

EFFECTS OF SEEDING DENSITY OF PINK SALMON, *ONCORHYNCHUS GORBUSCHA*, EGGS ON WATER CHEMISTRY, FRY CHARACTERISTICS, AND FRY SURVIVAL IN GRAVEL INCUBATORS

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ABSTRACT

We determined the effects of seeding density of pink salmon eggs in gravel incubators on water chemistry and on size, stage of development, and time of emergence of fry. Sixty days after fertilization, eyed eggs were placed in eight identical test incubators at five different densities (0 to 25,600 eggs per incubator). Test incubators had upwelling water (apparent velocity, 53 cm per hour); 0.015 m³ of gravel (size, 3-32 mm); and an average incubation temperature of 4.5° C (range, 3.5°-10.0° C). Total ammonia (NH₃ + NH₄⁺) production and oxygen consumption rates per alevin generally increased throughout incubation. Maximum total ammonia production at any density was about 8 × 10⁻⁴ mg/h per alevin. Maximum oxygen consumption was 0.028 mg/h per alevin. The rate of ammonia production and oxygen consumption per alevin increased with increased seeding density until the reduced oxygen concentration limited metabolism. Indications of stress—reduction in size of fry and early emergence—were evident only at the higher seeding densities, 12,800 and 25,600 eggs per 0.015 m³, and were either absent or unimportant at the lower seeding densities, 1,600 and 6,400 eggs per 0.015 m³. Un-ionized ammonia (NH₃) concentrations did not reach lethal levels. The stress at higher seeding densities, 12,800 and 25,600 eggs per 0.015 m³, was probably caused by depletion of oxygen to concentrations below 6 mg/l. Sublethal ammonia concentrations and low dissolved oxygen concentrations were probably synergistic.

Gravel incubators with upwelling water are being tested at hatcheries in the Pacific Northwest for production of fry from eggs of pink salmon, *Oncorhynchus gorbuscha*, chum salmon, *O. keta*, and sockeye salmon, *O. nerka*. To operate most economically, these incubators must be stocked with optimum numbers of eggs and be supplied with a flow of water consistent with production of good quality fry. Frugal use of water is important to hatcheries in Alaska where long, cold winters limit free-flowing water. However, densities of eggs that are too high for water flows result in oxygen depletion or ammonia buildup—stressing conditions that produce undersized fry or early emerging alevins. Both undersized fry and early emerging alevins are believed to survive poorly if released unfed (Bams and Simpson 1977).

Acute ammonia toxicity may not be a significant problem in Alaska where waters typically have low temperature and low pH. Ammonia equilibrates in water to form dissolved, un-ionized NH₃ and ionized NH₄⁺ (NH₃ is more toxic than NH₄⁺), and low temperature and low pH shift the equilib-

rium toward NH₄⁺ (Emerson et al. 1975). At lower temperatures, however, salmon incubation times are longer than at higher temperatures so that increased cumulative exposure to NH₃ could have adverse effects.

In this paper we describe effects of seeding densities of pink salmon eggs in gravel incubators on 1) oxygen consumption, 2) ammonia production, 3) physical characteristics of fry, 4) survival of fry, and 5) time of volitional emergence. The production limits of the gravel incubators are also defined.

METHODS

Experimental gravel incubators were seeded with different densities of eggs. Temperature, pH, dissolved oxygen, and total ammonia (NH₃ and NH₄⁺) concentrations were measured in water entering and leaving the incubators. Rates of oxygen consumption and ammonia production per egg or alevin were estimated during incubation. We monitored numbers and size of fry and time of emergence of fry to identify stressful conditions. Chemical data were compared with biological data to determine maximum seeding densities for the gravel incubators and to define limits of oxygen

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and ammonia concentrations that do not produce stressful conditions.

On 16 September 1971, pink salmon eggs were collected from spawners in Sashin Creek on Baranof Island, southeastern Alaska. The eggs were immediately fertilized, water hardened, and placed in Heath² trays. On 4 November, we transported the eyed eggs from Sashin Creek to Auke Creek near Juneau, Alaska, and on 16 November, the eggs were placed in eight gravel incubators at five seeding densities—0, 1,600, 6,400, 12,800, and 25,600 eggs/incubator (Table 1).

Each incubator (inside measurements, 30 cm × 30 cm × 30 cm, Bailey and Heard 1973) contained 0.015 m³ of gravel. A 25 mm layer of bird's-eye gravel (particle size, 2-4 mm) covered the sides and bottom. The remainder of the gravel was larger (particle size, 13-32 mm). We installed airtight covers on the incubators to prevent exchange of gases between atmosphere and water. Water was introduced into each incubator from the bottom in an upwelling flow of 0.8 l/min (apparent velocity, 53 cm/h).

Numbers of eggs were estimated by displacement (Burrows 1951). Precision of the seeding densities, given by ±2 times their estimated standard error (Table 1), was based on appropriate expansion of variation in egg counts from ten 100 ml samples. In previous studies of incubation at this hatchery (Bailey and Taylor 1974), the eggs hatched in late December or early January, 100-120 d after fertilization; and the fry emerged in April, 205-225 d after fertilization. In this study, we expected the eggs to hatch and the fry to emerge at similar times.

