

# Influence of Water Temperature and Salinity on Sand Ejection of Manila Clam, *Ruditapes philippinarum*

Kyoung Ho Kang, Jae-Min Kim and Young Hun Kim

Division of Aqua Life Science, Yosu National University, Yosu, Korea

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## ABSTRACT

In this study, the influence of water temperature and salinity on sand ejection of Manila clams, *Ruditapes philippinarum*, was investigated. The result showed that, under different water temperature, the highest quantity of sand ejection was at 20°C, the value of which was 0.091 g, while the smallest one was only 0.058 g at 0°C. The highest releasing rate, 95.79%, could be seen in 20°C group, while 0°C group was the lowest one, 85.29%. Under different salinity, 30 psu group had the highest value, 0.057 g, and 0 psu (freshwater) group was the lowest one, only 0.026 g. At the same time, the highest releasing rate appeared at 25 psu, which was 90%. 0 psu group was the lowest one in releasing rate, 60.47%. According to these results, we recommend that the clams should be placed into the clear seawater with about 25 psu of salinity at near 20°C to let the clams perform the self-depuration. Better quality of clams containing less impurity in the aquaculture and commerce could be obtained from this pre-treatment.

**Keywords:** *Ruditapes philippinarum*, Sand ejection, Temperature, Salinity

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## INTRODUCTION

*Ruditapes philippinarum*, which is a species of filter-feeding bivalve living in soft bottoms, subjects to Mollusca, Lamellibranchia, Heterodonta, Veneroidea,

Veneridae. It widely distributes in China, Korea, Japan, *etc.* The clam is edible and its protein and inorganic salt contents can reach 7.4% and 2.9% respectively. Furthermore, the shell can be burned to ashes or made into the feedstuff (Wang, 1993). Hence it is commercially important and valuable in the aquiculture. As to the relative studies of *Ruditapes philippinarum*, Sorokin and Giovanardi (1995) investigated the trophic characteristics of *R. philippinarum*. Saurian *et al.* (1998) reported that a new method of distributing live algae food in marine coastal ponds was used for this clam intensive culture. Cima *et al.* (2000) used various methods of cytochemical staining to investigate the haemocytes of *R. philippinarum*, and the result showed that four cell types: granulocytes, hyalinocytes, haemoblast and serous cells were found in the haemolymph. Some researchers suggested that the temperature was a key factor in determining the excretion rate of *R. philippinarum* (Zhu, *et al.*, 1999). Gouletquer *et al.* (1999) reported that rearing of this clam had only a limited effect on the environmental sediment parameters, such as water percentage, phaeopigment and silt level, from a spatiotemporal point of view. Caers *et al.* (1999) performed an experiment to research the dietary impact of algal and artificial diets, fed at different feeding rations, on the growth and fatty acid composition of *R. philippinarum*. Regarding to the depuration of invertebrate, the main concentration was focused on the depuration of some contaminants or compounds. Sericano *et al.* (1996) investigated the accumulation and depuration of organic contaminants by the American oyster, *Crassostrea virginica*. Some researchers performed

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Corresponding author: Kang, Kyoung Ho

Tel: (82) 61-659-3165 e-mail: mobidic@yosu.ac.kr

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some experiment to seek the relation between the sample size and the depuration. Gilek *et al.* (1996) reported the influence of body size on the uptake, depuration, and bioaccumulation of polychlorinated biphenyl coneners by Baltic Sea blue mussels, *Mytilus edulis*. As the depuration of sand in the clam will directly impact the quality of clams in aquiculture and commerce, we designed this experiment to do some primary study on the appropriate temperature and salinity for the sand ejection.

## MATERIALS AND METHODS

### 1. Clams breeding

The *Ruditapes philippinarum* individuals were

collected by boat from inshore area of Yosu City, Korea. Then they were immediately put into breeding tanks containing filtered seawater at various salinity and temperature. Here, every 20 clams were bred in one tank, the size of which was about 40 × 30 × 20 mm. The salinity was designed from 0 psu to 50 psu at intervals of 5 psu, and the water temperature was from 0°C to 30°C at intervals of 5°C. (Table 1 and Table 2)

### 2. Examining the sand ejection

Without feeding the samples for 4 h, 8 h, 12 h and 24 h, the quantity of sand released from the shellfish was examined. In examining the sand weight, the membrane filter was used. At first, the clear dry

**Table 1.** Measurement of *Ruditapes philippinarum* used in the experiment for sand releasing at different water temperature.

