Comparison of Feeding Regime and Diet on Compensatory Growth of Hybrid Bluegill

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Abstract: We conducted 2 experiments to evaluate the potential of feeding schedules designed to elicit compensatory growth and increase growth of hybrid bluegill (F_1: male bluegill *Lepomis macrochirus* x female *L. cyanellus*). The first experiment evaluated a commercially prepared pellet and consisted of 3 treatments: fish fed every day and fish starved for 2 or 4 days after cessation of hyperphagia. The second experiment evaluated 2 diets, mealworms and commercial pellets, fed every day and on a 2-day starvation schedule. Growth and feed consumption in starvation treatments did not significantly exceed that of controls in either experiment. Our results contradict those of earlier studies that showed increased growth and consumption with similar feeding methods. Our results suggest that increasing growth rate using feeding schedules designed to elicit compensatory growth may not be practical when feeding an artificial pelleted diet, and feeding strategies of this type may be difficult to implement for large-scale hybrid bluegill production. However, our results suggest that hybrid bluegill do not need to be fed every day to optimize growth and that alternative feeding regimes could significantly reduce labor costs.

Proc. Annu. Conf. Southeast. Assoc. Fish and Wildl. Agencies 55:346–352

Compensatory growth is a period of rapid weight gain following a period of food deprivation. Compensatory growth has been observed in invertebrates, mammals, birds, and fish (Wilson and Osbourn 1960, Broekhuizen et al. 1994, Jobling 1994); however, the mechanisms underlying this phenomenon are not fully understood. Most studies suggest a physiological change, whereby organisms reduce their basal metabolic rate, increase food conversion efficiency, and begin excessive consumption (hyperphagia) once food supplies are available (Miglavs and Jobling 1989, Russell and Wootton 1992, Wieser et al. 1992, Jobling 1994).

Regardless of the mechanism, compensatory growth has potential for increasing commercial production in aquaculture. Hayward et al. (1997) were the first to show that compensatory growth occurred in hybrid bluegill fed mealworms *(Tenebrio molito)* on various feeding schedules. We attempted to duplicate their experiment using a commercially produced diet. Our first experiment used a commercially prepared pelleted diet and 3 treatments used by Hayward et al. (1997): fish fed every day, fish starved for 2 days and fish starved for 4 days. Because we were unable to duplicate the results reported by Hayward et al. (1997), we performed a second experiment to try to more closely duplicate their protocol and experimental design, as well as to directly compare mealworms to a commercial pellet diet.

We thank M. G. Walsh, G. Cummings, and M. Willis for assistance with laboratory work. Additional thanks to G. Gebhart for help with laboratory equipment and J. Parsons for feed analyses. Financial support was provided by the Federal Aid in Sport Fish Restoration Act under Project F-41-R of the Oklahoma Department of Wildlife Conservation and Oklahoma State University through the Oklahoma Cooperative Fish and Wildlife Research Unit (cooperators: Oklahoma Department of Wildlife Conservation, Oklahoma State University, U.S. Geological Survey Biological Resources Division, and the Wildlife Management Institute).

Methods

Experiment 1

We acclimated approximately 70 hybrid bluegill to laboratory conditions (14 hours light: 10 hours dark photoperiod and 20 C water) for 2 weeks and trained them to consume a pelleted diet. During September 1999, fish were selected for size uniformity (2.90–3.27 g) and individuals were placed into 15 20-liter aquaria. Each aquarium was equipped with an airstone, a 100-watt aquarium heater, and visual dividers to prevent agonistic behavior from influencing neighboring fish. These fish were acclimated for 8 days prior to the initiation of feeding trials. During acclimation fish were fed to satiation and water temperatures were elevated to 24 C, the water temperature used by Hayward et al. (1997).

The experiment consisted of 3 treatment groups, selected on the basis of their performance as reported in Hayward et al. (1997): a continuously fed control (PC) and 2 treatments consisting of 2- and 4-day starvation periods following feeding (P2 and P4, respectively). The feeding schedule of each group followed the procedures described by Hayward et al. (1997) with 1 exception. Hayward et al. (1997) removed food items after 24 hours; however, because pellets dissolve, we removed excess pellets after 2 hours so we could make accurate estimates of consumption. Our observations indicated that fish consumed food primarily during the first 30 minutes after feeding and rarely consumed additional food when fed twice per day. Therefore, we concluded that a 2-hour feeding period was sufficient to estimate daily consumption. The experimental diet was a commercially produced 2.4-mm floating pellet (EXTR 450, Rangen Inc., Angleton, Texas). All feedings took place between 0800 and 0830 hours daily. Aquaria were cleaned and quarter volume water changes were made every 3–5 days to maintain water quality.

