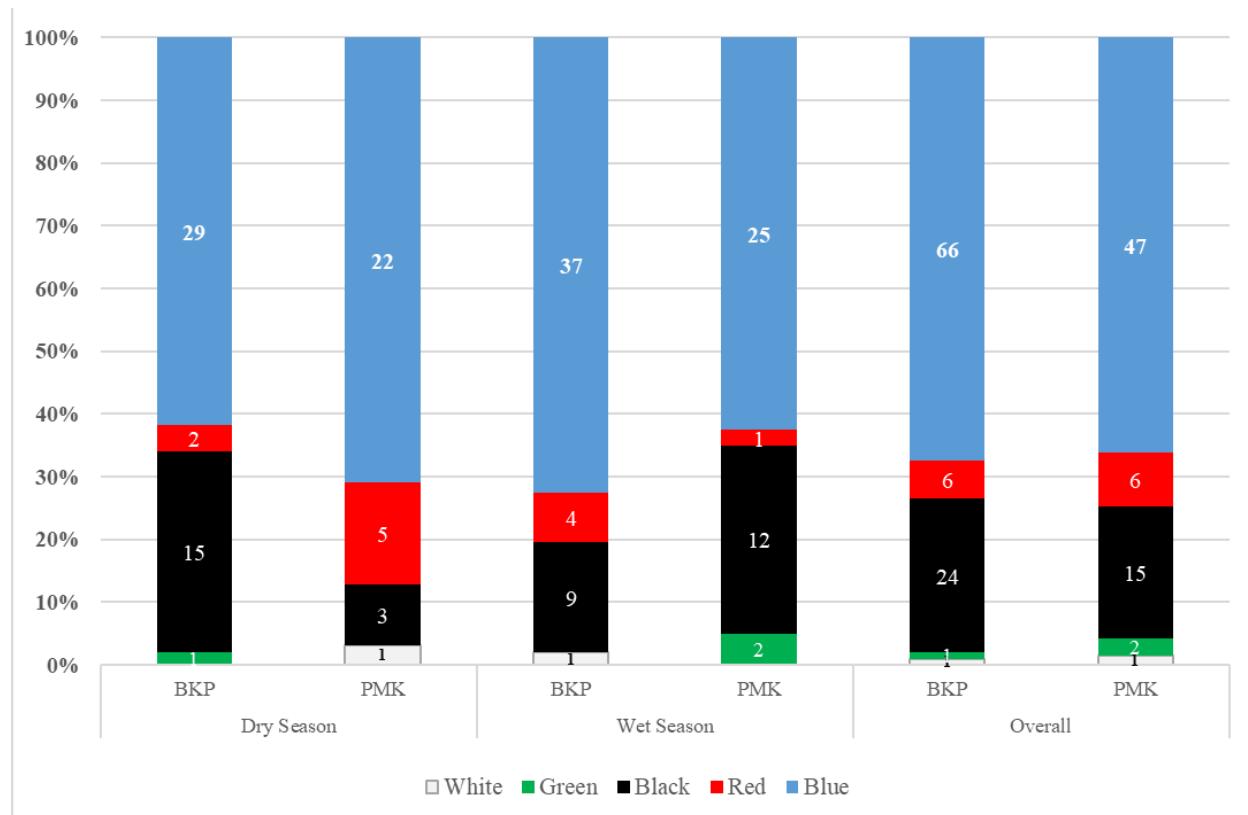


Figure 13. Proportion of colors composition of microplastics found at the study sites; Bang Pakong River (BKP) and Mae Klong River. (PMK)

