

Review article





# Results of integrated multitrophic aquaculture on seaweed plants in Primorsky krai (Sea of Japan)

### **Abstract**

The article presents the results of cultivating commercial invertebrates with two species of alge (kelp): Saccharina japonica and Saccharina cichorioides on seaweed plantations in Primorsky Krai, as well as keeping salmon: masu salmon Oncorhynchus mason and chum salmon Oncorhynchus keta in a Bridgestone fish cage. It was found that masu salmon is more suited for cage keeping than chum salmon. Cage culture of Japanese scallop Mizuhopecten yessoensis and its non-transplantation culture in rarefied collector - cages (CC) at the Whale Bay kelp farm showed the advantage of non-transplantation culture. In CC, in addition to M. yessoensis, the Pacific mussel Mytilus trossulus reached a commercial size, but the Swift's scallop Swiftopecten swiftii and the sea urchin Strongylocentrotus intermedius reached a viable size, suitable for sowing on the sea floor. Co-cultivation of invertebrates and algae has a mutually beneficial effect on the production indicators of these mariculture objects.

**Keywords:** cultivation of *M. yessoensis*, *M. trossulus*, *S. swiftii*, *S. intermedius*, *S. japonica* and *S. cichorioides* on algae plantations of Primorsky Krai, Sea of Japan.

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# Introduction

Seafood farming at sea (mariculture) is a rapidly growing industry in which China has achieved outstanding results.<sup>1,2</sup> Moreover, seaweeds (especially the algal kelp Saccharina japonica) are the leaders in production volume.3 In recent decades, the area and density of cultivation have increased so much that the negative impact of animals on the environment due to self-pollution has been recognised. An increased concentration of metabolites in shallow marine ecosystems has led to eutrophication, decreased biodiversity, reduced growth, death of cultured organisms, 4,5 and even a decrease in production.<sup>6,7</sup> In China, to reduce the negative impact of mariculture on the environment, intensive monoculture has moved to polyculture. The first successful attempt was made in 1975, when S. japonica and the mussel Mytilus edulis were co-cultured; the metabolites of M. edulis were consumed by S. japonica, and dissolved organic matter secreted by the algae was consumed by mollusks. 8,9 Soon, polyculture in China developed such that valuable (fish and shrimp) and low value species (bivalves and seaweed) were co-cultured, with the low

value species serving as a recycling agent for the valuable species. <sup>10,11</sup> Such integrated multitrophic aquaculture (IMTA) is practiced both in China<sup>9,12</sup> and elsewhere. <sup>13-16</sup> High growth rates of kelp and mussels near fish farms, compared to reference sites, reflect higher availability of food and energy. <sup>17</sup> Reduced cultivation intensity, better hydrodynamic conditions, and scallop-kelp polyculture may explain the healthy benthos in Sanggou Bay. <sup>18</sup> Seaweeds absorb dissolved nutrients and this function has been well studied in effluents from fish, shrimp, and shellfish mariculture. <sup>19</sup>

In the late 1970s, the Experimental Mariculture Base (EMB) was created in Whale Bay, Russia, (43°06'36" N and 134°19'60" E) (Figure 1) for the cultivation of four species of valuable seafood: the algae (kelp) *S. japonica*, the holothurian *Apostichopus japonicus*, and the salmon *O. mason* and *O. keta*. By the mid-1980s, 100 hectares of marine plantations (Figure 2) for growing *S. japonica* had been created. In 1982, a broodstock of *M. yessoensis* formed on the sea floor due to the transition to a bottom lifestyle of juveniles raised on *S. japonica*.<sup>20</sup>

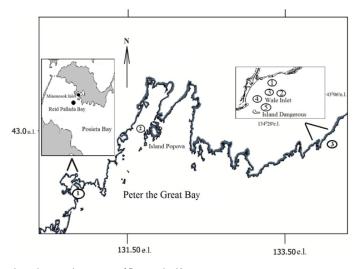


Figure I Map-scheme of stations located on the sea plantations of Primorsky Krai.







