

10-1-2008

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Zydlewski, Gayle; Gale, W.; Holmes, J.; Johnson, J.; Brigham, T.; and Thorson, W., "Use of Electroshock for Euthanizing and Immobilizing Adult Spring Chinook Salmon in a Hatchery" (2008). *Marine Sciences Faculty Scholarship*. 140.
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Use of Electroshock for Euthanizing and Immobilizing Adult Spring Chinook Salmon in a Hatchery

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Abstract.—This study evaluated the use of electroshock as an alternative to traditional techniques for immobilizing and euthanizing hatchery fish. We used a commercially available electroanesthesia unit at the U.S. Fish and Wildlife Service's Carson National Fish Hatchery (Carson, Washington) to euthanize adult spring Chinook salmon *Oncorhynchus tshawytscha* and to sort and collect gametes of fish at maturation. During euthanization by electroshock, the response of each fish was observed, muscular and vertebral hemorrhaging was quantified, and electrical settings were optimized accordingly. During gamete collection, fish were either electroshocked or exposed to tricaine methanesulfonate (MS-222); hemorrhaging, egg viability, egg size and quantity, and resultant fry quality were examined for each treatment group. Electroshocked fish had a higher likelihood of injury during gamete collection than did fish exposed to MS-222. On average, each electroshocked fish had less than two hemorrhages on both fillets examined. The size of each hemorrhage was less than 0.10% of the fillet surface. Fecundity and egg and fry quality were not affected by either immobilization method. Electroshock was a viable and efficient means of euthanizing adult spring Chinook salmon or sorting the fish and collecting their gametes. However, equipment settings must be optimized based on site-specific (e.g., water conductivity) and species-specific (e.g., fish size and seasonal state of maturation) factors.

In most hatchery programs that produce spring Chinook salmon *Oncorhynchus tshawytscha*, the adults spend several months in the hatchery before they are mature and ready to spawn. At most facilities, adults are collected throughout the migratory season (April–August), so their progeny represent genetic contributions from all run times. Extended hatchery residence before gamete collection often makes it necessary to

handle these fish on multiple occasions. For example, diseased or injured fish may have to be separated from other broodstock and fish health is often managed by prophylactic injection with antibiotics. It is imperative that handling operations minimize stress and physical impacts to the fish to ensure their survival and collection of gametes for hatchery purposes.

At times, the number of returning adults can greatly exceed hatchery goals and excess fish must be removed to prevent overcrowding. Under these circumstances, fish must be processed humanely while maximizing worker safety. To avoid wastage, excess fish are often given to tribal groups, food banks, and other entities (e.g., federal prison systems) for use as an additional food source. Euthanizing fish for consumption requires consideration of fish and human welfare as well as the effect of the procedure on fillet quality.

Current methods of fish preparation for consumption and gamete collection vary widely. It was beyond the scope of this project to review all such methods.

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Received May 24, 2007; accepted November 11, 2007

Published online October 30, 2008

However, two commonly used techniques—application of carbon dioxide (CO₂) and tricaine methanesulfonate (MS-222)—provide examples of limitations that are inherent to current technology. Application of CO₂ to holding tanks results in acute acidosis and spasms that are injurious to fish (G. K. Iwama, University of British Columbia, unpublished), therefore deviating from humane slaughter methods (Van de Vis 2003). For human consumption purposes, traditional chemical anesthetics are usually not permissible. For example, MS-222 requires a 21-d withdrawal period before consumption or release into the wild. In many cases, the application of CO₂ or MS-222 requires an additional blow to the head to kill the fish. This method is of increasing concern to animal protection groups and governments (Lambooij et al. 2007). Furthermore, results from tests on percussive stunning are inconclusive in terms of agreement with optimal slaughter methods, which require the fish to be rendered “unconscious until death without avoidable excitement. . .” (Van de Vis et al. 2003).

Electroshock has been used as a viable and humane means of processing large numbers of adult salmonids (Tipping and Gilhuly 1996; Tesch et al. 1999; Roth et al. 2002; Van de Vis et al. 2003). Evidence suggests that when electroshock is administered at carefully chosen levels, it renders the fish unconscious immediately (Van de Vis et al. 2003), causing minimal injury to adult fish and indiscernible effects on their progeny. Small-scale studies of northern pike *Esox lucius* and brook trout *Salvelinus fontinalis* ($n = 1-3$ fish/trial) have reported that egg viability and fry growth did not differ between parents treated with MS-222 and those treated with electroshock (Walker et al. 1994; Redman et al. 1998). Walker et al. (1994) also reported no physical damage to the adults when pulsed DC was used. However, deleterious effects of electroshock on individual parents (e.g., Arctic grayling *Thymallus arcticus* and cutthroat trout *O. clarkii*) have been associated with negative effects on progeny (Roach 1999; Dwyer et al. 2001). These conflicting results make it necessary to compare methods of immobilization and euthanization for each considered species and scale of operation.

