ワカサギ卵の孵化に及ぼす水温と塩分の影響

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Effects of Temperature and Salinity on Egg Hatch of the Pond Smelt *Hypomesus olidus*

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Responses of eggs of the pond smelt *Hypomesus olidus* to a series of temperature and chlorinity combinations were measured for percentages of total- and viable-hatch and time to 50% hatch.

Hatch occurred at the combinations of temperature range of 10 to 22.5°C and chlorinity range of 0 to 10‰. The relationships between hatching rate and two variables were expressed by the second order equations:

$TH=37.9974+6.2171\ T-0.3220\ T^2-3.1775\ C-0.6297\ C^2+0.3355TC$

$VH=55.4947+3.1276\ T-0.2344\ T^2-5.3836\ C-0.5386\ C^2+0.4340TC$

where TH=total-hatch (%), VH=viable-hatch (%), T=temperature(°C), C=chlorinity (‰). Response surfacee analysis suggests that the viable-hatch occurred safely at temperature lower than 19.0°C and at chlorinity lower than 6.7‰.

Time required to 50% hatch varied between 7.3 and 23.3 days after fertilization, and was inversely and exponentially related to the temperature. The effect of chlorinity on the time was statistically insignificant within the hatchable range. The relationship equation was third order:

$D=131.3675-19.5853\ T+1.0729\ T^2-0.0202\ T^3$

where D=days to 50% hatch.

Key words: temperature, salinity, fish egg, pond smelt, *Hypomesus olidus*

Temperature and salinity are factors which have a major effect on the development of teleosts eggs.[1,2] This is one of our papers concerning the effects of these variable on the early life stages of the fishes. Those papers which have been published refer to threeline grunt *Parapristipoma trilineatum*,[3,4] amago salmon *Oncorhynchus masou macrostomus*,[5] ayu *Plecoglossus altivelis*,[6] and rainbow trout *Salmo gairdneri*.[7]

The present paper deals with the pond smelt *Hypomesus olidus* which is widely distributed in the coastal and inland water regions throughout Japan, and furnishes an important catch for both fisheries and recreational fishing located there. This study was designed to determine the percentages of total- and viable-hatch and time to 50% hatch of the eggs at various combinations of temperature and salinity. In this connection, there are few reports on the present species other than those which HIGURASHI & NAKAI,[8] NAKAI,[9] and YAMAMOTO[10] have noted for the temperature effect only.

Materials and Methods

The adult pond smelts were caught from the lake Suwa, Nagano Prefecture, on April 9, 1984. The eggs,

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obtained from several ten ripe fish, were fertilized routinely and attached to collectors of china palm at temperature of 10°C. The collectors kept at the same temperature were transported to our laboratory at Tsu and subdivided there into small pieces which contained average of 390 (range 189–793) eggs.

The experimental design used for the present study was a 2 factor 7×6 factorial with a temperature range of 10 to 25°C in 2.5°C intervals and a chlorinity range of 0 to 10‰ in 2‰ intervals (Table 1). A subsample of eggs was put into a 50 mm diameter glass beaker with 100 m*l* water. A set of six beakers with each test chlorinity was soaked in each test temperature bath, constantly controlled within ±0.3°C. The beakers were checked more than two times every day, and dead eggs in each beaker were counted and removed from it. The test chlorinities were obtained by dilution of Allen's synthetic sea salts* with freshwater. The test water was changed every three days to keep the desired chlorinities. A slight aeration was used in all these incubations.

The percentages of total- and viable-hatch and the time to 50% hatch were determined. The objects of observation for viable-hatch were limited to the normal larvae hatched without any abnormalities such as the curvatures of tail or notochord and the lack of eyes. The time to 50% hatch was calculated by interpolation. For these data, the statistical treatments followed the analysis of variance of two-way layout,[11] and the relationships between hatching rate, time and variables were calculated by the methods of orthogonal polynomials and shown as the response surfaces.[12]

Results

Percentages of total-hatch

Results in Table 1 show that the total-hatch occurred at the combinations of temperature range of 10 to 22.5°C and chlorinity range of 0 to 10‰. The percentages varied between 0 and 70.1% at all combinations. The analysis of variance for these data proved to be highly significant for both temperature and chlorinity (Table 2). The relationship between hatching rate and variables was expressed by the second order equation:

Table 1. Percentages of Total and Viable Hatch of the Pond Smelt Eggs Incubated at 42 Temperature-Salinity Combinations

Temp. (°C)	Chlorinity (‰)											
	0		2		4		6		8		10	
	TH	VH	TH	VH	TH	VH	TH	VH	TH	VH	TH	VH
25.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
22.5	5.4	2.8	4.8	2.3	3.3	1.2	0.9	0.4	0.0	0.0	0.0	0.0
20.0	43.6	11.1	52.1	37.4	54.9	33.7	39.9	24.4	15.5	15.2	0.0	0.0
17.5	55.8	38.6	57.0	48.3	55.0	46.2	39.9	35.2	24.1	19.3	6.0	2.8
15.0	56.4	55.0	57.1	52.2	62.3	48.4	46.2	39.7	35.8	23.3	8.7	4.4
12.5	58.1	57.3	70.1	62.6	61.1	53.1	48.5	42.5	37.4	27.2	12.0	6.0
10.0	60.3	53.7	60.6	53.7	58.4	52.5	51.2	37.3	36.0	13.1	4.4	1.2

TH: percentages of total-hatch. VH: percentages of viable-hatch.

* Formula for the salts (Cl=20.127‰): NaCl 28.17 g, $MgCl_2$ 2.55 g, KCl 0.77 g, $CaCl_2$ 1.20 g, $MgSO_4$ 3.50 g, $NaHCO_3$ 0.22 g, Water to 1,000 ml.

$TH=37.9974+6.2171\ T-0.3220\ T^2-3.1775\ C-0.6297\ C^2+0.3355TC$

where TH=total-hatch (%), T=temperature(°C), and C=chlorinity (‰). From this equation, the response surface was constructed and shown as the isopleths of percentages for a selected level such as 60, 50,…and 0% (Fig. 1). The maximum percentage was 68% at temperature of 10°C coupled with chlorinity of 0‰ (point S in Fig. 1). Both increases and decreases in the two variables from these values reduced the percentages of total-hatch. The limiting temperature (LT_{50}) and chlorinity (LC_{50}) for hatch were determined as the values at the isopleth of 34% hatch which is a half of the maximum percentage (dotted line in Fig. 1). The highest LT_{50} and LC_{50} were 20.6°C and 8.0‰, respectively.

Table 2. Analysis of Variance for the Data of Total-Hatch

SV	SS	DF	MS	F
Temperature	15,139.51	6	2,523.25	24.40**
Linear	11,955.10	1	11,955.10	115.62**
Quadratic	2,040.88	1	2,040.88	19.74**
Remainder	1,143.53	4	285.88	2.76^{ns}
Chlorinity	8,044.70	5	1,608.94	15.56**
Linear	6,361.93	1	6,361.93	61.53**
Quadratic	1,658.09	1	1,658.09	16.04**
Remainder	24.68	3	8.23	0.08^{ns}
Interaction				
Quadratic	1,378.95	1	1,378.95	13.34**
Residual	3,102.03	30	103.401	
Total	26,286.4	41		

**: highly significant ($p<0.01$). ns: not significant.

Percentages of viable hatch

The percentages of viable-hatch varied between 0 and 62.6% (Table 1). The results of analysis of variance showed little difference from the those of the total-hatch (Table 3). The fitted equation was:

$VH=55.4947+3.1276\ T-0.2344\ T^2-5.3836\ C-0.5386\ C^2+0.4340TC$

where VH=viable-hatch (%).

The highest LT_{50} and LC_{50} were 19.0°C and 6.7‰, respectively, and these values were lower 1.6°C and 1.3‰ than those of total-hatch. The hatch of the pond smelt eggs seems to occur safely below these values.