Figure 2 Plantations for growing Saccharina japonica.

For the cultivation of salmon fish, five Bridgestone fish cages, each 5x5x4 m, and a pilot plant for feed production were purchased. A workshop with pools and thermal treatment of fresh and salt water was built for growing algal kelp seedling colonies (thallus), sea cucumber larvae and fish fry. In 1985, 100 garlands of scallop collector-cages (CC)<sup>21</sup> were suspended between ropes with the seaweed. Similar work on the co-cultivation of seaweed and shellfish was carried out in the 1980s in the Stark Strait (Popov Island) (42°98'02" N and 131°75'63" E) and Reid Pallada Bay (Posyeta Bay) (42°59'14" N and 130°83'75" E). This publication reports on the results obtained from the co-cultivation of seaweed algae and other marine life.

### **Material and methods**

For breeding masu salmon *Oncorhynchus mason* and chum salmon *Oncorhynchus keta* in a sea cage, in the autumn of 1982 and 1984, the spawners were caught, incubated, raised with artificial feeds and transferred to sea water. At the end of August, they were placed in a cage in Whale Bay where they were fed salmon eggs. In the spring of 1984, thallus of the kelp (algae) *Saccharina cichorioides* were taken from the bottom near Popov Island, woven into 6 mm nylon ropes and suspended on horizontal ropes in the Stark Strait (Figure 1) near this island. Several designs of mussel collectors and collector-cages (CC) with and without a cover were simultaneously exposed. In the summer of 1985, the Pacific mussel *Mytilus trossulus* was removed from three ropes containing of this kelp. Over the course of two years, the growth of *M. trossulus* that had settled on the thallus and collectors was monitored.

S. japonica was grown in Whale Bay and Reid Pallada Bay in a two-year cycle using 6-metre-long nylon ropes. <sup>22</sup> During the kelp harvest year in Whale Bay, <sup>20</sup> one nylon rope containing algae was removed monthly, and the length, width, and weight of each thallus were measured. At least 25 thalli were used for analysis. In September 1987 and October 1988, in Minonosok Inlet, Posyet Bay (42°36'55" N and 130°50'48" E), the nylon ropes were placed in water containing spores extracted from S. japonica thalli collected in Peter the Great Bay. After the spores had settled, the ropes were suspended vertically on horizontal ropes in Reid Pallada Bay (Figure 1). In June 1989, plastic cages with yearling M. yessoensis were placed on adjacent ropes and between the ropes with spores. After 3–6 months, one rope

was raised to the surface and the length, width, thickness, and weight of each thallus were measured. At the same time, *M. yessoensis* and *M. trossulus* molluscs growing on artificial substrates and on *S. japonica* rhizoids were counted and measured. Minonosok Inlet, located near Reid Pallada Bay, was used to compare the production indices of the mollusks.

In Whale Bay, to determine the optimal time for collector exposure, 25-30 M. yessoensis spawners were captured every 10 days using diving equipment and their gonadal index was measured using the method.<sup>23</sup> A decrease in the gonadal index by 9-12% meant the end of spawning. One week after spawning, plankton samples were collected at five stations using a Judy net with 100µm mesh size. Plankton samples were fixed with 4% formaldehyde. The abundance of M. yessoensis larvae in the sample was counted in a Bogorov camera under an MBS-9 microscope. Knowing the volume of water filtered by the plankton net, the resulting number of larvae was recalculated per 1 m3. The height of the larval valves was measured with an ocular micrometer with an accuracy of ±5μm. Simultaneously with catching the spawners and collecting plankton samples at three horizons (0, 5 and 10m), the water temperature was measured. After the appearance of scallop larvae with a shell height of 200-250µm, CC with a cover having 5mm holes and mussel collectors made of nylon net were suspended at five stations of algae farms between the ropes with algae. In addition, at each station there was one garland of CC with double the spacing between the cones and the with cover holes of 15mm diameter. After the juvenile scallops reached 8-10 mm in shell height, part of the CC with 5mm holes in the cover was lifted to the surface. Japanese scallops M. yessoensis, Pacific mussels M. trossulus, Swift's scallops Swiftopecten swiftii and sea urchins Strongylocentrotus intermedius were removed from the collectors. The living and dead individuals were measured and counted. The results were expressed per 1m<sup>2</sup>. Due to the harsh weather conditions in Whale Bay, the main transfer of juvenile M. yessoensis from CC to plastic cages was carried out in the summer of 1986. The growth and survival of animals transferred to cages and not transferred to CC were monitored for four years. For the analysis, 25-30 specimens of cultured mollusks were used. The number, weight, and shell height of mussels sitting in rhizoids (roots) of kelp were determined.

