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Behavioral responses of captive-bred post-hatchling and juvenile sea turtles to different colors of single-use plastic film

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Abstract

Sea turtles face significant threats from plastic ingestion, yet their behavioral responses to plastics remain poorly understood. We observed the responses of eight 4-year-old juvenile and twenty-seven 10-week-old post-hatchling captive-bred hawksbill turtles (*Eretmochelys imbricata*) to single-use plastic films with different colors and toughness, types commonly found in the digestive tracts of wild turtle carcasses. Each turtle's biting or touching toward the introduced plastic item was recorded. Juveniles frequently bit or touched light-colored (transparent or white) packaging and plastic bags, black plastic bags, and yellow snack packaging, but ignored blue bottle labels. Their preference may reflect sensitivity to light colors (e.g., white), and to softer materials (i.e., plastic bags over packaging). These findings align with previous studies on sea turtle responses to undyed or dyed jellyfish (*Rhopilema esculentum*), and plastics in the digestive tracts of carcasses. However, post-hatchlings showed no significant differences in responses, likely due to limited prey recognition at an early life stage. This study represents the first assessment of sea turtle behavioral responses to single-use plastics as visual cues. While post-hatchlings may ingest plastic randomly, older turtles are vulnerable to light-colored, soft plastics, putting sea turtles at risk of plastic ingestion throughout all life stages. Action is needed for targeted management of high-risk debris.

Keywords: Behavioral experiment; Hawksbill turtle; Marine debris; Plastic ingestion; Visual cue

1. Introduction

Plastic production has surged globally, reaching 400 million tons in 2022, with widespread applications in packaging, construction, and electronics[1]. However, plastic waste enters the ocean, being prevalent in coastal areas and mid-water areas, and on seabeds[2,3]. Marine organisms, including sea turtles, are affected by plastic ingestion, entanglement, and exposure to plastic-associated chemicals[4]. Notably, sea turtles have shown elevated plastic ingestion levels compared to other marine fauna[5]. Plastic film products such as bags and food packaging are commonly found in various sea turtle species[6]. These common single-use plastics are characterized by a short usage cycle and high usage amount[7]. A recent review revealed that a large proportion of light-colored plastics were found in sea turtles globally[8]. Clear-colored plastics were one of the preferred colors ingested by sea turtles compared to beach debris[9], such that the consistent presence of light-colored plastic film could pose an ongoing threat to them.

Researchers have examined whether sea turtles ingest plastic because of its vast abundance in the environment or whether they selectively target it for consumption. For example, a study on plastic in the gastrointestinal tract of sea turtles indicated a preference for soft plastics, which was attributed to its resemblance to jellyfish[10]. This preference may stem from visual or olfactory attraction. Visual cues are suggested to be crucial for sea turtles in detecting prey[11]. A study using animal-borne video cameras demonstrated that sea turtles exhibit similar movement patterns toward artificial debris and gelatinous prey[12]. Additionally, sea turtles can detect plastic through chemical cues; they have been observed to respond to airborne odorants released from biofouled plastic in ways that resemble their responses to food odors in controlled experiments[13].

Despite the known threat of plastic ingestion to sea turtles, few behavioral studies have specifically investigated sea turtle interactions with plastic debris. To date, only two experimental behavioral studies have addressed this issue[13,14]. Studying wild sea turtle behavior is ideal for understanding their interaction with plastic debris, but it presents significant practical challenges. A primary obstacle is their life cycle, which involves extended periods in the open ocean where they are difficult to access. Access to sea turtles for studies of their behavior is often limited to early post-hatchling stages. Therefore, we know of only one experimental study[13] of sea turtle behavior specific to plastic stimuli. This study was conducted on 5-month-old loggerhead turtles (*Caretta caretta*) that were collected from the beach after nest emergence and reared in captivity. Another challenge is the use of rescued turtles, which are often limited in number and may have health issues or low biological activity, which could bias behavioral results[15].

Given these challenges, captive-bred turtles provide a valuable alternative. While their tank environment differs from the wild, their consistent availability and known foraging histories allow for controlled experiments. Furthermore, these turtles can be tracked via satellite after release, providing a link to their wild behavior[16]. Our previous study[14] used captive-born juveniles and long-term captive juveniles and adults to examine their selectivity for light-colored, using dried dyed jellyfish (*Rhopilema esculentum*) as a proxy. The results indicated diet-related preferences, particularly for colors resembling aquarium feed. Building upon these findings, we conducted a follow-up experiment using commonly encountered plastic debris to investigate whether the color of plastic affects sea turtle foraging behavior, thereby providing insights into the sensory mechanisms driving plastic ingestion.

