UTAH DIVISION OF WILDLIFE RESOURCES
FISHERIES EXPERIMENT STATION
1465 WEST 200 NORTH
LOGAN, UT 84321

The Ichthyogram

VOLUME 18, NUMBER 2

OCTOBER 2007

INSIDE THIS ISSUE:

Comparison, at two temperatures, of heat and pressure shock production-scale triploidy induction in rainbow trout and subsequent hatchery performance

Introduction

Triploid rainbow trout have been a valuable tool to fish managers around the state of Utah over the past five years. Because the triploid rainbows are sterile, the possibility of crossbreeding with cutthroat can be avoided, so it allows for continued rainbow stocking meeting sport fishing needs, while at the same time allows for the management of various cutthroat trout subspecies. The use of triploid rainbow trout is not the answer to all management problems however. With triploid trout, research has found elevated mortality during the egg and sac fry staged (Happe et al. 1988), and poor survival when reared in chronically high temperatures (21° C; Ojolick et al. 1995). Several hatchery mangers in Utah have suggested that triploid rainbow don't survival as well as their diploid counterparts during stressful events such as crowding and stocking, and there also seems to be some very anecdotal evidence, within the Utah hatchery system, that triploid rainbows reared in waters above 15° C survive better than they might at cooler temperatures. To address these issues and possibilities, a cohort of diploid and triploid rainbow trout were, were reared at the Fisheries Experiment Station (FES) to analyze for differences in hatchery performance, when reared at two different water temperatures.

Methods

During the course of a spawning day at the Egan State Brood Hatchery, three separate lots of eggs were produced each consisting of gametes from approximately 13 males and eight females. Each of those three lots were further divided into thirds, with one third being heat shocked, one being pressure shocked, and the last surviving as a diploid control. Heat shocked eggs were treated at 26.5°C at 20 min after fertilization for 20 min. Pressure treated eggs were pressurized to 9800 psi for 5 min at 20 min after fertilization. Controls were handled as if they were to be pressure treated, but no pressure was applied and temperature was at the hatchery's ambient level, 9°C. Three different lots were fertilized, generating 8,524 to 10,367 eggs per replicate per treatment. The lot replicates were kept in separate incubator trays, however when the sac-fry were put into troughs, all three trays from a given treatment were combined into one trough due to space limitations. When the fish were large enough to move to outdoor raceways (mean weight, 0.7 to 0.9 g among all raceways), the fish were divided into 12 raceways, 2700 per raceway. Each of the three treatment groups were assigned to two different temperatures (13.4 and 16.7°C) for rearing. Temperatures were based on those available in our well water system. Space was limited, so only two replicate raceways were used for each treatment.

Fish were reared for roughly five months during which time they were sampled monthly to determine mean weight for estimating feed rations. Weight estimates were derived from 3 subsamples from each raceway. Mortalities were enumerated during the study. Feed conversion ratios were calculated as a ratio of feed fed to weight gain of fish. Fish were fed a commercial trout diet (Silvercup, Salt Lake City, Utah). Specific growth rates were determined for fish in each raceway using the formula, [$\log_e$(final weight in g)- $\log_e$(initial weight in g)]/days on feed *100.

Fin measurements were made on day 48 and 133 for 15 fish from each raceway and transformed to relative fin index values using the methods of Kindschi (1987). Necropsies were conducted at the end of the study (day 135) on 10 fish from each raceway using the Health and Condition Profile (HCP) methodology (Goede and Barton

(Continued on page 2)

(Continued from page 1)
1990).

For verification of ploidy level, blood samples were collected from 20 putative diploids and from 30 fish in each of the triploid treatments. Samples were collected by caudal venipuncture and diluted with Alsere's anticoagulant in the syringes. The blood was shipped cold to a lab for determination of ploidy by flow cytometry (Thorgaard et al. 1982).