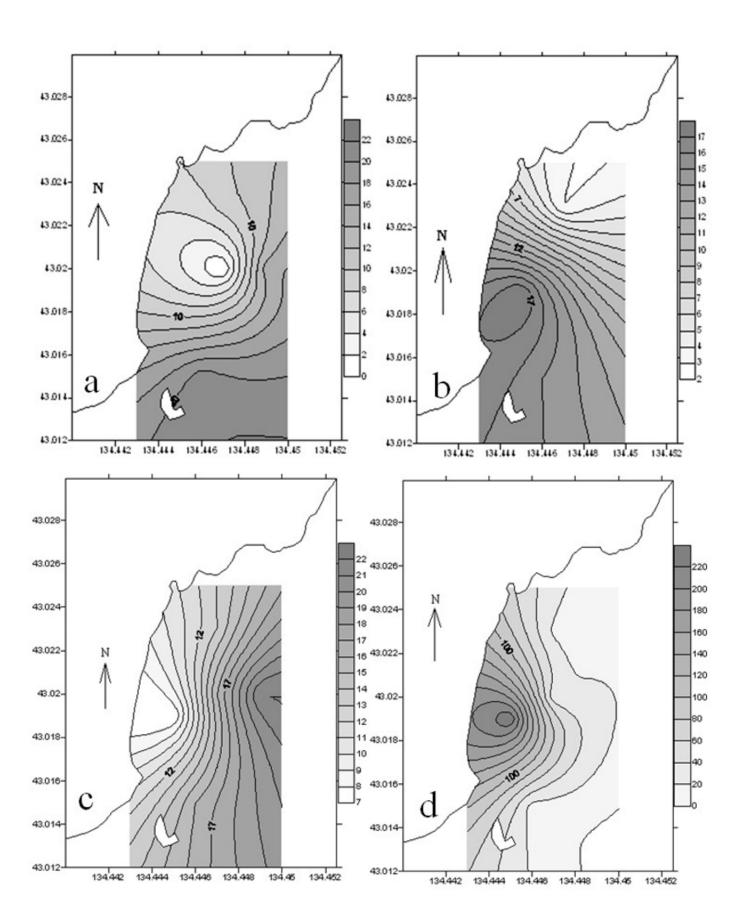
In 1985-1988, employees of the Far Eastern Hydrometeorological Institute analyzed water samples and determined the wind direction in Whale Bay. Statistical processing of the data was carried out using the STATISTICA 6 program<sup>24</sup> and tested at the level of  $\alpha=0.05$ . Graphic material for publication was built using Serfer 7 Golden Software, and Excel 2007.