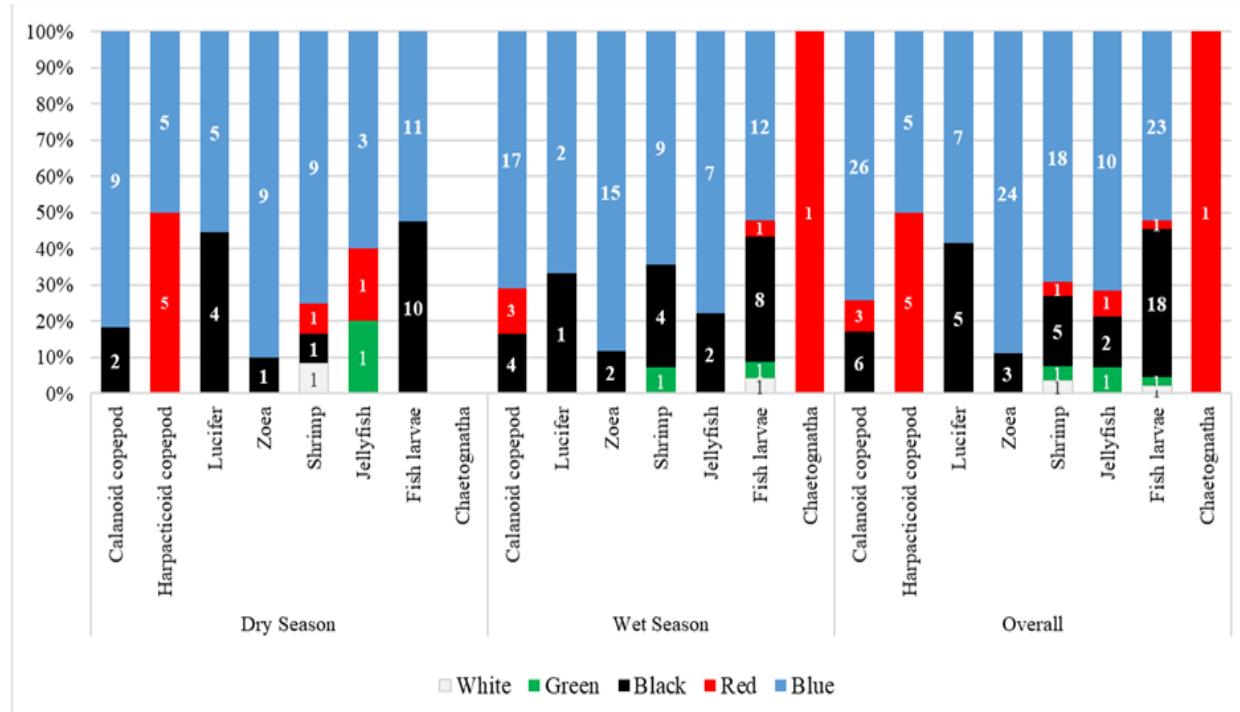


Figure 14. Proportion of colors composition of microplastics in zooplanktonic taxa

Polymer analysis indicated that cellulose (CEL) was the most frequently detected polymer (59.2%), followed by polyester (PES) at 21.3%, polypropylene (PP) at 10.7%, and polyethylene terephthalate (PET) at 7.1%. A minor proportion of nylon (NYL) and other polymers were also detected. The dominance of cellulose could be due to the prevalence of semi-synthetic fibers (such as rayon) derived from natural sources but categorized as microplastics (Figure 15).

The distribution of microplastic types (PET, PP, PS, CEL, NYL, and others) in various marine species from the Gulf of Thailand across the dry and wet seasons, as well as overall is showed in Figure 16. In both seasons, PET (blue) is the most dominant microplastic type, especially in calanoid copepods and

harpacticoid copepods, which show high contamination levels across all seasons. Polypropylene (PP) (orange) is also present but in lower proportions, while polystyrene (PS) (grey) and cellulose (CEL) (green) are less common. Zoa and shrimp show a relatively balanced distribution of microplastic types, with PET still being the most prevalent, but fewer PP and PS types. The overall trend indicates that PET dominates across species, with calanoid copepods having the highest microplastic contamination in both the dry and wet seasons. Additionally, other types are minimal, with very few occurrences in species like Lucifer and Chaetognatha. The data reflects a consistent presence of PET microplastics across species, seasons, and types (Figure 16).

