

## アユの蓄積脂質に及ぼす流速の影響

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## Effects of Water Velocities on Lipid Reserves in Ayu

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Ayu *Plecoglossus altivelis* of 12 cm in body length were raised in concrete tanks at four different water velocities (4 cm/s, 25 cm/s, 35 cm/s, and 45 cm/s) for 60 days. Rearing of the fish in water velocities 35 and 45 cm/s exerted favorable growth and feed efficiency. Rearing in current water, however, failed to depress lipid content of muscle, liver, and intraperitoneal fat body when compared to that in the standing water (4 cm/s).

Following the feeding experiment, the fish were starved for 13 days in indoor tanks. The body weight loss was suppressed in the fish reared in water velocities of 35 and 45 cm/s. Rearing in high water velocity (<45 cm/s) could improve lipid metabolism, especially in activation of utilization of lipid reserves for energy requirements.

In ayu culture, water velocity in the rearing tank should be controlled to 45 cm/s or more.

In recent years there have been several studies on the effects of sustained exercise on growth, survival, feed efficiency, and physiological condition of salmonids.<sup>1-7)</sup> A pronounced effect of moderate sustained exercise exerted a physiological condition associating to the elevation of muscle protein assimilation.<sup>4,9)</sup>

The culture of ayu *Plecoglossus altivelis* merits high market value and high growth rate. However, deterioration of the fish quality has been arisen in proportion to elevation of productivity. Ohya *et al.*<sup>9)</sup> reported that water drawing system with current could improve productivity and physiological condition in ayu. Therefore the purpose of the study was to examine the effects of different water velocities of rearing tanks upon the states of lipid reserves, which is an important factor for carcass quality.

### Materials and Methods

Juvenile ayu caught in the Biwa lake were transferred to Fisheries Laboratory of Kinki University (Shingu). About 8,000 fish with average body weight 24.3 g were divided into four groups and taken in concrete tanks (5×5×0.6 m) with different water velocities. The tanks were equally supplied with fresh water (300 l/m). A standing

water condition was created by vertical pouring through a perforated inlet pipe.<sup>9)</sup> Different levels of water velocity were created by forcing the water through horizontally mounted inlet pipes with or without water pump. Average water velocities of four groups were 4 cm/s (standing water), 25 cm/s (low velocity), 35 cm/s (medium velocity), and 45 cm/s (high velocity). These water velocities correspond to 0.24, 1.5, 2.1, and 2.7 body length/s (L/s) at the end of experiment, respectively. The water temperature ranged 12.7-18.5°C (average 15.6°C). The fish were fed with a commercial diet (Nihon Nosan Co., Ltd.; crude protein 54.1%) by automatic feeders twice a day at approximately 4-5% of body weight for 60 days.

After feeding experiment, twenty fish taken out from each group were submitted to a starvation test in indoor plastic tanks at 18°C for 13 days under the same condition.

At the end of feeding experiment and starvation test, muscle, liver, and intraperitoneal fat body (IPF) were analyzed for lipid content and lipid class composition. The analysis was carried out in the pooled sample obtained from five fish. Lipid was extracted with methanol-chloroform mixture.<sup>10)</sup> Lipid class composition was measured by an Iatroscan (Iatron Co., Ltd.). The analysis

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was carried out for the four parts of muscle, as shown in Fig. 1.

Mean values were compared by Student's *t*-test and differences at 5% level were considered significant. Significance of difference among groups were analyzed using Duncan's multiple range test.

### Results

The mortality of the fish in all groups was less than 11% during the feeding experiment (Table 1). The fish in standing water (4 cm/s) were swimming in all directions. Bursts of swimming in surprise were occasionally observed. The fish acclimated in more than 25 cm/s water velocities held their positions against the water current with keeping at regular spaces.

Table 1 presents growth and biological characteristics of fish at the end of feeding experiment. A significant difference was found in body length

as well as in body weight and condition factor among the groups. The body weight gain in higher water velocities exceeded that in the standing water. The best weight gain was in 35 cm/s and 45 cm/s. High body weight gain was accompanied by high feed efficiency (weight gain/total diet fed  $\times 100$ ). Water velocity had no effect on muscle ratio, hepatosomatic index, or IPF ratio.