Statistical analysis— SPSS version 13 was used for all computations and a significance level of 0.05 was used throughout. For analysis of percent mortality, values were arc-sine transformed prior to analysis. The Kolmogorov-Smirnov test was used to evaluate the assumption of normality for all response variables. If a variable was not normally distributed, non-parametric tests were used. This was necessary for analysis of feed conversion, for which values were rank transformed prior to use in a general linear model (GLM). The GLM used temperature and treatment (control, heat, or pressure) as fixed variables in a saturated design using type III sums of squares. For categorical data (fin, thymus and fat indices), a hierarchical log linear model was used to test the effects of temperature and treatment. For HCP fin data, the 3-way interaction term was significant, so separate partial tables (Fienberg 1980) were used for further chi-square analysis and using maximum likelihood estimators.

Results and Discussion

Data on egg survival indicated that pressure treatment was superior to heat shock. Survival to the eyed egg stage was significantly lower for heat shocked eggs (83.1%) than for pressure treated (89.6%) or control eggs (91.3%). Survival to hatching was lower for heat shocked fish (83.4%) than controls (91.4%), but percent hatch of pressure treated eggs (87.0%) did not significantly differ from either of the other two treatments. It has been demonstrated that triploid eggs and fry generally have lower survival than diploid counterparts (Happe et al. 1988).

Once the fish were transferred to outdoor raceways, growth differences attributable to temperature differences were significant, but there were few differences in hatchery performance and fish health among the triploidy induction methods. Fish in the higher temperature raceways were significantly heavier and had higher specific growth rates at the end of the study than fish at 13.4°C (Table 1). However, feed conversion and mortality rates did not significantly differ between the two temperatures (Table 1). Ploidy treatment effects were not significant for final weight, specific growth rate, feed conversion, or mortality. The data indicated that once the fish were transferred to outdoor raceways, there was no difference between diploid and triploid performance regardless of the two rearing temperatures.

Table 1. Hatchery performance of diploid and triploid rainbow trout reared at two temperatures; triploid groups eggs that were either heat shocked or pressure shocked.

	Control		Heat shocked		Pressure shocked	
Temperature (°C)	13.4	16.7	13.4	16.7	13.4	16.7
Specific growth rate (% per day)	2.27	2.35	2.29	2.38	2.21	2.43
	± 0.01	± 0.02	± 0.02	± 0.01	± 0.03	± 0.08
Final weight (g)	16.5	25.8	18.48	25.45	16.7	26.13
	± 0.16	± 0.77	± 0.42	± 0.53	± 0.68	± 2.89
Cumulative mortality (%)	0.72	2.29	2.64	2.33	2.03	1.88
	± 0.02	± 0.29	± 0.79	± 1.07	± 0.22	± 0.18
Feed conversion ratio (FCR)	1.13	1.12	1.00	1.04	1.08	1.04
	± 0.00	± 0.02	± 0.00	± 0.00	± 0.05	± 0.07

Analysis of the Health and Condition Profile variables indicated that length, weight, and condition factor were significantly higher at the higher temperature, but hematocrit, leucocrit, and plasma protein values did not significantly differ between temperature treatments. Eye, gill, pseudobranch, spleen, kidney, and liver tissues were all normal, unaffected by temperature or ploidy induction method. Thymus index values similarly did not differ among treatments. Fat levels were high in all treatments (score of 3 or 4 among all fish). Fin erosion was significantly higher (higher frequency of '2' score) in the pressure treated group ($P = 0.017$) and at 16.7°C for control fish ($P = 0.011$), but the distribution of fin index frequencies was not significantly different for fish in the heat shocked group ($P = 0.437$). In the pressure treated group, there were significantly fewer ($P = 0.027$) '0' scores at the higher temperature, but significantly fewer '2' scores.

Previous research has indicated that triploid rainbow may exhibit better fin condition than diploids when reared under identical conditions (Wagner et al. 2006). The fin index results from this study reinforce those previous ones. When making comparisons across all treatments, there were no significant differences found. Also, when data were lumped together to make a comparison of fish reared in 13.4 versus 16.7° C, no significant differences were found, with the exception of ventral fins, which were significantly better (P = 0.017), on the second sampling date, for fish reared in the warmer water. Comparisons of all triploids compared to diploids indicated uniformly better fins among the triploids for caudal, ventral, pelvic and pectoral fins by the second sampling date.

