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Effects of flow velocity on fitness-related behaviours of the sea urchin *Mesocentrotus nudus*: new information on stock enhancement

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Abstract

The effects of flow velocity on the fitness-related behaviours of *Mesocentrotus nudus* remain largely unknown, greatly hampering the efficiency of stock enhancement. To explore the appropriate velocities for stock enhancement, we investigated dislodgement and immobilization velocities up to 90 cm s⁻¹. The experimental results showed that *M. nudus* (test diameter of ~30 mm) were dislodged at 73.50 ± 7.7 cm s⁻¹ and that *M. nudus* movement occurred only when the flow velocity was less than 33.40 ± 2.7 cm s⁻¹. Three flow velocities less than 33.40 ± 2.7 cm s⁻¹ (2, 10 and 20 cm s⁻¹) were subsequently used to study the effects of flow velocities on covering behaviour and the righting response time of *M. nudus*. The downstream movement velocity of *M. nudus* was significantly larger than that upstream at 2 cm s⁻¹ (*P* = 0.016) and 10 cm s⁻¹ (*P* = 0.008), but not at 20 cm s⁻¹ (*P* = 0.222). The righting response time of *M. nudus* was significantly longer at 20 cm s⁻¹, preferably 2–10 cm s⁻¹, is probably appropriate for the stock enhancement of *M. nudus*. Notably, the current study is a laboratory investigation without considering the hydrographic complexity in the field. Further studies should be carried out to investigate the long-term effects of water flow on feeding and growth of *M. nudus* both in the laboratory and the field.

Introduction

Besides their ecological importance (Willoughby, 2018; Ling *et al.*, 2019), sea urchins are of commercial importance (Lauzon-Guay & Scheibling, 2007; Rahman *et al.*, 2014). Sea urchins have been overfished around the world because of the increasing market requirements (Cirino *et al.*, 2017), leading to a reduction in supply. Stock enhancement, which introduces small sea urchins into the field, is an important approach to meeting the increasing market requirements of sea urchins. Water flow greatly affects a series of fitness-related behaviours, including movement (Morse & Hunt, 2013), grazing (Kawamata, 1998; Tamaki *et al.*, 2009) righting (Challener & McClintock, 2017) and covering (James, 2000; Dumont *et al.*, 2007). Because these behaviours link to survival and growth, water flow is important for stock enhancement of sea urchins. Nevertheless, information is still limited on appropriate flow velocities for stock enhancement of sea urchins, although the effects of high water velocity were documented in the field (Lawrence, 1996).

Sea urchins use their tube feet to attach to the sea bottom, effectively resisting the dislodgement risk in water flow (Morse & Hunt, 2013). The adhesion of tube feet regulates the movement of sea urchins and thus plays an important role in their foraging and risk avoidance (Cohen-Rengifo *et al.*, 2017; Tamaki *et al.*, 2018). Further, righting behaviour is essential to maintain activity in water flow (Lawrence, 1975; Hagen, 1994). Besides, covering behaviour plays a role in the response to waves, by increasing their weight (Crook, 2003; Dumont *et al.*, 2007). However, quantitative studies of these fitness-related behaviours are still insufficient at certain flow velocities, despite their important implications for stock enhancement.

The sea urchin *Mesocentrotus nudus* is a commercially important species appropriate for stock enhancement, but not for long-line culture. In Japan, stock enhancement of *M. nudus* provides high-quality gonads and thus great commercial benefits (Agatsuma, 2020). Stock enhancement of *M. nudus* has also been important in China to meet the increasing market demands (~US\$ 20 kg^{-1} in Chinese markets) (Wang *et al.*, 2006; Ding *et al.*, 2020). Releasing size is an important concern in the stock enhancement of *M. nudus* (Agatsuma & Kawai, 1997). For example, *M. nudus* (test diameter >20 mm) were reseeded in April and harvested when the test diameter reached ~70 mm in Dalian, China. For the purposes of studying stock enhancement, we thus used *M. nudus* of test diameter of ~30 mm for the present study in April. The maximum flow velocity was ~90 cm s⁻¹ in the Changshan Islands, which is the major field for the stock enhancement of *M. nudus* in China (Zhang, 2019). Therefore, the maximum flow velocity was set as 90 cm s⁻¹ for the adhesive ability experiments. High flow velocity (>20 cm s⁻¹) significantly inhibits the feeding and foraging of *M. nudus* (Agatsuma, 2013), seriously affecting their survival and growth. Thus, we set 20 cm s⁻¹ as the maximum

flow velocity, 10 cm s^{-1} as the medium flow velocity and 2 cm s^{-1} was set as the control flow velocity for the experiments of movement, righting and covering behaviours.