The goal of this study was to examine whether electroshock is a viable and humane alternative for spring Chinook salmon immobilization (for up to 5 min; allowing subsequent recovery) and euthanization (humane death). First, we examined whether use of electroshock to euthanize excess adult fish affected fillet quality. Second, we examined whether electroshock could be used in place of MS-222 for sorting and gamete collection. Finally, we assessed how electro-

shock application to adults affected the survival and growth of progeny.

Methods

Adult spring Chinook salmon from the Wind River, Washington, ascend a fish ladder into adult holding ponds (662 m³) at the U.S. Fish and Wildlife Service (USFWS) Carson National Fish Hatchery (CNFH), Carson. Throughout the season (May–August), predetermined numbers of males and females are maintained for gamete collection (in August). Fish in excess of the number needed for hatchery operations are kept in a separate holding pond to ensure that enough fish are available for gamete collection. Excess fish are euthanized as necessary.

A Model EA-1000 electroanesthesia (EA) unit (Smith-Root, Inc., Vancouver, Washington) was used for this study. The unit consists of an electronic pulse generator and fiberglass tank. The fiberglass tank has two brailers (1.02 m long × 0.63 m wide) for moving shocked fish up to a sorting table. Plate electrodes are located at the tank ends and in the middle between the two brailers. The design produces a predictable, homogeneous electrical field. The unit can be operated with the end electrodes as cathodes and the central electrode as the anode (normal polarity) or vice versa (reversed polarity).

The unit generates a pulsed-DC waveform and has a two-stage operation; the first stage uses lower voltage settings (3.5–35.5 V) than the second stage (12–235 V). Both stages produce a constant-DC pedestal and a pulsed-DC waveform on the pedestal. The pulsed-DC waveform is a burst of three pulses at 240 Hz and 50% duty cycle (2.08-ms pulse width). This pulse train is repeated 15 times/s. The constant-DC pedestal is 45% of peak voltage in stage 1 and 20–45% of peak voltage in stage 2. In stage 2, the higher percentage is at the lower peak voltage and decreases with increasing peak voltage settings. Voltage duration in the original configuration could be applied from 0 to 128 s for both stages. The design of this waveform is based upon two separate waveforms known to cause a low level of fish injury. Also, constant DC is less injurious than pulsed DC (McMichael 1993; Dalbey et al. 1996; Ainslie et al. 1998). The pulsed waveform used for anesthesia is similar to the patented complex pulse system (CPS) waveform developed by Coffelt Manufacturing, Inc. (Sharber et al. 1994). Studies comparing CPS with traditional pulsed-DC waveforms have shown significantly lower rates of injury when CPS is used (Sharber et al. 1994).

Preliminary tests were conducted to determine the minimum voltage levels, electroshock duration, and number of fish that could be processed at one time to

achieve euthanization and immobilization while minimizing hemorrhaging. Immobilization was achieved when fish remained motionless on the sampling table for 5 min. Euthanization was achieved when fish did not recover from immobilization. Results from 31 individuals (11 single-fish trials and 3 multiple-fish trials) were used to establish methods for hatchery operations. Settings for euthanization were 19 V for 60 s and 298 V for 120 s. For sorting and gamete collection, settings were 19 V for 60 s and 130 V for 68 s. We did not optimize settings for other handling procedures, such as antibiotic injection. Control fish (i.e., those handled without EA or MS-222 treatment) were not used due to the impracticality of handling nonanesthetized animals. For purposes of this study, reported injury rates for MS-222-treated fish are considered to represent normal handling-related injury and any EA-related increase above the normal injury level represents the effect of electroshock treatment.

Euthanization of excess fish.—In 2002, 4,800 adult spring Chinook salmon were euthanized by CNFH staff over a 5-d period and 1 of every 100 fish ($n = 48$) was examined on each of the 5 d. In 2003, 5,800 fish were euthanized over 6 d and 15 fish were examined on each of 3 d.

All examined fish were measured postmortem, filleted (the entire length from operculum to tail), and sexed. The right and left fillets (and a metric scale) were photographed with a digital camera in lateral projection. Visual discrimination was used to separate fish into two mutually exclusive categories (modified from Reynolds 1996): (1) muscular injury (muscle hemorrhage that was not associated with the spinal column); and (2) vertebral injury (muscle hemorrhage that was associated with the spinal cord). Vertebral injury was measured as a hemorrhage that occurred near the vertebra, but actual vertebral injury was not assessed. In 2002, hemorrhages of euthanized fish were further analyzed using digital imaging software (Image J version 1.36b; Wayne Rasband, National Institute of Mental Health, Research Services Branch, Bethesda, Maryland). The area (in pixels) of each hemorrhage relative to the area of the fillet was determined ($n = 63$ hemorrhages from 17 fish [2 fillets/fish]).

