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Relative Return to Creel of Triploid and Diploid Rainbow Trout Stocked in Eighteen Idaho Streams

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Abstract.—Introductions of fertile nonnative hatchery trout have led to interspecific and intraspecific hybridization of native salmonid stocks throughout North America. Use of sterile triploid hatchery trout in stream-stocking programs could reduce genetic risks to native stocks while addressing public demand for consumptive fishing opportunity. Techniques to produce triploid salmonids are well developed, and triploid rainbow trout *Oncorhynchus mykiss are* readily available from commercial sources. However, there is no published information on the return to creel of triploid trout in stream recreational fisheries. We purchased mixed-sex triploid and diploid rainbow trout eggs from a commercial supplier and reared the resulting fish to catchable size. Flow cytometry was used to verify triploid induction rates in the triploid group. Estimated cost to produce a triploid catchable rainbow trout was about 15% higher than for a diploid fish. We jaw-tagged and stocked 300 triploid and 300 diploid fish into each of 18 streams throughout Idaho and used tag returns to assess relative return to creel and timing of returns for the two groups. In all, 1,849 tags were returned by anglers, 931 from triploid fish and 918 from diploid fish. Overall returns were not significantly different between groups (paired *t*-test: P = 0.80). Mean time to harvest also did not differ between groups (paired *t*-test: P = 0.35). These results suggest that triploid rainbow trout can provide stream angling opportunity equal to that provided by fertile diploid fish. Although there are other concerns regarding the stocking of hatchery trout in streams containing native trout, we suggest that using triploid rainbow trout in stream-stocking programs can help balance the demands for both consumptive fishing opportunity and conservation of native stocks.

The widespread interspecific and intraspecific hybridization of native salmonid stocks due to introductions of nonnative species or strains is well documented (Allendorf et al. 1980; Busack and Gall 1981; Campton and Johnston 1985; Krueger and May 1991; Hindar et al. 1993). Stocking hatchery trout, however, does not always result in significant hybridization (Krueger and Menzel 1979: Wishard et al. 1984: Marnell et al. 1987). Jones et al. (1996) hypothesized that more than 30 years of stocking hatchery brook trout *Salvelinus fontinalis* could have led to introgression in native gene pools in 33 eastern Canada waters; however, mitochondrial DNA and electrophoretic data suggest this did not occur. Nonetheless, Allendorf and Leary (1988) suggested that continued stocking of nonnative hatchery rainbow trout *Oncorhynchus mykiss* in the western United States would eventually result in the homogenization of all indigenous stocks of cutthroat trout *O. clarki* into a single "mongrel" species. Whether such a dire prediction will eventually come to pass is uncertain, but introgression has clearly been a negative consequence of stocking trout in some waters (e.g., Campton and Johnston *1985).*

Despite concern for genetic impacts and other interactions with wild trout, hatchery trout continue to play a significant role in the management of many trout streams (Wiley et al. 1993; Van Vooren 1995; Incerpi 1996). The continued stocking of hatchery trout in streams reflects the dual mission of many state fisheries agencies. In Idaho and many other states, resource managers are legislatively charged with perpetuating native species while providing harvestable surpluses for the public. These two charges can be mutually exclusive in many situations where low productivity trout streams cannot provide substantial harvest opportunity for native fish. In some cases, hatchery trout stocking can play a role in native trout management by redirecting consumptive angling effort to specific streams or stream reaches, facilitating

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public acceptance of more restrictive regulation; on other waters (Van Vooren 1995).

The Idaho Department of Fish and Game (IDFG) has changed its stream stocking programs substantially in the last decade. As recently as 1985, IDFG stocked 3,400 km of the state's 44,000 fishable stream kilometers with catchable-size rainbow trout (hereafter referred to as catchables). Since 1985, kilometers of Idaho streams stocked has been reduced by more than 50% to 1,450 km, or about 3.3% of the fishable total (Van Vooren 1995). Reductions in stocking were largely due to increased emphasis on wild trout management, genetic concerns, and the desire to eliminate stocking in sites with poor return rates. Despite these reductions, however, 40% of stream catchables continue to be stocked in waters where viable native or naturalized nonnative trout stocks exist (J. Dillon, Idaho Department of Fish and Game, unpublished data). Stocking continues in some native trout streams because of high public demand and political pressure to provide sport harvest opportunity above the levels wild fish can provide.