### **Results**

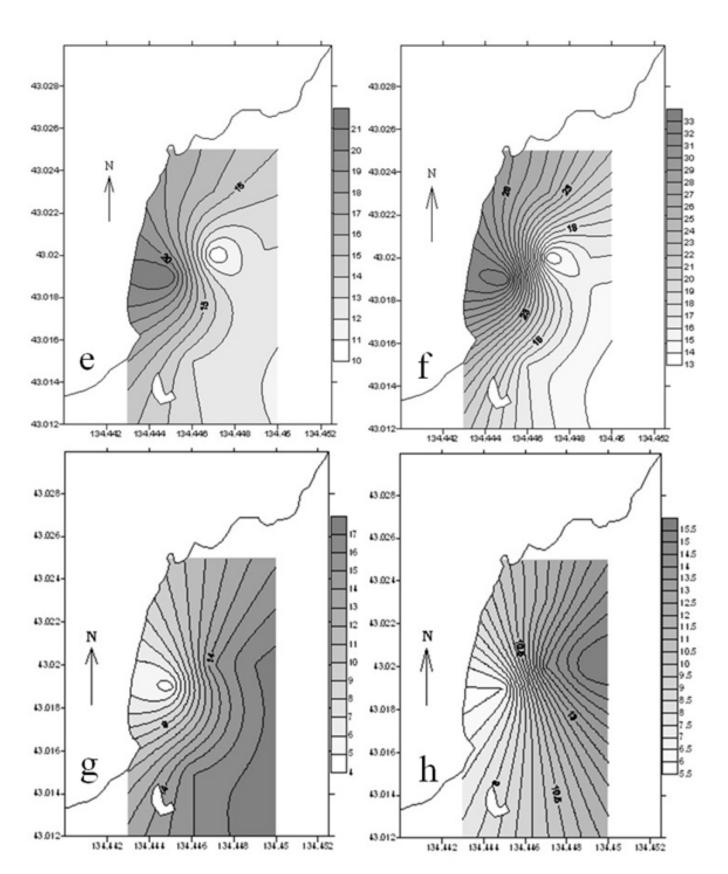
Salmon farming in Whale Bay ceased due to cage destruction. In 1983, the cage cover broke and the young fish swam away. In 1985, the cage frame collapsed during a storm because of faulty installation. However, the month-long stay of the young fish in the cage in 1985 showed that despite the painful transition to sea water, the cherry salmon *O. mason* adapts to captivity faster than the chum salmon *O. keta* and is more suitable for cage keeping. The biological peculiarity of the chum salmon is its early migration to the open sea; it rubbed its snout against the cage walls, was injured, fell ill and died.

The horizontal distribution of *M. yessoensis* larvae in Whale Bay showed a fairly clear seasonal variability. In June and July 1985, 1986 and 1988, the distribution of larvae was more similar than in August 1986 (Figure 3).

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**Figure 3** Results of catching *M. yessoensis* larvae in 1985, 1986 and 1988 in Whale Bay. a = 07.16.1985; b = 07.24.1985; c = 07.25.1986; d = 08.01.1986; e = 08.05.1986; f = 08.06.1986; g = 06.29.1988; h = 07.12.1988.

In July 1985, 1986 and 1988, the average wind direction was 170.1°; 159.9° and 161.4°, respectively, and in August 1986 it was 155.2°. The southerly wind direction and the currents caused by them in 1985 contributed to a specific circulation leading to the accumulation of M. yessoensis larvae in the northeastern part of Whale Bay and the maximum number of juveniles observed there on the collectors (Figure 4).