We monitored daily food consumption by feeding an excess number of pellets, counting and removing all pellets remaining after 2 hours, and multiplying the number consumed by the mean dry pellet weight (0.0121 g). Each fish was measured (nearest 1 mm) and weighed (nearest 0.01 g) weekly, 1 hour before feeding. At the conclusion of each feeding day, the daily weight-specific consumption for treatments P2 and P4 was evaluated. Fish in treatments P2 or P4 were fed on consecutive days until the daily weight-specific consumption no longer significantly exceeded that of the control group (Student's *t*-test; $P < 0.05$), then the predetermined starvation period (2 or 4 days) was initiated the following day. The experiment was scheduled to end at the conclusion of a feeding cycle on or beyond 80 days for both treatment groups.

Absolute growth rate (AGR) and gross growth efficiency (GGE) were calculated using the following formulae:

$$AGR = (W_f - W_i)/T \qquad GGE = (W_f - W_i)/CC$$

where W_f and W_i are the final and initial weights, respectively, T is the total number of days and CC is the cumulative consumption. Growth, consumption, and food conversion efficiency data were analyzed using analysis of variance (SAS 1998). Post hoc contrasts were made using least square means. Comparisons were considered significant if $P < 0.10$ and all probabilities are reported. We performed correlations between CC and AGR to estimate the effect of consumption on growth.

Experiment 2

We acclimated approximately 100 hybrid bluegill to the laboratory for 2 weeks. Thirty-two hybrid bluegill (11.25–14.75 g) were selected in the same manner as in Experiment 1, except we chose fish similar in size to Hayward et al. (1997; Table 1). These fish were randomly placed into individually numbered, 3.25-liter chambers (20 x 12.5 x 13 cm) of clear plexiglass. Each chamber was perforated with 32 holes (3.5 mm) on the sides to allow water circulation and fitted with a clear plexiglass lid. The chambers were placed side by side on a rack that elevated them 30 cm off the bottom of a 2.7 x 0.6 x 0.6-m flow through circulation tank. Municipal water, filtered through an organic filtration cartridge, filled the tank to within 1 cm of the top of the chambers. The tank was fitted with airstones and 8 300-watt submersible aquarium heaters to maintain water temperatures at 24 ± 1 C throughout the experiment. The lab was maintained at a constant photoperiod of 14 hours light: 10 hours dark.

This experiment consisted of 2 diet types and 2 feeding schedules. The diet types were a 4.8-mm floating pellet (EXTR 400, Rangen Inc., Angleton, Texas) and mealworms. The feeding schedules consisted of a control group fed every day and a 2-day starvation group. This resulted in 4 treatments: pellet control (PC), mealworm control (MC), pellet 2-day starvation (P2), and mealworm 2-day starvation (M2). The feeding protocols for the control and 2-day starvation groups were similar to Experiment 1. Due to differences in chamber size and feed type, pellets broke down more slowly in this experiment than in Experiment 1 and were allowed to remain for 7 hours after feeding. Feedings occurred between 1000 and 1015 hours daily. Consumption data was collected by weighing and feeding a known number of food items (pellets or mealworms), counting the number removed, and multiplying the number consumed by the mean weight of the fed items. Collection of fish live weights, statis-

Table 1. Sample size (N), total number of experimental days, mean (standard error) start and finish weights, absolute growth rates (AGR), gross growth efficiency (GGE), mean cumulative consumption, consumption per feeding day (FD), number of no-feed/refeed cycles completed and mean feeding days per deprivation days (DD) for treatment groups of hybrid bluegill. Treatment means with different upper case letters are significantly ($P < 0.10$) different from other treatments within that experiment.

Experiment	Treatment	N	Total days	Initial weight (g)	Final weight (g)	AGR (g/day)	GGE	Cumulative consumption	Consumption per FD	Total cycles	FD/DD
Experiment 1	PC	5	82	3.67 (0.17)	12.23 (2.04)	0.10 (0.02) A	1.13	7.3 (1.1) A	0.09 A		
	P2	5	82	3.53 (0.13)	9.12 (1.00)	0.07 (0.01) AB	1.23	4.4 (0.6) B	0.11 A	21	0.95
	P4	5	81	3.78 (0.21)	7.25 (0.57)	0.04 (0.01) B	1.08	3.2 (0.4) B	0.11 A	13	0.56
Experiment 2	MC	8	86	11.98 (0.19)	20.28 (1.95)	0.10 (0.02) A	0.35	21.6 (3.4) A	0.25 A		
	M2	8	86	12.17 (0.18)	23.79 (2.91)	0.14 (0.03) A	0.43	23.0 (3.9) A	0.48 B	19	1.26
	PC	6	82	11.76 (0.24)	20.96 (1.87)	0.11 (0.02) A	0.76	11.7 (1.4) B	0.14 A		
	P2	8	82	12.29 (0.23)	21.99 (1.03)	0.12 (0.01) A	0.83	11.5 (0.8) B	0.26 A	19	1.16
Hayward et al. (1997)	MC[a]	7	105	13.84 (1.16)	23.13 (3.43)	0.09 (0.02) A	0.31	27.5 (4.1)	0.26		
	M2[a]	7	105	13.99 (1.21)	32.70 (5.40)	0.18 (0.04) B	0.35	48.7 (9.8)	0.64	13	2.92
	M4[a]	7	105	13.64 (1.22)	26.04 (4.78)	0.12 (0.04) A	0.31	35.9 (9.2)	0.56	10	1.60