Logistic regression was used to model the relationship between euthanization date, fish size, or fish sex and the likelihood of combined injury (presence-absence of either hemorrhage type) using a generalized linear model. Assessment of a date effect on combined injury was important, since physiological status of the muscular tissue changed during reproductive maturation (Figure 1; note the pale appearance of tissue in the lower photograph). Data were analyzed and reported for combined injury and vertebral injury (muscular

injury alone was minimal and is not reported; however, it can be calculated by subtracting vertebral injury from combined injury). In the binary logit model, the response variable was likelihood of combined fish injury (hemorrhage present or absent), the explanatory value was size (fork length [FL]; size was correlated with response to electroshock in a study by Dalbey et al. [1996]), and the class variables were fish sex and euthanization date. All two-way interactions were considered in the initial model. We assessed the significance of the logistic regression models using a full-reduced-model likelihood ratio chi-square (χ^2) test. The likelihood of fish vertebral injury (presence or absence) was analyzed using the same logistic methods, explanatory variables, and class variables used for combined injury. A Kruskal-Wallis test was used to compare hemorrhage size (imaged from photographs) among sampling dates, as these data did not meet the assumptions of analysis of variance (ANOVA).

Immobilization of fish for gamete collection.—During two gamete collection days in 2002 and 2003, we compared the use of EA to the use of MS-222. Water temperature was approximately 10°C, and ambient conductivity was between 37 and 45 $\mu\text{S}/\text{cm}$. Adult Chinook salmon ($n = 12\text{--}14$) were crowded randomly from holding ponds and immobilized by use of either MS-222 or EA. The EA treatment was applied first, and then the treatment tank was filled with MS-222 (50 mg/L). The two treatments were alternated through the day, resulting in two MS-222 exposure groups and two EA exposure groups. The holding tank was drained and refilled with freshwater between treatments. Immobilized adults were sexed and checked for maturity. Ripe males were killed with a blow to the head. Ripe females were dispatched with a pneumatic guillotine and were allowed to bleed for approximately 3–5 min before egg collection. Approximately every other fish (males and females) was measured (FL) and filleted (operculum to tail). Both the right and left fillets were photographed, and blood vessel hemorrhaging was assessed as described previously. In 2002, photographs of individuals with hemorrhages were analyzed to estimate hemorrhage size relative to fillet surface area ($n = 155$ hemorrhages from 59 individuals). Adults that were not ripe were returned to the holding pond for future gamete collection. Unripe fish from each treatment group were marked with opercular punches (hole punches in the opercular plate); two punches were applied to MS-222-treated fish, and one punch was applied to EA-treated fish. On the second gamete collection date, males and females were paired within a treatment group (i.e., only EA \times EA and MS-222 \times MS-222 pairings were used).