Oxygen and total ammonia concentrations were measured weekly between 3 December 1971 and 11 April 1972. Dissolved oxygen concentrations were measured with the Winkler method to the nearest 0.01 mg/l. Total ammonia (NH₃ + NH₄⁺) in the water was measured with an autoanalyzer using a procedure modified from that of the U.S.

Environmental Protection Agency (1974). Our modification used a larger capacity heating bath and measured total ammonia to within 0.004 ppb. If temperature and pH are known, the amount of NH₃ can be calculated from tables by Emerson et al. (1975). When calculating periodic estimates of oxygen uptake and total ammonia production per individual, we corrected for the number of fry that had left the incubator. We assumed the number of alevins in the incubator equaled the final total of alevins emerging from the incubator less the number of alevins already emerged. Temperatures of incubator effluents were measured daily to the nearest 0.1°C, and pH was measured twice a week with a standardized Corning model 112 pH meter. Confidence intervals were calculated for each estimate and displayed graphically. These confidence intervals were computed under the assumptions of normality of variation and homogeneous variance, the latter holding both among incubators and over observation times.

The fry were sampled and counted daily (February through April 1972) as they voluntarily emerged from incubators. Samples of fry were preserved in 5% Formalin for 6 wk. Later we selected three samples of 50 fry from the daily samples of each incubator to represent the days when cumulative fry emergence was 25%, 50%, and 75% of the total emergent fry for each incubator. Fry in these selected samples were measured to the nearest millimeter (fork length) and weighed to the nearest milligram after they were blotted with a damp paper towel. Developmental index (K_D) was computed to determine efficiency of yolk conversion [$K_D = 10(\text{weight, milligrams})^{1/3}/(\text{length, millimeters})$, Bams 1972]. The K_D index was computed for unfed fry at the time of emergence. A high K_D indicates a large amount of unabsorbed yolk, whereas a small K_D indicates a small amount of yolk and a more developed fry. The sample of fry at 25% emergence from the incubator seeded with 25,600 eggs was lost.

TEMPERATURE, pH, AND TOTAL AMMONIA IN INCUBATOR EFFLUENT

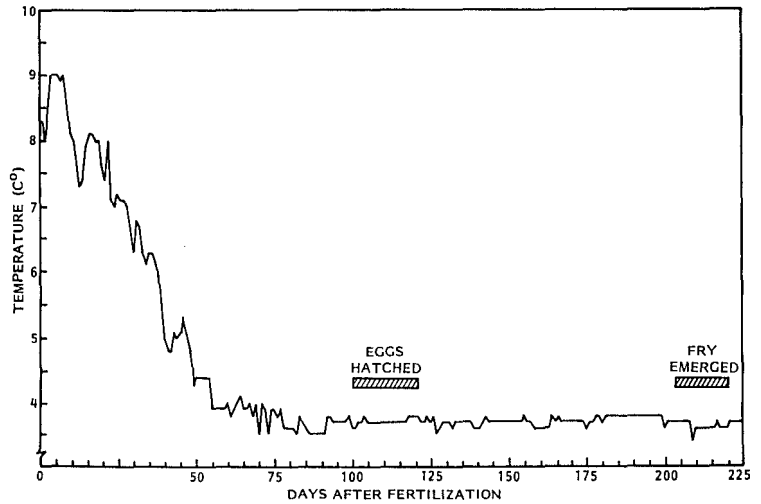
Temperature of the water source decreased as the experiment proceeded. Temperature was about 8°C when the eggs were fertilized 16 September 1971 (day 0, Figure 1), remained above 7°C until 14 October (day 28), and then gradually dropped to 3.6°C (range, 3.5°-3.8°C) by 16

²Reference to trade names does not imply endorsement by the National Marine Fisheries Service, NOAA.

TABLE 1.—Survival of pink salmon from eyed egg to migrant fry in gravel incubators seeded with indicated number of eggs (±2 SE).

Eyed eggs per incubator	Survival (%)	Eyed eggs per incubator	Survival (%)
1,600 ± 76	100	6,400 ± 302	92
6,400 ± 302	94	12,800 ± 604	100
6,400 ± 302	92	25,600 ± 1,209	50

FIGURE 1.—Incubation temperature of pink salmon eggs from fertilization, 16 September 1971 (day 0) until termination of the experiment, 28 April 1972 (day 225) after fry emerged from the incubators. Horizontal bars show when eggs hatched and fry emerged from the incubator seeded with 1,600 eggs.



November (day 61). It remained near 3.6° C from 16 November 1971 until termination of the experiment on 28 April 1972 (day 225). The daily temperature variation was <0.2° C.

During the incubation period, pH of the hatchery water supply changed little (pH, 6.13-6.39). Effluents of incubators with eggs had a pH from 6.08 to 6.36. Effluent from the incubator with the highest density of eggs (average pH, 6.19) was more acidic than the hatchery supply (average pH, 6.27).

