AQUA TIPS

Selective breeding for improved growth performance in the Pacific Threadfin, *Polydactylus sexfilis*

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Introduction

Aquaculture development of the Pacific threadfin (*Polydactylus sexfilis*) is gaining substantial momentum in Hawaii with the appearance of captive farmed product in local restaurants, retail markets, and sales to both mainland and international markets. The CTSA has fostered much of industry expansion through many years of research support and assisting in the transfer of hatchery and grow out technologies to the commercial sector. Recent adoption of cage culture technologies based on the joint Oceanic Institute/UH Sea Grant collaboration under the Hawaii Offshore Aquaculture Research Project has further expanded production capability in the sector. As the industry enters into its growth phase, it is critical that we continue to invest in technological advantages to secure the competitiveness and future success of the Pacific threadfin industry.

Advanced hatchery technologies are needed to ensure a continuous supply of high quality and healthy seedstock for growout operations. Currently, production methods for Pacific threadfin are based on egg generation from wild-collected broodstock and methods for controlling broodstock health, reproductive development, spawning, and egg quality are all rudimentary. Fish bred to accommodate the stresses of domestication will grow and survive better under captive culture which would improve overall production efficiencies. The development of select genetic strains will further ensure market competitiveness. Available estimates of heritable improvements in fish growth performance through genetic selection typically range from 10 to 23% per generation of selection amongst species examined to date. The majority of the costs associated with commercial operations (aside from feeds) are tied to the rates of production or growth. Thus, improvements in growth performance will reduce time to market and yield immediate gains in farm profitability.

Methodology

Project efforts began in 2000 with the establishment of a partially domesticated "founder" broodstock population composed of ten wild-collected males and ten F1 females (Figure 1). These stocks were maintained in 25m^3 outdoor broodstock tanks provided with a continuous supply of 26 to 27 °C saltwater derived from the OI/Sea Life Park wells. Stocks were fed to satiation daily with mixed diets of raw smelt and squid occasionally supplemented with pellets (Moore Clark Marine Grower). Reproductive development of developing stocks was tracked through daily examination of tank outflow for the appearance of eggs in egg collection nets and periodic tank rotation and maturation checks by way of gonadal biopsy.

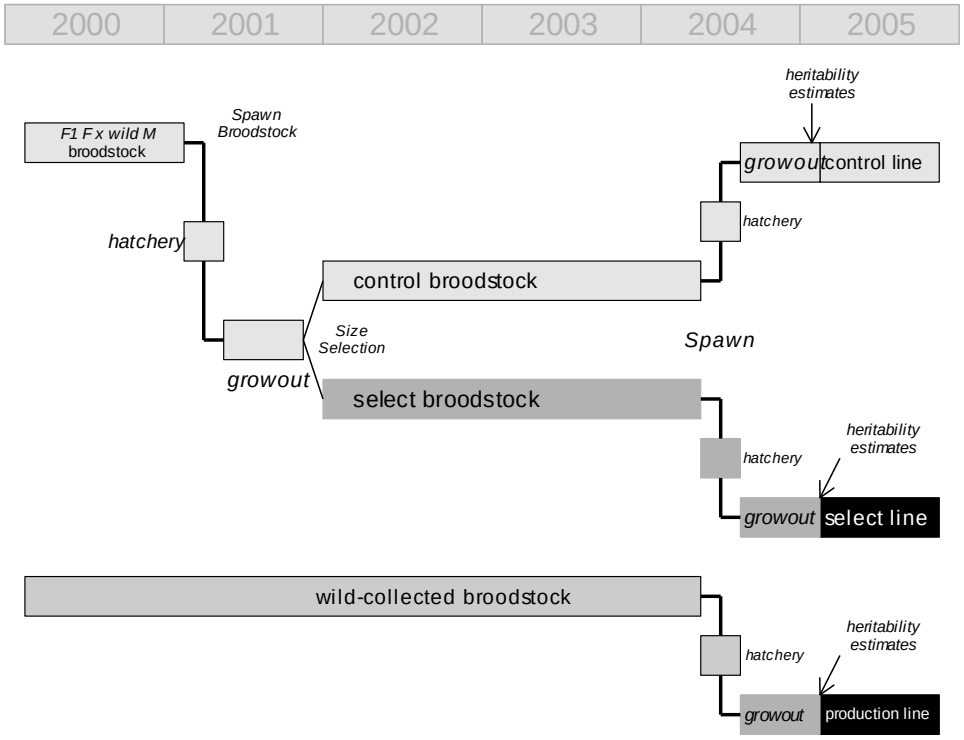

Figure 1. Schematic representation of the time line for a single round selective breeding for the Pacific threadfin at the Oceanic Institute research facilities at Makapu'u Point, Hawaii.