Lipid contents of muscle, liver, and IPF at the end of feeding experiment and starvation test are shown in Table 2. The analyses of muscle were carried out in four different parts as shown in Fig. 1. While there was no marked difference in lipid content of muscle lipid among the groups, the values of parts A and C were variable in response to the water velocities. The lipid level was the highest in part A which mainly consists of imperfect neural spine flesh, and extremely low in parts B and C. Lipid content of IPF was not influenced by water velocity, whereas the low

Table 1. Effect of water velocity on growth and characteristics of ayu

	n	Average water velocity (cm/s)			
		4	25	35	45
Body length (cm)	105	16.0 $\pm$ 1.0 <sup>a</sup>	16.4 $\pm$ 1.2 <sup>b</sup>	16.6 $\pm$ 1.0 <sup>b</sup>	16.6 $\pm$ 1.1 <sup>b</sup>
Body weight (g)	105	67.1 $\pm$ 13.6 <sup>a</sup>	72.6 $\pm$ 16.1 <sup>b</sup>	79.9 $\pm$ 15.2 <sup>c</sup>	78.0 $\pm$ 15.2 <sup>c</sup>
Condition factor	105	16.3 $\pm$ 1.9 <sup>a</sup>	16.1 $\pm$ 1.4 <sup>a</sup>	17.1 $\pm$ 1.1 <sup>b</sup>	16.9 $\pm$ 1.1 <sup>b</sup>
Muscle ratio (%)	5	46.7 $\pm$ 4.6	46.0 $\pm$ 1.5	47.8 $\pm$ 1.1	46.3 $\pm$ 2.5
Hepatosomatic index (%)	5	2.5 $\pm$ 0.4	2.9 $\pm$ 0.4	3.1 $\pm$ 0.4	2.6 $\pm$ 0.3
Intraperitoneal fat body ratio (%)	5	8.2 $\pm$ 2.7	8.8 $\pm$ 0.5	8.6 $\pm$ 1.0	8.3 $\pm$ 0.5
Feed efficiency (%)		35.8	43.4	46.6	46.0
Mortality (%)		10.5	9.2	9.8	6.1
Daily growth rate (%/day)		1.46	1.71	1.80	1.75

Mean  $\pm$  SD.

Initial fish size: body length 11.8  $\pm$  1.0 cm, body weight 24.3  $\pm$  6.6 g, condition factor 14.6  $\pm$  1.2.

Values in the same line followed by different superscript letters are significantly different ( $p < 0.05$ ).

Table 2. Effect of water velocity on lipid content\* of fed (F) and starved ayu (S)

	Average water velocity (cm/s)							
	4		25		35		45	
	F	S	F	S	F	S	F	S
Muscle								
Whole (%)	9.8	12.1	10.8	9.9	9.9	12.1	9.4	10.6
Part A (%)	14.6	22.0	25.8	17.7	15.5	23.1	11.3	17.9
Part B (%)	6.6	6.8	6.0	6.4	6.2	6.6	6.3	8.0
Part C (%)	7.4	7.1	2.9	4.5	5.3	6.3	5.9	6.9
Part D (%)	10.2	11.1	8.2	9.5	10.6	11.1	11.9	9.1
Liver (%)	8.3	9.3	10.5	8.6	8.7	8.7	6.7	8.2
Intraperitoneal fat body (%)	76.0	84.3	75.2	75.2	73.6	75.5	77.5	35.1

\* Wet basis. Pooled samples obtained from 5 fish were analyzed.

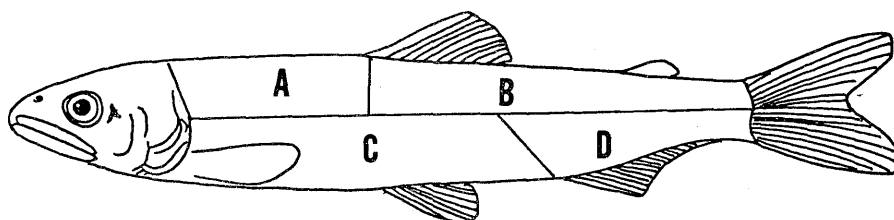


Fig. 1. Diagram showing different parts of muscle analyzed. A, anterior dorsal part; B, posterior dorsal part; C, anterior ventral part; D, posterior ventral part.

