North American Journal of Aquaculture 69:395–399, 2007

DOI: 10.1577/A06-048.1

[Article]

Alternatives to Freeze-Dried Krill in the Feed Training Phase of Largemouth Bass

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Abstract.—Fry of largemouth bass *Micropterus salmoides* are typically pond reared on natural foods until they reach approximately 4–6 cm. Fish that are to be raised on pellets are typically feed trained in tanks using freeze-dried krill (FDK), which is gradually replaced with prepared diet. The cost of FDK (retail >US$40 per kg) can be a significant expense in the feed training phase. An experiment was conducted to determine whether FDK could be eliminated by simple diet amendment. The largemouth bass fingerlings (mean weight $\pm$ SD = 2.2 $\pm$ 0.3 g) were stocked in 190-L tanks at 150 fish/tank. Four treatments with four replicates each were compared: (1) fingerlings were initially fed FDK and then were gradually weaned to a commercial pellet (1.5-mm floating trout feed; control [CTL], current practice); (2) fish were offered only the dry commercial pellet throughout (DRY); (3) commercial pellets were moistened with water before use (MST); and (4) commercial pellets were top-dressed with menhaden fish oil at 10% of diet by weight initially and at a gradually reduced percentage over time (OIL). After 18 d, there was no significant difference ($P > 0.05$) in the average weights of fish. However, in terms of percentage of fish that were successfully habituated to the final dry pellets, the DRY treatment resulted in a significantly lower ($P < 0.05$) success rate (82%) than other treatments. Training success was not significantly different ($P > 0.05$) among the CTL (98%), MST (93%), and OIL (92%) treatments. The best diet for feed training of largemouth bass will vary depending on the cost and availability of FDK and the economic importance of production maximization.

Largemouth bass *Micropterus salmoides* have been cultured in the United States since the 1890s and are considered one of the most popular warmwater sport fishes (JSA 1983). In recent years, interest in production of largemouth bass has increased (Brandt 1991) based on their increased use for corrective stocking (JSA 1983), fee fishing (Dupree and Huner 1984), managed trophy fisheries (JSA 1983), and live sales as food fish in ethnic Asian markets (Tidwell et al. 1996). In the United States, the Joint Subcommittee on Aquaculture (JSA 1983) listed determination of efficient grow-out procedures under intensive conditions as one of the research priorities for largemouth bass aquaculture.

While largemouth bass fingerlings have been produced for stocking programs by state and federal hatcheries for many years (Turner and Kraatz 1920), before the 1960s the fish were reared solely on live food, beginning with zooplankton and then switching to forage fish until the desired stocking size was reached. However, this extensive method requires large numbers of ponds for the largemouth bass and forage fish, as it takes approximately 5 lb (2.27 kg) of forage fish to produce 1 lb (0.45 kg) of largemouth bass (Nelson et al. 1974). As early as the 1960s, studies were conducted on the pond culture of largemouth bass fed prepared diets (Snow 1970). Several studies have examined the production of largemouth bass raised on formulated feeds (Snow 1968; Snow and Maxwell 1970; McCraren 1975; Snow and Wright 1976; Sloane 1993) and this is now a common practice for many federal, state, and private hatcheries.

Unlike channel catfish *Ictalurus punctatus* and rainbow trout *Oncorhynchus mykiss*, newly hatched largemouth bass fry (swim-up stage) cannot be trained to accept prepared diets (Brant et al. 1987; Williamson et al. 1993). Therefore, largemouth bass fry are usually raised in fertilized nursery ponds or in tanks until they reach 4–6 cm in length (Tidwell et al. 2000). At this

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Received June 29, 2006; accepted March 26, 2007
Published online September 13, 2007

size, the juveniles are crowded into tanks at high densities and are presented with highly palatable food items, such as freeze-dried krill (FDK), at frequent intervals (Kubitza and Lovshin 1997). Over a training period of about 10–14 d, the juveniles are gradually introduced to 100% dry diet. Several studies have described and tested different initial food items, such as groundfish, fish eggs, moist pellets, and FDK, for use in feed training of largemouth bass fingerlings (Snow 1965; Anderson 1974; Kubitza and Lovshin 1997).