In the present study, we exposed aquarium-bred hawksbill turtles (*Eretmochelys imbricata*) to different colors of plastic films that are commonly found in coastal waters and the digestive tracts of sea turtle carcasses[6,17]. These turtles were encountering these plastics for the first time in their lives. We explored whether behavioral responses to these plastic stimuli vary with age-related developmental differences by using hawksbill turtles at distinct early life stages: 4-

year-old juveniles and 10-week-old post-hatchlings. The post-hatchlings' young age is comparable to the turtles used in the previous study[13]. Additionally, we measured the color difference between the plastic stimuli and the turtles' aquarium food, such as small fish, to explore the relationship between food experience and their response to plastic. To our knowledge, this is the first study to investigate sea turtle behavioral responses to commonly used plastic materials (bags and packaging) as visual stimuli. Using recorded videos of sea turtle behavior, we analyzed frequency, sequence and response rate (%) of biting and beak touching in their interactions with plastic stimuli.

2. Materials and Methods

2.1 Study animals and housing environment

Eight 4-year-old (juvenile) hawksbill turtles and 27 10-week-old (post-hatchling) hawksbill turtles were used in this experiment (Supplementary Table S1). This species is classified as critically endangered on the International Union for Conservation of Nature (IUCN) red list[18]. All juveniles and post-hatchlings were born at the Aqua Planet aquarium (Yeosu, South Korea) through artificial breeding between October and December 2019 and between August and September 2023, respectively. All turtles were in normal health, as confirmed through periodic checks by aquarists and veterinarians. The 4-year-old juvenile hawksbill turtles used in this study had previously been used in a dyed-jellyfish experiment at three years of age[14].

Each juvenile hawksbill turtle was housed in a rectangular tank measuring 250 cm × 100 cm × 90 cm, and were fed fish (*Trachurus japonicus*) and squid (*Todarodes pacificus*) 5 days per week. The turtles were offered jellyfish (*Aurelia aurita*) on a few occasions approximately 1 year before the trials, not as a primary diet item. This was done to expose them to a naturally occurring prey type[19] and to provide a relevant context for their subsequent responses to jellyfish-like plastic stimuli. Each post-hatchling turtle was kept in a circular water tank (diameter, 30 cm; height, 30 cm) and fed vitamin and mineral brown tablets (Mazuri, 5B48) each morning and a mixture of fish and Antarctic krill (*Euphausia superba*) each afternoon.

2.2 Stimuli and behavior measurements

All behavioral experiments were conducted at the aquarium from May 10 to June 2, 2023. The experimental tanks were 125 cm × 100 cm × 90 cm for juveniles and 60 cm × 45 cm × 35 cm for post-hatchlings. Because plastic film products are frequently ingested by sea turtles[6], we used six types of plastic film as stimuli; these varied in color to compare sea turtle behavioral responses to items commonly used in commercial applications. The stimuli comprised plastic films in six colors—transparent food packaging (polyethylene, PE), blue bottle labels (polypropylene, PP), red ramen packaging (PP), yellow snack packaging (PP), white plastic bag (PE), and black plastic bag (PE) (Fig. 1). To ensure that the stimuli did not differ in olfactory cues, they were washed inside and out with detergent to remove any residual odors from the packaging contents, particularly for snack or ramen packaging. The stimuli were composed of two layers, which made them difficult to tear. To ensure submersion, weights (~25 g) were added between the layers of the plastic stimuli for experiments with juvenile hawksbill turtles, and the stimuli were hung on a stainless-steel bar using nylon lines.

Juvenile hawksbill turtles were starved overnight before each trial, in which they were exposed to six stimuli simultaneously, each measuring approximately 10 cm × 10 cm (Figs. 1, 2, and Supplementary Fig. S1). The stimuli were placed approximately 5 cm apart, and the trials were repeated three times at 1-week intervals using the same juveniles. Stimulus placement was randomized for each trial. Stimuli were presented simultaneously to assess preferences based on response frequency and order. Post-hatchlings were not fed for less than

12 h prior to each trial, in which they were exposed to one stimulus measuring approximately 2 cm × 2 cm (Figs. 1, 2, and Supplementary Fig. S1). As the post-hatchlings showed signs of agitation and avoidance when exposed to multiple stimuli at once, only one stimulus was presented per trial, and preferences were assessed solely on response frequency. These trials were conducted once per post-hatchling and were not repeated. Additionally, red ramen packaging was excluded from the post-hatchling trials based on the aquarist's observation that the turtles had been fed from a red basket since their initial feeding and consistently showed heightened activity upon seeing the basket, which could have biased their behavior toward the red stimulus. No control group was included in this study because the aim was to compare behavioral responses of turtles to different colors of plastic stimuli within the same individuals.

