

# The Ichthyogram

VOLUME 18, NUMBER 3-4

DECEMBER 2007

## INSIDE THIS ISSUE:

### Hatchery Performance of a Whirling-Disease Resistant Strain of Rainbow Trout

New Face at FES 2

The hatchery and field performance of a whirling disease resistant strain of rainbow trout (*Oncorhynchus mykiss*) was compared to a traditional Utah strain: Ten Sleep (TS). The resistant strain consisted of progeny from a German strain (GR) x Harrison Lake strain (HA) backcrossed with the German strain, resulting in fish that were  $\frac{3}{4}$  German strain x  $\frac{1}{4}$  Harrison Lake strain. The disease resistance of the GR and HA strains have been documented in laboratory studies by Hedrick et al. (2003) and Wagner et al. (2006). The GR strain was originally from the Gunnison River in Colorado, shipped to Germany in the 1880s and subsequently used for aquaculture. Now, over a century later, after exposure to *Myxobolus cerebralis* for generations, the strain is being evaluated for use in stocking programs in Colorado, Utah and other states.

WD Resistant Trout at Low/High Rations 3

Field Performance of WD Resistant Trout 5

New Findings of *Myxobolus cerebralis* 7

As part of an ongoing study to evaluate the hatchery and field performance of GR x HA, a second cohort of both TS and GR x HA were reared and stocked into two study reservoirs. This article summarizes the hatchery performance of the second cohort of GR x HA relative to TS.

#### Methods

Eggs of the GR-HA strain were generously provided by the Colorado Division of Wildlife and TS strain provided by the Egan State Fish Hatchery, Bicknell, UT. The GR-HA eggs were from 25 different females. These eggs were allocated to 7 different trays of a Heath incubator stack upon arrival as eyed eggs (27 Dec 06). These eggs hatched 31 Dec 2006. The TS eggs also arrived as eyed eggs to the Fisheries Experiment Station, Logan, UT, were allocated to 7 trays, and hatched 9 Jan 2007. Dead eggs were enumerated and removed from each tray, recording the number removed in each. Percent hatch was calculated as a ratio of the number of surviving eggs to the number of eyed eggs received. The percentage of crippled and dead fry within 2 days after hatch was expressed as a percentage of hatched eggs.

After 2 months of fry rearing, 10,000 fish of each strain were allocated to each of three grow-out raceways (11 m x 1.2 m x 0.6 m deep). Each strain was reared at similar densities (Piper density index < 0.32). Monthly sample counts were made ( $n = 3$  per raceway) to assess growth and project new rations based on an expected growth rate. Fish were fed 7 d/ week, adjusting amounts if waste feed was observed. Specific growth rates were calculated for each raceway using the formula:  $SGR = (\log_e(\text{final weight in g}) - \log_e(\text{initial weight})) / \text{days} * 100$ . Mortalities were recorded for each raceway and feed conversions were expressed as the ratio of feed fed (dry weight) to weight gain (wet weight).

#### Results and Discussion

The percent hatch averaged 91.3% for TS and 92.7% for GR x HA, and did not significantly differ ( $P = 0.415$ ). Specific growth rates ranged from 2.5 to 2.8 among all replicates and was significantly higher for GR x HA than for TS ( $P < 0.001$ ). Feed conversions were also significantly better for the hybrid than TS ( $P = 0.001$ , Table 1). Mortality was nearly significantly higher ( $P = 0.057$ ) for TS than GR x HA. Mortality was influenced by a bacterial infection during rearing, which was more severe in TS raceways.

(Continued on page 2)

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Table 1. Comparison of hatchery performance (mean  $\pm$  SE) between a whirling disease resistant strain of rainbow trout (GR x HA) and a traditional Utah strain (TS). Significant differences between strains are noted with an asterisk (*t*-test,  $P < 0.050$ ).

