NOTES

GAS-BUBBLE DISEASE: MORTALITIES OF COHO SALMON, *ONCORHYNCHUS KISUTCH*, IN WATER WITH CONSTANT TOTAL GAS PRESSURE AND DIFFERENT OXYGEN-NITROGEN RATIOS[1]

A review of the literature regarding gas-bubble disease can be found in a recent publication by Rucker (1972); one by the National Academy of Science (Anonymous in press); and an unpublished report by Weitkamp and Katz (1973).[2] Most discussions on gas-bubble disease have dealt with the inert gas, nitrogen—oxygen was given a secondary role. It is important to know the relationship of nitrogen and oxygen when we are concerned with the total gas pressure in water. Where water becomes aerated at dams or falls, oxygen and nitrogen are usually about equally saturated, however, many of the samples analyzed from the Columbia River indicate that nitrogen is often about 7% higher than oxygen when expressed as a percentage. When oxygen is removed from water by metabolic and chemical action, or when oxygen is added to the water by photosynthesis, there is a definite change in the ratio of oxygen and the inert gases (mainly nitrogen with some argon, etc.). This present study shows the effect of varying the oxygen and nitrogen ratio in water on fingerling coho salmon, *Oncorhynchus kisutch*, while maintaining a constant total gas pressure.

The primary purpose of these experiments was to determine differences in lethality of various gas ratios of oxygen and nitrogen at a constant total gas pressure of 119%. I also wished to determine whether there was a difference in susceptibility between sizes and stocks of juvenile coho. Also to be examined was the effect of reducing the oxygen while holding the nitrogen constant.

Methods

Juvenile coho salmon averaging 6 cm in length, obtained from the Quilcene National Fish Hatchery, Quilcene, Wash., and the Northwest Fisheries Center of the National Marine Fisheries Service, NOAA (National Oceanic and Atmospheric Administration), Seattle, Wash., were used during all the tests concerning differences in lethality of O_2/N_2 ratios. During these tests water temperatures were 13.6° + 0.1°C. Gas concentrations usually varied slightly from the desired ratios. The tank facility consisted of six troughs, two of which were used to hold experimental fish at normal saturation (100%) and two pairs of troughs used to test fish at different gas ratios.

Control of gas concentrations and the test apparatus is described in a subsequent section. During initial testing of the gas control system, I determined that a ratio of 114% O_2 to 121% N_2 could be achieved by merely allowing air to be sucked into the intake side of the recirculation pump. Since this gas ratio did not require injection of either oxygen or nitrogen, the resultant concentration (114% O_2 and 121% N_2) was used as a quasi control for comparison with the other gas ratios. Several replicates were completed at this concentration. Water saturated at this ratio and concentration was also used to test for differences in size and stock and to provide base line data in determining effect of reduced oxygen concentrations while maintaining a constant nitrogen level.

In all the tests free carbon dioxide was near normal, or about 2 ppm. Oxygen is expressed as "O_2" and the inert gases as "N_2."

The number of days required to kill 25% of the fish at the different gas levels is expressed as the lethal exposure—LE_{25} and to kill 50%— LE_{50}.

Apparatus shown in Figure 1 was used to supply water with a definite oxygen and nitrogen content. The tank (1) was divided so that two experiments could be carried on simultaneously with similar equipment. Water was circulated by a centrifugal pump (2) with a valve (3) on the effluent side to cause a controlled back pressure as read on a gauge (4). This created a vacuum on the inflow side (5) so that air could be introduced into the water with either oxygen or nitrogen (6)

[1]Research performed under contract with the U.S. Army Corps of Engineers.

[2]D. E. Weitkamp and M. Katz. 1973. Resource and literature review of dissolved gas supersaturation in relation to the Columbia and Snake River fishery resources. Submitted to Northwest Utilities Cooperative, c/o Idaho Power Co., Boise, Idaho, Apr. 3, 1973, by Seattle Marine Laboratories, Div. of Xelco Corp., Seattle, Wash., 55 p. (Typewritten.)