Concentrations of total ammonia in effluents were higher in seeded incubators than in control incubators and generally increased with more eggs (Figure 2). Concentrations of total ammonia

in the effluent from control incubators and in the water supply were nearly identical (maximum concentration about 0.011 mg/l). During the study, maximum concentrations of total ammonia in seeded incubators ranged from 0.03 mg/l for the incubator seeded with 1,600 eggs (March) to 0.32 mg/l for the incubator seeded with 25,600 eggs (January).

The rate of total ammonia production per individual was periodically measured in all of the incubators. As development progressed from the eyed-egg stage to the emerging fry stage, rate of total ammonia production per individual increased. For example, in the three incubators seeded with 6,400 eggs (Figure 3), the mean of total ammonia production 3 wk before hatching (89 d after fertilization) was $<2 \times 10^{-4}$ mg/h per egg. By hatching (110 d after fertilization), the mean of total ammonia production increased to nearly 4×10^{-4} mg/h per egg. At emergence, approximately 14 wk after hatching (208 d after fertilization), the mean of total ammonia production was almost 6×10^{-4} mg/h per alevin.

The rate of total ammonia production per egg or alevin increased with seeding density (Figure 4). The rates of total ammonia production per individual were meaningless for the incubator with 25,600 eggs (not shown in Figure 4) because many of the eggs and alevins had died and were decomposing and because many of the alevins had emerged 30-60 days early. At the other three seeding densities (1,600, 6,400, and 12,800 eggs), the rates of total ammonia production were higher at higher seeding densities, and the regression of average rates of total ammonia production per indi-

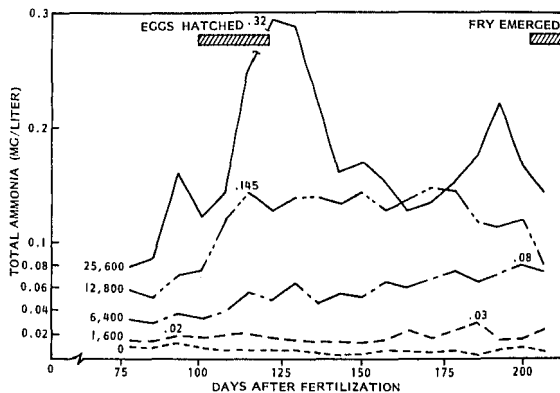


FIGURE 2.—Concentrations of total ammonia in hatchery water supply (0 eggs) and in effluents from incubators seeded with indicated numbers of pink salmon eggs. Horizontal bars show when eggs hatched and fry emerged in the incubator seeded with 1,600 eggs. Peak total ammonia concentration for each seeding density is indicated numerically.

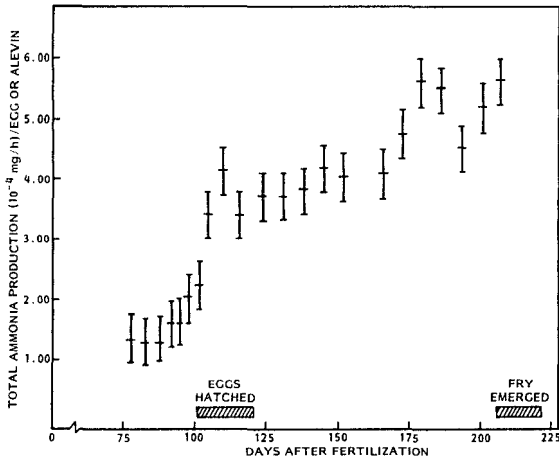


FIGURE 3.—Total ammonia production per individual pink salmon egg or alevin in the three gravel incubators seeded with 6,400 eggs. Ninety-five percent confidence limits for the periodic means were calculated using the error mean square from a one-way ANOVA for sampling periods. Horizontal bars show when eggs hatched and fry emerged.

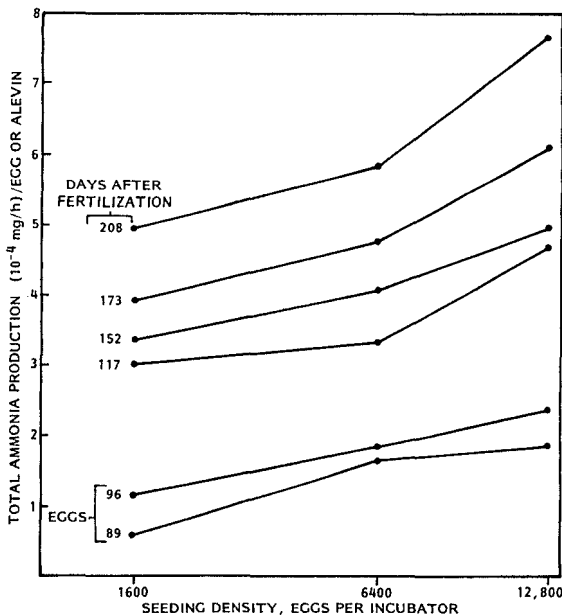


FIGURE 4.—The effect of seeding density on individual ammonia production rates during development of eggs to emerging alevins. Total ammonia was measured in incubator effluents and was corrected for emergence from the incubators. Eggs hatched 100-120 days after fertilization, and alevins from 1,600-egg density emerged about 203-220 days after fertilization.

vidual egg or alevin in the incubators on their seeding densities was significant ($P < 0.01$). The average rate of total ammonia production in each

incubator was the mean of 22 periodic rates measured as the eggs developed into emergent fry.