	GR x HA	TS
Final weight (g)	23.0 $\pm$ 0.33*	14.2 $\pm$ 0.09
SGR (%/d)	2.78 $\pm$ 0.02*	2.55 $\pm$ 0.01
Mortality (%)	0.79 $\pm$ 0.11	3.68 $\pm$ 0.74
Feed conversion	0.77 $\pm$ 0.01*	0.93 $\pm$ 0.01
Egg survival to hatch (%)	92.7 $\pm$ 1.5	91.3 $\pm$ 0.2

The data clearly indicated that the growth rate and feed conversion of the hybrid strain were superior to the traditional TS strain. Even when naturally challenged with a bacterial infection, the strain performed well. Egg survival was comparable between strains, with high survival for both. The data is in accord with the work conducted last year, which also indicated that the resistant strain had superior growth in the hatchery (Wagner et al. 2007).

### Literature Cited

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### New Faces at FES

The FES welcomes **Randy Oplinger** to the research team. Randy is from Boulder, Colorado. He has just completed his Master's degree at the University of Illinois, Champaign/Urbana, working on a project examining the influence of predators and maternal effects on the size and maturation of bluegill sunfish. His interests include fishing, camping, and hiking.

Randy replaces **Ronney Arndt**, who has moved to an open position in the fish culture program at FES.



## Whirling Disease Resistant Strain of Rainbow Trout Out-grows TenSleep Strain at Both Low and High Rations

Using a subset from the same lots of fish noted in the companion article, a 63 day feed trial was conducted to compare the performance of the whirling-disease resistant GR x HA strain and the traditional TS strain, fed either a low or high ration level. We were interested to see if the growth advantage observed in the GR x HA would continue if rations were significantly reduced. The strain might perform well in the hatchery at maximum ration, but how would they do in situations where they may not get all the food they could consume?

On 27 March 2007, 101 to 111 fish were placed into each of 12 circular tanks. Each strain received either a high ration (4.5% of body weight at the beginning of the experiment to 2.1% at the end) or low ration (initially 3.6% of body weight to 1.7% at the end) of a commercial feed (Silvercup). The initial weights for each strain are given in Table 1. The fish were fed several times a day, with the number of feedings varying according to the fish's ability to consume all the feed given in a feeding (see Figure 1). Also, more feedings were required for the high ration, which was expected in order to maximize efficiency. A maximum density of fish in the tanks during the study was 0.3 density index (Piper et al 1982). A general linear model with ration level and strain as fixed factors was used for statistical analysis of specific growth rate, final weight, feed conversion. A mink caused significant mortalities during the experiment, so the mortality rates were not compared statistically.

At the end of study, GR x HA had significantly higher specific growth rates than TS ( $P \leq 0.005$ ; Table 1). Even at low rations levels, growth was faster in the GR x HA (SGR = 2.64) than TS (SGR = 2.59). As expected, final weights and specific growth rates were significantly higher in the higher ration groups (Table 1;  $P \leq 0.001$ ). It was noted during the study that GR x HA were much more aggressive feeders than the TS strain and generally required fewer feedings each day (Figures 1 and 2). Feed conversion rate was good for each ration and was not significantly different between strains ( $P = 0.801$ ), but significantly differed between ration levels; as expected, the low ration treatments were more efficient at converting feed to biomass.

Table 1. Comparison of mean ( $\pm$  SD) hatchery performance variables between two strains of rainbow trout (GR x HA, TS) each receiving either a low or high ration of a commercial pelleted diet. SGR = specific growth rate.