When the turtles were looking in a different direction from that of the stimulus apparatus, the stainless-steel bar holding the stimuli was placed on the tank. The trial commenced when the turtle glanced at a stimulus; the trial period was 3 min for juveniles and 2 min for post-hatchlings. The entire trial was recorded using an action camera (HERO9 Black; GoPro, San Mateo, CA, USA). An investigator, who was blinded to the experimental conditions and purpose, measured the turtles' behavioral responses. Because biting and beak touching were interpreted as attention or an attempt to ingest the stimulus, their frequencies were quantified for each stimulus by reviewing recorded videos. Beak touches were recorded because, in hawksbill turtles, this behavior often occurs either as a failed attempt to bite or immediately before biting. The investigator also analyzed the response order to the plastic stimuli. To prevent ingestion, the nylon line holding each stimulus was briefly raised only when turtles engaged in active biting or sustained pulling. Following this adjustment, turtles occasionally moved backward or swam a short loop before resuming normal exploration. This safety measure prevented ingestion while minimally affecting the turtles' behavior.

2.3 Measurement of plastic stimulus characteristics

We measured the color difference (ΔE) between each plastic stimulus and sea turtle prey (squid and fish) provided at the aquarium and the tank interior for juvenile hawksbill turtles (Table 1), and between the plastic stimulus and prey (tablets and fish/krill mixture) and the tank interior for post-hatchling turtles (Supplementary Table S2), using a colorimeter (CR-400, D65 illuminant; Konica-Minolta Sensing Inc., Tokyo, Japan). Because white plastic bags and transparent food packaging are clear and the plastics were placed in the center of the water column, color differences were measured on the tank's outer rim, as the tank's background color remained visible, ensuring consistency. Both plastic and food had uneven coloration; therefore, these measurements were taken at three randomly selected points. For squid and fish, which have dark and light sides, each side was measured separately, and the average value was used as the representative color of the food.

2.4 Statistical analysis

All statistical analyses were performed using R version 4.4.2 (R Core Team, Vienna, Austria). A Kruskal–Wallis rank sum test followed by pairwise comparisons with Holm correction was used to assess the significance of differences in counts and response orders between stimuli. To assess whether responses were influenced by spatial location, we applied a permutation-based independence test to compare bite and touch order across the six stimulus positions.

2.5 Ethical statement

All experiments were reviewed and approved by the Animal Welfare Ethical Committee and the Animal Experimental Ethics Committee of the Korea Institute of Ocean Science and Technology (protocol no. 2023-01), in accordance with relevant national guidelines and regulations in Korea. Under Korean regulations, experiments involving animals, including species classified as Critically Endangered on the IUCN Red List, must be reviewed and approved by an institutional animal ethics committee prior to commencement. All methods were carried out in accordance with relevant guidelines and regulations, and the study is reported in accordance with the ARRIVE guidelines[20]. No anesthetic agents or euthanasia procedures were performed during or at the end of the study. After the experiments, all turtles were returned to their rearing tanks at the Aqua Planet aquarium.

3. Results

3.1 Juvenile hawksbill sea turtles

Among 24 trials (three repeated measures per turtle for eight individuals), juvenile hawksbill turtles bit or touched on transparent food packaging in 50% (n = 12) of trials, followed by yellow snack packaging (42%, n = 10), black plastic bags (38%, n = 9), white plastic bags (38%, n = 9), and red snack packaging (13%, n = 3); no juveniles bit or touched blue bottle labels (Fig. 3a). We assigned scores (1–6) to the plastic stimuli based on the order of bite or touch interactions, with the first stimulus interacted with receiving a score of 6, and the last a score of 1. These scores were averaged across trials for each individual to obtain a mean preference ranking for each stimulus in Fig. 3b. The ranking of plastic stimuli based on these scores mirrored the frequency (%) of biting or touching among trials, in the descending order transparent > yellow > black = white > red > blue (Fig. 3b). No differences were found in the ranking among the six stimulus locations (independence test, $p = 0.054$), indicating that turtles showed no tendency to select closer stimuli first. Response counts were highest for white plastic films (1.3 ± 0.49), followed by transparent (1.3 ± 0.34), yellow (0.88 ± 0.3), black (0.83 ± 0.29), red (0.17 ± 0.098), and blue (0) (Fig. 3c). Blue stimuli elicited significantly fewer responses, both in count and rank, than some other stimuli (Kruskal–Wallis test with holm correction; count: blue–transparent, $p = 0.002$; blue–white, $p = 0.04$; blue–yellow, $p = 0.04$; rank: blue–transparent, $p = 0.004$; blue–yellow, $p = 0.03$). Overall, the hawksbill turtles showed the highest response to light-colored (transparent and white) and black plastics, and the lowest response for blue plastic.