In theory, use of sterile fish in hatchery programs could potentially minimize genetic interactions with native trout stocks. One method to produce sterile trout is through induced triploidy (Thorgaard 1983). Sexual development of female triploids is greatly retarded, and secondary sex characteristics and spawning behavior have not been observed. Male triploids develop secondary sex characteristics and may exhibit spawning behavior, but sperm production and viability is greatly reduced.

Early interest in the use of triploid fish in recreational fisheries management focused on the predicted potential for enhanced growth and longevity (Allen and Stanley 1978; Thorgaard 1983; Simon et al. 1993), rather than on the potential advantage of sterility itself. Conservation geneticists subsequently recommended use of triploid fish in marine net-pen culture to inhibit escaped domesticated or nonnative stocks from hybridizing with native anadromous salmonids (Utter et al. 1983; Hindar et al. 1991). In these instances, the discussion has focused largely on net pen escapees not intended for general release into the environment.

Less attention has been paid to the use of triploid hatchery fish as a genetic conservation measure in resident (nonanadromous) salmonid fisheries. Rohrer and Thorgaard (1986) suggested that stocking triploid rather than diploid rainbow x cutthroat trout hybrids in Henry's Lake, Idaho, would help conserve genetic integrity of the indigenous cutthroat trout. Brock et al. (1994) also describe the use of triploid rainbow trout in lake stocking to protect wild-stock integrity. However, we know of no published literature evaluating use of triploid hatchery trout in stream fisheries and only one unpublished evaluation. In that evaluation, return to creel was 60-70% for triploid rainbow trout stocked in high-use, poor-habitat segments of the Similkameen and Kettle rivers in British Columbia (D. Smith, British Columbia Fisheries Branch, unpublished data), which is a high return rate for stream fisheries. However, triploid fish were not compared with diploid fish so relative performance is unknown.

Although evaluations of triploid trout in streams are limited, techniques to produce triploid rainbow trout are well-developed, and triploid rainbow trout eggs have been available from a number of commercial egg suppliers for more than a decade. Thus, a strategy to minimize genetic impacts of hatchery trout on native trout might be to stock only triploid hatchery trout into streams containing native trout populations.

Before implementing such policy, however, studies evaluating the performance of triploid trout in stream fisheries are necessary. Triploid trout must provide fisheries at a reasonable cost and be acceptable to the public if they are to be a useful management tool. Catchable rainbow trout stocked in streams usually exhibit poor overwinter survival (Shetter 1947; Miller 1958; Bachman 1984) and are typically caught within the first 2 months of planting (Smith and Smith 1943; Cooper 1953). For this reason, success of catchable trout stocking programs in streams is usually based on return to creel rather than population parameters, such as growth rates or longevity.

In this study the performance of commercially provided triploid and diploid rainbow trout is compared in 18 stocked stream fisheries in Idaho. We compare their relative return to creel and also evaluate relative timing of returns as an indirect comparison of catchability or survival. The costs and potential management applications of triploid catchable trout are also discussed.

Methods

In spring 1996 we purchased 20,000 triploid and 20,000 diploid rainbow trout eggs from Mt. Lassen Trout Farms. Inc., Red Bluff, California. Triploidy was induced by heat-shocking mixed-sex eggs shortly after fertilization, following established methods (Thorgaard 1983: D. Brown. Mt. Lassen Trout Farms, Inc., personal communication). Eyed eggs were shipped on 20 June to IDFG's Nampa Hatchery in Nampa, Idaho, where incubation and rearing took place. Hatch rates were assessed for both groups by enumerating hatched fry and dividing by the number of eyed eggs. To describe relative cost to produce a triploid and diploid catchable, we summed egg and feed costs to rear all fish in both groups to catchable size. To estimate cost per fish stocked in both groups. egg and feed costs were divided by the number of fish surviving to catchable size. We assumed the hatchery capital costs and required rearing manpower were the same for both groups.