	GR x HA		TS	
	High	Low	High	Low
Initial weight (g)	4.1	4.2	3.4	3.4
Final weight (g)	28.1 $\pm$ 7.4	20.4 $\pm$ 6.46	17.8 $\pm$ 7.16	16.7 $\pm$ 4.65
SGR (%/d)	2.83 $\pm$ 0.006	2.64 $\pm$ 0.029	2.67 $\pm$ 0.036	2.59 $\pm$ 0.018
Feed conversion ratio	0.95 $\pm$ 0.05	0.75 $\pm$ 0.06	0.91 $\pm$ 0.03	0.81 $\pm$ 0.04
Mortality range (%)	15.5 to 24.5	2 to 24.3	4.7 to 14.9	1.9 to 11.5

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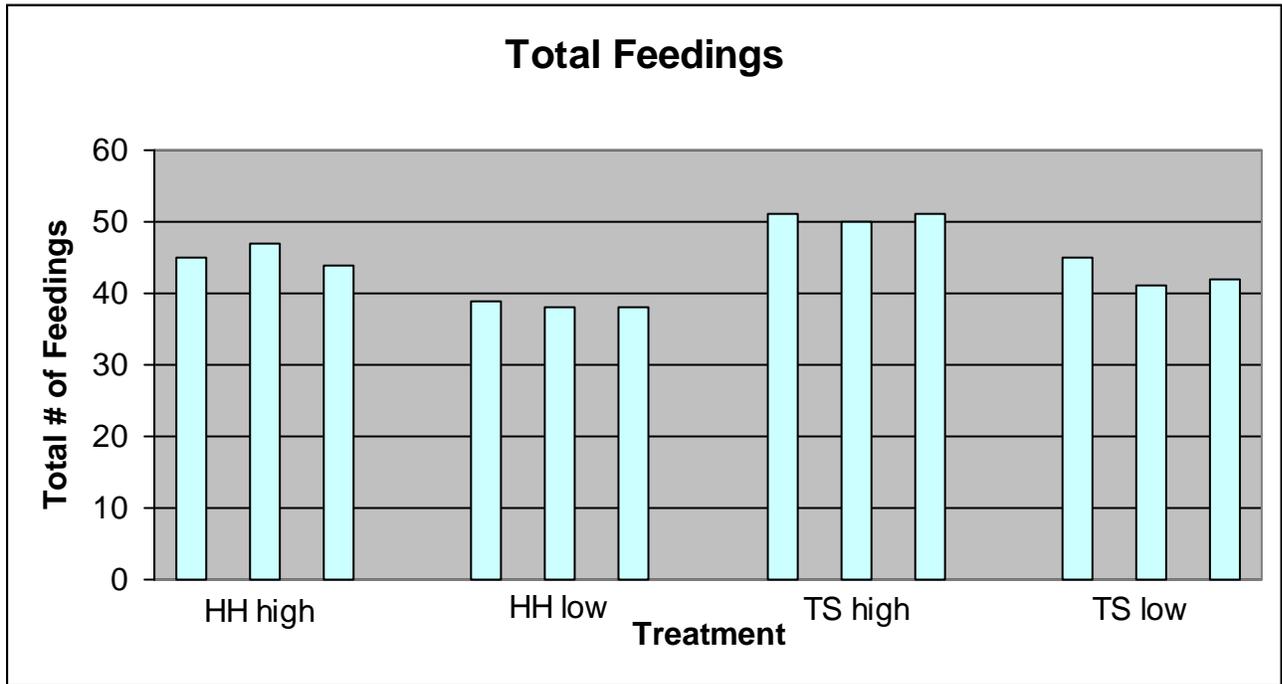


Figure 1. Total number of feedings each treatment received throughout the experiment.