Ploidy analysis by flow cytometry indicated that all putatative diploids were diploid and 96.6% and 100% of pressure treated and heat shocked groups, respectively, were triploid. In Utah, the heat shock methodology has been very reliable over the past five years with triploidy percentages ranging from 97-100%. Routine quality control checks are made of yearly production with samples taken across spawning dates and from the three main rainbow trout strains cultured.

Overall, it appears that temperature was more of a factor during this study than ploidy status. Pressure-induced triploids may have exhibited a slightly better early life history survival than the heat-induced fish, but this did not carry over to later life history stages. The pressure method is also limited in the number of eggs that can processed during spawning events compared to the heat shock method, so the justification of switching from heat shock to pressure to improve egg and fry survival may be weak due to the absolute number of eggs each method can be produced. Although it has been shown that triploid rainbow trout have a higher mortality when reared at chronic high temperature of 21° C, (Ojolick et al. 1995), rearing them within the temperature range we used, 13.4 - 16.7° C, should be without difficulty.

Ronney Arndt, Eric J. Wagner, M. Douglas Routledge, and Roger Mellenthin

Literature Cited

Fienberg, S.E. 1980. The analysis of cross-classified categorical data, 2nd edition. MIT Press, Cambridge, Massachusetts.

Goede, R.W., and B.A. Barton. 1990. Organismic indices and an autopsy-based assessment as indicators of health and condition of fish. Pages 93-108 *in* Adams, M. editor. Biological indicators of stress in fish. American Fisheries Society Symposium 8, Bethesda, Maryland:

Happe, A., Quillet, E. and B. Chevassus. 1988. Early life history of triploid rainbow trout (*Salmo gairdneri* Richardson). Aquaculture 71:107-118.

Kindschi, G.A. 1987. Method for quantifying degree of fin erosion. Progressive Fish-Culturist 49:314-315.

Leggatt, R. A., Scheer, K. W., Afonso L. O. B., and G. K. Iwama. 2006. Triploid and diploid rainbow trout do not differ in their stress response to transportation. North American Journal of Aquaculture 68:1-8.

Ojolick E. J., Cusack, R., Benfey, T. J., and S. R. Kerr. 1995. Survival and growth of all-female diploid and triploid rainbow trout (*Oncorhynchus mykiss*) reared at chronic high temperatures. Aquaculture 131:177-187.

Wagner, E. J., Arndt, R. E., Routledge, M. D., Latremouille, D., and R. F. Mellenthin. 2006. Comparison of hatchery performance, agonistic behavior, and poststocking survival between diploid and triploid rainbow trout of three different Utah strains. North American Journal of Aquaculture 68:63-73.

Coldwater Disease Vaccine: Promising Preliminary Results

A recent trial has demonstrated significantly reduced mortality in vaccinated rainbow trout challenged with *Flavobacterium psychrophilum*, the causative agent of bacterial coldwater disease. The disease is also known as 'rainbow trout fry syndrome' or 'peduncle disease' and is a significant disease of fishes around the world. Mortality is especially acute in younger fish, resulting in mortality rates of up to 90%. The clinical signs of disease can include necrotic lesions, skin darkening in the peduncle area, loss of equilibrium, dorsal swelling just posterior to the skull, spinal deformities, and spiral swimming. Antibiotics have traditionally been used to control the disease, but concerns about drug resistance and cost led to research into immunological approaches of control.

Several attempts have been made to produce an efficacious vaccine against the bacterium using whole cells (Holt 1987; Obach and Laurencin 1991; Madetoja et al. 2006). These authors and others (Rahman et al. 2002; LaFrentz et al. 2002) noted that survival and antibody titers were significantly higher in fish vaccinated with adjuvants. Adjuvants are compounds injected with the vaccine that help stimulate the immune system. Further studies have shown that use of immunogenic whole cell fractionations provide better protection than whole cell vaccines (Rahman et al. 2002; Gudmundsdottir and Magnadottir 1997). The fractions that appear to be most useful are those with larger molecular weight. (Velji et al. 1992; LaFrentz et al. 2004).