Table 3. Lipid class composition (%)\* of muscle, liver, and intraperitoneal fat body after feeding (F) and starvation (S)

Velocity	CE		TG		FA		Cho		PG		PL	
	F	S	F	S	F	S	F	S	F	S	F	S
Muscle												
4 cm/s	tr	tr	93.9	95.8	0.7	0.8	0.6	0.4	tr	tr	4.8	3.0
25 cm/s	tr	tr	93.5	93.2	1.0	1.8	0.9	0.5	tr	tr	4.7	4.6
35 cm/s	tr	tr	93.8	94.8	0.7	0.8	0.5	0.7	tr	tr	5.1	3.8
45 cm/s	tr	tr	93.3	95.7	0.5	0.8	0.8	0.6	tr	tr	5.5	2.9
Liver												
4 cm/s	1.9	2.1	43.8	14.0	53.8	61.1	0.5	5.2	tr	tr	tr	17.6
25 cm/s	2.6	3.7	44.1	16.5	32.4	57.8	4.7	4.6	tr	tr	16.2	17.4
35 cm/s	1.2	0.7	36.5	12.9	61.9	56.4	0.4	3.1	tr	tr	tr	26.9
45 cm/s	0.8	1.4	15.4	28.2	69.2	54.8	3.0	1.6	tr	tr	12.6	14.0
Intraperitoneal fat body												
4 cm/s	tr	tr	98.6	96.1	tr	2.1	1.0	1.2	tr	tr	0.4	0.6
25 cm/s	tr	tr	97.6	96.0	tr	2.0	2.4	2.0	tr	tr	tr	tr
35 cm/s	tr	tr	96.5	96.7	tr	1.3	2.2	1.3	tr	tr	1.3	0.7
45 cm/s	tr	tr	96.2	97.8	tr	1.1	3.2	1.1	tr	tr	0.6	tr

CE, cholesterol esters; TG, triglycerides; FA, fatty acids; Cho, cholesterol; PG, partial glycerides; PL, phospholipids; tr, trace.

\* Pooled samples obtained from 5 fish were analyzed.

Table 4. Effect of water velocity on characteristics of ayu after starvation

	n	Average water velocity (cm/s)			
		4	25	35	45
Body weight (g)	10	46.3±5.6 <sup>a</sup> (83.7)*	56.2±9.5 <sup>b</sup> (79.7)*	58.0±9.0 <sup>b</sup> (89.8)	54.4±10.6 <sup>b</sup> (97.8)
Condition factor	10	12.7±0.6 (85.8)*	13.0±0.9 (86.7)*	13.0±0.4 (90.3)*	13.1±0.6 (89.7)*
Muscle ratio (%)	5	50.8±3.1 <sup>a</sup> (109)	46.5±2.8 <sup>a,b</sup> (101)	49.7±3.4 <sup>a</sup> (104)	45.7±1.5 <sup>b</sup> (98.7)
Hepatosomatic index (%)	5	0.9±0.2 (36.4)*	0.9±0.1 (30.8)*	0.7±0.1 (23.8)*	0.8±0.3 (30.7)*
Intraperitoneal fat body ratio (%)	5	7.3±0.8 (89.7)	8.6±1.4 (97.9)	7.1±1.0 (82.5)*	7.4±1.1 (88.7)

Mean±SD.

Parentheses mean percentage when the value of fed ayu expressed as 100.

Values in the same line followed by different superscript letters are significantly different ( $p<0.05$ ).

\* Significantly different from the value before starvation ( $p<0.05$ ).