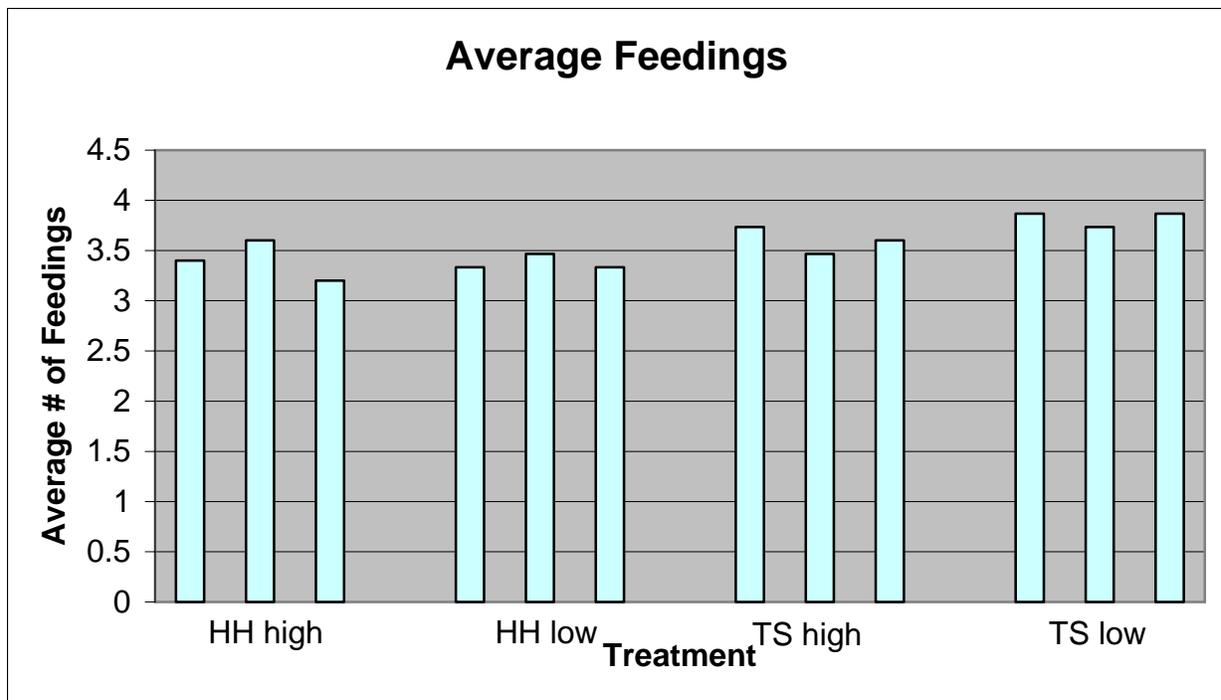


Figure 2. Average number of feedings required each day to maximize the efficiency of feed.

Eric Wagner and Ronney Arndt

## Comparison of the Field Performance of a Whirling-Disease Resistant Rainbow Trout Strain with the TenSleep Strain: 2007 Data

A whirling disease resistant rainbow trout strain (GRxHA) was obtained from the Colorado Division of Wildlife (see companion article on hatchery performance). Field evaluation of the strain, relative to the domesticated TenSleep (TS) strain of rainbow trout, has been ongoing for the last two years. In 2006, many of the fish regenerated their fins after marking and strains were not identifiable. In 2007, the fish were spray-marked with fluorescent grit (red = GRxHA, green = TS; Nielson 1990) prior to stocking. A week after marking, samples of 15 to 19 fish per raceway were checked for mark retention. Fish were stocked into Porcupine and Hyrum Reservoirs on May 8 and 9, 2007, respectively (Table 1). A portion of the fish of each strain was kept at the hatchery to stock at a larger size and later date (Table 1). These fish, already spray-marked, were given a pelvic fin clip (left= TS, right = GR x HA) to differentiate them from fish stocked in May.

In October 2007, 4 gill nets (39 m x 1.7 m, 5 panels with mesh from 3.5 to 12 cm) were set overnight in each of the three study reservoirs, Hyrum, Porcupine, and the control reservoir, Causey. The recaptured fish were examined for deformities and heads from up to 60 fish of each salmonid species were examined for myxospores by pepsin-trypsin digest methods (USFWS and AFS-FHS 2005). Counts of myxospores were made on all 9 squares (1 mm<sup>2</sup> each) of each side of a hemocytometer. The average of the two sides was subsequently used to extrapolate to the number of spores per head based on the volume in the sample.