Time to 50% hatch

The data of time to 50% hatch varied between 7.3 and 23.3 days at the combinations of temperature of 10 to 20°C and chlorinity of 0 to 8‰ (Table 4). The analysis of variance for these data proved to be significant for the temperature only (Table 5). The relationship between time and temperature was expressed by the third order equation:

$D=131.3675-19.5853\ T+1.0729\ T^2-0.0202\ T^3$

where D=days to 50% hatch. The regression curve in Fig. 3 showed that the time was inversely and exponentially related to the temperature.

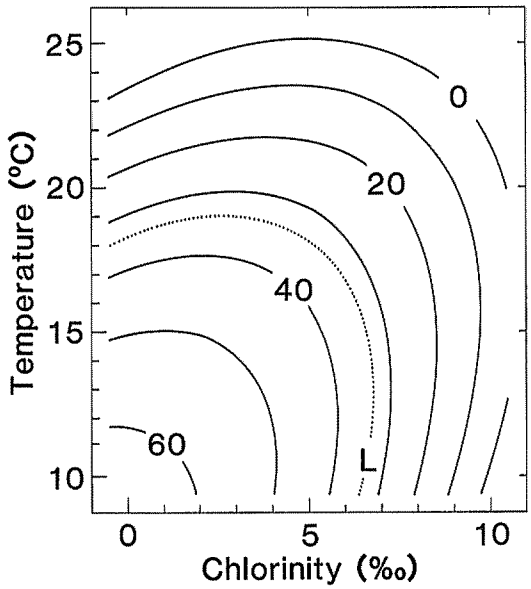

Fig. 1. Response surface showing the isopleths of percentages of total-hatch in relation to water temperature and salinity. Dotted line shows the levels of limiting temperature (LT_{50}) and chlorinity (LC_{50}) for hatch.

Table 3. Analysis of Variance for the Data of Viable-Hatch

SV	SS	DF	MS	F
Temperature	10,692.81	6	1,782.14	16.58**
Linear	8,878.41	1	8,878.41	82.59**
Quadratic	1,082.11	1	1,082.11	10.07**
Remainder	732.29	4	183.07	1.70^{ns}
Chlorinity	6,224.75	5	1,244.95	11.58**
Linear	4,936.65	1	4,936.65	45.92**
Quadratic	1,212.89	1	1,212.89	11.28**
Remainder	75.21	3	25.07	0.23^{ns}
Interaction				
Quadratic	2,307.58	1	2,307.58	21.45**
Residual	3,224.85	30	107.50	
Total	20,142.41	41		

**: highly significant ($p<0.01$). ns: not significant.

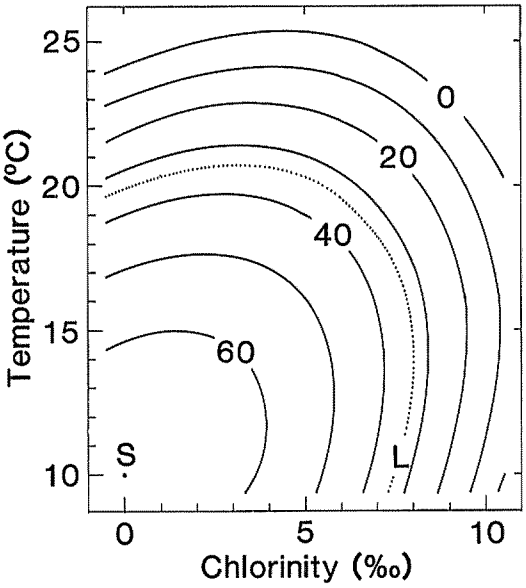

Fig. 2. Response surface showing the isopleths of percentages of viable-hatch in relation to water temperature and salinity. Dotted line shows the levels of limiting temperature (LT_{50}) and chlorinity (LC_{50}) for hatch.