Based on the studies noted above, a vaccine that used an adjuvant and immunogenic fractions injected intraperitoneally (i.p.) appeared to be the best approach to pursue. The objective of this study was to evaluate the efficacy of a subunit (>50 kD) vaccine for reducing mortality after challenge with *F. psychrophilum*. The vaccine efficacy was compared among four treatments: (1) vaccine alone, (2) vaccine mixed with Freund's Incomplete Adjuvant, (3) a phosphate buffered saline (PBS) positive control, and (4) a PBS negative control.

Methods

Experimental Fish and Rearing Conditions — Rainbow trout fry were obtained from the Fisheries Experiment Station Hatchery, Logan, Utah. For each treatment, triplicate groups of 20 fish were maintained in 12˚C water in aerated 75 L aquaria. The aquaria were part of a recycle system in which water was collected in a sump, pumped to a head box, then distributed via a manifold to each aquarium. Each group of three aquaria was on a separate exchange system which had 19 L of hatchery well water exchanged per day. Fish were fed to apparent satiation two times per day with a pelleted commercial trout feed. These fish were certified specific pathogen free, but were not tested specifically for the presence of *F. psychrophilum*.

Bacterial culture — A virulent strain of *F. psychrophilum* (CSF-259-93) cultured on tryptone yeast extract salts (TYES) agar plates and in MAT broth was used for the vaccine and challenge trials.

Vaccine preparation — For the immunization trials, bacteria l cultures were processed in a manner that allowed for the retention of all cellular fractions >50kDa. Protein concentration was determined and samples were appropriately diluted with either PBS or with Freund's Incomplete Adjuvant (FIA) to achieve a dosage for each fish of 40 µg protein per 50 µL volume.

Vaccination and sampling — Fish were anesthetized by immersion in tricaine methane sulfonate (MS-222) and triplicate groups of 20 were i.p. immunized with either (1) PBS (positive and negative control treatments) (2) 40 µg of the >50kDa fraction (vaccine-only treatment), or (3) 40 µg of the >50kDa fraction + FIA (vaccine + adjuvant treatment). Intraperitoneal injections were administered just posterior to the pelvic fins using a 27-gauge needle. All fish received booster immunizations after six weeks, following the original dose and treatment regime of the primary injection.

At both six and twelve weeks, five fish from each group were lethally sampled and serum was collected for

(Continued on page 5)

specific antibody assays. Blood was collected into tubes after severing the caudal peduncle, was. The bBlood was allowed to clot overnight at 4°C, was then centrifuged and serum was removed and stored at -20°C. Serum samples were collected from 50 randomly selected fish at the start of the trial to provide a baseline sample for testing by enzyme linked immunosorbent assay (ELISA).

Bacterial Challenge — Twelve weeks after initial immunization, the remaining fish (average weight 14.54 g) from the vaccination trial were challenged with either PBS (negative control) or live *F. psychrophilum* bacteria (positive control and vaccinated groups). Bacterial culture (as described above) was resuspended in PBS to an optical density of 0.4 measured at 525 nm (OD_{525}) approximating 5 x 10^7 colony forming units per mL (Holt 1987). Fish were anesthetized and injected with 50 µL of the bacterial suspension subcutaneously immediately posterior to the dorsal fin. Mortalities were recorded for 14 days and 20% of the fish were examined for the presence of *F. psychrophilum* after spleen inoculation onto TYES agar.

Statistical analysis — The percent mortality was arc-sine transformed prior to analysis. The normal distribution assumption was tested using the Kolmogorov-Smirnov and Shapiro-Wilkes tests (SPSS 1996, version 13.0). Differences in the percent cumulative mortality among treatments were tested with one-way analysis of variance. A significance level of 0.05 was used for all tests.

Results

Mortality was significantly lower in fish vaccinated with the subunit vaccine-adjuvant mix (19.7%) than in fish vaccinated without adjuvant (79.6%) or not vaccinated at all (90.0%; positive control) (Fig. 1). Blood samples have yet to be analyzed for specific antibody titer (data is pending). Lesions were observed on the positive controls and many of the mortalities at the injection site for the challenge. Gram stains of the cultured bacteria indicated that they were long, thin, $G^{(-)}$ rods. The biochemical profile of the isolates was also consistent with *F. psychrophilum,* indicating that the bacterium was the cause of the mortalities.