The main purposes of the present study were to investigate (1) the mean flow velocities to trigger dislodgement and to stop the movement of *M. nudus* and (2) the effects of velocities (2, 10 and 20 cm s⁻¹) on movement, righting and covering behaviours of *M. nudus*. A better understanding of behavioural responses to water flow can provide valuable information on stock enhancement of *M. nudus*.

Materials and methods

Sea urchins

Mesocentrotus nudus were collected locally in the intertidal of Heishijiao in August 2017 and were cultured in the Key Laboratory of Mariculture & Stock Enhancement in North China's Sea, Ministry of Agriculture and Rural Affairs, China, until the experiments started in April 2018. One-third of the seawater in the tanks was changed every 3 days. The sea urchins were fed fresh kelp (Saccharina japonica) and wakame (Undaria pinnatifida). We used different sea urchins for each behavioural experiment. Before the experiment, test diameter and height were measured by a digital Vernier calliper (16EWR, Mahr Co., Germany). They were weighed using an electronic balance (JJ1000Y, G&G Co., USA). A total of 45 sea urchins were involved in the righting, covering and movement experiments. There was no significant difference in test diameter $(29.97 \pm 0.30 \text{ mm})$, test height $(14.52 \pm 0.24 \text{ mm})$ and body weight $(10.69 \pm 0.35 \text{ g})$ (P = 0.221 for test diameter, P = 0.771 for test height, P = 0.556for body weight).

Experimental facility

The experimental facility was a one-level tank made by acrylic material with eight separate runways (length × width × height = $420 \times 60 \times 80$ mm) to exclude potential interference between individuals (Figure 1). Water flow regulators were independent to ensure the consistency of water flow in each of the eight separated runways. The experimental area was limited between the two nets in the runway (length × width × height = $240 \times 60 \times 80$ mm) to ensure flow stability (Figure 1). Flow velocity was controlled by a ball valve and measured using a flow meter (JDC Electronic SA Co., Switzerland). Water flow was unidirectional. Water circulation was supported by a water pump (200 W, 20,000 l h⁻¹, Jebao, China). We investigated the dislodgement and immobilization velocities within the maximum flow velocity (90 cm s^{-1}) in a single runway. Further, three flow velocities $(2 \text{ cm s}^{-1}, 10 \text{ cm s}^{-1} \text{ and } 20 \text{ cm s}^{-1})$ were used for the experiments of movement, righting and covering behaviours.

Dislodgement and immobilization velocity

We individually assessed the velocity that dislodged *M. nudus* from the bottom (Tuya *et al.*, 2007). After sea urchins adhered to the bottom and moved normally, we increased the flow velocity from 0 cm s^{-1} to that which detached sea urchins (the maximum was 90 cm s^{-1}) within 3 min. The flow velocity was constantly accelerated for each trial. The flow velocity that detached sea urchins was recorded as the dislodgement velocity (N = 10). If the maximum flow did not dislodge the sea urchin, we recorded the dislodgement velocity as 90 cm s^{-1} .

To measure the flow velocity that prevents the movement of *M. nudus*, we recorded the flow velocity that caused the sea urchin to stop moving. After the individual adhered to the bottom and



Fig. 1. Diagram of the facility used for measuring behaviours of *Mesocentrotus nudus*. The area of a runway is $24 \times 6 \times 8$ cm (length × width × height), with two nets to limit the experimental area. The lid can be opened to put sea urchins into the facility. It was closed during the experiment.

moved normally, flow velocity was gradually increased until the sea urchin stopped moving. The immobilization velocity was that at which the sea urchin did not move within 15 s (N = 5).

Movement

We individually recorded the movement direction and distance and subsequently calculated the movement velocity of sea urchins at 2, 10 and 20 cm s⁻¹ using a camera (FDR-AXP55, SONY Co., Japan). The experimental duration was 15 min. Since the water flow was unidirectional, we recorded the movement of sea urchins along with the water flow as downstream movement and against the water flow as upstream movement, respectively. Total movement distance, movement time, the distance of downstream and upstream movement were recorded using a video in the movement experiment. Movement velocity, upstream and downstream movement velocity were subsequently calculated (N = 5).