Table 4. Time (days) to 50% Hatch of the Pond Smelt Eggs

Temp. (℃)	Chlorinity (‰) 0	2	4	6	8
20.0	8.0	7.5	7.3	7.3	7.3
17.5	10.0	9.0	9.3	9.3	9.5
15.0	10.5	10.5	10.5	10.5	10.8
12.5	15.5	15.3	15.3	14.8	14.5
10.0	21.5	21.8	23.0	23.3	23.3

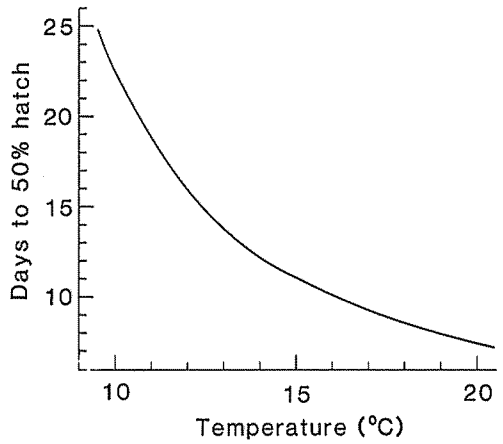

Fig. 3. Regression line showing the time to 50 % hatch in relation to temperature.

Table 5. Analysis of Variance for the Time to 50% Hatch

SV	SS	DF	MS	F
Temperature	726.70	4	181.68	661.48**
Linear	642.97	1	642.97	2,341.05**
Quadratic	75.09	1	75.09	273.40**
Cubic	7.14	1	7.14	26.01**
Remainder	0.31	1	0.31	1.14^{ns}
Chlorinity	0.27	4	0.07	0.25^{ns}
Residual	4.39	16	0.27	
Total	731.37	24		

**: highly significant ($p<0.01$). ns: not significant.

Discussion

Responses of teleost eggs to changing temperature and salinity vary widely with fish species.[1,13] In the present species, HIGURASHI & NAKAI[8], NAKAI, and YAMAMOTO[10] noted in regard to the effect of temperature that hatch occurred safely within the ranges from 6 to 19°C. Our results, in which the limiting temperature for the viable-hatch is lower than 19°C, are well in accordance with their descriptions and newly propose that the limiting chlorinity is lower than 6.7‰.

On the time to hatch, YAMAMOTO[10] reported that the days were 19.6, 13.5, 10.0 and 7.2 days after fertilization when the eggs were incubated at temperatures of 10.4, 13.3, 15.6, and 19.4°C, respectively, and then suggested that the time was inversely and exponentially related to the temperature. The results in this study follow his suggestion. On the other hand, in the present study, the effect of salinity on the time was statistically insignificant within the hatchable range of the eggs. The same effect has been reported on the amago salmon,[5] the rainbow trout,[7] and Pacific herring *Clupea pallasi*[14] while some acceleration or retardation of the time in changing salinities has also been reported on several fish, such as English sole *Parophrys vetulus*,[15] the threeline grunt,[3] Atlantic herring *Clupea harengus*,[16] Pacific cod *Gadus macrocephalus*,[17,18] petrale sole *Eopsetta jordani*,[19] red sea bream *Pagrus major*,[20] and yellowtail flounder *Limanda ferruginea*.[21] It is generally seen that the effect of salinity is small in comparison with that of temperature.[2,22]

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ワカサギ卵の孵化に及ぼす水温と塩分の影響

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ワカサギの受精卵を水温7段階（範囲 10～25°C，間隔 2.5°C）と塩素量6段階（範囲 0～10‰，間隔 2‰）からなる42組の試水中で孵化させた。

その結果，孵化がみられた水温と塩素量はそれぞれ 22.5°C 以下および 10‰ 以下であった。総孵化率TH（%），正常孵化率VH（%），水温T（°C），塩素量C（‰）の関係は次の2次式で表わされた。

$TH=37.9974+6.2171T-0.3220T^2-3.1775C-0.6297C^2+0.3355TC$

$VH=55.4947+3.1276T-0.2344T^2-5.3836C-0.5386C^2+0.4340TC$

これらの式から作られた応答曲面によれば，供試卵の半数が正常に孵化する水温と塩素量の上限はそれぞれ 19°C および 6.7‰ と推定された。

孵化時間は，水温とは高度に有意な負の相関関係（指数関数的）が認められたが，塩素量との関係は有意でなかった。水温 10～25°C における半数孵化時間D（日）と水温T（°C）の関係は次の3次式で表わされた。

$D=131.3675-19.5853\,T+1.0729T^2-0.0202T^3$