3.2 Post-hatchling hawksbill sea turtles

Among 27 trials (i.e., one trial per each of 27 individuals), post-hatchling hawksbill turtles most frequently bit or touched black plastic bags (96%, n = 26), followed by blue bottle labels (78%, n = 21), white plastic bags (70%, n = 19), yellow snack packaging (63%, n = 17), and transparent food packaging (63%, n = 17) (Fig. 4a). The count of responses did not differ significantly among stimuli, including blue (5.7 ± 1.3), black (5.3 ± 0.84), transparent (3.7 ± 0.83), yellow (3.6 ± 0.89), and white (3.4 ± 0.78) films (Fig. 4b).

4. Discussion

A global review of plastic ingestion across all sea turtle species found that white (34%) and transparent (31%) plastics were most frequently detected in their digestive tracts[8]. In particular, four species of sea turtles, including both juveniles and adults, from Korean waters primarily ingested transparent (50%) and white (26%) plastic films[6]. In this study, when juvenile hawksbill turtles were exposed to various plastic films, they most frequently bit transparent food packaging and white plastic bags, followed by yellow snack packaging and black plastic bags (Fig. 3). These results are consistent with a previous study that used dried jellyfish as stimuli and found color preferences in the order white (control) > yellow > black > red > blue, showing stronger attraction to prey-like colors and higher ingestion rates for soft dried jellyfish than for rigid jellyfish[14]. Jellyfish are a key component of the natural diet of some wild sea turtles, such as leatherback (*Dermochelys coriacea*) and loggerhead turtles[19]. Although adult hawksbill turtles are primarily spongivorous, juvenile hawksbill turtles in certain locations—including the Cayman Islands—have been observed consuming jellyfish occasionally[19]. Consistent with this, Australian hawksbill turtles may remain in the oceanic stage for up to five years, during which they feed mainly on macroplankton[19]. These ontogenetic shifts suggest that juvenile hawksbill turtles may retain sensory biases toward gelatinous prey, making it plausible that they visually respond to light-colored or translucent plastic films that resemble jellyfish. In captivity, the turtles in this study had previous exposure to jellyfish as part of their diet, further supporting that their responses to jellyfish-like plastic films represent ecologically relevant foraging responses to plastics that visually resemble natural prey. This interpretation is consistent with the hypothesis that sea turtles are inclined to interact with soft plastics resembling jellyfish in color, especially those that are clear or white[10]. Similarly, a previous study[21] investigated whether bait color influenced the behavioral responses of sea turtles in laboratory experiments, and they found that 2-year-old loggerhead and Kemp's ridley turtles (*Lepidochelys kempii*) preferred undyed squid over red- or blue-dyed squid.

Although yellow snack packaging had higher color contrast with prey compared to red ramen packaging or blue packaging, the sea turtles frequently bit or touched yellow packaging. This behavior could be explained by the high contrast with the tank, which may have made it more visually noticeable[14] (Table 1). Alternatively, sea turtles may be physiologically more sensitive to yellow, as they are well adapted to detecting light within the visible wavelength range (400–700 nm), particularly yellow (580 nm)[22]. A study[23] conducted an experiment on juvenile loggerhead turtles aged 6–25 months, in which the turtles were conditioned by prey rewards to bite enrichment items; the juveniles showed a higher biting frequency for yellow items and a lower frequency for blue items. In summary, the heightened response to white and transparent plastic films may reflect their resemblance to prey, whereas the increased response to yellow could stem from color-specific visual sensitivity and the contrast between the tank environment and the plastic. This pattern may reflect foraging behavior in wild juvenile turtles and may differ in wild adult turtles that primarily forage on sponges, which may therefore be more responsive to plastic colors similar to those of sponge prey.