Covering behaviour

Covering is the behaviour in which sea urchins hold material (for example, particles and debris) on their aboral surface with their tube feet (Lawrence, 1976). Twenty clear, white, round polished shells of small scallops (*Mizuhopecten yessoensis*, 20 ± 0.22 mm in diameter) were randomly placed in the experimental area for each experiment. The number of shells used for covering was measured for each sea urchin 15 min after the beginning of the experiment at each flow velocity (N = 5).

Righting response time

Righting response time refers to the time needed for a sea urchin to resume its normal posture with the aboral side up after being inverted to the aboral side down (Lawrence, 1976). We measured the righting response time of *M. nudus* exposed to the three flow velocities within 5 min. When the individual did not right, we recorded the righting response time as 300 s (N = 5).

Statistical analysis

Normal distribution and homogeneity of variance were analysed using Kolmogorov–Smirnov test and Levene's test, respectively. One-way ANOVA was used to compare the movement velocity between the three flow velocities. Least-Significant Difference



Fig. 2. Total movement time (A) and movement velocity (B) of *Mesocentrotus nudus* at 2, 10 and 20 cm s⁻¹ flow velocity (N = 5, mean ± SE). Different letters above the bars refer to the significant difference among experimental groups.





Fig. 4. Movement velocity of *Mesocentrotus nudus* upstream and downstream at 2, 10 and 20 cm s⁻¹ flow velocity (N = 5, mean \pm SE). Different letters beside the bars refer to the significant difference among experimental groups.

Fig. 3. Movement distance of Mesocentrotus nudus during 15 min at 2, 10 and 20 cm s^{-1} flow velocity (N = 5, mean \pm SE).

(LSD) test was used for *post hoc* comparisons when significant differences were found in the ANOVA. We analysed righting response time, covering and movement using the Kruskal–Wallis test and movement direction in a flow velocity using the Mann–Whitney *U* test, because of the heterogeneity of variance and/or abnormal distribution of the data. Statistical analyses were performed using SPSS (Version 22.0). A probability level of P < 0.05 was considered significant.

Results

Dislodgement and immobility velocities

The mean dislodgement velocity of *M. nudus* was 73.50 ± 7.7 cm s⁻¹ (N = 10). The mean flow velocity that inhibited *M. nudus* movement was 33.40 ± 2.7 cm s⁻¹ (N = 5).

Movement

Flow velocity significantly affected movement time (P = 0.041, Figure 2A) and movement velocity (P < 0.001, Figure 2B), but

not movement distance (P = 0.842, Figure 3). The movement time of *M. nudus* exposed to 20 cm s⁻¹ was significantly longer than that at 2 cm s⁻¹ (P = 0.036), while there was no significant difference between 2 cm s⁻¹ and 10 cm s⁻¹ (P = 0.965). Movement velocity of *M. nudus* exposed to 2 cm s⁻¹ (P < 0.001) and 10 cm s⁻¹ (P < 0.001) was significantly higher than that at 20 cm s⁻¹. However, there was no significant difference of movement velocity between 2 cm s⁻¹ and 10 cm s⁻¹ (P = 0.117).

Neither movement distance (P = 0.405 and 0.482), movement velocity (P = 0.400 and 0.803), nor movement time (P = 0.664 and 0.082) varied significantly at 2 cm s⁻¹, 10 cm s⁻¹ and 20 cm s⁻¹ in downstream and upstream movement. However, the velocity in the downstream movement was significantly higher than that in the upstream at 2 cm s⁻¹ (P = 0.016) and 10 cm s⁻¹ (P = 0.008) (Figure 4). There was no significant difference in movement velocity between downstream and upstream at 20 cm s⁻¹ (P = 0.222).

Covering behaviour

There was no significant difference in the number of shells used for covering among 2 cm s^{-1} , 10 cm s^{-1} and 20 cm s^{-1} (P = 0.073, Figure 5A).



Fig. 5. Number of shells used for covering (A) and righting response time (B) of *Mesocentrotus nudus* at 2, 10 and 20 cm s⁻¹ flow velocity (N = 5, mean ± SE). Different letters above the bars refer to the significant difference among experimental groups.