**Table 5.** Effect of water velocity on lipid reserves in fed (F) and starved ayu (S)

Lipid reserves (g/fish)	Average water velocity (cm/s)							
	4		25		35		45	
	F	S	F	S	F	S	F	S
Muscle	3.07	2.85	3.61	2.59	3.78	3.49	3.39	2.64
Liver	0.14	0.04	0.22	0.05	0.22	0.03	0.09	0.04
Intraperitoneal fat body	4.16	2.86	4.78	3.62	5.04	3.60	5.02	1.41
Total	7.37	5.75	8.61	7.28	9.04	7.12	8.50	4.09

The amounts of lipid reserves were calculated from the values in Tables 1, 2, 3, and 4.

water velocity resulted in a slight elevation in liver lipid.

After 13 days starvation, the lipid content did not always seem to decrease. However, the decrease of IPF lipid was remarkable, especially in 45 cm/s.

Table 3 shows lipid class composition of muscle, liver, and IPF at the end of feeding experiment and starvation test. In the table, the value of whole muscle is presented, because the composition represented little difference in every part of muscle of which lipids comprised more than 88% of triglycerides (TG). Liver lipid was characterized by high proportion of free fatty acids (>32%). The proportion of TG was more than 43% in the standing water and 25 cm/s, and lowered with increasing water velocity. Over 96% of TG were included in IPF which was main energy reserves. The proportion was not affected by water velocity.

While the lipid class composition did not change in muscle by starvation, degradation of TG and increase of free fatty acids were found in liver. As a result, the proportion of free fatty acids exceeded TG in liver. In IPF, small amounts of free fatty acids appeared after starvation.

Effects of starvation on some biological parameters are shown in Table 4. Body weight loss during starvation was quite different among the groups. The body weight decreased significantly in the groups reared in the standing water and the low velocity. A significant decrease of condition factor was observed in all groups. The muscle ratio was not affected. Hepatosomatic index of all groups significantly decreased by starvation, but IPF ratio decreased in different manner in response to the water velocity. Significantly high IPF loss was found only in the medium velocity.

Table 5 shows the total amount of lipid in a

fish. After 13 days starvation, the lipid reserves markedly decreased. The starvation represented some differences in lipid mobilization among the groups. The fish reared in the high velocity greatly lost lipid of IPF and minimized body weight loss during starvation. The response of lipid level to starvation varied depending on parts of muscle.

### Discussion

In our previous work, it was suggested that drawing system of rearing tank which caused water current improved growth, body condition and some physiological conditions in ayu.<sup>9)</sup> The merits of the water current would relate to water quality including oxygen supply. Furthermore, current may contribute to suspend pelleted feed which influences the efficiency of feed utilization.

A number of studies on the effects of exercise in fish have been carried out in muscle morphology and muscle protein turnover.<sup>3-5,8,11-14)</sup> Increase in protein synthesis and hypertrophy of muscle resulted from exercise exerted growth.<sup>1,3,8,15,16)</sup> Although the increase in muscle mass was not achieved by increased velocity in this experiment, lipid metabolism was improved at higher velocities (>35 cm/s). Increasing swimming speed resulted in a decrease in muscle lipid.<sup>7)</sup> However, in this experiment, water velocity which might elevate energy expenditure could not depress lipid reserves, as reported by Poston *et al.*<sup>17)</sup> The decrease in muscle lipid might not only be a response to increasing energy expenditure, but also be associated with increasing assimilation rate of dietary protein. The assumption could be supported by the fact that fish acclimated in running water could accumulate higher energy reserves than those in still water.<sup>3,18)</sup>

Retention of muscle protein and exhaustion of

reserved lipids during starvation were relatively high in the fish raised in high current condition. High water velocity seemed unlikely to suppress lipid accumulation, nevertheless the reserved lipids would be active to be mobilized and could be preferentially utilized as energy prior to muscle protein. In the previous paper,<sup>9)</sup> activity of lipid mobilization was higher in fish reared in running water than standing water. Therefore, rearing with a certain water velocity may be advantageous in growth, feed efficiency, and carcass quality, considering the fact that wild ayu can survive in higher water current (> 100 cm/s). However, some expenses such as electricity for controlling water velocity would be a restricting factor.

Recently, improvement of carcass quality without depression of growth and feed efficiency has been requested. In ayu culture, the water velocity in rearing tanks should be controlled to 45 cm/s or more.

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