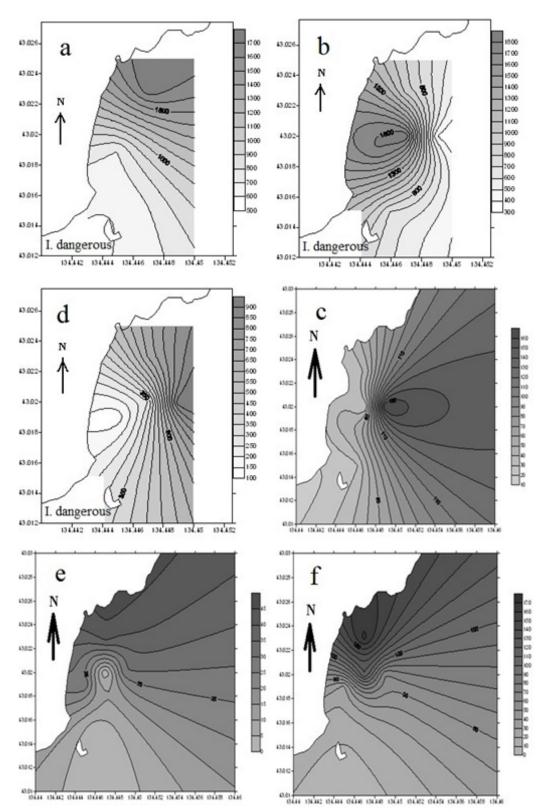


Figure 4 Results of distribution of juveniles of three bivalve species on the collector - cage (CC) in 1985 and 1986 in Whale Bay. a - M. yessoensis in 1985; b - M. trossulus in 1985; d - S. swiftii in 1985; c - M. yessoensis in 1986; e - M. trossulus in 1986 and f - S. swiftii in 1986.

A two-year survey of CC collectors at five stations in Whale Bay showed that, despite strong currents, juveniles exhibit both interspecific and interannual spatial differences. The warm-waterloving M. trossulus is more common in CCs closer to the shore, while the cold-water-loving S. swiftii is more common at those far from the shore (Figure 4). Differences are also observed in the vertical distribution of these mollusks in CCs. In the top 10m, M. yessoensis and M. trossulus showed discordant vertical distribution patterns, below 10meters they were similar. As they approach the sea floor, the abundance of juveniles increases, with S. swiftii being most noticeable (Figure 5).

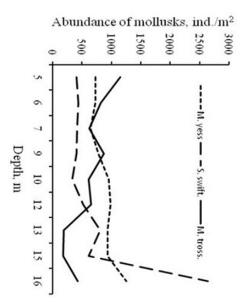


Figure 5 Abundance of juvenile bivalve mollusks of three species in CC at various depths in Whale Bay.

Apparently, M. yessoensis that were transplanted from CC to cages grew slower than those not transplanted, possibly due to being exposed to air during transplantation (Figure 6).

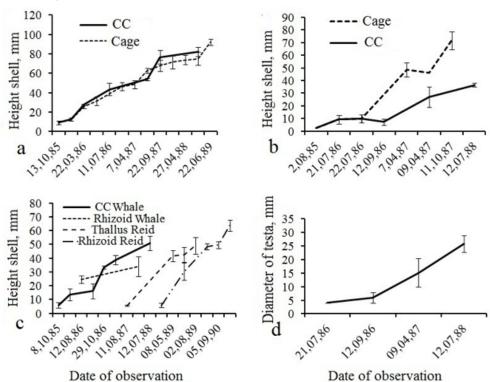


Figure 6 Growth rate of four species of marine invertebrates on artificial substrates and algae in Whale Bay and Reid Pallada Bay: a - growth rate of M. yessoensis in rarefied (cones double spaced) CC without transplantation into cages and transplanted into cages, b - growth rate of S. swiftii in rarefied CC and in cages with M. yessoensis, c - growth rate of M. trossulus in rarefied CC and rhizoids of S. japonica in Whale Bay, and also on thallus and rhizoids of S. japonica in Reid Pallada Bay, d - growth rate of S. swiftii in rarefied CC in Whale Bay

The results of three-year cultivation of *M. yessoensis* in Whale Bay and Posyeta Bay showed that not transplanting mollusks into cages or putting them on the sea floor increases the productivity of the

cultivation technology due to the low survival rate of mollusks on the sea floor and their loss during transplantation (Table 1).

Table I Productivity indicators for M. yessoensis cultivation in Whale Bay and Posyet Bay over three years using three technological schemes

Cultivation technology and Bay	Scallop shell height, mm	Survival, %	The general weight with shell, g	Weight the soft tissue, g	Weight muscle, g	Production, g/m <sup>2</sup>	
CC1 Whale	82.3+1.8	75	58.1+4.2	21.7+1.8	9.5+0.8	14728	
CC <sup>1</sup> Posyeta	84.1+1.7	30.3	76.7+3.9	23.7+3.2	6.0+0.8	11045	
Transplanted <sup>2</sup> Whale	75.1+6.5	55.2	48.6+4.3	18.2+1.7	7.6+1.8	4050	
Transplanted <sup>2</sup> Posyeta	84.4+8.6	56	79.5+8.1	31.8+9.5	13.5+1.5	7950	
Sea floor <sup>3</sup> Posyeta	88.9+1.5	9.9	79.3+4.1	39.8+1.8	15.1+0.7	397	

<sup>1</sup>grown in CC without transplantation; <sup>2</sup>transplanted from CC to cages; <sup>3</sup>yearlings own on sea floor.