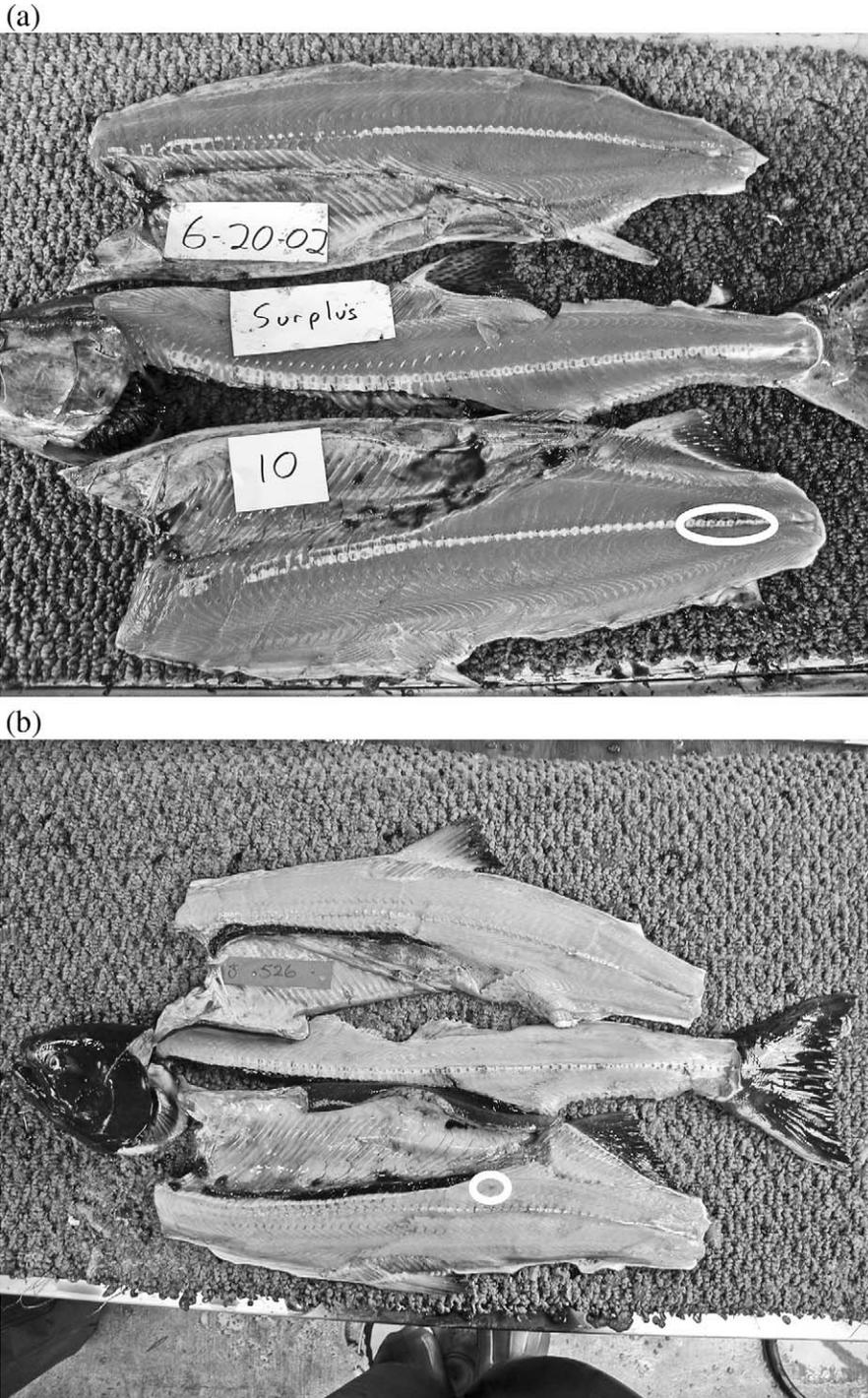


FIGURE 1.—Photographs illustrating hemorrhages (indicated by white ovals) in muscle (fillets) of spring Chinook salmon subjected to electroanesthesia at Carson National Fish Hatchery, Carson, Washington: (a) a male (96 cm fork length [FL]) euthanized on 20 June 2002, exhibiting a hemorrhage in the tail region along the vertebrae (fillet at bottom); and (b) a female (96 cm FL) anesthetized prior to gamete collection on 21 August 2002, exhibiting one hemorrhage in the bottom fillet.

Eggs were removed from females by abdominal incision, milt was collected from males, and fertilization was achieved using standard hatchery practices. A 1:1 random pairing of males to females was maintained. Eggs from an individual cross were placed in a plastic colander suspended in a bucket (7.6 L) supplied with single-pass spring water (7.6 L/min; 7°C). To prevent fungal growth, eggs were treated three times weekly with formalin (1,667 mg/L, 15-min exposure). Eggs from 60 females (30 fish/treatment group) were physically shocked via gentle transfer between two containers to reveal dead eggs (those that were white and opaque), which were counted and removed by hand. The total number of live eggs was calculated by weight based on a sample weight of 500 eggs/female. Fecundity was calculated as the sum of live and dead eggs for each female. In 2002, the eggs used for the sample weight for each female were transported to USFWS Abernathy Fish Technology Center (AFTC), Longview, Washington, to further track hatching and fry growth. Data on hatching and fry growth were not collected in 2003.

Logistic regression was used to model the relationship between immobilization treatment, gamete collection date, fish size, or fish sex and the likelihood of combined injury (presence-absence) using a generalized linear model. Treatment, fish sex, and date were class variables in the binary logit model. A model that incorporated all two-way interactions was used. Fish size was initially included as an explanatory variable but was removed from the final model due to its strong correlation with sex. The likelihood of vertebral injury (presence-absence) was analyzed using the same logistic methods and explanatory variables that were used for the combined injury analysis. A two-way ANOVA on ranked data (since data were not normally distributed and did not have equal variances) was used to determine date and treatment effects on hemorrhage size as determined from photographs.

Progeny growth and survival.—Upon arrival at AFTC, eggs were loaded into vertical tray incubators at a density of 500 eggs/tray (each tray contained eggs from a single female). Flow in the incubators was 11.4 L/min for eyed eggs and 18.9 L/min for fry. During incubation, the water was 12.5°C and saturated with oxygen. To control fungus, formalin treatments (1,600 mg/L for 15 min) were performed daily until hatch. At the swim-up stage, fish from each egg take were divided randomly among six tanks (3 tanks/treatment). Fish were fed an ad libitum ration of BioDiet (BioOregon, Warrenton, Oregon) for 2 months to allow acclimation to the feeding process and tank environment.