Primarily due to its effectiveness and ease of use, FDK has become one of the most popular feed training diets for largemouth bass. However, the typically high cost of FDK (>US$40 per kg; Argent Chemical Co., Redmond, Washington) can result in an expensive feed training phase. This experiment was designed to evaluate the modification of a commercial dry pellet to see if its acceptance as an initial training diet could be improved, thus allowing FDK use to be reduced or eliminated.

Methods

Approximately 2,000 pond-raised largemouth bass fingerlings were obtained from the Pfeiffer Fish Hatchery in Frankfort, Kentucky, and were held in a raceway for 2 d before stocking into experimental tanks. Initially, 150 fish were randomly sampled from the raceway with a dip net and were individually measured (total length [TL]) and individually weighed with an electronic balance (Ohaus Corporation). The initial average (±SD) TL was 5.6 ± 0.5 cm, and average weight was 2.2 ± 0.3 g. Largemouth bass were randomly stocked into 16 polyethylene, cone-bottom, semisquare tanks (190 L each; Polytank, Inc., Litchfield, Minnesota) at a rate of 150 fish/tank. Four feed training treatments were randomly assigned to four replicate tanks each. The experimental system was a flow-through system. Each tank contained a center standpipe with a 2,000-μm screen and was supplied with dechlorinated municipal water at approximately 1.5 L/min. Each tank was equipped with a medium-pore air stone supplied by a regenerative blower. In all treatments, fish were fed three times daily at 10% of initial body weight per day (IBW/d) divided equally among feedings. The dry diets and FDK were weighed to the nearest gram before each feeding and before the addition of water (treatment 3) or oil (treatment 4). Remaining uneaten feed was removed by siphon from each tank once daily between feedings.

In the control (CTL), fingerlings were initially fed FDK and then were gradually introduced to a commercial floating pellet containing 45% protein and 12% fat (Silver Cup, Inc.; Steelhead Grower [closed formulation], 1.5-mm floating pellet) according to a schedule modified from Kubitza and Lovshin (1997). On days 1–3, fish were fed only FDK (10% IBW/d divided equally among three feedings). On days 4–6, they were fed a mixed ration containing 75% FDK and 25% commercial diet (75:25 ration). A 50:50 ration was administered on days 7–9, a 25:75 ration (FDK : commercial) was administered on days 10–12, and the commercial diet only was administered on days 13–18. In the second treatment, only the dry commercial pellet was fed throughout the experiment (DRY).

For treatments 3 and 4, a modification of the "gradual feed ingredient transition" method described by Kubitza and Lovshin (1997) was followed. In treatment 3, the commercial pellets were moistened with water before use (MST). For this treatment, diet soaking duration decreased over time: 10 min on days 1–3, 7.5 min on days 4–6, 5 min on days 7–9, and 2.5 min on days 10–12. On days 13–18, fish were given the dry commercial diet. In treatment 4, pellets were top-dressed with menhaden fish oil (Rangen, Inc., Buhl, Idaho; $0.80 per kg) to soften the pellet and potentially increase palatability (OIL). The percentage (by weight) of top dressing in the diet was decreased over time: 10% on days 1–3; 7.5% on days 4–6; 5% on days 7–9; 2.5% on days 10–12; and 0% (i.e., dry diet) on days 13–18.

Water temperature and dissolved oxygen were measured twice daily with a YSI 85 meter (Yellow Springs Instrument Company, Yellow Springs, Ohio). Total ammonia, nitrite, pH, and alkalinity were measured three times per week using a Hach DR/2500 spectrophotometer (Hach Company, Loveland, Colorado).

At the conclusion of the experiment, all fish in each tank were removed, individually weighed, and measured for total length. Before the harvest date and through routine observation, a small number of surviving fish in each tank were identified as emaciated and were assumed to be nonfeeders. To validate the number of feed trained fish in each tank, fish were fed on the morning of the harvest date and each fish was then euthanized and eviscerated to document the presence or absence of feed. Percentage of feed-trained fish was calculated as 100 times the number of fish containing feed pellets divided by the number stocked.