Comparison of the growth rates of *M. yessoensis* in cages on the seaweed farms of Whale Bay and Reid Pallada Bay showed a positive

effect of seaweed on this indicator, since it was lower on the scallop farm in Minonosok Inlet (Table 2).

Table 2 Growth rates of four-year-old M. yessoensis in three bays of Primorsky Krai

Nº	Date	H <b>, mm</b>	W gen. g	W soft tiss. g	W muscle, g	W gonad, g
I	03.06.89.	94.0 <u>+</u> 9.07	84,9 <u>+</u> 23,27	27,63 <u>+</u> 7,23	9,76 <u>+</u> 2,86	2,44 <u>+</u> 0,71
2	09.06.89	89.5 <u>+</u> 4.26	84.6 <u>+</u> 11.18	34.88 <u>+</u> 4.77	15.95 <u>+</u> 2.39	2.53 <u>+</u> 0.82
3	22.06.89	92.4 <u>+</u> 8.62	85.5 <u>+</u> 26.6	33.01 <u>+</u> 14.07	12.13 <u>+</u> 5.34	4.65 <u>+</u> 3.82

Note: I - Reid Pallada Bay, 2 - Minonosok Inlet, 3 - Whale Bay

S. swiftii, like other animals, showed density-dependent growth. It was accidentally transplanted together with M. yessoensis into cages, and the low density improved its living conditions and the growth rate exceeded those in the rarefied CC (Figure 6b). M. trossulus sitting on rope with S. japonica in Reid Pallada Bay (Figure 7) showed the highest growth rate compared to those sitting on scallop cages and mussel collectors. The shell height of two-year-old individuals

was  $49.3 \pm 2.13$ ;  $37.2 \pm 3.09$  and  $35.6 \pm 3.74$  mm, respectively. On rope with the harvested *S. japonica*, the rhizoids did not deteriorate for another year and *M. trossulus* achieved record results there. The height of three-year-old individuals was  $63.65 \pm 4.22$  mm. In Whale Bay, *M. trossulus* on *S. japonica* thallus had a similar growth rate to those in the rarefied CC and outpaced those in the rhizoids of *S. japonica* (Figure 6).



Figure 7 M. trossulus living in the rhizoids of S. japonica.

Despite the harvesting of the spawners in Whale Bay, the larvae of the sea urchin *S. intermedius* successfully colonized CC, where the abundance of juveniles at deep-water station 2 reached 353.2 ind./m². They attacked juvenile bivalves in groups, and their shell diameter on the rarefied CC reached 30 mm over three years (Figure 6).

The indentations on the thallus of *S. cichorioides* facilitate the colonization and growth to commercial size of the *M. trossulus* larvae, since the thallus do not disintegrate for three years. The secretions of this alga (kelp) are apparently food for the mollusks, since, despite the high settlement density, *M. trossulus* were larger than those sitting on artificial substrates (Table 3).

Table 3 Comparative analysis of several collector designs and S. cichorioides thallus on the abundance and growth of M. trossulus

Time of observations	Tallus of S. cichorioides		Artificial substrates							
			collector - cage with cover		Collector – cage without cover		Mussel collector			
	Abundance M.trossulus ind./ m <sup>2</sup>	altitude shell <i>M.trossulus</i> mm	Abundance M.trossulus ind./ m²	altitude shell <i>M.trossulus</i> mm	Abundance M.trossulus ind./m²	altitude shell <i>M.trossulus</i> mm	Abundance M.trossulus ind./m²	altitude shell <i>M.trossulus</i> mm		
09.08.84	20336,6	6,1+0,6	15120,0		10566,6	_	19831,0			
16.10.84		19,5+1,0		15,1+1,1		19,3+1,4		18,7+1,9		
31.05.85	9242,4	36,2+1,6			550,0	33,8+2,0				

*M. trossulus* on the thallus of *S. cichorioides* stimulated the growth rate of this alga, and by mid-June the length of the uncleaned thallus exceeded those cleared of it. However, the following year, the grown

*M. trossulus* covered the thallus completely, which led to their growth retardation (Figure 8).