After the growth trial was initiated, fish were fed BioDiet Starter 3 (first week of the trial) and BioDiet

1000 (remainder of the trial) in accordance with the manufacturer's feeding guidelines. Feeding rates (percent body weight per day) varied from 4.1% at the beginning of the trial to 2.6% at the end. The total tank biomass was determined every 2 weeks by weighing all individuals collectively, and ration amount was adjusted accordingly. The FL and weight of 30 randomly sampled fish from each tank were determined at the beginning and end of the trial. The absolute growth rate for each tank was calculated based on the following formula (Busacker et al. 1990): $(Y_2 - Y_1)/t$, where Y_2 is the individual mean weight at the end of the trial, Y_1 is the individual mean weight at the beginning of the trial, and t is the number of days in the trial.

Initial and final FLs, initial and final wet weights, and absolute growth rate were analyzed using a three-way ANOVA with tank, egg take, and treatment as the explanatory variables. When the ANOVA indicated significant differences, Tukey's multiple comparison technique was then used.

Results

Euthanization of Excess Fish

In 2002, 55 fish (36 males and 19 females) were sampled; mean (\pm SE) FL was 79.0 \pm 3.1 cm for males ($n = 23$) and 76.0 \pm 0.6 cm for females ($n = 11$). In 2003, 45 fish (17 males and 28 females) were sampled; mean FL was 81.20 \pm 2.02 cm for males ($n = 16$) and 79.10 \pm 1.26 cm for females ($n = 29$).

In 2002 and 2003, electroshock resulted in hemorrhage injury to 33–71% of euthanized fish (Table 1). Nearly all injured individuals had hemorrhages associated with the vertebrae (e.g., Figure 1a). The relation between likelihood of combined injury or vertebral injury and euthanization date, sex, or fish size was not significant in either year (logistic regression: all $P > 0.10$).

The number of hemorrhages per fish ranged from 0 to 8 (2002: 1.07 \pm 0.27 hemorrhages/fish; 2003: 1.38 \pm 0.16 hemorrhages/fish). In 2002, median hemorrhage size was 0.07% of the fillet surface area (minimum = 0.01%; maximum = 0.33%). Hemorrhage size did not vary with date ($P = 0.28$, $n = 63$ hemorrhages).

Immobilization of Fish for Gamete Collection

In 2002, 120 fish (55 males and 65 females) were sampled for injury assessment during the first egg take and 122 fish (61 males and 61 females) were sampled during the second egg take. Mean (\pm SE) FL was 79.0 \pm 0.8 cm for males and 76.0 \pm 0.4 cm for females. In 2003, 89 fish (45 males and 44 females) were sampled for injuries during the first egg take and 98 fish (49

TABLE 1.—Percentages of male, female, and all spring Chinook salmon (*n* = number of fish examined) that exhibited one or more hemorrhages in muscle after euthanization by electroshock at Carson National Fish Hatchery, Carson, Washington. Vertebral injury was hemorrhaging associated with the spinal cord; combined injury was any muscle hemorrhage regardless of association with the spinal cord (i.e., vertebral injury plus muscle injury, as defined in Methods).

Date	Male injury			Female injury			Total injury		
	Vertebral (%)	Combined (%)	<i>n</i>	Vertebral (%)	Combined (%)	<i>n</i>	Vertebral (%)	Combined (%)	<i>n</i>
2002									
6 Jun	57	57	7	50	50	8	53	53	15
12 Jun	44	56	9	50	50	4	46	54	13
19 Jun	50	58	12	40	40	5	47	53	17
26 Jun	50	50	2	80	80	5	71	71	7
8 Jul	NA	NA	NA	67	100	3	67	100	3
2003									
3 Jun	14	29	7	37	37	8	27	33	15
10 Jun	20	20	5	70	90	10	53	67	15
1 Jul	50	50	4	27	27	11	33	33	15

males and 49 females) were sampled during the second egg take. Mean FL was 84.0 ± 1.1 cm for males and 82.0 ± 0.8 cm for females.

In 2002 and 2003, EA resulted in combined injury (Figure 1b) or vertebral injury (Figure 1a) to 24–71% of all processed fish, regardless of sex or number of times shocked (Table 2). Most hemorrhage injuries were associated with vertebrae. The use of MS-222 resulted in a 2–16% combined injury rate that was also primarily related to vertebrae. Relations between combined injury or vertebral injury and gamete collection date, treatment, or sex were significant for 2002 but not for 2003 (Table 3). Fish exposed to EA had significantly higher levels of combined injury than those exposed to MS-222. Furthermore, combined or vertebral injury level was significantly higher on the second gamete collection date than on the first collection date; the gamete collection date × treatment

interaction was significant for the combined injury data. In 2002, males had a higher rate of combined injury than females.