Growth performance parameters were calculated as follows: specific growth rate (SGR, %BW/d) was calculated as $[(\log_e W_f - \log_e W_i)/t] \times 100$, where W_f = final weight (g), W_i = initial weight (g), and t = time in days. Feed conversion ratio (FCR) was calculated as total diet fed (g) divided by total wet weight gain (g). Treatments were compared statistically by analysis of variance (ANOVA) using Statistix version 8.0 (Statistix Analytical Software 2000). If significant differences

TABLE 1.—Mean (±SD) weight (g), percent weight gain, specific growth rate (SGR), condition factor (K), feed conversion ratio (FCR), percent survival, and percent feed trained (averaged across all fish) for pond-reared juvenile largemouth bass after 14 d of feed training with either freeze-dried krill (control, CTL), a commercial diet only (DRY), the commercial pellet moistened with water (MST), or the oil-dressed commercial pellet (OIL). Within a row, values followed by different letters are significantly different (ANOVA: $P \leq 0.05$).

Variable	Treatment			
	CTL	DRY	MST	OIL
Average weight (g)	5.8 ± 0.0	5.4 ± 0.3	5.2 ± 0.1	5.2 ± 0.2
Percent weight gain	275.5 ± 1.3	256.8 ± 12.6	243.6 ± 4.2	245.4 ± 7.4
SGR (% BW/d)	7.2 ± 0.0	6.7 ± 0.4	6.4 ± 0.1	6.4 ± 0.2
K	1.8 ± 0.0	1.7 ± 0.1	1.6 ± 0.0	1.6 ± 0.1
FCR	0.9 ± 1.0	1.4 ± 1.2	1.3 ± 0.1	1.2 ± 0.1
Percent survival	99.9 ± 0.6 z	94.6 ± 1.4 y	99.0 ± 0.6 z	97.4 ± 0.6 zy
Percent feed trained	99.8 ± 0.2 z	87.3 ± 3.0 y	94.0 ± 1.4 z	94.7 ± 1.0 z

were found, treatment means were separated using Fisher's least-significant-difference test (Steele and Torrie 1980). All percentage and ratio data were arcsine transformed before analysis (Zar 1984). However, data are presented untransformed for ease of comparison.

Results and Discussion

Over the 18-d study period, averages (±SD) for water quality variables were as follows: 25.6 ± 1.0°C for water temperature; 6.3 ± 0.6 mg/L for dissolved oxygen; 0.5 ± 0.2 mg/L for total ammonia-N; 0.02 ± 0.01 mg/L for un-ionized ammonia-N; 0.1 ± 0.0 mg/L for nitrite-N; 7.8 ± 0.3 for pH; and 87.3 ± 22.7 mg/L for alkalinity. These values represent conditions well suited for largemouth bass growth and health (Tidwell et al. 2000).

Survival was significantly higher ($P < 0.05$) for the CTL and the MST treatments than for the DRY treatment (Table 1). Survival in the OIL treatment was not significantly different ($P > 0.05$) than that in other treatments (Table 1). There was no significant difference ($P > 0.05$) among fish in the four treatments in terms of average individual weight (overall mean = 5.4 g), percent weight gain (255%), SGR (6.7% per d), condition factor (1.7), or FCR (1.2; Table 1). However, in terms of percentage of fish that were successfully feed trained, the DRY treatment had a significantly lower ($P < 0.05$) success rate (82%) than other treatments (Table 1). Although the highest percentage of feed-trained fish was produced by the CTL treatment, training success did not differ significantly ($P > 0.05$) among the CTL (98%), MST (93%), and OIL (92%) treatments.

In this study, the water-softened pellets performed better than the dry commercial pellet. These data are in agreement with previous findings that semimoist or soft diets perform better than dry or hard diets during feed training of largemouth bass (Snow 1965; Lovshin and Rushing 1989; Sloane 1993). However, our results differ from those of Kubitza and Lovshin (1997), who reported no performance differences between semimoist or soft pellets and dry or hard pellets. We examined a different diet than other published research, which might account for differences.