When test fish reached adequate size for blood sampling (50—75 mm, 3 months), 70 triploid and 10 diploid fish were sacrificed to confirm ploidy. We collected blood from individual fish by severing the caudal peduncle, and fixed the blood in Alsever's solution. Samples were shipped on ice to the Washington State University Veterinary Sciences Laboratory, where each sample was evaluated for ploidy using flow cytometry (Thorgaard et al. 1982; Utter et al. 1983).

From 20 May to 27 July 1997 we stocked each of 18 streams throughout Idaho with 300 triploid and 300 diploid rainbow trout (Figure 1). All fish were anesthetized with carbon dioxide, tagged with size-8 monel jaw tags, and held in hatchery raceways 8 h to 2 d before transport and stocking. Jaw tags were sequentially numbered to identify individual streams and treatment groups and were stamped "RTN IFG." Total length (TL), to the nearest millimeter, was measured for a subsample of each stocked group during the tagging process. The few mortalities observed between tagging and transport were replaced with fish from the appropriate treatment group. To characterize the range of sizes and productivities of study streams into which the fish were stocked, we measured channel width at a minimum of four sites near the stocking location, and quantified total dissolved solids using a digital meter.

To encourage tag returns, signs were placed along stocked sections of each stream informing anglers of the presence of tagged fish and providing mailing instructions. We specifically requested information on date and location of catch and angler address and phone number. As an incentive anglers were offered one chance at three gift certificates worth up to U.S. $200 for each tag returned. Because we sought only to compare relative returns for triploid and diploid fish, tag return data were not adjusted for nonresponse bias. Tag return data were compiled through the 1997 fishing season that ended on 30 November. Data were also compiled from tags returned during the 1998 fishing season, but because sample sizes were small, these data were not analyzed statistically.

Data Analysis

We completed an a priori power analysis for paired *t*-tests as part of the experimental design process (Cohen 1988; Peterman 1990). To choose an effect size we subjectively assumed that most fishery managers would elect to use rainbow triploid trout to reduce genetic risks to native stocks if triploids return to the creel at 75% the rate of diploid fish (effect size of 0.25). We further assumed tag return rates among streams would be 10—70% and that return of triploid and diploid fish within streams would be highly correlated (r = 0.80). The probability of a type I error (α) was set at 0.10. Based on these assumptions, the design with 18 paired stockings would provide a 98% chance of avoiding type II error for the effect size.

We used a two-sample *t*-test (Zar 1974) to compare mean total lengths at stocking (mm) for triploid and diploid fish in each stream stocked. We compiled tag return data (through 30 November 1997) for each group by stream and by time (d) between stocking and harvest. A paired t-test (Zar 1974) was used to test the hypothesis that the mean difference in tag returns from triploid and diploid fish was not significantly different from zero. In addition, we derived an estimate of mean time to harvest (d) for each group and stream using stocking dates and the harvest dates provided by anglers. A paired t-test was also used to test for a significant difference in mean time to harvest for the two groups. Data used in t-tests were tested for normality using the Komolgorov-Smirnov test (Zar 1974). Both tag return and timing data used in the paired *t*-tests were normally distributed ($P > 0.05$), and no data transformations were required.

Results

Survival to hatching was lower for triploid eggs (59.7%) than for diploid eggs (87.7%). Reduced survival of triploid fish occurred primarily in the first 2 months; survival between groups was similar thereafter. Estimated costs to catchable size, including only egg and feed costs and accounting for survival differences, were $0.38 for triploids and $0.33 for diploids.