Water temperature (°C)	Shell length (mm) ± SD	Shell height (mm) ± SD	Shell breadth (mm) ± SD	Total weight (g) ± SD
0	42.63 ± 0.88	30.30 ± 1.20	19.80 ± 1.13	17.13 ± 1.93
5	41.75 ± 1.27	31.10 ± 1.06	21.40 ± 1.13	18.31 ± 0.90
10	41.50 ± 0.14	29.55 ± 0.64	20.00 ± 0.64	17.55 ± 1.51
15	45.80 ± 1.34	30.83 ± 0.25	20.10 ± 0.78	18.49 ± 0.88
20	44.15 ± 5.44	30.80 ± 0.28	21.50 ± 0.85	17.82 ± 2.86
25	43.05 ± 1.34	32.22 ± 0.39	20.53 ± 0.53	18.93 ± 0.27
30	42.10 ± 1.27	29.60 ± 0.42	19.93 ± 0.60	17.24 ± 0.45

Data presented as mean ± SD

**Table 2.** Measurement of *Ruditapes philippinarum* used in the experiment for sand releasing at various salinity.

Salinity (psu)	Shell length (mm) ± SD	Shell height (mm) ± SD	Shell breadth (mm) ± SD	Total weight (g) ± SD
0	36.50 ± 1.41	27.28 ± 0.81	18.45 ± 0.28	12.63 ± 0.09
5	37.63 ± 1.38	27.80 ± 0.64	19.10 ± 0.22	13.19 ± 0.69
10	38.78 ± 1.17	27.68 ± 0.11	18.75 ± 0.36	13.07 ± 1.13
15	39.20 ± 0.99	28.80 ± 0.50	18.70 ± 0.42	13.70 ± 1.13
20	42.10 ± 4.10	31.13 ± 1.17	21.28 ± 1.31	18.39 ± 2.99
25	40.73 ± 3.50	29.55 ± 3.32	23.15 ± 5.73	16.65 ± 4.71
30	40.00 ± 0.57	27.70 ± 0.28	18.75 ± 0.35	15.19 ± 3.08
40	42.50 ± 2.12	30.40 ± 1.41	20.05 ± 0.64	17.85 ± 1.06
45	39.10 ± 0.42	26.63 ± 0.32	16.58 ± 1.73	17.78 ± 0.98
50	42.65 ± 2.37	27.75 ± 1.52	18.10 ± 0.84	18.85 ± 1.02

Data presented as mean ± SD

membrane filter was weighed, and then the seawater, which was used in breeding the clams, was filtrated through it. After the membrane filter was dried in the warm drier and weighed, the sand weight could be figured out. When examining the sand remained in digestive diverticula of *Ruditapes philippinarum*, first, the digestive diverticula were taken out by dissection. Then the saturated NaClO was used to erode the flesh for about 3-4 hours. Dilute the solution, after the filtration and dryness, the sand remained in digestive diverticula could be obtained. The releasing rate can be calculated by the following formula: Releasing rate (%) = sand released / (sand released + sand remained) × 100.

## RESULTS

### 1. At different water temperature

Under different water temperature and breeding time, the maximum sand released from clams appeared in 20°C-4 h group, 15°C-8 h group, 30°C-12 h group and 30°C-24 h group, the values of which were

0.039 ± 0.003 g, 0.022 ± 0.003 g, 0.023 ± 0.003 g, 0.016 ± 0.003 g. The minimum was at 0°C-4 h, 0°C-8 h, 0°C-12 h, and 10°C-24 h, only 0.016 ± 0.004 g, 0.012 ± 0.001 g, 0.015 ± 0.001 g, and 0.009 ± 0.003 g, respectively. In total quantity, 20°C group had the highest sand released, which was 0.091 ± 0.004 g. 0°C group had the smallest value, only 0.059 ± 0.003 g (Table 3). As to the sand remained in digestive diverticula of the clams, 0°C group had the highest value, which was 0.010 g, while the 20°C group had the smallest remainder, which was 0.004 g (Table 4). The highest releasing rate appeared in 20°C group, valued in 95.79%. The lowest rate was 85.29%, which was in 0°C group (Fig. 1).

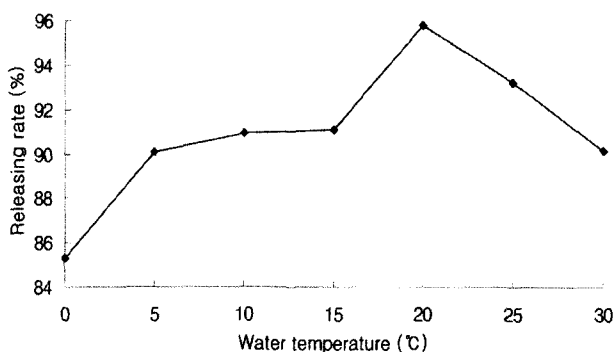
### 2. At different water salinity

Under different water salinity with breeding time of 4 h, 8 h, 12 h, and 24 h, the highest sand released groups at each breeding time were 30 psu-4 h, 30 psu-8 h, 15 psu-12 h, and 30 psu-24 h, the values of which were 0.028 ± 0.003 g, 0.012 ± 0.003 g, 0.013 ±