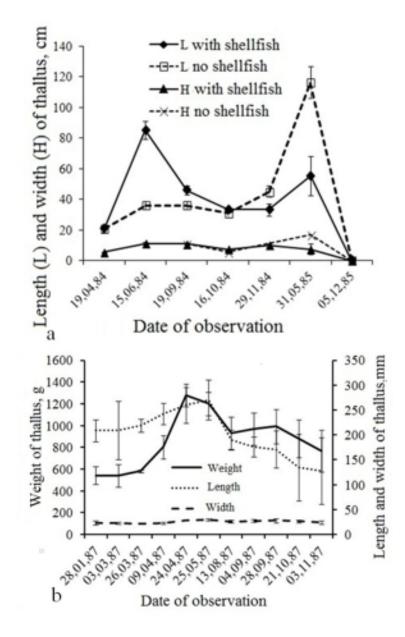


Figure 8 a - growth rate of thallus of S. cichorioides with and without M. trossulus, b - growth rate of S. japonica on a marine plantation in Whale Bay.

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Favorable conditions for the existence of algae caused by the presence of animals in the rhizoids near the algae growth zone are confirmed by the fact that the transplantation of *S. japonica* to new ropes in Reid Pallada Bay twice led to the complete death of the thallus. Only the unreplanted and sparse thallus survived.

During the process of cultivating mollusks in Whale Bay, the concentration of phosphorus, silicon, and nitrites in the water increased (Table 4).

Table 4 Results of hydrochemical observations in Whale Bay in 1985-1988

Date	T °C, 0 <b>m</b>	SALINITY, PSU	0,		pΗ, μ <b>g/l</b>	Alk, μg/l	Po <sub>4</sub> -P <b>, μg/l</b>	Pcom., µg/l	SiO₃-si, μg/l	NO <sub>2</sub> .Ν, μg/l	NO₃-N, μg/l	NO <sub>4</sub> .N, μg/l
			ml/l	%	_							
22.07.85	16.1	30.44	6.94				0.5	1.5	300	0,3	7,2	44,0
21.08.85	19,1	31,82	6,46	120,7	7,46	2,075	0	4,5	0	1	0	44,0
11.09.85	18,0	33,75	6,37	118	8,11	2,028	9	10	160	0,9	13,0	44,0
10.10.85	12,8	32,62	5,99	99,1	8,09	2,242	4,5	6	0	2,7	6,3	0
31.07.86	12,3	35,41	6,53				0,0	16,0	430	0,6	4,7	0
19.08.86	16,2	32,95	6,73	119,8	8,68	2,315	0,0	5	800	2,1	30,0	0
11.09.86	16,6	33,49	6,5 I	117,1	8,80	2,419	2	4	580	1,7	15,0	0
10.10.86	13,5	33,58	7,06	119,5	8,88	2,344	1	5	0	0,9	16,0	0
30.07.87	16,6	34,22	8,79				0,0	11,0	400	0,0	3,0	163
12.07.88	15,6	33,46	6,28				4,2	5,5	550	0,3		2,0

It is likely that the active consumption of biogens from the water contributed to the fact that the commercial thallus in Whale Bay reached record sizes (Figure 8). By the time of harvesting of *S. japonica*, the weight of thallus from 40 ropes (1ha) was 170 tons, waste from processing the raw material was 12%, and the marketable output reached 150 tons with an average rope weight of 42kg.