Flow cytometry analysis confirmed that all (N = 70) of the putative triploid fish were triploid (*P.* Wheeler. Washington State University personal

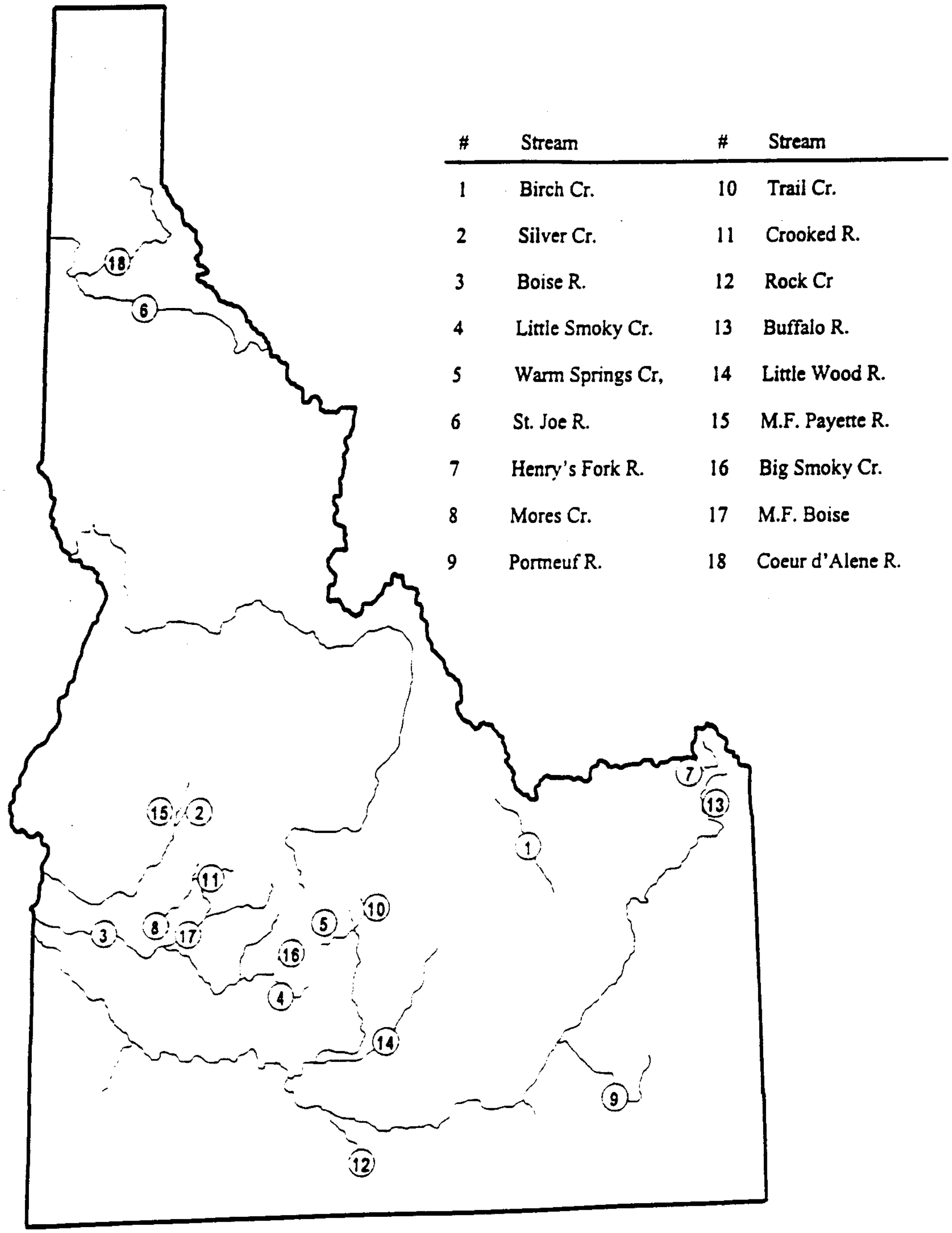

FIGURE 1.-Locations of 18 study streams used to compare performance of triploid and diploid catchable rainbow trout in recreational fisheries.

communication). The 10 control fish were all confirmed to be diploid.