However, in contrast to these patterns observed in juvenile turtles, hawksbill post-hatchlings did not show significant differences in biting or touching behaviors toward different colors of plastic film (Fig. 4). Response occurrence (%) was highest for black plastic bags, but no distinct response pattern was observed. Additionally, the turtles even attempted to bite bubbles created by their own movement or breathing, a behavior rarely seen in juvenile hawksbill turtles. This behavior suggests that younger hawksbill turtles tend to bite any object they see at random.

Few studies of sea turtle behavior toward plastics have included post-hatchlings of a similar age to those included in the present study (10 weeks). Most such research has focused on the sensitivity of post-hatchlings to light wavelengths either on land or in water. For example, detection thresholds of hawksbill post-hatchlings were measured and found to be highest at 555 nm (green), while sensitivity was lowest at 660 nm (red)[24]. In the case of loggerhead and flatback (*Natator depressus*) post-hatchlings have been observed to avoid red light on land and respond more frequently to shorter wavelengths such as green and blue[25]. However, direct comparisons with the present study are difficult, as the previous experiments were conducted on land using light stimuli, whereas the present study examined responses to physical objects in water. A study[26] found that prey selection in 6-month-old green (*Chelonia mydas*) post-hatchlings was influenced by tank color rather than the contrast between the tank and the prey-containing dish. In the gray tank, the turtles showed no dish color preference, whereas in yellow, red, and blue tanks, they favored prey in blue dishes. These findings suggest that green turtle post-hatchlings are more sensitive to short-wavelength light, such as blue. In contrast, the 10-week-old hawksbill post-hatchlings used in this study did not exhibit a clear preference pattern. Although they were slightly more responsive to black and blue plastic films, these differences were not statistically significant.

Thus, juvenile hawksbill turtles tended to respond more frequently to light-colored plastic films that displayed minimal color contrast with their food. In contrast, post-hatchlings indiscriminately responded to plastic films of all colors. Reptiles adapt their feeding behavior based on previous experiences with diverse types of food, forming food preferences through learning[27], and can be trained to select specific stimuli through food rewards[28]. Therefore, the 10-week-old post-hatchling hawksbill turtles in this study may not have had enough time to adapt to the food provided in the aquarium. Unlike the 4-year-old juvenile turtles, which may have developed feeding preferences, the younger turtles responded to any object they saw, regardless of the type of plastic. A comparable pattern has been observed in hatchling snakes (*Coleognathus helena*), which, being entirely naive to prey size or type, initially responded with minimal selectivity to novel prey and gradually refined their foraging behavior through experience[29]. This suggests that early-stage reptiles may exhibit broad, stimulus-driven responses before developing more selective feeding strategies. Another possibility is that post-hatchlings may be more responsive to other visual cues such as motion, or to olfactory cues, or a combination of both, rather than to color differences alone. Previous studies have shown that loggerhead post-hatchlings can detect both waterborne and airborne odors and exhibit increased responsiveness to food-related chemical cues[30,31]. The plastics used in this study were all washed with detergent and rinsed, ensuring that olfactory stimuli were the same. Further research is needed to confirm whether post-hatchlings rely more on non-color-based cues or non-visual stimuli, and whether such mechanisms also apply to juvenile turtles.

This study, while providing valuable insights, has a limitation using artificially bred individuals rather than wild sea turtles. Studies on the responses of wild sea turtles, rather than those artificially housed in tanks, to different colors have focused on reducing bycatch by modifying bait colors or investigating the role of colors in inducing biting behavior. However, no significant differences were found between undyed bait and red or blue bait[21], undyed bait and blue bait[32], or yellow, red, and blue fabric sacks[33] in loggerhead and Kemp's ridley turtles. These results differ from some tank-based experiments that reported color preferences. The lack of clear preferences in wild or rehabilitated individuals may reflect differences in prior foraging history[33] or life stage (e.g., unknown in wild turtles vs. known juveniles in captivity) or other environmental factors.

While captive-reared individuals may differ from wild sea turtles in terms of their prior experiences and rearing conditions, it is important to note that the plastic items used in this study are representative of debris commonly found in the digestive tracts of wild sea turtles. Therefore, the biting responses observed in our captive-bred juveniles are ecologically relevant, especially considering that these individuals were encountering plastic for the first time in a controlled environment. This finding gains further significance in the context of conservation, as captive-bred turtles are routinely released into the wild in Korea, and satellite tracking has shown that they display migratory patterns similar to those of wild populations[16,34], suggesting their behavior largely align with those of wild populations.