DISSOLVED OXYGEN

Dissolved oxygen concentration in the supply water declined gradually from 9.16 mg/l (70% saturation) on 14 December 1971 (day 89), about 2 wk before the eggs hatched, to 8.08 mg/l (62% saturation) on 11 April 1972 (day 208, Figure 5). This decline was normal because the lake source is usually covered with ice in winter.

Generally, as the eggs developed into fry, dissolved oxygen concentrations in the seeded incubators decreased more in incubators seeded with more eggs (Figure 5), except in the incubator with 25,600 eggs, massive early emergence of fry left fewer alevins in the incubator than in the incubator initially seeded with 12,800 eggs. After the early emergence in the incubator initially seeded with 25,600 eggs, the effluent of the incubator with 12,800 eggs had the lowest dissolved oxygen concentration of the study—3.8 mg/l dissolved oxygen (29% saturated) on 7 March (day 173).

Generally, oxygen consumption per egg or alevin increased steadily during development. At 7-d intervals during incubation, we estimated oxygen consumption rates in each of the three incubators seeded with 6,400 eggs (Figure 6) and averaged these rates. The average rate of oxygen consumption about 2 wk before hatching was about 0.003

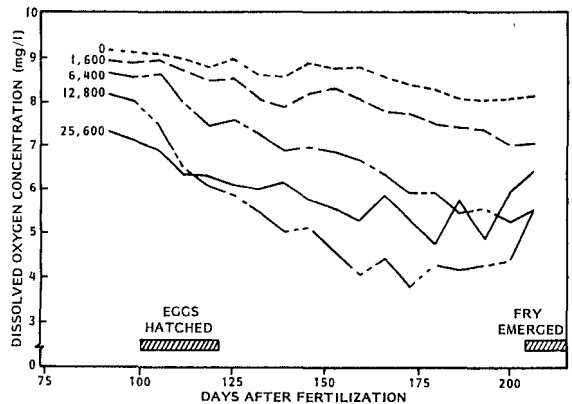


FIGURE 5.—Dissolved oxygen concentration in hatchery water supply (0 eggs) and in effluents from incubators seeded with indicated numbers of pink salmon eggs, December 1971-April 1972. Horizontal bars show when eggs hatched and fry emerged in the incubator seeded with 1,600 eggs.

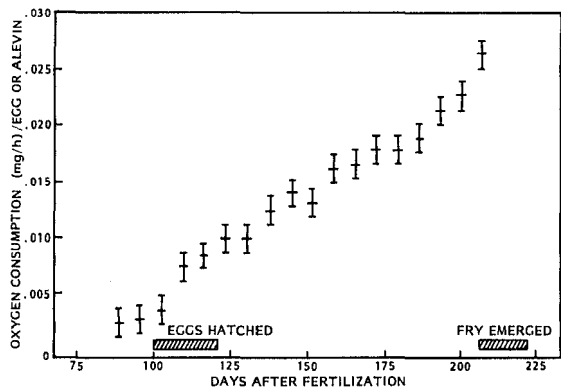


FIGURE 6.—Oxygen consumption per individual pink salmon egg or alevin in three gravel incubators seeded with 6,400 eggs. Ninety-five percent confidence limits for the periodic means were calculated using the error mean square from a one-way ANOVA for sampling periods. Horizontal bars show when eggs hatched and fry emerged.

mg/h per egg; by hatching, oxygen consumption increased to about 0.010 mg/h per alevin. The transient peak of oxygen consumption at the end of hatching is probably associated with increases in metabolism due to increases in activity during the hatching process and activity of alevins as they redistribute themselves within the incubator. These transient increases during hatching would have been more significant if the large numbers of eggs in the incubators had hatched synchronously over a day rather than over a 2-wk period. By the time of emergence, oxygen consumption had increased to a mean of 0.027 mg/h per alevin in the incubators seeded with 6,400 eggs. In the incubators seeded with other densities of eggs, oxygen consumption per individual also increased as eggs developed into alevins. The increase in oxygen consumption by eggs and alevins during development was in response to growth and not in response to increased temperature. Temperature remained nearly constant (about 3.6° C) from 2 wk before hatching until all alevins emerged.