Righting response time

Righting response time of *M. nudus* exposed to 20 cm s⁻¹ was significantly longer than that at 2 cm s⁻¹ (P = 0.015). However, there was no significant difference of righting response time among *M. nudus* at 2 cm s⁻¹ and 10 cm s⁻¹ (P = 1.000, Figure 5B).

Discussion

Sea urchins use tube feet to attach to the sea bottom to prevent dislodgement at high water flow velocities (Santos & Flammang, 2008). This is essential for the survival and consequently for stock enhancement of M. nudus. In the present study, we found that the dislodgement velocity was $73.50 \pm 7.7 \text{ cm s}^{-1}$ for M. nudus (~30 mm test diameter). The dislodgement velocity of M. nudus was greater than that of the sea urchin Diadema antillarum (test diameter of 22-27 mm), but less than that in the sea urchin Arbacia lixula (test diameter of 23-29 mm) (Tuya et al., 2007). This suggests that differences of adhesion ability exist among sea urchin species with different spine size, orientation and the number of tube feet available for attachment (Sharp & Gray, 1962; Tuya et al., 2007). Alternatively, the disparity may be partially due to the test diameter which is slightly larger in the present study, considering adherence is size-independent according to the safety factor evaluated by the attachment ability of the sea urchin A. lixula (Santos & Flammang, 2007). The current result indicates that tube feet of M. nudus could not adhere at a flow velocity of \sim 70 cm s⁻¹ and is thus easily dislodged (Agatsuma, 2013). Therefore, the reseeding of *M. nudus* is probably not appropriate in the areas with a flow velocity of more than $\sim 70 \text{ cm s}^-$

Movement occurs when the flow velocity is in the safe range (Santos & Flammang, 2008). In the present study, 33.40 ± 2.7 cm s⁻¹ was the flow velocity that inhibited the movement of *M. nudus*. This result generally agrees with the finding of Kawamata (1998) that *M. nudus* ceased movement at >40 cm s⁻¹. The partial disagreement between the two studies is probably because of the difference of test diameter, which is ~30 mm in the present study and 55–86 mm in Kawamata (1998). These results indicate that small *M. nudus* could be reseeded in areas with a flow velocity below 30 cm s⁻¹. This is because movement is the basis for the other fitness-related behaviours such as foraging (Agatsuma, 2013) and sheltering (Tamaki *et al.*, 2018) of sea urchins (Santos & Flammang, 2008; Morse & Hunt, 2013).

To better understand the behavioural responses of *M. nudus* to water flow, we investigated their movement, displacement

direction, covering and righting behaviours at flow velocities below 33.40 ± 2.7 cm s⁻¹ (2, 10 and 20 cm s⁻¹). In the present study, movement velocity was reduced by more than half at 20 $\mathrm{cm}\,\mathrm{s}^{-1}$, compared with that at 2 $\mathrm{cm}\,\mathrm{s}^{-1}$. This result is consistent with the report that movement velocity significantly decreased under high-velocity water currents in M. nudus (Kawamata, 1998; Agatsuma, 2013) and Strongylocentrotus droebachiensis (Morse & Hunt, 2013). A reasonable explanation is that sea urchins probably use more energy to maintain body stability at high water velocity (Morse & Hunt, 2013). There was no significant difference in movement distance among the three flow velocities. However, movement time was longer at 20 cm s^{-1} than those at 2 cm s⁻¹ and 10 cm s⁻¹. Shorter movement time probably impacts the capacity to find shelter and consequently increases the risk of predation in the field, although M. nudus can retain movement and other behaviours, including sheltering (Tamaki et al., 2018) and grazing (Kawamata, 1998) at 20 cm s⁻¹. Further, flow velocity affected the movement velocity of the sea urchins in the two directions. Downstream movement velocity of *M. nudus* was significantly higher than upstream movement velocity at 2 cm s and 10 cm s^{-1} (P = 0.016 and P = 0.008), but not at 20 cm s⁻¹ (P = 0.222). This result indicates that *M. nudus* tends to move downstream at low flow velocities, while this trend decreases at high flow velocities. This result is consistent with the finding of Morse & Hunt (2013) that sea urchins tended to move downstream and upstream at low and high flow velocities, respectively. Twenty cm s^{-1} significantly limits the movement direction of *M*. nudus and is probably detrimental to its escape from adverse environments. This suggests that flow velocity above 20 cm s^{-1} markedly affects the movement of M. nudus and thus probably is not appropriate for reseeding.