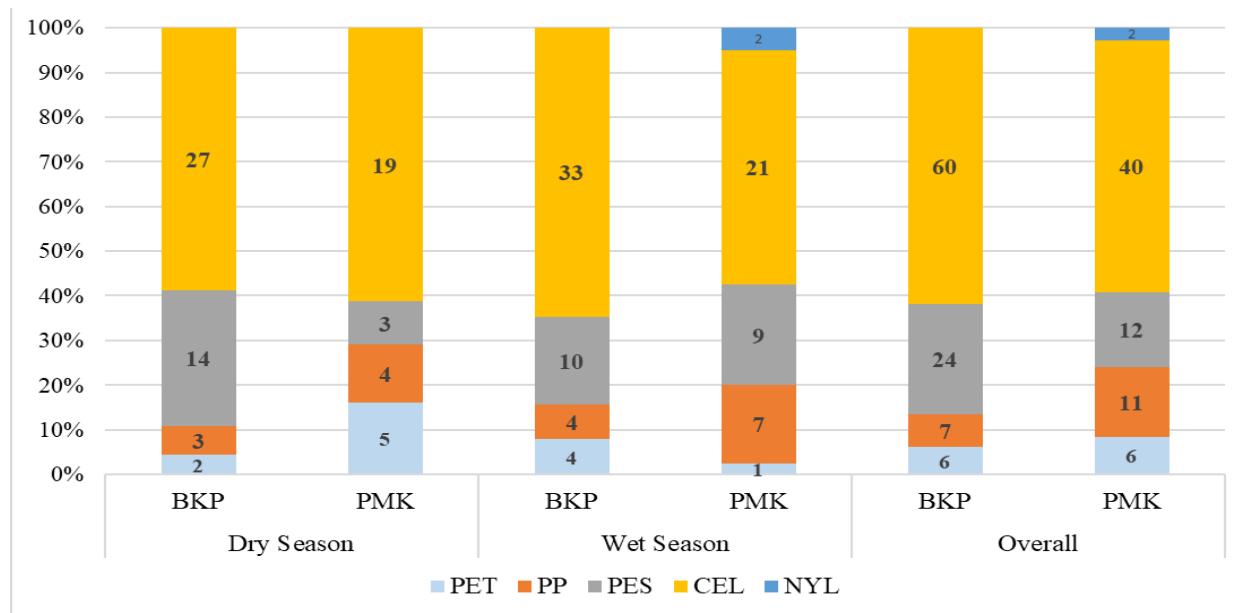


Figure 15. Proportion of polymer types found at the study sites; Bang Pakong River (BKP) and Mae Klong River. (PMK)