While rehabilitated individuals could also be utilized, they often require treatment and stabilization[15], reduced biological activity in some individuals, making it difficult to substitute their behavior for that of wild counterparts. On the other hand, wild individuals are relatively less available for experimental use. Given these challenges, and despite increasing concern about marine plastic pollution, behavioral studies on plastic interactions remain scarce across all types of sea turtle populations, including wild, rehabilitated, and captive-bred individuals.

Therefore, as it is widely recognized that combining behavioral data from multiple sources is beneficial[35], providing information from various species and age stages of rehabilitated and artificially bred individuals alongside wild animal data will contribute to a more comprehensive understanding of the impact on marine animals and better inform policy decisions.

Furthermore, some captive studies have demonstrated color-based preferences, and our results also suggest similar tendencies. Globally, sea turtles have been found to predominantly ingest white or transparent plastics[8]. Further research is needed to determine whether sea turtles actively select light-colored plastics or if these plastics are simply the most available in their environment. As most studies have focused solely on inshore debris, there is a lack of research examining plastic debris throughout the entire water column, where sea turtles are more likely to encounter it. This gap hinders our understanding of how sea turtles encounter plastic debris in the marine environment[6]. Research on plastic ingestion by post-hatchlings has also shown that light-colored (white or transparent) plastics are the most frequently ingested, accounting for more than half of all ingested debris[36-38]. These findings highlight the importance of investigating the characteristics of environmental plastic pollution to better understand its impact on sea turtles.

Plastic products also differ in properties such as flexibility and gloss, which are often deliberately modified. These factors should not be overlooked when studying turtle responses to plastics. Additionally, packaging materials such as red ramen packaging, yellow snack packaging, and blue bottle labels are not composed of a single uniform color but rather have a dominant color that is mixed with various other hues. This combination of colors may create different visual stimuli compared to single-color objects. Notably, red ramen packaging tends to include contrasting hues such as black, white, and yellow (Fig. 1). In this study, 4-year-old juvenile hawksbill turtles showed a lower response rate to red ramen packaging compared to the response to red-dyed jellyfish when they were three years old[14]. Our results indicate that color plays a particularly important role among these factors as a visual cue, regardless of whether it is presented as a single color or as the dominant color within a mixture.

To our knowledge, this is the first study to investigate sea turtle behavioral responses to actual plastic films, which are commonly discarded as single-use materials, as visual stimuli. Despite some limitations, our findings provide a valuable foundation for future studies on species-specific perceptions and interactions with plastic debris. We observed that sea turtles actively bit or touched plastic films; if ingestion had not been discouraged by hanging the stimuli on a nylon line above the tank or through human intervention, it might have resulted in plastic ingestion. These findings suggest that prolonged exposure to plastics resembling their prey may increase the likelihood of plastic ingestion in juvenile sea turtles. Based on the finding that juvenile turtles show a preference for lighter-colored plastics, we recommend reduced production of plastic films such as plastic bags, and that packaging should favor colors such as red or blue over lighter shades such as yellow, to minimize the risk of plastic ingestion by juvenile sea turtles. Furthermore, post-hatchlings, which tend to ingest objects indiscriminately, may be susceptible to consuming a wider variety of plastics, including microplastics. Microplastics can be divided into two categories: primary microplastics, which are intentionally produced at sizes smaller than 5 mm; and secondary microplastics, which are unintentionally generated from the breakdown of larger plastic items[39]. While primary microplastics are regulated in some nations, greater efforts are needed to control microplastic pollution globally[39]. As secondary microplastics originate from larger plastic debris such as plastic bags, their management should be prioritized accordingly.

From the post-hatchling stage to the juvenile stage, sea turtles may be at risk of falling into an evolutionary trap[36], in which their natural foraging instincts or prior feeding experiences lead them to mistakenly ingest plastic. As plastic debris continues to impact marine ecosystems, appropriate actions must be taken to manage plastic pollution and mitigate its ecological consequences.

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7. Author contributions

Y.M., H.-J.N., and S.H.H. contributed to conceptualization and methodology. Y.M., G.M.H., and E.V.C. conducted investigation. Y.M. performed formal analysis and visualization, and wrote the original draft. H.-J.N. and S.H.H. contributed to writing (review and editing). S.H.H. and W.J.S. contributed to project administration and supervision, and funding acquisition. E.V.C. provided resources. All authors reviewed the manuscript.