Densities of alevins in the incubators influenced the individual oxygen consumption rates (Figure 7). Before hatching, the oxygen consumption rates per egg (days 89 and 96) were about the same in incubators of different densities. After hatching and to the time approaching emergence (days 117, 152, and 173, Figure 7), oxygen demand by individual alevins increased with increased seeding density (excluding the incubator with 25,600 eggs). The incubator seeded with 25,600 eggs (not

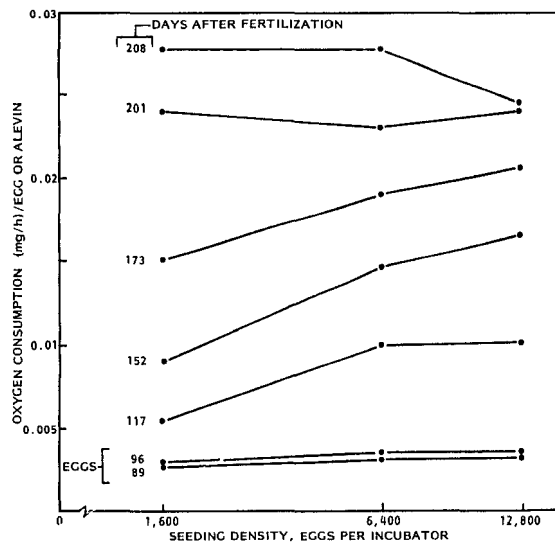


FIGURE 7.—The effect of seeding density on individual oxygen consumption rates during development of eggs to emerging alevins. Oxygen measurements were taken from incubator effluents and corrected for emergence to milligrams/alevin per hour. Eggs hatched 100-120 days after fertilization, and alevins from the 1,600-egg density emerged about 203-220 days after fertilization.

shown in Figure 7) contained a large number of dead eggs and alevins, which were decomposing. As emergence approached (days 201 and 208), oxygen consumption per alevin did not tend to increase with alevin density.

The increased production of ammonia and consumption of oxygen by alevins with increased seeding densities indicate increased metabolic rates caused by more frequent stimulation and interaction of neighboring alevins.

QUANTITY AND QUALITY OF FRY PRODUCED

Egg-to-Fry Survival and Time of Emergence

Survival from eyed eggs to fry in the incubator seeded with 25,600 eggs (50%) was markedly lower than survival in all other incubators ($\geq 92\%$, Table 1). Survival was almost 100% in the incubator seeded with 12,800 eggs; the incubator seeded with 12,800 eggs produced almost as many live fry as the incubator seeded with 25,600 eggs. Survival in the incubators seeded with 1,600 eggs and 6,400 eggs ranged from 92% to 100%.

Alevins emerged markedly earlier from incubators seeded with $>6,400$ eggs (Figure 8). If the

time of 50% emergence in the incubator with 1,600 eggs is used as a standard, 50% of the fry in incubators seeded with 6,400 eggs emerged on the same day (15 April, day 212); 50% of the fry in the incubator seeded with 12,800 eggs emerged 7 d early (8 April, day 205); and 50% of the fry in the incubator seeded with 25,600 eggs emerged 82 d early (24 January, day 130).

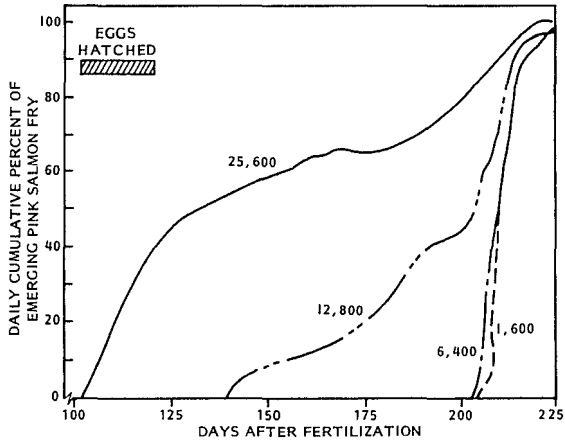


FIGURE 8.—Effect of seeding density on daily cumulative percentages of emergence of pink salmon fry from gravel incubators, December 1971-April 1972. Horizontal bars show when eggs hatched and fry emerged from incubator seeded with 1,600 eggs. Number beside each line is the number of eggs seeded in each incubator.

Size of Fry and Stage of Development

In the incubators with seeding densities above 6,400 eggs, fry emerged earlier and were shorter, lighter, and less developed (higher K_D) than fry in incubators with lower seeding densities.

During the time we monitored emergence, alevins emerging from the incubator seeded with 25,600 eggs were substantially smaller than alevins emerging from the other incubators. At 50% emergence, the alevins in the incubators seeded with 25,600 eggs were in an earlier stage of development (higher K_D) (Figures 9-11) than alevins in other incubators.