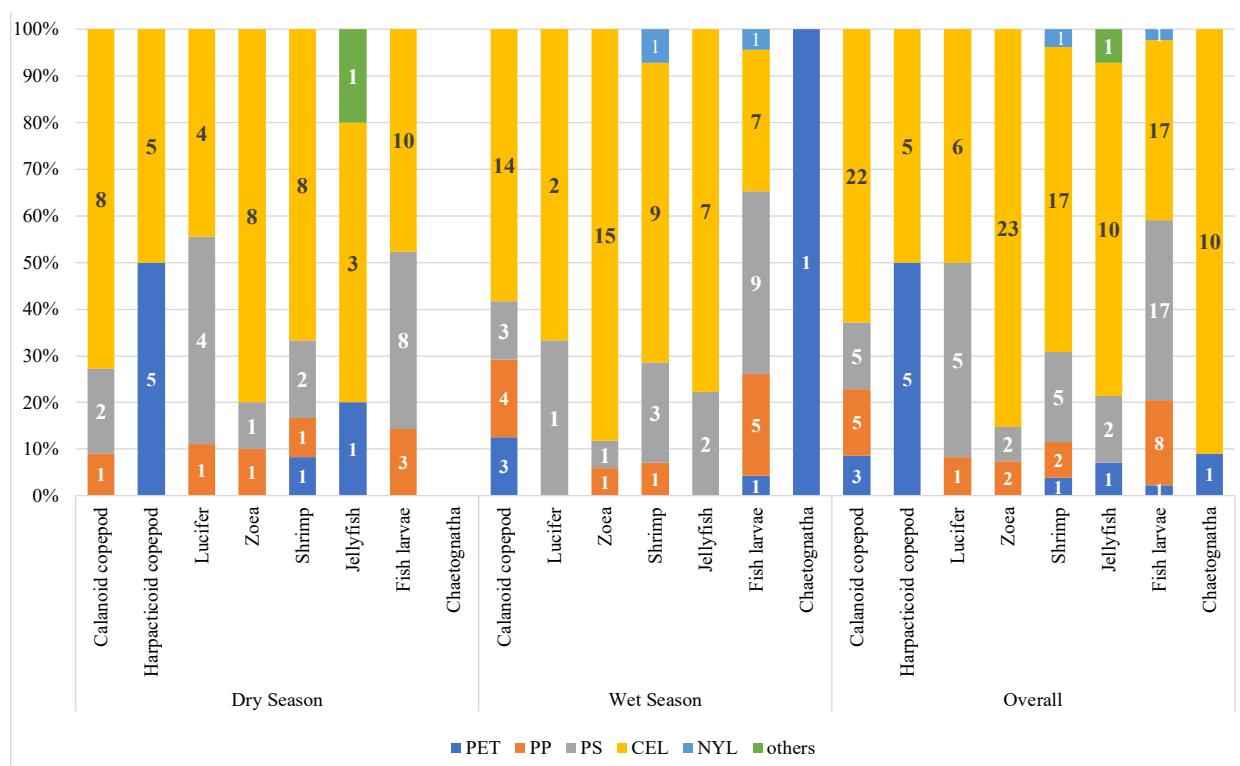


Figure 16. Proportion of polymer types in zooplanktonic taxa

4. Discussion

The size of microplastic particles ranged from 25.02 to 7,304.09 μm , with an average of $\sim 1,099 \mu\text{m}$. Differences in particle size among taxa were significant, with jellyfish containing significantly larger particles than copepods and fish larvae. This observation may reflect passive or low-selectivity feeding strategies and larger oral apparatus in jellyfish, enabling them to capture larger debris (Cole et al., 2013; Sambolino et al., 2022; Valdez-Cibrián et al., 2024). The detection of large-sized microplastics raises concern about potential gut blockage and reduced energy intake, as reported by previous laboratory studies (Cole et al., 2015; Sun et al., 2023; Arat, 2024).

In terms of shape, fibers overwhelmingly dominated (88.2%) the microplastics identified in zooplankton samples, followed by films and fragments. This fiber dominance is consistent with other marine studies and is typically attributed to synthetic textile shedding during laundry, which enters aquatic systems via wastewater treatment plants (Browne et al., 2011; Andrade, 2011; Arat, 2024; Espincho et al., 2024). The prevalence of blue and black fibers (66.9% and 23.1%, respectively) also mirrors global patterns, as these colors are commonly found in fishing nets, ropes, and synthetic fabrics (Lusher et al., 2017; Sun et al., 2023; Rodríguez-Torres et al., 2024).

Polymer identification by FTIR spectroscopy revealed that cellulose (CEL), polyester (PES), and polypropylene (PP) were the most common types, with CEL accounting for 59.2% of all particles. While cellulose is semi-natural, it is often classified as microplastic when processed into rayon and other regenerated fibers. The presence of such fibers underscores the complex nature of defining and monitoring "natural vs. synthetic" microplastic pollution (Hidalgo-Ruz et al., 2012; Arat, 2024; Espincho et al., 2024). The frequent detection of PES and PP further implicates household and fishing-related sources, as these polymers are widely used in textiles and packaging (Sun et al., 2023; Rodríguez-Torres et al., 2024).