a. Treatments MC, M2, and M4 correspond to C, D2, and D4 in Hayward et al. (1997).

tical analyses, and termination of the feeding trials were performed identically to Experiment 1. Two fish were excluded from the PC treatment on day 45 of the experiment due to apparent illness, and were not included in the final analyses. None of the remaining fish exhibited signs of illness and remained healthy throughout the experiment.

Results

Experiment 1

Differences in mean final weights (F = 3.46; df = 2, 12; P = 0.065) and absolute growth rates (F = 3.42; df = 2, 12; P = 0.067) were significant (Table 1) among treatments. Pairwise comparisons indicated treatment P4 had a significantly lower final weight and absolute growth rate ($P < 0.05$) than the control, whereas treatment P2 was not significantly different ($P > 0.10$) than either group. No significant difference occurred between mean gross growth efficiencies (F = 0.96; df = 2, 12; P = 0.4104) (Table 1).

Growth was positively correlated with cumulative consumption (R^2=0.95, $P <$ 0.01). Differences in cumulative consumption were significant among treatments (F = 7.98; df = 2, 12; P = 0.0063). Controls consumed significantly more food than both the P2 and P4 treatments (1.7 and 2.3 times more than P2 and P4, respectively, $P <$ 0.05, Table 1). Mean consumption per feeding day did not differ among treatments (F = 0.63; df = 2, 12; P = 0.5473, Table 1).

The mean feeding period following starvation for treatments P2 and P4 was 1.9 and 2.2 days, respectively. This resulted in feeding day to deprivation day ratios less than 1.0 (Table 1). For treatment P2, 7 of the 21 no-feed/refeed cycles did not induce hyperphagia and resulted in a 1-day feeding period. Treatment P4 followed each starvation period with at least 1 day of hyperphagia.

Treatments PC, P2, and P4 concluded on days 82, 82, and 81 of the experiment, respectively. Because only 1 day separated the conclusion of the 3 groups, the final weights were compared without adjusting for the additional day; however, all other calculations were based on the total number of days. Water temperatures fluctuated throughout the experiment (mean temperature = 23.4 C, range = 19.5–29.0 C) and daily temperature fluctuations may have influenced daily consumption. This problem was alleviated in the second experiment.

Experiment 2

Absolute growth rates did not differ among treatments (F = 0.41; df = 3, 26; P = 0.75, Table 1). Mean gross growth efficiencies (Table 1) were significantly higher for pellet diets (treatments P2 and PC) than mealworm diets (treatments M2 and MC) (F = 60.0; df = 1, 26; P = 0.0001). Pellets consisted of 40.0% protein and 6.1% moisture, while mealworms consisted of 21.6% protein and 58.9% moisture. These differences in diet composition may have resulted in the differences in utilization efficiencies.

Growth was positively correlated with cumulative consumption (pellet R^2 = 0.95 and mealworms R^2 =0.97, $P < 0.01$). There was no significant difference in cumulative consumption between treatments M2 and P2 and their corresponding control groups ($P > 0.10$, Table 1). Mean consumption per feeding day was significantly different among the 4 groups (F = 7.64; df = 3, 26; $P = 0.0008$). Pairwise comparisons revealed that M2 consumed a significantly greater amount per feeding day than all other groups ($P < 0.01$). Differences in consumption per feeding day were not significant for MC, P2, and PC (Table 1). Feeding periods for treatments M2 and P2 averaged 2.5 and 2.3 days, respectively. There were 19 cycles during the experiment and hyperphagia was not induced during 2 cycles for treatment M2 and 3 cycles for treatment P2.

Treatment groups fed mealworms and those fed pellets concluded on day 86 and 82 of the experiment, respectively. Because of the difference in termination dates, only absolute growth rates were statistically analyzed.

