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Effect of Temperature and Salinity on Egg Hatch of the Rainbow Trout *Salmo gairdneri*

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Responses of eggs of the rainbow trout *Salmo gairdneri* to a series of temperature and salinity combinations were measured for percentages of total- and viable-hatch and time to 50 % hatch.

Hatch occurred at the combinations of temperature range of 8.0 to 16.0 $^{\circ}$ C and chlorinity range of 0 to 6.0 $^{\circ}$. Response surface analysis suggests that the maximum percentages of total- and viable-hatch (*i. e.*, optimum) may be found at 10.0 $^{\circ}$ C combined with 0 $^{\circ}$.

Time required to 50 % hatch varied 18.5 and 44.6 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 of eggs.

Key words : Temperature, Salinity, Fish egg, Rainbow trout, Salmo gairdneri

Temprature and salinity are factors which have a major effect on the survival of brackish and marine teleosts (KINNE 1964, BLAXTER 1969). This is one of our papers concerning the effects of these variables on the early development of the fishes. Those papers which have been published refer to threeline grunt *Parapristipoma trilineatum* (KASHIWAGI *et al.* 1984), amago salmon *Oncorhynchus masou macrostomus* (LOPES *et al.* 1985), and ayu *Plecoglossus altivelis* (KASHIWAGI *et al.* 1986).

The present paper deals with the rainbow trout *Salmo gairdneri* which is widely cultured in the inland water regions throughout Japan, and determines the percentages of total-and viable-hatch and time to 50 % hatch of the eggs at the various combinations of temperature and salinity.

Materials and Methods

The eggs obtained on December 12, 1986, soon after fertilization from the Shiga

Prefectural Samegai Trout Farm, and then transported to our laboratory at a temperature of 10.2 $^{\circ}$ C, the same as the value at fertilization.

The experimental design was a 2 factor 7×5 factorial with a temperature range of 8.0 to 20.0 °C in 2 °C intervals and a chlorinity range of 0 to 8.0 ‰ in 2.0 ‰ intervals (**Table** 1). Twenty eggs were put into each 50 mmdiameter glass beaker with 100 ml water. Two sets of five beakers each with chlorinity were soaked in the seven test temperature baths, constantly controlled within ± 0.3 °C. When the beakers were checked more than two times every day, any dead eggs were counted and removed. Two replicates of percentages of total- and viable-hatch and the time to 50 % hatch were determined. Here, 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.

Experimental chlorinities were obtained by dilution of ALLEN's synthetic sea salts* with fresh water. The test water was changed every three days to keep the desired chlorinities. Slight aeration was used in all incubations.

For the experimental data, statistical treatments followed the analysis of variance, and the relationships between percentages of total-and viable-hatch and two variables were calculated by the methods of orthogonal polynomial and then shown as the response surfaces (ALDERDICE 1972).

Results

Percentages of total-hatch

Results in **Table 1** show that the total-hatch occurred at the combinations of temperature range of 0 to 16 $^{\circ}$ C and chlorinity range of 0 to 6.0 $^{\circ}$. Analysis of variance for these data proved to be significant for both temperature and chlorinity (**Table 2**). The relationship between total-hatch (TH %), temperature (T $^{\circ}$ C), and chlorinity (C $^{\circ}$) was expressed by the second order equation :

 $TH = 7.7071 + 17.3304T - 0.8705T^2 - 13.4500C$

but where the term of interaction, TC, was omitted because it was statistically insignificant as shown in ANOVA (**Table 2**). From this equation, the response surface was constructed and shown as 90, 80, 70,, and 0%(Fig. 1). The highest percentage was 94.0 % at 10.0 °C and 0% (point S in Fig. 1). Both increases and decreases in the two variables from these values reduced the percentages of total-hatch.

Percentages of viable-hatch

The percentages of viable-hatch showed little difference from those of total-hatch (**Tables** 1 and 3). The fitted equation was :

* Formula for the salts (Cl=20.127 ‰) : NaCl 28.17 g, MgCl₂ 2.55 g, KCl 0.77 g, CaCl₂ 1.20 g, MgSO₄ 3.50 g, NaHCO₃ 0.22 g, Water to 1,000 ml.

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Temp					Chlori	nity (9	60)			
(℃)		0	2		4	ŀ	6	; ;	8	
	TH	VH	TH	VH	TH	VH	TH	VH	TH	VH
20.0	0	0	0	0	0	0	0	0	0	0
	0	0	0	0	0	0	0	0	0	0
18.0	0	0	0	0	0	0	0	0	0	0
	0	0	0	0	0	0	0	0	0	0
16.0	90	80	0	0	0	0	0	0	0	0.
	80	70	0	0	0	0	0	0	0	0
14.0	80 80	80 80	65 30	50 15	45 40	40 35	0	0 0	0 0	0 0
12.0	95	95	80	80	40	35	10	10	0	0
	80	80	45	35	0	0	0	0	0	0
10.0	95	90	90	90	75	70	5	5	0	0
	75	75	80	80	50	30	0	0	0	0
8.0	85	75	80	80	65	55	0	0	0	0
	75	75	80	75	5	0	0	0	0	0

Table 1. Percentages of total and viable hatch of the rainbow trout eggs incubated at 35 temperature-salinity combinations

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

Table 2. Analysis of variance for the data of percentages of total-hatch at the combinations of temperature range of 8.0 to 16.0 °C and chlorinity range of 0 to 6.0 %.