Although no statistically significant seasonal differences were observed in microplastic abundance or size, the ecological implications remain concerning. Zooplankton are central to the marine food web and play a critical role in biogeochemical cycling, particularly through the biological carbon pump. As microplastics are incorporated into fecal pellets, they may alter sinking rates and reduce vertical carbon flux (Kvale et al., 2020; Sambolino et al., 2022; Rodríguez-Torres et al., 2024). Furthermore, the trophic transfer of microplastics from zooplankton to fish and other predators introduces risks of bioaccumulation and potential toxic effects in higher organisms, including those consumed by humans (Farrell & Nelson, 2013; Rochman et al., 2013; Botterell et al., 2019; Valdez-Cibrián et al., 2024).

This study provides strong evidence that microplastics are already embedded in the base of the estuarine food web in Thailand. The high incidence of ingestion by early life stages of fish and other zooplankton raises ecological concerns, particularly in regions where fisheries are economically and socially important. The results emphasize the urgent need for improved waste management, public awareness, and regulatory action to address the growing threat of microplastic pollution.

This study also reveals compelling evidence of microplastic contamination in zooplankton communities inhabiting two major estuarine systems in Thailand: the Bang Pakong River and the Mae Klong River. Microplastics were detected in over a quarter of all zooplankton individuals sampled, with a wide range of particle types, sizes, and polymer compositions observed. The consistent presence of microplastics across both dry and wet seasons, and in both study sites, suggests continuous and year-round pollution likely driven by anthropogenic activities such as wastewater discharge, urban runoff, and fishing operations (GESAMP, 2015; Jambeck et al., 2015; Arat, 2024; Espincho et al., 2024).

Taxon-specific differences in microplastic ingestion were observed, with fish larvae and Lucifer showing the highest accumulation rates. These results emphasize the vulnerability of early developmental stages and indiscriminate feeders

to microplastic exposure (Setälä et al., 2014; Botterell et al., 2019; Sun et al., 2023; Valdez-Cibrián et al., 2024). The predominance of fibers, especially those composed of cellulose, polyester, and polypropylene, underscores the significant contribution of domestic wastewater and fishing-related sources (Browne et al., 2011; Lusher et al., 2017; Arat, 2024; Rodríguez-Torres et al., 2024). The wide range of particle sizes, including ingestion of large microplastics by jellyfish, further indicates that particle size selectivity may vary by species and feeding strategy (Cole et al., 2015; Sambolino et al., 2022).

The ecological implications of these findings are profound. Zooplankton represent a foundational component of aquatic food webs and play a crucial role in biogeochemical processes such as the biological carbon pump. The ingestion and accumulation of microplastics by zooplankton not only threaten individual health and population dynamics but may also facilitate the upward transfer of microplastics and associated contaminants to higher trophic levels, including commercially important fish and eventually humans (Farrell & Nelson, 2013; Rochman et al., 2013; Rodríguez-Torres et al., 2024; Valdez-Cibrián et al., 2024).

A number of key recommendations are proposed:

1. Comprehensive tracking systems should be introduced in Thailand's estuarine and coastal zones to evaluate microplastic contamination across both living organisms and physical environmental elements. To achieve data consistency and enable comparison among studies, unified protocols should be applied. Recent research underscores the value of including polymer typology, geographic representation, and temporal variations for building effective monitoring strategies.

2. Efforts to curb the discharge of synthetic fibers from household and industrial sources should prioritize enhancements in filtration mechanisms within wastewater treatment facilities. Educational initiatives promoting responsible plastic usage and improved laundry habits can also aid in minimizing primary inputs of microplastics.

3. controls on disposable plastic products, cosmetic microbeads, and fishing gear management. Domestic policies should be consistent with broader international initiatives, such as the objectives laid out in the United Nations Sustainable Development Goal 14.

4. It is important to expand studies assessing the chronic impacts of microplastic consumption on zooplankton, including their biological functions, reproduction, and longevity. These investigations should also consider synergistic effects from co-occurring stressors like chemical pollutants and climate shifts. Experimental designs ought to reflect species-specific traits and natural environmental dynamics to better inform ecological risk assessments.

5. Research should concentrate on tracing the passage of microplastics through aquatic food webs to assess potential risks of bioaccumulation and biomagnification. This is especially critical in areas where marine organisms form a substantial part of human diets.

Acknowledgements

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