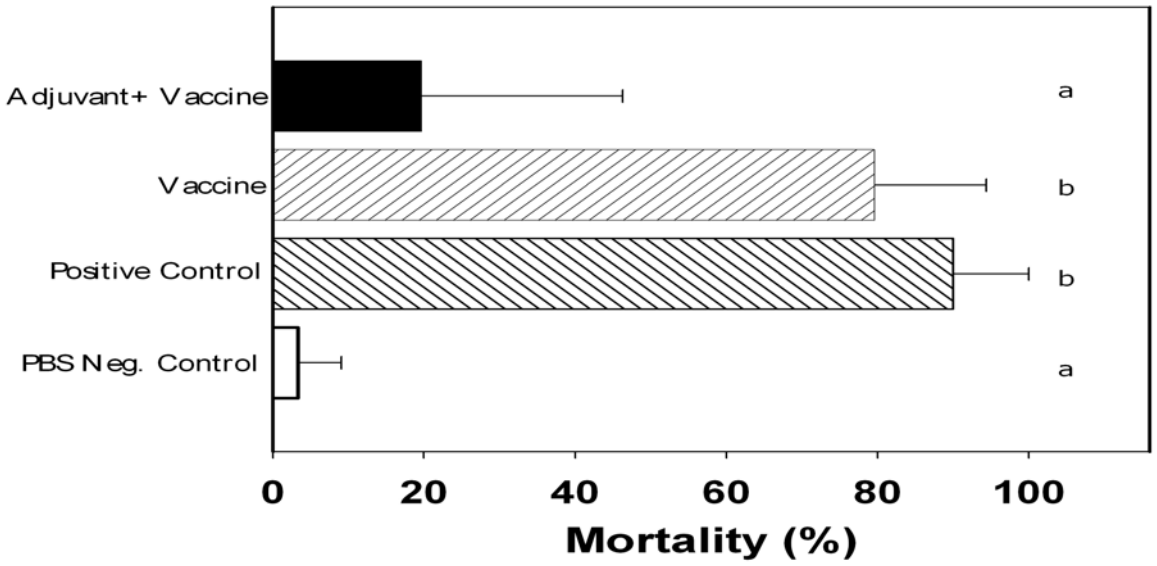

Fig. 1. Percent mortality for rainbow trout injected with either phosphate buffered saline (PBS), a subunit vaccine, or a vaccine mixed with Freund's Incomplete Adjuvant. All fish except negative controls were challenged with an intramuscular injection of *F. psychrophilum*.

(Continued on page 6)

(Continued from page 5)

Literature Cited

Gudmundsdóttir, B.K., and B. Magnadóttir. 1997. Protection of Atlantic salmon (*Salmo salar* L.) against an experimental infection of *Aeromonas salmonicida* ssp. Achromogenes. Fish and Shellfish Immunology 7:55-69.

Holt, R.A. 1987. *Cytophaga psychrophila*, the causative agent of bacterial cold-water disease in salmonid fish. Doctoral Thesis, Oregon State University, Corvallis.

LaFrentz, B.R., S.E. LaPatra, G.R. Jones, J.L. Congleton, B. Sun, and K. Cain. 2002. Characterization of serum and mucosal antibody responses and relative per cent survival in rainbow trout, *Oncorhynchus mykiss* (Walbaum), following immunization and challenge with *Flavobacterium psychrophilum*. Journal of Fish Diseases 25:703-713.

LaFrentz, B.R., S.E. LaPatra, G.R, Jones, and K.D. Cain. 2004. Protective immunity in rainbow trout *Oncorhynchus mykiss* following immunization with distinct molecular mass fractions isolated from *Flavobacterium psychrophilum*. Diseases of Aquatic Organisms 59:17-26.

Madetoja, J., L.-G. Lönnström, C.Björkblom, G. Uluköy, G. Bulund, C. Syverstsen, K. Gravningen, E.A. Norderhus, and T. Wiklund. 2006. Efficacy of injection vaccines against *Flavobacterium psychrophilum* in rainbow trout, *Oncorhynchus mykiss* (Walbaum). Journal of Fish Diseases 29:9-20.