Analysis of variance of average lengths of fry at the three lower seeding levels—1,600, 6,400, and 12,800 eggs—indicated significant and changing differences ($P < 0.001$), i.e., interaction, in fry length among seeding densities and sampling times (Table 2, Figure 9). At the first sampling time (about 25% emergence), fry emerging from the incubator seeded with 12,800 eggs were substan-

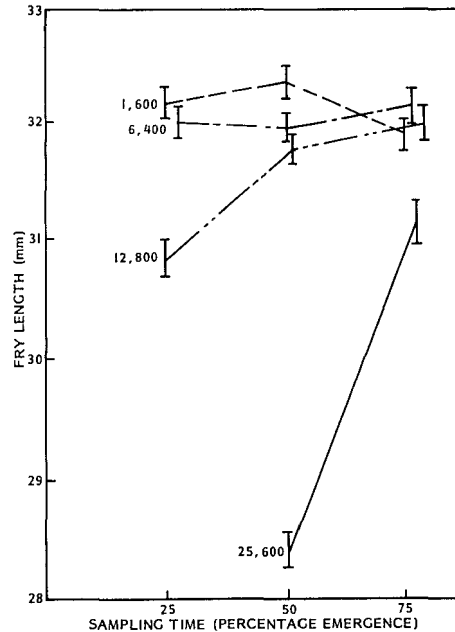


FIGURE 9.—Average length of pink salmon fry that emerged from gravel incubators seeded with indicated numbers of eggs. Samples of 50 fry were taken from each incubator when 25%, 50%, and 75% of emergence from that incubator had occurred. Bars represent 95% confidence limits. The sample for 25% emergence at the 25,600 density was lost.

TABLE 2.—Analysis of variance of average lengths of pink salmon fry, with variation among seeding levels within sampling times partitioned out.

Source	df	SS	MS	F
A Sampling times	2	0.18437	0.09218	—
B Seeding levels	2	0.63706	0.31863	—
Levels in time 1	2	1.21253	0.60626	78.33***
Levels in time 2	2	0.17941	0.08970	11.59**
Levels in time 3	2	0.04821	0.02410	3.11ns
A × B Interaction	4	0.80309	0.20077	25.93***
Within	6	0.04641	0.00774	
Total	14	1.67093		

*** $P < 0.001$.

** $P < 0.01$.

ns = not significant.

tially smaller than fry emerging from incubators seeded with 1,600 and 6,400 eggs. At the second sampling (about 50% emergence), fry emerging from the incubator seeded with 12,800 eggs were still smaller than fry emerging from incubators seeded with <12,800 eggs, but the difference was not as large as at the first sampling. By the third sampling (about 75% emergence), differences were not statistically significant ($P > 0.05$).

Analysis of variance of average weights of the same fry from the three lower seeding densities detected differences among seeding densities (Ta-

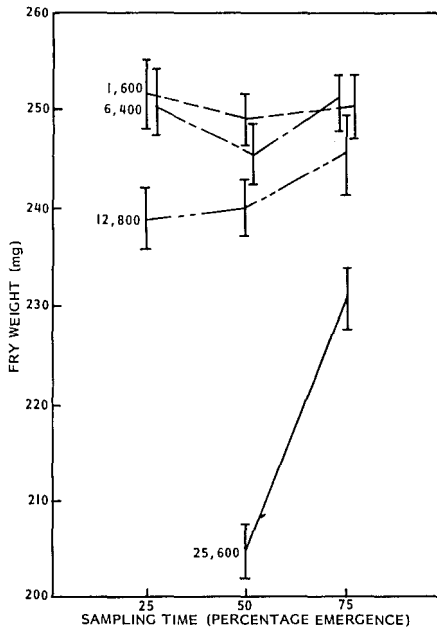


FIGURE 10.—Average weight of pink salmon fry that emerged from gravel incubators seeded with indicated numbers of eggs. Samples of 50 fry were taken from each incubator when 25%, 50%, and 75% of emergence from that incubator had occurred. Bars represent 95% confidence limits. The sample for 25% emergence at the 25,600 density was lost.

ble 3, $P < 0.01$). Differences in weights were particularly evident at first sampling (about 25% emergence, Figure 10), although no change in these differences in average weight with time was detectable, i.e., interaction was not significant ($P > 0.05$). Mean weight of fry in the incubator seeded with 12,800 eggs was considerably less than the mean weight of fry in the incubators seeded with 1,600 or 6,400 eggs.

Analysis of variance of average developmental index of these fry from incubators seeded with 1,600, 6,400, and 12,800 eggs determined that differences among incubators varied with time, i.e., interaction is significant (Table 4, $P = 0.05$). At the first sampling time, fry from the incubator seeded with 12,800 eggs had a substantially larger mean developmental index (were less developed) than fry from incubators seeded with fewer eggs. Yolk was still visible through the transparent abdominal sutures of fry in the incubator seeded with 12,800 eggs. At the second and third sampling times, developmental indices did not vary significantly among these lower densities ($P > 0.05$, Figure 11). However, early-emerging alevins from the incubator seeded with 25,600 eggs were

TABLE 3.—Analysis of variance of average weights of pink salmon fry.

Source	df	SS	MS	F
A Times	2	65.836	32.918	—
B Seeding levels	2	150.0017	75.00085	11.308**
A × B Interactions	4	19.4129	4.853225	0.7317ns
Within	6	39.7934	6.63223	
Total	14	275.0440		

** $P < 0.01$.

ns = not significant.

TABLE 4.—Analysis of variance of average developmental index of pink salmon fry with variation among seeding levels within sampling times partitioned out.