Table 1. Size and number of two strains of rainbow trout (GR x HA or TS) stocked into study reservoirs.

Reservoir Strain	May 2007		July 2007	
	Number	Mean weight (g)	Number	Mean weight (g)
Hyrum				
GR x HA	13,008	22.7	3,384	63.0
TS	14,309	14.1	2,778	44.9
Porcupine				
GR x HA	9,998	23.5	1,502	63.0
TS	9,995	14.4	1,496	44.9
Causey	3119	149.2		

A creel survey was conducted to estimate angler harvest of rainbow trout from both Porcupine and Hyrum reservoirs. The survey was divided into three seasons: Winter (15 December 2006 to 31 March 2007), Spring (1 April to 31 May 2007), and Summer (June 1 to September 3, 2007). The survey was further divided into weekend or holiday days and weekdays. About 10% of the days were randomly sampled within each stratum for each reservoir. Surveys were conducted between about 10:30-11:00 am until sunset. During the summer survey, the survey was concluded before sunset, usually by 6:30 to 7 pm. Anglers were asked a series of questions to determine the number of fish kept, number released, time spent fishing, whether the angler fished from shore, on ice, or by boat, and the angler's zip code. Any fish harvested were weighed and measured for total length and any fin clips, spray-marks, or deformities noted. Total angler counts were also made at 2 h intervals.

### Field performance

The numbers of rainbow trout recovered in the fall gill netting for each strain are given in Table 2. Prior to stocking, the retention of the spray-mark averaged 97.9% ( $n = 48$ ) for fish given the red pigment (GR x HA) and 88.5% ( $n = 52$ ) for the green (TS). After adjusting marked numbers for mark retention, there were significantly more GR x HA recovered than TS in both Hyrum ( $P = 0.002$ ) and Porcupine ( $P < 0.001$ ) reservoirs for both the May 2007 cohort and for all fish combined ( $P \leq 0.001$ ; Table 2). For the July 2007 cohort, significantly more

(Continued on page 6)

(Continued from page 5)

GR x HA than TS were recovered in Hyrum Reservoir ( $P=0.042$ ), but in Porcupine Reservoir there was no significant difference in survival between strains in this cohort ( $P = 0.284$ ) or for the 9 fish recaptured from the 2006 cohort ( $P = 0.375$ ; Table 2).

Table 2. Number of GR x HA and TS strain rainbow trout recovered in each of two study reservoirs and a control reservoir (Causey) stocked with rainbow trout. Prevalence of *Myxobolus cerebralis*, mean length and weight ( $\pm$ SD) are given for each strain and stocking cohort. Within each reservoir and row, significant differences are noted with an asterisk. For Porcupine Reservoir, the prevalence of *Myxobolus cerebralis* was lower for GR x HA than in TS for all

	Porcupine		Hyrum		Causey
	GR x HA	TS	GR x HA	TS	RT
May 2007 stock					
Number recovered	51*	15	16*	3	1
Mean length (mm)	235.6* (9.1)	221.3 (14.6)	248.8* (12.1)	230.0 (21.8)	294.0
Mean weight (g)	128.7* (16.8)	111.2 (23.3)	154.7 (22.9)	124.7 (33.3)	358
Prevalence (%)	13.7	33.3	0	0	0
Spores per fish	14,968 (17,540)	17,858 (25,629)	0	0	0
July 2007 stock					
Number recovered	9	5	3	0	
Mean length (mm)	230.4 (19.0)	219.0 (4.4)	251.3 (12.6)		
Mean weight (g)	121.6 (29.4)	106.6 (13.1)	153.7 (17.2)		
Prevalence (%)	11.1	40.0	0		
Spores per fish	911	30,522 (39,299)	0		
2006 stock					
Number recovered	11	7	2	1	
Mean length (mm)	248.9 (31.4)	239.6 (40.3)	402.5 (55.9)	393	
Mean weight (g)	161.9 (92.2)	150.4 (102.9)	655.0 (288.5)		
Prevalence (%)	9.1	57.1*	100.0	100.0	
Mean spores per fish	911	64,233 (111,032)	21,411 (9,664)	155,800	