Obach, A., and F.B. Laurencin. 1991. Vaccination of rainbow trout (*Oncorhynchus mykiss* against the visceral form of coldwater disease. Diseases of Aquatic Organisms 12:13-15.

Rahman, M.H., A. Kuroda, J.M. Dijkstra, I. Kiryu, T. Nakanishi, and M. Ototake. 2002. The outer membrane fraction of *Flavobacterium psychrophilum* induces protective immunity in rainbow trout and ayu. Fish and Shellfish Immunology 12:169-179.

Velji, M.I., L.J. Albright, and T.P.T. Evelyn. 1992. Immunogenicity of various *Vibrio ordalii* lipopolysaccaride fractions in coho salmon *Oncorhynchus kisutch*. Diseases of Aquatic Organisms 12:97-101.

Wade Cavender and Christine Swan

Transitions at FES

The last few months have seen an unusual turnover of fish culture personnel at the Fisheries Experiment Station.

Roger Mellenthin has filled the position of assistant hatchery supervisor, replacing Chad Hill who transferred to Ft. Green hatchery. Congratulations to Roger!

The 3 new amigos: Travis Dees, Jason Tull and Roger Mellenthin.

Replacing Roger as wildlife specialist is **Jason Tull**, who transferred to FES from Mantua hatchery. Jason graduated from Arizona State Univ. with a BS in Conservation Biology. Welcome Jason!

Jared Smith has left Utah for greener pastures at the Auburn state fish hatchery at Star Valley, Wyoming. Best wishes to him!

Replacing Jared is **Travis Dees**, who worked formerly as a technician during the summer. Travis is a senior, majoring in Wildlife and Fisheries at USU.

Decapsulated Artemia as an Initial Feed item for Colorado River Cutthroat Trout

Colorado River cutthroat trout *Oncorhynchus clarkii pleuriticus* are routinely cultured at the Fisheries Experiment Station (FES), Logan, to meet the needs of both conservation and sportfishing efforts. For production, eggs are taken and fertilized using wild brood fish and then shipped to the FES, where eye-up and hatch are generally above 85%. However, mortalities for the first month or two after first feeding may exceed 25%. Newly hatched *Artemia* nauplii can be a good source of nutrition to larval fish, and have been a mainstay of marine-fish larval culture for several decades (Sorgeloos et al. 2001). Previous research at the FES has demonstrated that a feeding cycle using *Artemia* nauplii can improve the survival of Colorado River cutthroat trout (Arndt and Wagner 2007). The culture and maintenance of live *Artemia* cultures can be a time consuming endeavour, however. By feeding dried decapsulated *Artemia* cysts to chub, *Leuciscus cephalus* (L.), Harzevili et al. (2003), obtained superior survival compared to other feeding regimes including live *Artemia* nauplii. By decapsulating cysts and subsequently drying them, a good source of larval nutrition could be obtained at a labor savings compared to live nauplii culture. The purpose of this research was to determine whether or not dried, decapsulated *Artemia* cysts would be a good source of nutrition to first-feeding cutthroat trout.

The source of the fish for this test was from a wild brood lake, Sheep Creek Lake, located on the north slope of the Uinta Mountains, which ultimately drains to the Colorado River. The eggs were fertilized on site and then brought to the Fisheries Experiment Station, where they further developed until hatching. When approximately 50% of the recently hatched fish had absorbed their yolk sac and were swimming in the water column they were randomly distributed (200/tank) into twelve 26 L tanks that had a flow of 0.5-0.7 L/min. All four treatments were represented in triplicate. This tank system was housed inside a hatchery building provided with 10 h artificial light per day and with additional, incidental light from a nearby window. Mortalities were removed daily in between feeding events. Beginning at 0700 through 1600, the fish were offered feed hourly. The amount of feed was not quantified as wasted feed is ascertained at this early stage of development. The feed treatments consisted of a commercial swim up diet (Silver Cup, Nelson and Sons, Inc.), live Artemia nauplii, dried decapsulated Artemia cysts, and a second commercial swim up diet (Skretting, Vancouver, Canada). The Silver Cup diet served as a negative control of sorts because it had been used previously with poor results, and the Skretting diet was being tested as a higher end larval diet whose use had been suggested by biologists from the U.S. Fish and Wildlife Service.