The number of hemorrhages (along the entire fillet surface) per fish ranged from 0 to 8 (EA: 1.33 ± 0.17 hemorrhages/fish in 2002, 1.61 ± 0.21 hemorrhages/fish in 2003; MS-222: 0.27 ± 0.09 hemorrhages/fish in 2002, 0.22 ± 0.16 hemorrhages/fish in 2003). In 2002, the median hemorrhage area was 0.06% (minimum = 0.01%; maximum = 0.48%) of the fillet surface area (e.g., Figure 1b). Hemorrhage size did not vary between dates or treatments (two-way ANOVA: date *P* = 0.45, treatment *P* = 0.356, date × treatment interaction *P* = 0.71; *n* = 151 hemorrhages). Fecundity, progeny survival to eye-up, and progeny survival from eye-up to swim-up did not differ between females that were immobilized with MS-222 and those immobilized with EA (Table 4).

TABLE 2.—Percentages of male, female, and all spring Chinook salmon (*n* = number of fish examined) that exhibited one or more hemorrhages in muscle after being immobilized with electroanesthesia (EA) or tricaine methanesulfonate (MS-222) during gamete collection at Carson National Fish Hatchery, Carson, Washington. Ripe fish were euthanized (males by a blow to the head; females by pneumatic guillotine), and fillets were examined for hemorrhages immediately after gamete collection. Vertebral injury was hemorrhaging associated with the spinal cord; combined injury was any muscle hemorrhage regardless of association with the spinal cord (i.e., vertebral injury plus muscle injury, as defined in Methods).

Date	Method	Male injury			Female injury			Total injury		
		Vertebral (%)	Combined (%)	<i>n</i>	Vertebral (%)	Combined (%)	<i>n</i>	Vertebral (%)	Combined (%)	<i>n</i>
2002										
14 Aug	EA	46	46	28	6	6	34	24	24	62
	MS-222	7	11	27	6	6	31	7	9	58
21 Aug	EA	70	87	30	50	53	30	60	70	60
	MS-222	19	19	31	10	13	31	15	16	62
2003										
14 Aug	EA	26	39	23	23	32	22	24	36	45
	MS-222	5	14	22	0	5	22	2	9	44
21 Aug	EA	61	68	31	74	74	31	68	71	62
	MS-222	6	6	18	6	6	18	6	6	36

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TABLE 3.—Results (P -values of maximum likelihood ratio tests) of logistic regressions examining the effects of immobilization treatment method (electroanesthesia or tricaine methanesulfonate), collection date (14 or 21 August), and sex on the presence of vertebral injury (muscle hemorrhage associated with the spinal cord) or combined injury (any hemorrhage regardless of association with the spinal cord; i.e., vertebral injury plus muscle injury, as defined in Methods) in spring Chinook salmon used for gamete collection at Carson National Fish Hatchery, Carson, Washington, 2002–2003.

Explanatory variable	Vertebral injury		Combined injury	
	2002	2003	2002	2003
Date	<0.001	<0.024	<0.001	0.308
Treatment	<0.001	<0.001	<0.001	<0.001
Sex	0.002	0.592	<0.001	0.504
Date \times treatment	0.308	0.581	0.034	0.047
Date \times sex	0.199	0.265	0.47	0.341
Treatment \times sex	0.277	0.402	0.045	0.519

Progeny Growth and Survival

Mean FL and body weight of fry at the time of transfer from trays to tanks did not differ between progeny of MS-222- and EA-treated broodstock (Table 5). Furthermore, the growth rate measured in the 57-d growth trial did not differ between progeny of the two immobilization treatment groups (Table 5). No relationship between egg take or tank and initial fish size or growth rate was apparent. Although initial size and absolute growth rate did not differ significantly, the progeny of the EA-treated group were slightly larger than the progeny of the MS-222-treated group at the end of the growth trial (weight: $P = 0.02$; FL: $P = 0.01$).

Discussion

Electroshock is a viable alternative to MS-222 for euthanizing and immobilizing adult spring Chinook salmon. When administered properly, electroshock can provide a safe and efficient means of euthanizing excess adult Chinook salmon while maintaining

satisfactory flesh quality. We found no discernable effect of electroshock exposure (immobilization) on spring Chinook salmon reproductive performance (fecundity) or progeny (eggs and fry) survival and growth. Mortality rates for EA- and MS-222-treated fish did not differ between the first and second spawning dates, and there were no obvious EA effects during interim periods. Adults subjected to electroshock had a significantly higher likelihood of combined injury than adults receiving the MS-222 treatment. However, the median area of injury per fish was less than 0.06% of the representative viewed fillets.