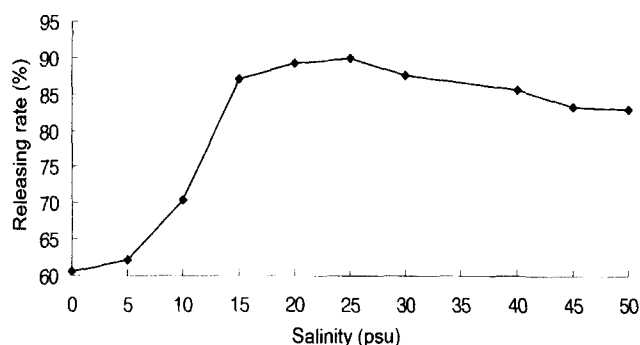
**Table 3.** Sand releasing (g) of *Ruditapes philippinarum* at different water temperature.

Time (hour)	Water temperature (°C)						
	0	5	10	15	20	25	30
4	0.016 ± 0.004	0.022 ± 0.003	0.034 ± 0.003	0.030 ± 0.003	0.039 ± 0.002	0.034 ± 0.003	0.026 ± 0.003
8	0.012 ± 0.001	0.020 ± 0.003	0.017 ± 0.003	0.022 ± 0.003	0.022 ± 0.003	0.020 ± 0.001	0.018 ± 0.003
12	0.015 ± 0.001	0.019 ± 0.003	0.020 ± 0.003	0.018 ± 0.003	0.020 ± 0.001	0.018 ± 0.003	0.023 ± 0.003
24	0.015 ± 0.002	0.021 ± 0.003	0.009 ± 0.003	0.012 ± 0.003	0.010 ± 0.003	0.010 ± 0.002	0.016 ± 0.003
Total	0.058 ± 0.003	0.082 ± 0.003	0.080 ± 0.003	0.082 ± 0.003	0.091 ± 0.004	0.082 ± 0.002	0.073 ± 0.003

Data presented as mean ± SD



**Fig. 1.** Sand releasing rate (%) of *Ruditapes philippinarum* at different temperature.



**Fig. 2.** Sand releasing rate (%) of *Ruditapes philippinarum* at different salinity.

0.002 g, and  $0.015 \pm 0.003$  g respectively. Under the same experiment, the smallest values were at 10 psu-4 h, 0 psu-8 h, 0 psu-12 h, and 5 psu-24 h, which were  $0.011 \pm 0.002$  g,  $0.004 \pm 0.004$  g,  $0.003 \pm 0.001$  g, and  $0.004 \pm 0.003$  g, respectively. In total, 30 psu group showed the maximum sand released, which was  $0.057 \pm 0.003$  g, while the minimum appeared at 0 psu group, only  $0.026 \pm 0.004$  g (Table 5). Under the same condition, the sand remained had the highest value, 0.017 g, in 0 and 5 psu groups. 20 and 25 psu group had the lowest value, 0.006 g (Table 6). Releasing rate using the formula mentioned above showed that 25 psu group had the highest releasing rate, 90%. As the salinity decreased or increased from 25 psu of salinity, the sand releasing rate decreased continuously. The lowest rate appeared at 5 psu group, valued in 62.22% (Fig. 2).

## DISCUSSION

*Ruditapes philippinarum* is a familiar species of aquacultural clams. This clam grows fast and moves slowly. It is easy to be cultivated. As the consumption of this clam increases day by day, in many areas, the cultivation of this clam plays an important role in the aquatic culture. Usually, it inhabits on the soft bottom with high content of sand (70-90%). It cave-dwells at the depth of 3-10 cm. It has a strong adaptability to the temperature ranging about 5-35°C, and the optimum is 18-30°C. When the temperature is higher than 36°C or lower than 0°C, the feeding will cease. At 44°C, the death rate will be 50% and at 45°C, 100% (Wang, 1993).

In our experiment, at different temperature, the quantity of sand ejection varied. Under different exposure time, the highest quantity of sand ejection

**Table 4.** Sand remained (g) in digestive diverticula of *Ruditapes philippinarum* used in the sand releasing experiment at different water temperature.