Source	df	SS	MS	F
A Sampling times	2	0.00108893	0.000544465	—
B Seeding levels	2	0.00028737	0.000143685	—
Levels in time 1	2	0.00165333	0.000826665	8.24*
Levels in time 2	2	0.00013653	0.000068265	0.68ns
Levels in time 3	2	0.00031413	0.000157065	1.57ns
A × B Interaction	4	0.00181662	0.000454155	4.53*
Within	6	0.00060201	0.000100335	
Total	14	0.00379493		

* $P < 0.05$.

ns = not significant.

clearly less developed at the second sampling than early-emerging alevins in the other incubators (Figure 11). (The first sample from the incubator seeded with 25,600 eggs was lost.)

WATER QUALITY AND FRY PRODUCTION IN RELATION TO SEEDING DENSITY

The maximum concentration of total ammonia (0.32 mg/l) detected during the experiment occurred shortly after hatching in the effluent from the incubator seeded with 25,600 eggs. Even in combination with the highest pH encountered during our study (6.4), this total ammonia concentration is equivalent to only 0.092 $\mu\text{g/l}$ NH_3 (0.092 ppb). Concentrations of NH_3 13 times greater (1.2 ppb) inhibit growth of emergent fry after 60-d exposures in their late alevin stages, and concentrations 100 times greater stimulate early emergence of alevins (Rice and Bailey 1980a). Concentrations of $\text{NH}_3 \leq 0.4$ ppb have no discernible effect on either size or emergence of the alevins in late stages. Ammonia concentrations in this experiment were not stressing even though alevins nearing emergence are more sensitive to NH_3 than earlier alevin stages (Rice and Stokes 1975; Rice and Bailey 1980a).

Similarly, Rice and Bailey (1980b) did not find toxic concentrations of ammonia in samples of intragravel water taken in late March from a streambed where alevin densities were much

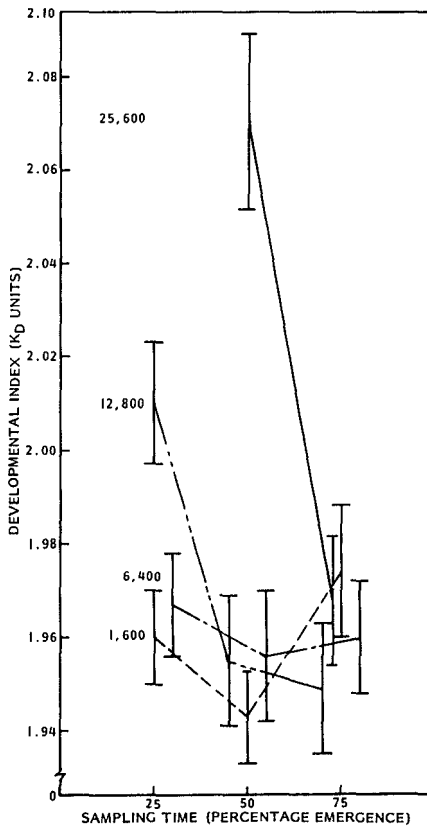


FIGURE 11.—Average developmental indices (Bams 1972) of pink salmon fry that emerged from gravel incubators seeded with indicated numbers of eggs. Samples of 50 fry were taken from each incubator when 25%, 50%, and 75% of emergence from that incubator had occurred. Bars represent 95% confidence limits. The sample for 25% emergence at the 25,600 density was lost.

lower than the highest densities in our experimental incubators. Densities of pink salmon alevins in the stream sampled ranged from 0 to 352 alevins/0.1 m² (mean, 21 alevins/0.1 m²), and the highest concentration of NH₃ was 0.1 ppb. For comparison, the alevin densities in our experiment were >1,700 alevins/0.1 m².

Oxygen consumption rates per individual increased with time. The highest rate observed (0.028 mg/h per alevin) occurred near the end of incubation when 25% of the fry had emerged from the incubators seeded with 1,600 and 6,400 eggs. Further increases in rate of oxygen consumption per alevin might have occurred before emergence was complete, but this potential for stress was eliminated because fry left the incubators.

The respiration rates of salmon eggs and alevins are oxygen dependent (Fry 1957); consequently,

low dissolved oxygen concentrations in the water will limit normal metabolic rates. Our data suggest that oxygen concentrations below about 6 mg/l may decrease the normal metabolism of pink salmon alevins, although survival to emergence may not be noticeably affected. Alevins in the incubator seeded with 12,800 eggs were probably stressed and began to emerge early, about 60 d earlier than alevins in lower density incubators (Figure 8), when oxygen concentrations decreased to about 5 mg/l (Figure 5). Just prior to peak emergence (201 and 208 d after fertilization), alevins in the incubators seeded with 6,400 and 12,800 eggs probably had their oxygen consumption limited because oxygen concentrations had decreased to <6 mg/l at a time when their demand was greatest. If oxygen concentrations were not limiting, oxygen consumption rates (Figure 7) would have been greater than the rates for alevins in the 1,600-egg incubator as in the earlier measurements.

Although ammonia (NH₃) alone did not increase to concentrations that decreased size at emergence or reduced survival, it may have acted synergistically with relatively low oxygen concentrations to create stressful conditions in our study. In earlier studies, survival time of fish exposed to ammonia (NH₃) was reduced at adequate but low oxygen levels (Wuhrmann 1952; Downing and Merckens 1955).