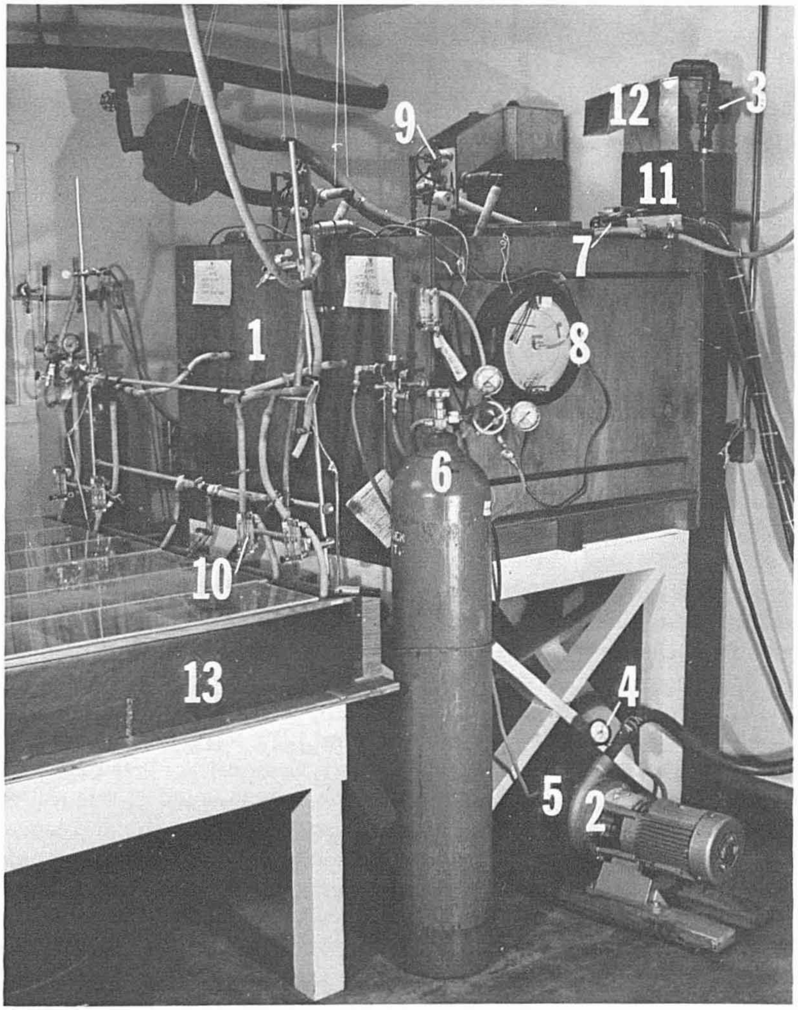

FIGURE 1.–Apparatus for subjecting fish to constant-temperature, flowing water with a definite oxygen and nitrogen content.

through a "Y" tube. Circulation of the water caused an increase in temperature which was maintained at approximately 13.6°C by means of a refrigeration system (7) and recorded on a thermograph (8). Water level in the tank was maintained by float valves (9). Each trough was supplied with 1 liter per minute of water regulated with flow meters (10). The water used was from the municipal supply, was soft, and was passed through activated charcoal to remove the chlorine. A greater depth of water was needed for absorption of the gases than was afforded by the tank (1), so two towers (11) were added to the system. The spout (12) at the top of the towers was to direct possible overflow water back into the system.

Inside dimensions of troughs in the fish holding area (13) were 104.5 × 23.5 × 20 cm high. Water depth was maintained at 14 cm. Each trough could be separated into three compartments with screens–"A" was at the inflow end of the trough, "B" middle, and "C" outflow end. In a few cases a compartment was divided longitudinally so that two groups of fish could be subjected to almost identical conditions.

Results

Effect of Variation in O_2/N_2 Ratios on Mortality

Times to death (LE_{25} and LE_{50}) of juvenile coho salmon at various concentrations of O_2 and N_2 during constant total gas saturation of 119% appear in Table 1 are shown graphically in Figure 2. All tests were run in duplicate with 50 fish per test, except one at 229% O_2 and 90% N_2 which involve 50 fish but one test. With one exception (192% O_2 and

TABLE 1.—Time to death of groups of juvenile coho salmon (about 6 cm long) in 13.6°C water with total gas pressure of 119% of saturation and different ratios of O_2 and N_2.