Discussion

Our results differ dramatically from those of Hayward et al. (1997, 2000), which indicated that hybrid bluegill fed mealworms on a 2-day no-feed/refeed schedule would significantly outgrow continuously fed controls. Initially, we wished to evaluate feeding schedules that elicited compensatory growth (Hayward et al. 1997) using a commercially prepared fish feed. We expected that fish fed a pelleted diet would increase their growth rate in response to starvation schedules and that the response might differ in magnitude to that of fish fed mealworms. However, we could not duplicate the results of Hayward et al. (1997, 2000) with either mealworms or a commercially prepared diet.

The differences in results between our study and those of Hayward et al. (1997, 2000) are probably due to differences in duration of hyperphagia. In 2 previous experiments (Hayward et al. 1997, 2000) fish were hyperphagic for approximately 6 days after a 2-day starvation period. Fish in our experiments did not exhibit these prolonged periods of hyperphagia. Hybrid bluegill growth is directly correlated to consumption (Wang et al. 1998, and the present study); therefore, reduced feeding periods in our experiments negatively influenced growth.

We have no explanation for the differences in the duration of hyperphagia between the studies. The only major deviation between our second experiment and previous work was the length of time food remained in the water. Because commercial pellets dissolved, it was not practical to allow the feed to remain in the aquaria for a 24-hour period as was done by Hayward et al. (1997, 2000). Based on our observations, however, it appears that most feeding occurred during the first 30 minutes after food was introduced, and we do not feel that the shortened feeding period greatly reduced daily consumption.

Social interactions have been shown to negatively affect growth (Jobling and Reinsnes 1986, Jobling and Baardvik 1994) and may be detrimental to feeding strategies of this type. Hayward et al. (2000) tested 2-day no-feed/refeed schedules on group-held hybrid bluegill in the laboratory and found that the growth of treatment groups did not significantly exceed that of continuously fed controls. The effects of social interactions on feeding regimes designed to elicit compensatory growth need further evaluation before these strategies are implemented for large-scale production.

To be useful for aquacultural production, compensatory growth must be easy to induce and monitor, practical in a production setting, and occur over a range of environmental conditions. Biomass and consumption must be estimated daily to accurately estimate hyperphagia and this would not be practical in a production setting. Additionally, our experiments indicated that compensatory growth may be difficult to induce. Finally, experiments holding fish in groups indicate that compensatory growth may be overcome by social interactions (Hayward et al. 2000). Although we did not increase absolute growth rate, our results suggest that fish do not need to be fed daily to optimize growth and alternative feeding strategies could significantly reduce labor costs associated with feeding. Our results, as well as Hayward et al. (1997, 2000) suggest that feeding strategies could be useful to fish producers and are worth further study.

Literature Cited

Broekhuizen, N., W. S. C. Gurney, A. Jones and A. D. Bryant. 1994. Modeling compensatory growth. Functional Ecol. 8:770–782.

Hayward, R. S., D. B. Noltie, and N. Wang. 1997. Use of compensatory growth to double hybrid bluegill growth rates. Trans. Am. Fish. Soc. 126:316–322.

______, N. Wang, and D. B. Noltie. 2000. Group holding impedes compensatory growth of hybrid sunfish. Aquaculture 183:299–305.

Jobling, M. 1994. Fish Bioenergetics. Chapman and Hall, London. 195pp.

______ and B. M. Baardvik. 1994. The influence of environmental manipulations on inter- and intra-individual variation in food acquisition and growth performance of Arctic charr, *Salvelinus alpinus*. J. Fish Biol. 44:1069–1087.

______ and T. G. Reinsnes. 1986. Physiological and social constraints on growth of Arctic charr, *Salvelinus alpinus* L.: an investigation of factors leading to stunting. J. Fish Biol. 28:379–384.

Miglavs, I. and M. Jobling. 1989. Effects of feeding regime on food consumption, growth rates and tissue nucleic acids in juvenile Arctic charr, *Salvelinus alpinus*, with particular respect to compensatory growth. J. Fish Biol. 34:947–957.

Russell, N. R. and R. J. Wootton. 1992. Appetite and growth compensation in the European minnow, *Phoxinus phoxinus* (Cyprinidae), following short periods of food restriction. Environ. Biol. Fishes 34:277–285.

SAS Institute, Inc. 1998. SAS/STAT User's guide for personal computers, Version 8.0 ed. SAS Institute, Inc., Cary, N.C.

Wang, N., R. S. Hayward, and D. B. Noltie. 1998. Effect of feeding frequency on food consumption, growth, size variation, and feeding pattern of age-0 hybrid sunfish. Aquaculture 165:261–267.

Wieser, W., G. Krumschnabel, and J. P. Ojwang-Okwor. 1992. The energetics of starvation and growth after refeeding in juveniles of three cyprinid species. Environ. Biol. Fishes 33:63–71.

Wilson, P. N. and D. F. Osbourn. 1960. Compensatory growth after undernutrition in mammals and birds. Biol. Rev. 35:324–363.