The brine shrimp (Great Salt Lake Artemia franciscana) were cultured as outlined by Treece (2000), and the decapsulted, dried cysts were prepared as outline by Harzevili et al. (2003). The fish were fed the above diets for a total of 18 days, after which the trial was ended and fish were inventoried from all tanks.

Over the course of the 14-day test, survival was not impacted one way or another by treatment application. All treatments exhibited an average survival of 90% (± 1.4). However, growth was

(Continued on page 8)

(Continued from page 7)

influenced by treatment type. Fish fed the Skretting diet had the highest individual fish weight, 0.32 g/fish, compared to 0.24 for the Silver Cup diet, 0.18 for fish fed dried cysts, and 0.14 for fish fed *Artemia* nauplii. Growth values for each treatment were significantly different from the other treatments ($P < 0.001$). The amount of time and labor involved was not quantified, but it was evident that larger batches of the dried cysts could be produced more quickly than producing live *Artemia* nauplii.

For the culture of fresh water species, *Artemia* can play an important role in early life survival. When European chub *Leuciscus cephalus* were fed decapsulated *Artemia* cysts, *Artemia* nauplii, rotifers and Daphnia, and an artificial diet, they grew best on the *Artemia* nauplii diet (Harzevili et al. 2003), but survival was better within the group fed the dried decapsulated cysts. Because *Artemia* are a live prey item, it is possible that larval cutthroat trout may prefer them to feed particles. Live nauplii survive for 1-2 hours when placed into fresh water. During that time they actively swim and being positively phototactic, may be more inclined to orient themselves in mid water column or even close to the surface. Commercial feed items may only remain suspended momentarily in the water column after which they fall to the tank bottom where they may remain uneaten by the fish. The theory that larval fish prefer live or active feed items has been supported by the work of Fernández-Diaz et al. (1994) who determined gilthead sea bream larvae *Sparus aurata* ingested more living prey items compared with inert particles. The concluding results from this small test indicated that either commercial diet exhibited similar survival and superior growth to either *Artemia* treatment. The dried, decapsulated *Artemia* were superior to live nauplii with respect to growth, so their use may be desirable in conditions where it is known that the use of brine shrimp is necessary.

Ronney Arndt and Eric Wagner

Citations

Arndt, R. E., and E. J. Wagner. 2007. Enriched *artemia* and probiotic diets improve survival of Colorado River cutthroat trout larvae and fry. North American Journal of Aquaculture 69:190-196.

Fernández-Diaz, C., Pascual, E., and M. Yufera. 1994. Feeding behaviour and prey size selection of gilthead seabream, *Sparus aurata*, larvae fed on inert and live food. Marine Biology 118:323-328.

Harzevili, A. S., De Charleroy, D., Auwerx, J., Vught, I., and J. Van Slycken. 2003. Larval rearing of chub, *Leuciscus cephalus* (L.), using decapsulated *Artemia* as direct food. Journal of Applied Ichthyology 19:123-125.

Sorgeloos, P., Dhert, P., and P. Candreva. 2001. Use of the brine shrimp, Atremia spp., in marine fish larviculture. Aquaculture 200:147-159.

Treece, G. D. 2000. *Artemia* production for marine larval fish culture. Southern Regional Aquaculture Center, Publication 702, Stoneville Mississippi.

Utah Division of Wildlife Resources
Fisheries Experiment Station
1465 West 200 North
Logan, UT 84321

Phone:435-752-1066
Fax: 435-752-6977
Email: chriswilson@utah.gov

The Ichthyogram is a quarterly publication of the Fisheries Experiment Station, Utah Division of Wildlife Resources, Logan Utah 84321.
Editor:
Chris Wilson (chriswilson@utah.gov)
Contributors:
Doug Routledge (dougroutledge@utah.gov)
Eric Wagner (ericwagner@utah.gov)
Wade Cavender (wadecavender@utah.gov
This newsletter is distributed electronically at http://wildlife.utah.gov/fes/ichthyogram.html
Send comments to: chriswilson@utah.gov

We're on the web!
http://wildlife.utah.gov/fes/

Culture • Fish Health • Research