8. Data availability statement

The data from the findings of this study are provided herein and in the Supplementary Material files. Raw data files are available from the corresponding author upon reasonable request.

9. Competing interests

The authors declare no competing interests.

Figure 1. Images of intact plastic film products and plastic stimuli used in the experiments for juvenile and post-hatchling turtles. For each plastic type, the leftmost item is the original product, the middle item was used as a stimulus for juveniles, and the rightmost item was used as a stimulus for post-hatchlings. Each small yellow and black bar on the scale indicates 1 cm, and the large bar indicates 1 inch.

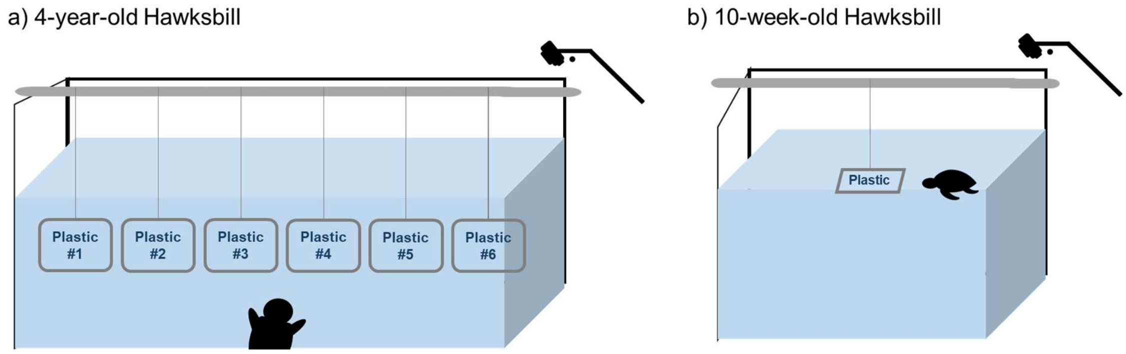

Figure 2. Simultaneous exposure to plastic stimuli among (a) 4-year-old (juvenile) and (b) 10-week-old (post-hatchling) hawksbill sea turtles.