Gas concentration (% saturation)		Time to death (days)			
		25% of all fish		30% of all fish	
O_2	N_2	Range	Average	Range	Average
50	138	1.8- 1.9	1.9	3.2-4.0	3.7
75	131	1.8- 2.7	2.3	3.5-4.3	3.9
114	121	3.2- 4.1	3.8	6.3-7.3	6.9
159	109	3.2- 5.3	4.5	6.5-9.1	8.2
173	105	33.5-35.3	34.4	—	(1)
192	100	—	(2)32.0	—	(3)
229	90	—	(4)	—	(4)

(1)Not reached, 28% mortality in 39 days.
(2)One replicate reached 24% mortality in 30 days; the other, 25% in 32 days.
(3)Not reached, test concluded at 33 days.
(4)Not reached, 20% mortality in 35 days.

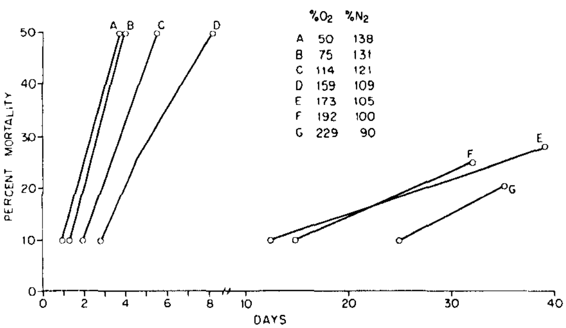

FIGURE 2.—Mortality pattern of 6-cm coho salmon reared at different O_2/N_2 levels at 13.6°C with a 119% total gas pressure.

100% N_2), all increases in ratio of O_2 to N_2 resulted in increased tolerance to the total gas saturation. A marked increase in tolerance to total gas pressure occurred between concentrations of 159%/109% and 173%/105% saturation of O_2 and N_2 (Figure 3).

Effect of Size and Stock of Fish on Mortality

A number of tests were carried out in the water containing 114% O_2 and 121% N_2 to determine effect of size and stock of fish on susceptibility to gas supersaturation (Table 2). Two groups of 3.8-cm coho from the Northwest Fisheries Center which had just started feeding were initially tested. One group of 96 fish reached LE_{25} in 22.9 days and LE_{30} after 30 days. The other group of 50 fish reached LE_{25} in 10.9 days. No further losses occurred until the 27th day. Loss at 30 days was 34%. Averages of the two groups placed LE_{25} at 16.9 days. Average loss at 30 days was 32%.

Two groups of 50, 4.6-cm fish from the Quilcene National Fish Hatchery were also tested. These tests produced LE_{25} of 15.1 and 18.3 days. LE_{50} was

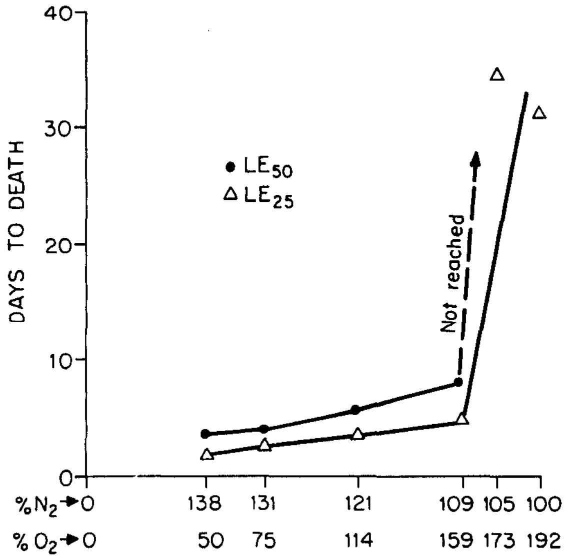

FIGURE 3.—Relationship between O_2/N_2 levels and time to death of 6-cm coho salmon fingerlings at 13.6°C and total gas concentration of 119%.