cohorts, but the difference was significant only for the 2006 cohort (Table 2). In Hyrum Reservoir, the fish from both the May and July 2007 cohorts did not have any myxospores; for the 2006 cohort only 3 fish were recovered, but each harbored spores.

Mean spores per fish were calculated for positive fish only. Mean spores per fish were compared between strains for each cohort within each reservoir. For Porcupine Reservoir, spore counts did not significantly differ (Mann-Whitney U Test) between strains for all three cohorts (Table 2). In Hyrum Reservoir, only three positive marked fish were recovered; all were from the 2006 cohort. The marked fish in the other Hyrum cohorts did not harbor myxospores.

Creel Survey

(Continued on page 7)

In the creel survey, only six of the harvested fish from Hyrum Reservoir had fin clips. Three of these were TS and three were GR x HA, indicating similar capture probabilities. In Porcupine Reservoir, no marked fish were noted in the creel. It is suspected that the fins that were clipped regenerated in most of the fish. No fluorescent marks were observed, but difficulties in using the blacklight in the field may have influenced the results. Over the entire course of the creel survey, 363 rainbow trout were harvested from Hyrum Reservoir and 37 were harvested from Porcupine Reservoir. The majority of the rainbow trout in Hyrum Reservoir were harvested in the spring (61.1%), but in Porcupine Reservoir, 78.4% were harvested in winter.

Overall, as in 2006, the hatchery data indicated that the whirling disease resistant strain performed well, even at low ration levels. The field data indicated that the survival of the GR x HA either matched or surpassed that of the traditional strain. Spore counts and prevalence data, especially in Porcupine Reservoir, also supported previous laboratory observations concerning resistance to infection by *M. cerebralis*. The reduction in spore loads is hoped to reduce the risks to naturally reproducing salmonids in Utah and the region.

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## **Latest *Myxobolus cerebralis* Findings**

Biologists at Utah's Fisheries Experiment Station have recently discovered the whirling disease parasite, *Myxobolus cerebralis*, in trout collected from several locations throughout the state. These locations include Garkane Impoundment, Seven Mile Creek, Beaver River (Beaver County) and the East Fork of the Sevier River in the South and the Middle Fork of the Ogden River to the North. Even though all these findings are unfortunate, none were unexpected.

The Main **Garkane Impoundment** was suspected of being stocked with MC infected fish in early 2000. As a result, continued efforts have been underway to determine if the parasite had become established in the reservoir. This recent finding suggest stocking of potentially infected fish may have contributed to the parasite's introduction at this location.

**Seven Mile Creek** is part of the Fremont River Drainage near Johnson Reservoir. Seven Mile Creek runs adjacent to UM Creek and is a tributary to the Fremont River, both of which have previously been determined to contain the whirling disease parasite. Close proximity to infected waters and in-stream movement of infected fish has likely contributed to this finding.

Detections in the **East Fork of the Sevier River** and **Beaver River** were expected because both sites are down stream from previously contaminated areas. The **Middle Fork of the Ogden River** is connected to the South Fork of the Ogden River through Pineview Reservoir. The South Fork was previously determined to be a source of MC infection.

Dispersion of the whirling disease parasite through recreational use, avian vectors and in-stream movement of infected fish are difficult variables to control. As a result, efforts through the state's whirling disease survey continue to be used as a way to identify these vectors and provide valuable information that can be used to reduce further dispersion.

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*The Ichthyogram* is a quarterly publication of the Fisheries Experiment Station, Utah Division of Wildlife Resources, Logan Utah 84321.

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