Stressful conditions associated with low dissolved oxygen concentrations alone, or in combination with ammonia, probably not only reduced survival and caused premature emergence of fry but may have also reduced the ability of alevins to use yolk for growth. In incubation studies at similar temperatures, the developmental indices decreased and lengths increased just before emergence of pink salmon fry (Bams 1972; Bailey and Taylor 1974). A decrease of 0.01 in *K_D* corresponds to about 0.2 mm increase in length (Bams 1972; Bailey and Taylor 1974). In our study, the developmental index decreased to about 1.96 at emergence in the incubator seeded with 1,600 eggs (Figure 11). The fry in the incubator seeded with 25,600 eggs were 28.44 mm, mean length, at 50% emergence and had a mean developmental index of 2.07. If these fry had remained in the incubator until their developmental index decreased to 1.96, they would have been only 30.64 mm long, 1.52 mm shorter than the fry emerging from the incubator seeded with 1,600 eggs.

Survival from egg to fry exceeded 90% in in-

cubators seeded with <25,600 eggs, but survival was only 50% in the incubator seeded with 25,600 eggs. We attribute this poor survival to crowding, possibly low dissolved oxygen concentrations, or the combined effects of these and elevated NH_3 concentrations. Ammonia, as a single factor, is unlikely to reach levels harmful to pink salmon alevins in gravel incubators supplied with slightly acidic water, as the water in Auke Creek. However, if the pH were 7.75 rather than 6.4, then the highest concentration of total ammonia, 0.32 mg/l, would be equivalent to 2.1 ppb of NH_3 and would reduce survival.

Much remains to be learned before we can define combinations of seeding density and water flow for efficient production of healthy, unfed fry. Seeding densities of 1,200-1,800 eggs/0.015 m³ of gravel and an apparent water velocity of 70-300 cm³/h per cm² can be used (Bams 1972; Bailey et al. 1976). Bams and Simpson (1977) suggested that 1,965 eggs/0.015 m³ with a water velocity of 200 cm/h is safe. In our study, increasing seeding density from 1,600 eggs/0.015 m³ to 6,400 eggs/0.015 m³ at a water flow of 53 cm/h apparently increased swimming activity of the alevins and also increased oxygen consumption and ammonia production. However, the average length, weight, developmental index, emergence time, and survival of these alevins were not importantly affected.

Under our experimental conditions, a seeding density of 6,400 eggs/0.015 m³ appears to be acceptable, although perhaps a nearly maximum seeding density. Our test incubators were operated at a water flow of only 0.8 l/min (apparent velocity, 53 cm³/h per cm²). If an apparent water velocity of 200 cm³/h per cm² were used as recommended by Bams and Simpson (1977), acceptable seeding densities might be higher.

SUMMARY

Pink salmon eggs were seeded in gravel incubators at four different densities (from 1,600 to 25,600 eggs/0.015 m³ of gravel) and incubated until fry voluntarily emerged. Dissolved oxygen and total ammonia concentrations of the incubator effluent were monitored periodically, and the emerged fry were counted, sampled, and measured. The rate of total ammonia production per egg or alevin increased with time after seeding at all densities. At seeding densities of 6,400 eggs/0.015 m³ the rate of total ammonia production increased from 2×10^{-4} mg/h per egg 3 wk before

hatching, to 4×10^{-4} mg/h per alevin at hatching, to 6×10^{-4} mg/h per alevin at emergence. The rate of total ammonia production per individual also increased with seeding density. Because of low pH and low temperature, NH_3 concentrations did not reach toxic or lethal concentrations in any incubator; however, NH_3 concentrations would have become toxic in the incubator seeded with the most eggs (25,600 eggs/0.015 m³) shortly after hatching if the pH had been 7.75 rather than 6.4.

Rate of oxygen consumption per egg or alevin increased during incubation. In the incubator seeded with 6,400 eggs/0.015 m³, it increased from 0.003 mg/h per egg 3 wk before hatching, to 0.007 mg/h per egg at hatching, to 0.028 mg/h per alevin at emergence. Probably because of increased interaction between adjacent alevins, rates of oxygen consumption per hour per alevin increased, until emergence, with increased seeding density. In incubators seeded with >6,400 eggs/0.015 m³, dissolved oxygen concentrations dropped to stressful levels (<6 mg/l) that limited metabolism. At seeding densities >6,400 eggs/0.015 m³, stressful conditions caused early emergence of premature fry and reduced the ability of alevins to convert yolk for growth. Additionally, survival was reduced at 25,600 eggs/0.015 m³. At an apparent water velocity of 53 cm³/h per cm², a seeding density of 6,400 eggs appeared to be marginally acceptable for the production of healthy pink salmon fry.

ACKNOWLEDGMENTS

The pH determinations were made by James Knull, oceanographer at the Auke Bay Laboratory. We wish to thank Don Alderdice, Environment Canada, for his helpful review of this manuscript.

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