# **Discussion**

Integrated multitrophic aquaculture (IMTA) is a production method based on an ecosystem approach that was developed to address the problems of marine pollution caused by fish farming. It is a polyculture system that combines the production of forage species as the primary commodity (e.g. pelagic fish), organic extractive species (e.g. benthos feeding on sediments and feces), and inorganic extractive species (e.g. seaweed). The method of culturing blue mussels *M. edulis* and *M. trossulus* in close proximity to salmon cages in IMTA systems will be a useful practice for the disposal of solid waste and feces. The use of seaweeds allows for the creation of a balanced forage ecology taking into account the carrying capacity, site selection and food safety. If

The interannual and interspecific variability of the spatial distribution of larvae in Whale Bay are apparently related to the peculiarities of the direction of the prevailing winds and different thermopathies of bivalve mollusks. In July 1985, 1986 and 1988 the average wind direction was 170.1°; 159.9° and 161.4°, respectively. It is possible that the "southern" direction of the winds and the currents caused by them contributed to the specific circulation causing the concentration of late larvae in 1985 in the northeastern part of Whale Bay and the maximum number of juveniles observed there on the collectors. <sup>27</sup> Spawners of the *M. yessoensis* also live on the bottom there. <sup>28</sup> The record number of scallop larvae on August 1, 1986, near Cape Misty (Figure 3) was apparently also caused by the wind direction during this period. Due to the river runoff and mixing, there are more biogens and, accordingly, phytoplankton in this area, which can increase the survival rate of the settled larvae. <sup>29</sup>

The variability of the vertical distribution of bottom animals – inhabitants of coastal waters – is the result of specific adaptation of their cells to the temperature of the environment and is associated with

differences in protein structure.<sup>30</sup> Apparently, intensive mixing in the open Whale Bay contributes to the fact that the vertical distribution of juveniles on the collectors is more uniform than in Posyeta Bay.<sup>28</sup>

Due to the poor viability of M. trossulus, it accumulates away from the sea floor in calmer waters often found with artificial substrates.<sup>31</sup> Therefore, near ports and marine plantations there are more of its mature individuals producing more larvae around anthropogenic structures.<sup>32</sup> Indentations on the thallus of S. chichorioides, create microscale turbulence,<sup>33</sup> which facilitates the attachment of larvae and high abundance of mussels (Table 3). These results allow us to recommend this alga, as well as the rhizoids of S. japonica, as substrates for collecting and growing M. trossulus. After the harvest of S. japonica, the algal plantation remains unused until spring. During this time, two-year-old M. trossulus in the rhizoids of S. japonica in Whale Bay will reach a commercial size of 40mm, since already in the fall the shell height of two-year-old individuals there was  $33.29 \pm 1.3$ mm, and its total weight was 32 tons per hectare.<sup>34</sup>

Scallop collectors of Japanese design are an ecological trap for other species. In the CC in Whale Bay, in addition to *M. yessoensis*, the species *M. trossulus*, *S. swiftii* and the sea urchin *S. intermidius* reached great abundance. At the time of transplantation of *M. yessoensis* into cages, these other species are very small and are discarded during sorting, since, for example, urchins become suitable for sowing on the bottom only in the third year of life, when the shell reaches 15mm in diameter.<sup>35</sup> In CC with 15mm cover holes they, together with *M. yessoensis*, reach commercial or viable size, thereby increasing the efficiency of the culture process.

The combined cultivation of algae and mollusks optimizes the process of cultivating of *S. japonica*, since the yields we obtained are higher than previous mono-culture the previous yields in Whale Bay. Previously, the length of the thallus in March fluctuated from 166 to 180 cm, the weight from 270 to 340 g.,<sup>22</sup> and the yield was 60-100 tons per hectare.<sup>36</sup> At this time, our average length was 219 cm, the weight was 582 g (Figure 8), and the yield was 170 tons per hectare.

We conclude that mollusk producers in Primorsky Krai have a unique opportunity to increase production by replacing mono culture of marine species with poly culture of sympathetic species.

# **Acknowledgement**

None.

### **Conflicts of interest**

We declare that there is no conflict of interest of any kind.

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