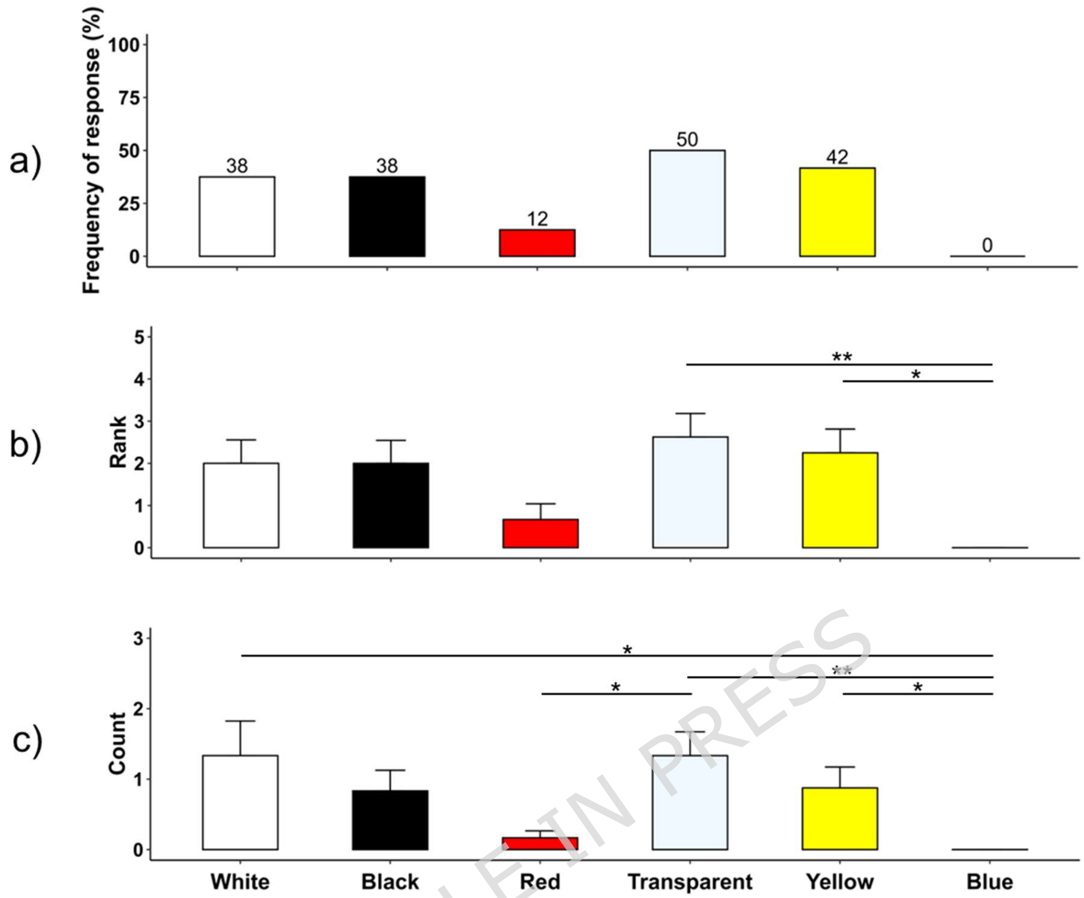

Figure 3. (a) Frequency of response (%), (b) rank, and (c) count of biting and touching behaviors by juvenile hawksbill turtles in response to simultaneous exposure to plastic stimuli (*$p < 0.05$, **$p < 0.01$, Kruskal–Wallis test with Holm correction). The error bars represent the standard error of the mean.

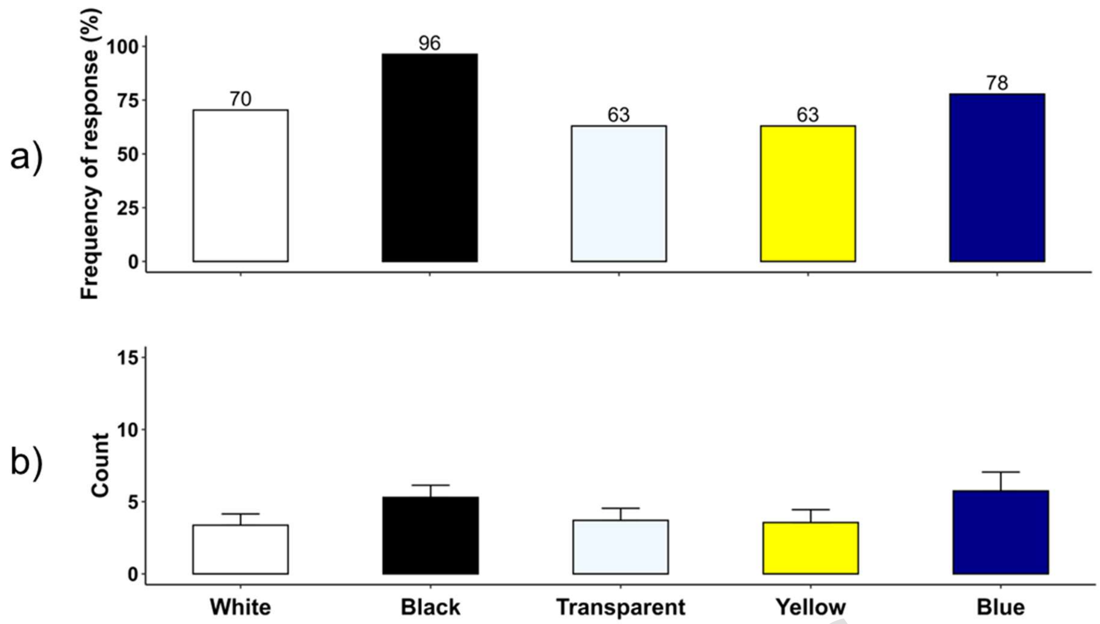

Figure 4. (a) Frequency of response (%) and (b) counts of biting and touching behaviors in post-hatchling hawksbill sea turtles in response to exposure to separate plastic stimuli. We detected no significant differences among stimuli ($p > 0.05$, Kruskal–Wallis test with Holm correction). The error bars represent the standard error of the mean.

Table 1. Color differences (△E) between plastic stimuli and the tank or prey of 4-year-old (juvenile) hawksbill turtles.

Stimuli	Tank	Fish	Squid
Black plastic bag	45.03	31.94	26.20
Blue bottle label	38.40	24.91	21.15
Red ramen packaging	84.75	47.83	39.99
Transparent food packaging	37.00	26.13	23.42
White plastic bag	61.39	24.32	30.82
Yellow snack packaging	114.2	65.75	50.68