Mean size at stocking differed slightly for the two groups ($P < 0.01$). Mean total length at stocking (all stockings combined) was 271.9 mm (SE 3.4 mm) for triploids and 256.2 mm (SE 4.5 mm) for diploids.

Relative tag returns for triploids and diploids varied by location across a broad range of stream sizes and productivities but was similar overall (Table I). Diploid fish returns exceeded triploid returns in nine streams, triploids exceeded diploids in seven streams, and triploid and diploid returns were equal in two streams. Of the total tag returns for all 18 streams, 931 were from triploid fish and 918 from diploid fish. The paired *t*-test indicated that the overall tag return rates for the two groups were not significantly different ($P = 0.80$). A total of 1,349 tags were returned from the 10,800 tagged fish stocked for an overall first-year return rate of

TABLE 1.-Stocking date, stream width, total dissolved solids (TDS), and first-year tag returns from each of 18 Idaho streams stocked with 300 fertile (diploid) and 300 sterile (triploid) catchable rainbow trout.

				Tag returns			
Stream	Stocking date	Width (m)[a]	TDS (mg/L)	Diploid	Triploid	Total	% return
Birch Creek	5 May	8	238	137	118	255	42.5
Silver Creek	25 Jun	11	26	78	82	160	26.7
Boise River	9 Jul	16	42	73	73	146	24.3
Little Smoky Creek	1 Jul	7	116	70	62	132	22.0
Warm Spring	1 Jul	9	101	63	63	126	21.0
St. Joe River	16 Jul	28	37	45	76	121	20.2
Henry's Fork River	20 May	45	67	56	54	110	18.3
Mores Creek	10 Jul	11	66	39	64	103	17.2
Portneuf River	15 Jul	17	310	51	46	97	16.2
Trail Creek	27 Jul	11	227	43	46	89	14.8
Crooked River	25 Jun	8	35	44	43	87	14.5
Rock Creek	2 Jul	7	109	34	42	76	12.7
Buffalo River	20 Jun	39	69	36	38	74	12.3
Little Wood River	10 Jul	8	149	39	32	71	11.8
MF Payette River	25 Jun	24	27	35	23	58	9.7
Big Smoky Creek	21 Jul	18	95	28	23	51	8.5
M.F. Boise River	15 Jul	32	--	28	20	48	8.0
Coeur d'Alene	16 Jul	36	43	19	26	45	7.5
Totals				918	931	1.849	17.1

[a] Stream width was a mean of at least four measurements taken at the fish planting location.

17.1% (Table 1). Because tag returns were not adjusted for nonresponse bias, true return rates are unknown.

For both stocked groups, most of the fish that returned to the creel during the first year were harvested relatively quickly. For all streams combined, the time for returns to reach 50, 75, and 90% of the cumulative total was 24, 41, and 57 d, respectively. Timing of returns for triploid and diploid fish was similar overall (Figure 2). The mean (± SE) time for 1997 returns was 28.1 ± 8.7 d for triploid fish and 30.2 ± 9.7 d for diploid fish. This difference was not significant ($P = 0.35$).

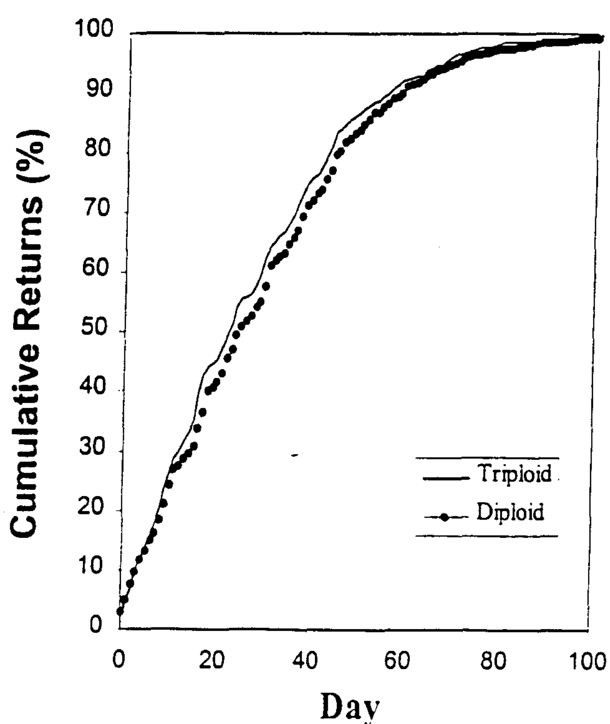

FIGURE 2.-Cumulative first-year returns to creel over time (100 d poststocking) for triploid and diploid hatchery rainbow trout in 18 streams combined.