Covering behaviour supports the sea urchin *Lytechinus aname*sus to stabilize under wave exposure (Lees & Carter, 1972). Sea urchins cover themselves with objects to increase their weight, partly because light individuals are susceptible to strong water flow (Crook, 2003). In the present study, however, no significant difference in covering behaviour was found between 2, 10 and 20 cm s⁻¹. This suggests that 20 cm s⁻¹ may not affect the stability of *M. nudus*. Righting response time of *M. nudus* is significantly longer at 20 cm s⁻¹ (16.60 ± 3.11 s) than at 2 cm s⁻¹ (193.80 ± 51.26 s, P = 0.015). This indicates that righting behaviour is weakened at high flow velocities. However, this result is not consistent with the finding of Challener & McClintock (2017) that *L. variegatus* improved righting behaviour at high flow velocity. This disagreement is probably because of the difference of the sediment and flow type, which was a smooth surface and unidirectional water flow $(2-20 \text{ cm s}^{-1})$ in the present study but sand surface and in waves (60 cm s^{-1}) in Challener & McClintock (2017). Notably, the present study is a laboratory investigation and probably different from the hygrographic conditions in the field.

Conclusions

In conclusion, a velocity below 30 cm s⁻¹ would be indicated to be appropriate for reseeding of *M. nudus* (test diameter of ~30 mm) to avoid dislodgement and to ensure their movement. Flow velocity of 20 cm s⁻¹ significantly affected the righting (P = 0.015) and movement behaviours (P < 0.001) of *M. nudus*. Sea areas with a flow velocity less than 20 cm s⁻¹, preferably 2–10 cm s⁻¹, would be expected to be appropriate for stock enhancement of *M. nudus*. The present study provides valuable information for stock enhancement of *M. nudus*. Future investigations, however, should be carried out on the effects of long-term water flow on feeding and growth of *M. nudus* both in the laboratory and the field.

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References

- Agatsuma Y (2013) Strongylocentrotus nudus. In Lawrence JM (ed.), Sea Urchins: Biology and Ecology, 3rd Edn. San Diego, CA: Academic Press, pp. 449–460.
- Agatsuma Y (2020) Stock enhancement of regular sea urchins. In Lawrence JM (ed.), *Sea Urchins: Biology and Ecology*, 4th Edn. San Diego, CA: Academic Press, pp. 299–313.
- Agatsuma Y and Kawai T (1997) Seasonal migration of the sea urchin Strongylocentrotus nudus in Oshoro Bay of southwestern Hokkaido, Japan. Nippon Suisan Gakkaishi 63, 557–562.
- **Challener R and McClintock J** (2017) *In situ* measurements of righting behaviour in the common sea urchin *Lytechinus variegatus*: the importance of body size, substrate type, and covering material. *Aquatic Biology* **26**, 33–40.
- **Cirino P, Ciaravolo M, Paglialonga A and Toscano A** (2017) Long-term maintenance of the sea urchin *Paracentrotus lividus* in culture. *Aquaculture Reports* 7, 27–33.
- Cohen-Rengifo M, Moureaux C, Dubois P and Flammang P (2017) Attachment capacity of the sea urchin *Paracentrotus lividus* in a range of seawater velocities in relation to test morphology and tube foot mechanical properties. *Marine Biology* **164**, 79.
- Crook AC (2003) Individual variation in the covering behaviour of the shallow water sea urchin *Paracentrotus lividus*. *Marine Ecology* **24**, 275–287.
- Ding J, Zheng D, Sun J, Hu F, Yu Y, Zhao C and Chang Y (2020) Effects of water temperature on survival, behaviours and growth of the sea urchin *Mesocentrotus nudus*: new insights into the stock enhancement. *Aquaculture* **519**, 734873.
- **Dumont CP, Drolet D, Deschênes I and Himmelman JH** (2007) Multiple factors explain the covering behaviour in the green sea urchin, *Strongylocentrotus droebachiensis. Animal Behaviour* **73**, 979–986.