Electroshocking excess fish for euthanization resulted in acceptable fillet quality and satisfactory working conditions. A Yakama Nation representative judged the excess electroshocked fish to be appropriate for both tribal consumption and ceremonial purposes (C. James, Yakama Nation, personal communication). By Alaska Seafood Marketing Institute standards, the fillets would

TABLE 4.—Mean (\pm SE) egg weight (calculated from a sample weight of 500 eggs/female), fecundity (total number of eggs [live or dead] per female), progeny survival to eye-up (%), and progeny survival from eye-up to swim-up (%) for female spring Chinook salmon subjected to immobilization with electroanesthesia (EA) or tricaine methanesulfonate (MS-222) during gamete collection at Carson National Fish Hatchery, Carson, Washington. Data on survival after eye-up were not collected in 2003.

Date	Method	Egg weight (g)	Fecundity (eggs/female)	Survival to eye-up (%)	Survival from eye-up to swim-up (%)	Number of females
2002						
14 Aug	EA	0.21 \pm 0.004 ^a	4,144 \pm 155	80.5 \pm 4.1 ^b	95.2 \pm 1.1	31
	MS-222	0.22 \pm 0.007 ^b	4,337 \pm 170	81.1 \pm 2.9	95.3 \pm 1.1	31
21 Aug	EA	0.21 \pm 0.005	4,425 \pm 158	89.9 \pm 3.3 ^a	95.1 \pm 1.1 ^c	30
	MS-222	0.21 \pm 0.005	4,236 \pm 150	92.9 \pm 2.1	95.7 \pm 0.7	31
2003						
21 Aug	EA	0.24 \pm 0.005	4,504 \pm 119	93.2 \pm 1.7		31
	MS-222	0.21 \pm 0.006	5,106 \pm 290	95.6 \pm 1.1		17

^a Calculated from the progeny of 29 females.

^b Calculated from the progeny of 30 females.

^c Calculated from the progeny of 28 females.

TABLE 5.—Mean (\pm SE) initial fry weight and fork length (FL), final weight and FL after a 57-d trial, absolute growth rate, and number of fish sampled (n) to examine growth performance in progeny of female spring Chinook salmon subjected to immobilization with electroanesthesia (EA) or tricaine methanesulfonate (MS-222) during gamete collection at Carson National Fish Hatchery, Carson, Washington, in 2002.

Method	Initial		Final		Growth rate (g/d)	n
	Weight (g)	FL (mm)	Weight (g)	FL (mm)		
EA	1.2 \pm 0.02	52.2 \pm 0.30	5.4 \pm 0.08	77.8 \pm 0.35	5.5 \pm 0.60 ^a	180
MS-222	1.2 \pm 0.02	51.9 \pm 0.30	5.1 \pm 0.08	76.5 \pm 0.40	4.9 \pm 0.30 ^a	180

^a Calculated from six tanks.

be grade 2, defined as being acceptable for canning, mincing, or breeding.

The EA device reduced labor requirements; previous operations at CNFH required a total of eight staff members, whereas electroshock required only three staff members. Electroshock eliminated the need to euthanize fish with a blow to the head, which is time consuming and physically demanding for personnel.

The effects of EA use in hatcheries on the long-term survival and injury rates of exposed fish have not been closely examined. However, a large body of evidence has accumulated that details the effects of electrofishing on individual injury and performance (Reynolds 1996; Snyder 2003). Several researchers have noted an alarmingly high rate of internal hemorrhaging and spinal injuries caused by both backpack (Hollender and Carline 1994) and boat (Sharber and Carothers 1988; Sharber et al. 1994; Thompson et al. 1997) electrofishing. Schill and Elle (2000) report that it can take up to 3–5 weeks for electrofishing-induced hemorrhages to heal and that the hemorrhage severity often increases through the first 2 weeks postexposure before declining. Furthermore, high mortality in eggs from backpack-electroshocked spring Chinook salmon females has been reported (Cho et al. 2002). The effects of electroshock on adult injury and on the progeny of treated fish in hatcheries have received little attention. Our study partly addresses this information gap by comparing hemorrhaging rates between EA and MS-222 groups and examining performance of the resultant progeny.

Our results are in agreement with previous findings that describe growth and survival of progeny from salmonids exposed to electroshock for purposes of gamete collection. For example, when EA is administered properly (i.e., when settings are systematically and carefully determined before production use), egg and fry survival is high and comparable with that associated with CO₂ use for adult steelhead *O. mykiss*, fall Chinook salmon (Tipping and Gilhuly 1996), and chum salmon *O. keta* (Tesch et al. 1999). Redman et al. (1998) also reported that progeny of brown trout *Salmo*

trutta that were exposed to EA and MS-222 had equivalent survival and growth.