Water temperature (°C)	0	5	10	15	20	25	30
Sand remained	0.010	0.009	0.008	0.008	0.004	0.006	0.008

**Table 5.** Sand released (g) of *Ruditapes philippinarum* at various salinity.

Time (hour)	Salinity (psu)									
	0	5	10	15	20	25	30	40	45	50
4	$0.012 \pm 0.002$	$0.012 \pm 0.001$	$0.011 \pm 0.002$	$0.014 \pm 0.002$	$0.015 \pm 0.002$	$0.023 \pm 0.002$	$0.028 \pm 0.003$	$0.020 \pm 0.002$	$0.012 \pm 0.002$	$0.019 \pm 0.002$
8	$0.004 \pm 0.004$	$0.006 \pm 0.002$	$0.005 \pm 0.002$	$0.006 \pm 0.002$	$0.011 \pm 0.002$	$0.007 \pm 0.002$	$0.012 \pm 0.003$	$0.010 \pm 0.002$	$0.008 \pm 0.002$	$0.011 \pm 0.002$
12	$0.003 \pm 0.001$	$0.006 \pm 0.002$	$0.008 \pm 0.002$	$0.013 \pm 0.002$	$0.012 \pm 0.002$	$0.011 \pm 0.002$	$0.012 \pm 0.003$	$0.010 \pm 0.002$	$0.011 \pm 0.002$	$0.005 \pm 0.002$
24	$0.007 \pm 0.003$	$0.004 \pm 0.003$	$0.007 \pm 0.002$	$0.014 \pm 0.002$	$0.014 \pm 0.002$	$0.013 \pm 0.002$	$0.015 \pm 0.003$	$0.008 \pm 0.002$	$0.009 \pm 0.002$	$0.009 \pm 0.002$
Total	$0.026 \pm 0.004$	$0.028 \pm 0.002$	$0.031 \pm 0.002$	$0.047 \pm 0.002$	$0.050 \pm 0.002$	$0.054 \pm 0.002$	$0.057 \pm 0.003$	$0.048 \pm 0.002$	$0.040 \pm 0.002$	$0.039 \pm 0.002$

Data presented as mean  $\pm$  SD

**Table 6.** Sand remained (g) in digestive diverticula of *Ruditapes philippinarum* used in the sand releasing experiment at different salinity.

Salinity (psu)	0	5	10	15	20	25	30	40	45	50
Sand remained	0.017	0.017	0.013	0.007	0.006	0.006	0.008	0.008	0.007	0.008

was  $0.039 \pm 0.003$  g,  $0.022 \pm 0.003$  g,  $0.023 \pm 0.003$  g,  $0.021 \pm 0.003$  g in 20°C-4 h, 20 and 15°C- 8 h, 30°C-12 h, and 5°C-24 h group respectively. As time passed by, the sand ejection decreased. In the total quantity, 20°C group had the highest value, 0.091 g. Concerning the releasing rate, 20°C group had the highest sand releasing rate, too. As the temperature decreased or increased from 20°C, the value decreased continuously. 0°C group had both the smallest sand released and the lowest releasing rate, only 0.058 g and 85.29%. As to the reason, we suggest that keep the clam at 20°C, which is normal living temperature of *R. philippinarum* in natural environment. The clams will live well under this temperature, and the metabolism will be active. Under the temperature of 0°C, the clam can be still alive, but the metabolism level will be very low. It is reported that the mortality would be about 10% after two weeks at 2-3°C (Wang, 1993). The low metabolism results in the weak sand depuration.

Under different salinity, in total sand ejection, 30 psu group had the highest value of 0.057 g, while 25 psu group had the highest releasing rate, 90%. With regard to other groups, the higher or lower the salinity than 25 psu, the lower the releasing rate was. This trend is similar to the result of different temperature. The smallest one was only 0.026 g, which is less than a half of the value in 30 psu group, appeared at 0 psu group. This group also had the lowest releasing rate, 60.47%. At natural environment, *R. philippinarum* is liable to inhabit the area with a certain inflow of freshwater, so usually, the salinity of its habitat, which is 20-30 psu, is a little lower than the normal seawater (31-35 psu). But under the freshwater environment, the clam cannot survive for a long time. After a short time, the clam will close its shells to prevent the freshwater flowing into its inside, so the activity of sand ejection will be very low. It is reported that under the freshwater condition, the clams can only survive for about 66 hours. After 71 hours, they may all die (Wang, 1993).

Furthermore, in our experiment, as to every temperature and salinity group, the values of 4 h were higher than those of other sampling time. It can be

inferred that, the clam would perform the self-depuration soon after it was transferred to the clear water.

According to the result herein before, we suggest that in the aquaculture and commerce, cleansing shellfish by purification in the relatively clear environment may be practical after gathering the clams. They can be put into the seawater of about 25 psu salinity under 20°C, or near this condition, and the clams with better quality containing less impurity can be obtained.

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