- Hagen NT (1994) Is righting response a useful indicator of functional wellbeing in the green sea urchin *Strongylocentrotus droebachiensis*? In Bruno D (ed.), *Echinoderms Through Time*. Rotterdam: Balkema, pp. 693–698.
- James DW (2000) Diet, movement, and covering behaviour of the sea urchin *Toxopneustes roseus* in rhodolith beds in the Gulf of California, México. *Marine Biology* 137, 913–923.
- Kawamata S (1998) Effect of wave-induced oscillatory flow on grazing by a subtidal sea urchin Strongylocentrotus nudus (A. Agassiz). Journal of Experimental Marine Biology and Ecology 224, 31–48.
- Lauzon-Guay J-S and Scheibling RE (2007) Seasonal variation in movement, aggregation and destructive grazing of the green sea urchin (*Strongylocentrotus droebachiensis*) in relation to wave action and sea temperature. *Marine Biology* 151, 2109–2118.
- Lawrence JM (1975) The effect of temperature-salinity combinations on the functional well-being of adult *Lytechinus variegatus* (Lamarck) (Echinodermata, Echinoidea). *Journal of Experimental Marine Biology* and Ecology 18, 271–275.
- Lawrence JM (1976) Covering response in sea urchins. Nature 262, 490-491.
- Lawrence JM (1996) Mass mortality of echinoderms from abiotic. In Jangoux M (ed.), *Echinoderm Studies*, vol. 5. Boca Raton, FL: CRC Press, pp. 103–137.
- Lees DC and Carter GA (1972) The covering response to surge, sunlight, and ultraviolet light in *Lytechinus anamesus* (Echinoidea). *Ecology* 53, 1127–1133.
- Ling SD, Kriegisch N, Woolley B and Reeves SE (2019) Density-dependent feedbacks, hysteresis, and demography of overgrazing sea urchins. *Ecology* 100, e02577.
- Morse BL and Hunt HL (2013) Effect of unidirectional water currents on displacement behaviour of the green sea urchin *Strongylocentrous droebachiensis. Journal of the Marine Biological Association of the United Kingdom* **93**, 1923–1928.
- Rahman MA, Arshad A and Yusoff FM (2014) Sea urchins (Echinodermata: Echinoidea): their biology, culture and bioactive compounds. In International Conference on Agricultural, Ecological and Medical Sciences (AEMS-2014) 3–4 July 2014 London (UK). International Institute of Chemical, Biological & Environmental Engineering.
- Santos R and Flammang P (2007) Intra- and interspecific variation of attachment strength in sea urchins. *Marine Ecology Progress Series* 332, 129–142.
- Santos R and Flammang P (2008) Estimation of the attachment strength of the shingle sea urchin, *Colobocentrotus atratus*, and comparison with three sympatric echinoids. *Marine Biology* **154**, 37–49.
- Sharp DT and Gray IE (1962) Studies on factors affecting the local distribution of two sea urchins, Arbacia punctulata and Lytechinus variegatus. Ecology 43, 309.
- Tamaki H, Kusaka K, Fukuda M, Arai S and Muraoka D (2009) Undaria pinnatifida habitat loss in relation to sea urchin grazing and water flow conditions, and their restoration effort in Ogatsu Bay, Japan. Journal of Water and Environment Technology 7, 201–213.
- Tamaki H, Muraoka D and Inoue T (2018) Effect of water flow on grazing by the sea urchin (*Strongylocentrotus nudus*) in the presence of refuge habitat. *Journal of Water and Environment Technology* **16**, 30–39.
- Tuya F, Cisneros-Aguirre J, Ortega-Borges L and Haroun RJ (2007) Bathymetric segregation of sea urchins on reefs of the Canarian Archipelago: role of flow-induced forces. *Estuarine, Coastal and Shelf Science* 73, 481–488.
- Wang Q, Wu H, Fang J and Wang S (2006) Stock enhancement and sea ranching: practices at Zhangzidao Island in the northern Yellow Sea. In *Third International Symposium on Stock Enhancement and Sea Ranching*, Seattle, WA, pp. 18–21.
- Willoughby L (2018) News feature: can predators have a big impact on carbon emissions calculations? *Proceedings of the National Academy of Sciences* USA 115, 2260–2263.
- Zhang B (2019) Observation and analysis of sea currents in the sea area of Changshan Island in Dalian, China. Scientific and Technological Innovation 11, 8–10. (In Chinese).