The OIL diet also performed better in producing feed-trained fish than did the dry diet. Feed consumption is influenced by several factors, including flavor, taste, and smell. Many natural ingredients, such as meals, hydrolysates, and oils derived from fish and crustaceans, are highly palatable and can increase feeding response and feed intake (Barrows 2000). The added oil may have improved both the texture and palatability of the training diet. Even though feed training percentages were higher for MST and OIL than for FDK, these modified diets may well be economically advantageous due to lower feed cost ($0.80 per kg for menhaden fish oil versus $40.00 per kg for FDK). The OIL treatment may be preferable to MST, as it would reduce the likelihood of nutrients leaching out of the pellets during water softening.

Training success in this study ranged from 82% to 98%, which is considerably better than that in many published reports. Sloane (1993) was one of the first to use FDK as a training diet for largemouth bass; however, only 13% of the fish were successfully introduced to the dry diet (feed trained). Kubitza and Lovshin (1997) reported 27% feed training success when using only FDK to introduce largemouth bass fingerlings to a dry diet. In another experiment, Kubitza and Lovshin (1997) reported 77% success rate when they incorporated FDK into specially formulated training diets using gradual feed ingredient transition. We found that a commercially available dry diet, with no amendment or augmentation, resulted in 82% feed training success.

Training success can be related to many variables, such as initial fish size and condition, acceptability of the training diet, suitability of the training system, and genetic potential of the particular strain (Williamson 1983). The size of fingerlings at the onset of feed training is an important factor in training success (Snow 1965). In our trial, the initial size of the largemouth bass fingerlings was approximately 2.2 g, whereas 1.0-g fish were used by Kubitza and Lovshin (1997). The larger size of the fish in our study could have positively influenced their trainability.

Another difference between our experiment and others is that we used a floating commercial diet, whereas most others have used a sinking or a slow-sink pellet. Young largemouth bass generally only strike at pellets on the surface or as they slowly sink and do not readily eat pellets on the tank bottom (McCraren 1974; Flickinger et al. 1976; Willis and Flickinger 1981). Kubitza and Lovshin (1997) indicated that the tendency to float was one of the positive attributes of freeze-dried krill as a training diet. The fish in our study often ingested and rejected pellets many times before actually consuming them. Floating pellets thus remained accessible for an extended period.

Little research has been conducted on genetic aspects of the culture traits of largemouth bass. One possible explanation for the better trainability of fish in our experiment relative to other experiments could be domestication. Williamson (1983) compared the feed training success of two genetic strains of largemouth bass and found an 84% difference (23% versus 43%). A recent experiment compared feed training success of two strains (wild and domestic) of largemouth bass fry and found that survival was greater in the domestic stain (60%) than in the wild strain (34%) (Aquaculture Research Center, Kentucky State University [KSU], unpublished data). Snow (1965) reported increased survival in offspring from pellet-trained broodstock and recommended a "genetic approach" to developing a strain of largemouth bass more readily adapted to the controlled intensive culture environment. The fish we used were progeny from a line of largemouth bass broodstock that had been pellet raised for several generations (M.L., personal observation). It seems likely that domestication may, at least in part, explain the high feed training success in this study.

Based on our results, it appears that FDK may not be essential to the successful feed training of largemouth bass. Use of a floating diet rather than a sinking pellet may be advantageous, and pellet soaking in water or coating in oil appears to be beneficial. Use of these alternatives may, however, yield different results in other genetic strains of largemouth bass. Future research should address the genetic improvement of largemouth bass used in intensive production situations. The best diet for feed training of largemouth bass will vary based on the cost and availability of FDK and the economic importance of production maximization.

Acknowledgments

We thank Russell Neal, Kyle Schneider, and Jamie Greene for assistance in daily care of the culture systems. This research was partially funded by a U.S. Department of Agriculture Cooperative State Research, Education, and Extension Service grant to KSU. Additional support was provided by Kentucky's Regional University Trust Fund to the Aquaculture Program as KSU's Program of Distinction.

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