Anglers caught few tagged fish during the second season after stocking. Tags from 29 diploids and 23 triploids were returned in 1998. These returns equate to 3.1% and 2.4%, respectively, of the first-year returns for each group.

Discussion

Numerous researchers have documented, from past stocking practices of hatchery trout, a range of genetic impacts on native fish, ranging from no detectable introgression (Krueger and Menzel 1979; Wishard et al. 1984: Jones et al. 1996) to virtually complete genetic admixtures or hybrid swarms (Campton and Johnston 1985; Gyllentsen et al. 1985). Fishery managers today more clearly recognize potential genetic risks than in the past, but still attempt to balance wild fish conservation with public and political pressures to provide consumptive angling opportunities (Van Vooren 1995). If triploid trout can provide the same quality of fishing as diploid hatchery trout, they will be a valuable tool for managers to use in addressing both issues. Our results provide strong evidence that, for stream fisheries, triploid rainbow trout can provide put-and-take harvest opportunities comparable to diploid fish.

Although our paired stocking design with 18 streams provided a relatively broad-based evaluation, relative return rates could have been confounded by differences in mean size at stocking between the two groups. Mean size at stocking for triploids (272 mm) was slightly higher (*P* < 0.01) than for diploids (256 mm), a difference of 16 mm. Because return to creel in streams is sometimes positively correlated with size at stocking (Mullan 1956; Walters et al. 1997), our results could have been affected by these slight differences in mean size. However, statistical difference does not necessarily equate to biological significance (Gold 1969; Steidl et al. 1997). Subsequent research on the effect of a larger (49 mm) average size difference on returns of catchable trout in the same 18 study streams demonstrated that size at stocking did not influence returns (D. Teuscher, Idaho Department of Fish and Game, unpublished data). Therefore, the much smaller difference in mean size at stocking in our experiment is unlikely to have influenced our results.

Our primary evaluation criterion was relative return to creel; we did not directly assess long-term survival, growth, or behavioral differences between triploid and diploid groups. Nonetheless, timing of first-year returns suggests that survival and catchability were similar. In both groups, more than 90% of the first-year returns occurred within 57 d of stocking. Because the rainbow trout used in this experiment were highly domesticated, we did not expect significant long-term or overwinter survival in our study streams (Shetter 1947; Miller 1958; Bachman 1984). This appeared to be the case for both triploids and diploids, only 2–3% of the total tag returns coming in the second year. Such tag-return data should equate to annual survival (Ricker 1975: 102). Although sample sizes are small, this suggests that overwinter survival of triploid and diploid fish in this experiment was low and similar between groups. However, behavioral differences, if they occur, could mean that triploid fish could have unexpected interactions with wild fish. We suggest that future evaluations monitor long-term survival and behavioral differences between triploid and diploid groups to clearly describe interactions with wild fish.

We specifically requested mixed-sex triploid and diploid groups for this experiment because Idaho currently uses mixed-sex hatchery rainbow trout in many stocking programs statewide. However, all-female rainbow trout are the triploid fish most commonly available from commercial sources. Although all triploid salmonids are functionally sterile, triploid males still undergo some sexual differentiation and may exhibit spawning behavior (e.g., migration), whereas triploid females do not (Warrillow et al. 1997; Benfey 1999). Theoretically, if triploid males survive to attempt spawning, they could displace fertile males and spawn with wild females. Because triploid females develop no secondary sex characteristics and exhibit no spawning behavior, they have virtually no potential to affect recruitment of native fish. Although we found no published literature documenting triploid males attempting to spawn with native trout females, we suggest future evaluations of stream-stocked triploid rainbow trout should include all-female triploids. Still, given the poor overwinter survival of stream-stocked hatchery trout observed in this study and others (Shetter 1947; Miller 1958; Bachman 1984), the risk of triploid male hatchery trout measurably reducing native fish recruitment appears low.