The long-term effects of electroshock on broodstock, especially fish that are maintained for long periods after treatment or that are handled multiple times, are not well documented. Tesch et al. (1999) reported survival rates of eggs from adult chum salmon exposed once to electrical shock, but they did not examine the effects of low DC voltage on the adults themselves. Redman et al. (1998) demonstrated that brown trout held for 6 months after EA treatment had a significantly higher mortality rate than fish treated with MS-222. In a study by Tipping and Gilhuly (1996), the recovery rate (hatchery returns) of EA-treated adult steelhead that were released into the wild was lower than the recovery rate of fish treated with CO₂. Those authors speculated that the lower recovery rate was due to an increased rate of delayed mortality for EA-treated fish.

Although our data show that EA resulted in more injuries than did MS-222, all hemorrhages (2002 data; regardless of treatment) were a small percentage of the fillet surface. However, when associated with vertebrae these hemorrhages could have an effect on subsequent performance. Injuries due to EA (i.e., those not visible upon external examination) could explain the negative results observed previously for this method (Tipping and Gilhuly 1996; Redman et al. 1998). Conversely, any increase in internal injury or stress during handling procedures could affect long-term survival of adult broodstock. These findings suggest that the use of electroshock must be carefully considered for broodstock programs in which repeated fish handling is necessary. However, due to the semelparous reproductive strategy of Pacific salmon, EA is a viable alternative for use in most hatchery programs for these species. The exception would be any steelhead program that attempts to recondition kelts for use as spawners in later years.

Optimal methods for immobilizing and euthanizing fish have been defined as those that cause the fish to become unconscious and remain so until death without avoidable excitement or pain (Van de Vis et al. 2003).

Many electroshocked individuals in this study exhibited some level of muscular hemorrhaging, but the method rendered fish immobile within seconds. While data suggest that electroshock is a useful tool for adult spring Chinook salmon immobilization and euthanization, fish pain perception is poorly understood and currently available studies are conflicting (UFR Committee 2004; Iwama 2007). Some evidence indicates that since pain is a psychological experience and fish lack the neurological structure associated with its perception, their detection of pain is a neurological impossibility (Rose 2002, 2007). Given the equivocal evidence of pain perception in fish, the best euthanization approach is to render fish unconscious (as with electrical stunning; Van de Vis et al. 2003) and prevent recovery. Our study concurs with previous studies showing that electrical stunning can be used to humanely slaughter salmon and other fish when conducted properly (Van de Vis et al. 2003; Lambooi et al. 2007). However, further research is needed to assess various methods of immobilization and euthanization to select the best technique for achieving hatchery goals.

An important aspect of EA application is the need to optimize the voltage and pulse settings based on site-specific differences in water conductivity, treatment chamber size, fish size, and species. Optimizing settings in the first year can be a significant commitment in labor and time. Settings must then be reviewed annually with established, relatively simple assessments, as optimal settings may change from year to year due to annual changes in fish size, condition, and water quality. Various authors have outlined the importance of identifying the best settings (voltage and pulse width) for individual hatchery situations. Tipping and Gilhuly (1996) advocated the use of electroshock, but they also emphasized the importance of using low voltage because of the notable damage to shocked adults at high voltages. Tesch et al. (1999) provided the caveat that each facility must optimize electrical settings based on water quality and proximity of the gamete collection area to the egg take area. Walker et al. (1994) outlined the importance of species specificity (particularly size at anesthesia) in electrical settings and the differences between AC and DC use. Roth et al. (2004) showed that a square AC wave inflicted more injury (spinal column and hemorrhaging) on Atlantic salmon *Salmo salar* than did a sinusoidal AC wave. Defined endpoints of fish health and quality are needed for all assessments. We suggest annual examination of internal hemorrhaging as a convenient and quantifiable method of monitoring EA effects on fish quality. Research focusing on the duration of EA-induced

injury and the presence and severity of long-term effects is needed.

Acknowledgments

The findings and conclusions in this manuscript are those of the authors and do not necessarily represent the views of the USFWS. The use of commercially available equipment does not denote an endorsement by the authors or the USFWS. The authors gratefully acknowledge the CNFH staff, especially Randy Berge and Jeff Blaisdell, and AFTC staff members Jeff Poole and Megan Hill for their help with data collection and fish handling. We thank Yakama Nation tribal member and U.S. Bureau of Indian Affairs representative C. James, Jr., for assistance in examining the fillet quality of euthanized fish. Lastly, we are grateful to Rich Johnson from the USFWS Fishery Resources Pacific Region, who helped to secure funding for this project.

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