Source	SS	DF	MS	F	-
Treatments	50,165.00	19	2,640.26	11.30**	
Temperature	6,040.00	4	1,510.00	6.46**	
Linear	4,061.25	1	4,061.25	17.37**	
Quadratic	1,358.04	1	1,358.04	5.81*	
Remainder	620.71	2	310,36	1.33 ^{ns}	
Chlorinity	36,275.00	3	12,091.67	51.73**	
Linear	36,180.50	1	36,180.50	154.78**	
Remainder	94.50	2	47.25	0.20 ^{ns}	
Interaction	7,850.00	12	654.17	2.80*	
Quadratic	72.25	1	72.25	0.31 ^{ns}	
Residual	4,675.00	20	233.75		
Total	54,842.00	39			

Asterisk : significant (*p<0.05, **p<0.01). ns : not significant.

Table 3. Analysis of variance for the data of percentages of viable - hatch at the commbinations of temperature range of 8.0 to 16.0 $^\circ\!C$ and chlorinity range of 0 to 6.0 ‰.

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Source	SS	DF	MS	F
Treatments	46,606.88	19	2,452.99	9.89**
Temperature	5,685.00	4	1,421.25	5.73**
Linear	3,920.00	1	3,920.00	15.80**
Quadratic	1,358.04	1	1,358.04	5.47*
Remainder	406.96	2	203.48	0.82 ^{ns}
Chlorinity	33,741.88	3	11,247.29	45.33**
Linear	33,670.13	1	33,670.13	135.70**
Remainder	71.75	2	35.88	0.14^{ns}
Interaction	7,180.00	12	598.33	2.41*
Quadratic	196.00	1	196.00	0.79 ^{ns}
Residual	4,962.50	20	248.13	
Total	51,569.38	39		

Asterisk : significant (*p<0.05, **p<0.01). ns : not significant.





Fig. 1. Response surface showing the isopleths of percentages of total-hatch in relation to temperature and chlorinity.



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VH=2.1571+17.3929T-0.8706T²-12.975C

where VH=viable-hatch (%). The highest percentage was 89.0 % at 10.0 °C and 0 ‰ (Fig. 2). The temperature and salinity combinations for the maximum percentages of total-and viable-hatch coincided with each other.

Time to 50 % hatch

The data varied between 18.5 and 44.6 days at the combinations where hatch occurred (**Table 4**). Analysis of variance for these data proved to be significant for temperature only (**Table 5**). The relationship between time (D days) and temperture (T $^{\circ}$ C) at the temperature range of 8.0 to 16.0 $^{\circ}$ C combined with chlorinity of 0 $^{\circ}_{0}$ was expressed by the second order equation :

 $D = 112.9329 - 11.5079T + 0.3518T^{2}$

The regression curve in Fig. 3 showed that time was inversely and exponetially related to the temperature.

Temp.	Chlorinity (%)				
(°C)	0	2	4	6	_
16.0	18.6 18.5	,			
14.0	21.1 22.1	21.4 20.1	21.1 21.5	 	
12.0	26.0 23.8	26.3 24.9	24.0	27.5	
10.0	33.0 33.0	32.6 33.0	32.6 33.1	31.6	
8.0	43.5 43.5	43.0 44.6	41.1 42.8		

Table 4. Time (days) to 50 % hatch of the rainbow trout eggs

Source	SS	DF	MS	F
Temperature ^{\$1}	1,928.00	4	482.00	508.58**
Residual	20.85	22	0.95	
Chlorinity ^{\$1}	37.61	3	12.54	0.15^{ns}
Residual	1,911.24	23	83.10	
Total	1,948.85			
Regression ^{\$2}	809.28	4	202.32	345.86**
Linear	751.54	1	751.54	1,284.68**
Quadratic	55.44	1	55.44	94.77**
Remainder	2.31	2	1.15	1.97 ^{ns}
Residual	2.93	5	0.59	
Total	812.21	9		

Table 5. Analysis of variance for the time to 50 % hatch

 $^{\$1}$ One-way lay out. $^{\$2}$ for the data at temperature of 8.0 to 16.0 $^\circ\!C$ combined with chlorinity of 0 $\%_0$.

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



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

Discussion

Responses of teleosts egg to changing temperature and salinity vary widely with species (KINNE 1963, 1964). In the present species, KAWAJIRI (1927) and MOORE (1940) noted in regard to the effect of temperature that hatch occurred safely within the ranges from 6 to 11 °C and from 3 to 13 °C, respectively. Our results, in which the hatch occurred at almost all tested temperature ranges frome 8.0 to 14.0 °C, are well in accordance with their descriptions, and newly propose that the range of chlorinity for hatchability was within 4‰.

On the time to hatch, KAWAJIRI (1927) reported that the days were 47-51, 32-33, 28, and 21 days after fertilization when the eggs were incubated at temperatures of 7.8, 10.6, 12.0, and 15.0 °C, respectively, and then suggested that the time was inversely and exponetially related to the temperature. The result in this study follows his suggestion. On the other hand, the effect of salinity on the time to 50 % hatch was statistically insignificant within the hatchable range of eggs. The same effect has been reported on the amago salmon (LOPES et al. 1985) and Pacific herring Clupea pallasi (ALDERDICE and VELSEN 1971), while some acceleration or retardation of the time in changing salinities has also been reported on several fishes, such as English sole Parophrys vetulus (ALDERDICE and FORRESTER 1968), the threeline grunt (KASHIWAGI et al. 1984), Atlantic herring Clupea harengus (HOLLIDAY and BLAXTER 1960), Pacific cod Gadus macrocephalus (FORRESTER and ALDERDICE 1965, ALDERDICE and FORRESTER 1971 b), petrale sole Eopstta jordani (ALDERDICE and FOR-RESTER 1971 a), red sea bream Pagrus major (APOSTOLOPOULOS 1976), and yellowtail flounder Limanda ferruginea (LAURENCE and HOWELL 1981). It is generally seen that the effect of salinity is small in comparison with that of temperature (BLAXTER 1969, ROTHENTHAL and ALDERDICE 1976).

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