Triploid rainbow trout are available from commercial sources, but a 100% triploidy rate is not guaranteed for eggs at the time of purchase. Although all of our tested fish in this experiment (*N* = 70) were confirmed triploids, commercial producers typically assure 85–100% triploid rates. Because ploidy is most commonly evaluated with flow cytometry of blood samples, triploid rates in individual egg lots received from commercial facilities have not been verified. Ideally, triploidy rates should be confirmed for all commercially provided triploid eggs; however, the relatively high cost of flow cytometric evaluation may preclude this approach if use of triploid fish becomes common. If ploidy verification of commercial fish consistently indicates sterility rates above 90%, periodic sampling would probably suffice.

Increased production cost is also a consideration that could affect applicability of triploid fish in stocking programs. In this experiment, costs for triploid rainbow trout eggs were 2.3 times the cost of diploid eggs, and hatch rate for the triploid group was lower. Other researchers have documented lower hatch rate for triploid eggs (Solar et al. 1984; Chourrout et al. 1986). Most of the expense of rearing catchable-size trout, however, is feed costs rather than egg costs, and our estimated total cost difference between diploid and triploid catchables was only about \$0.05. This value includes the differences in egg costs and survival between the two groups. With only one replicate and no direct measurement of food conversion for the two groups, this assessment of relative cost should be considered preliminary.

If triploid rainbow trout were to make up a significant portion of hatchery production, differences in rearing costs would need to be accounted for by either increasing hatchery budgets or by slightly reducing total production and stocking rates. Fishery managers and policy makers must assess the tradeoffs of higher stocking costs or decreased stocking rates versus the ability to afford genetic protection to wild fish. Agencies can also minimize costs of triploid hatchery trout by inducing triploidy in eggs from their own broodstocks rather than purchasing eggs from commercial suppliers. Based on the results of this study, Idaho has begun inducing triploidy in 600,000 rainbow trout eggs annually, enough to meet statewide demand for stream stocking of catchables.

In addition to genetic concerns, there are other biologically based issues regarding hatchery and wild trout interactions. Some biologists suggest that direct competition with hatchery trout has negative effects on wild trout, but the evidence to date on competition appears equivocal (Miller 1958; Marnell 1986; Vincent 1987; Petrosky and Bjornn 1988). Given recent experience with whirling disease in western U.S. trout fisheries, disease transmission from hatchery to wild fish is also a concern. Such concerns must be addressed in agency fish culture and stocking policies and include stringent regulations prohibiting release of fish carrying harmful pathogens.

Assuming our study results are accurate and replicable, triploid salmonids may have utility in a variety of stream fisheries nationwide. For example, Morgan and Danzman (1997) noted widespread introgression of wild brook trout stocks from hatchery brook trout introductions in eastern U.S. streams. Several authors have discussed the deleterious effects of continued rainbow trout stocking on imperiled Gila trout *O. gilae,* Apache trout *O. apache,* and Rio Grande cutthroat trout *O. clarki virginalis* (Dowling and Childs 1992; Stumpff and Cowley 1997), the former authors calling for studies to identify barriers to gene exchange with introduced rainbow trout. Use of triploid fish in stocking programs would seem to have potential in such situations.

Given the history of genetic impacts from hatchery fish introductions and the likelihood that public demand for consumptive stream fisheries will continue, fishery managers must find innovative ways to meet competing agency mandates. Triploid hatchery trout represent a potentially valuable tool that can help managers balance public demand for harvest opportunity with sound conservation strategies for wild trout. Additional research and management evaluation is needed to more fully explore this potential.

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