TABLE 2.—Time to death of groups of juvenile coho salmon of different body length and stock composition in 13.6° ± 0.1°C water with gas concentrations of 114% O_2 and 121% N_2.

Average body length and (in parentheses) stock of fish	Time to death (days)	
	25% of all fish	50% of all fish
3.8 cm (Seattle)	16.9	Not reached in 30 days
4.6 cm (Quilcene)	16.7	27.4
10 cm (Seattle)	2.1	2.6
10 cm (Quilcene)	2.9	4.2

reached in 24.7 and 30 days. Averages of the above placed LE_{25} at 16.7 and LE_{50} at 27.4 days.

Five groups of fish (8, 12, 16, 16, and 16 in number), and approximately 10 cm long, from the Northwest Fisheries Center were then tested. The average for all groups gave an LE_{25} of 2.1 days and an LE_{50} of 2.6 days.

Three groups of 12 fish each approximately 10 cm long from the Quilcene National Fish Hatchery were similarly tested. Averages were 2.9 days for LE_{25} and 4.2 days for LE_{50}.

These results indicate that the larger fingerlings approximately the same age are definitely more subject to harm from excess air in the water than the smaller fish. These data agree with those of Meekin and Turner (1974) and Dawley et al.[3]

[3]E. Dawley, B. Monk, M. Schiewe, and F. Ossiander. 1974. Salmonid bioassay of supersaturation of dissolved gas in water. Northwest Fish Cent., Natl. Mar. Fish. Serv., NOAA, Seattle, Wash., unpubl. manuscr.

Although the data are limited, there appears to be little difference between susceptibility of the Montlake and Quilcene stocks.

Effect of Reduced O_2 Concentration on Mortality

Fish held in compartments in a trough utilize oxygen so that the water in compartment C (outflow end) would have less oxygen than in compartment A (inflow end). Compartment B in the central part of the trough would have O_2 levels somewhere between those in A and C. Nitrogen levels in these compartments, however, were the same. To demonstrate the effect of reduced oxygen in relation to gas-bubble disease, 48 coho of 8.5 cm fork length were randomly distributed into compartments A, B, and C. Two additional replicates of the C compartment tests were run using 32 coho (8.5 cm) in each trial. These are listed as C_1 and C_2 in Table 3.

TABLE 3.—Time to death of groups of juvenile coho salmon (about 8.5 cm long) in 13.6°C water with 121% N_2 and different concentrations of O_2.

Trough compartment	Gas concentration (% saturation)		Time to death (days)	
	O_2	Total pressure	25% of all fish	50% of all fish
A	113	119	2.5	3.3
B	110	118	3.6	5.3
C	105	117	3.8	5.3
C_1	105	117	4.2	6.6
C_2	105	117	5.4	6.6

Inspection of these data indicates that when 121% N_2 is maintained, oxygen plays a more significant role above 110% than below 110%.

Some of the data obtained when the oxygen-nitrogen ratio tests were done also illustrated the effect of reduced oxygen on the mortality rate. This was apparent in the experiment using 173% O_2 and 105% N_2. At 173% O_2 there were losses of 26 and 30% in 39 days, whereas slightly larger fish at the lower end of the troughs subjected to 169% O_2 had losses of only 7% in 39 days.

Pathology

Generally the fish died suddenly in the higher nitrogen concentrations. Never was tissue damage or any progressive pathology demonstrated. The fish always seemed to die from gas embolism, restricting the flow of blood through the gills. When the nitrogen was near normal and the oxygen high, the fish were moribund for many days before succumbing. These fish had blebs in the mouth which interfered with feeding and caused emaciation.

Summary

Coho salmon fingerlings were subjected to a total gas pressure of 119% at 13.6°C with the O_2/N_2 varying from 50%/138% to 229%/90%. The small fish (3.8 to 6 cm) were the most resistant and the larger fish (8 to 10 cm) the least resistant to gas-bubble disease at the gas concentrations used. A drastic decrease in lethal effect of individual ratios of O_2 to N_2 occurred between 159% O_2/109% N_2 and 173% O_2/105% N_2 at the same total gas pressure (119%).

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