The founder broodstock population initiated spawning activity in December 2000, although initial attempts to rear eggs were unsuccessful due to poor hatch rates. In February 2001, a relatively small batch of fertile eggs was successfully reared through hatchery and nursery phases to generate approximately 600 juveniles for growout and size selection. This domesticated line was then stocked in a single 20m^3 outdoor growout tank provided with a continuous supply of OI/SLP saltwater at approximately 8 to 10 turnovers per day. Fish were grown on a marine grower diet (Moore Clark) originally developed at OI for growout of mahi-mahi. Diet was provided by way of

automatic feeders for the first two months, followed by daily satiation feeding by hand until harvest in August 2001, at approximately six months of age.

At harvest all fish were weighed and measured and 50 control fish (mean weight 377g) were randomly selected to form the "control" line followed by selection of the 50 largest fish (mean weight 516g) to form the "growth-selected" line. These fish were individually implanted with passive inductive transducers (PIT tags) for individual identification and stocked in separate broodstock tanks (control and growth selected) for maturation and eventual spawning. During this maturation period stocks were periodically (approximately each half-year) inspected for growth and reproductive development. The maturation of domesticated stocks proved much slower than anticipated resulting in significant delay in obtaining viable eggs from both control and select lines.

In July 2004, both the domesticated and growth selected broodstock populations spawned viable eggs in the same month, generating seedstock for growth performance evaluations. Although spawns were small, broodstock generated sufficient numbers of eggs to stock 1,000-L larval rearing tanks. Larvae from each of the lines were successfully reared using standard Pacific threadfin hatchery and nursery production procedures yielding 716 domesticated control fingerlings and 1,030 growth selected fingerlings. In addition, another 722 fingerlings from wild collected broodstock (wild controls) were recruited for comparison with the domesticated lines. Each of these fingerlings lines was kept in separate $20m^3$ growout tanks until three month of age at which time they had reached sufficient size (>10cm) to survive the stressors of the tagging process. Each fish was triple tagged, once with coded-wire-tags for specific identification and twice with color-coded visible elastomer implants (Figure 2) in the adipose tissue behind each eye for easy visual identification.

After tagging, fish were split into two replicated $20m^3$ growout tanks with evenly mixed juveniles from each of the three treatment groups for subsequent growout to market size. Fish were randomly sampled monthly to monitor changes in weight and length among lines. To ensure the most accurate assessment of growth performance possible, all fish were individually weighed and measured at six, seven, and eight months of age.

Figure 2. Photographs of the fingerling tagging process. Top left: Technical staff at the Oceanic Institute in the process of tagging over 2000 Pacific threadfin fingerlings. Top right: Juveniles are lightly anaesthetized and implanted with individual coded wire tags. Bottom left: Fish then received fluorescent elastomer implants in the adipose tissue behind the eye marking the treatment groups. Bottom right: fingerlings swimming in the recovery tanks immediately following implantation. Note the highly apparent coloration behind the eye of each fish.

Results and Discussion

Broodstock maturation. Control and fast-growth fish were selected from a post-growout population at approximately six months of age at a mean weight/length of 358g/25.6cm. From this population a 50 animal "control group" with mean weight/length of 378g/25.6cm and a 50 animal "select" group with mean weight/length of 516g/28.9cm were established (Table 1). The calculated selection differential (D) between selected and the population from which they came was 44.1%.

The control and select groups were then monitored for growth and reproductive development for another three years at which time these broodstock finally yielded sufficient numbers of viable eggs to generate the next generation. Maturation checks at 11 months of age revealed that in addition to maintaining significant size advantage over controls (31%), the growth selected fish also entered the primary male phase of reproductive development much quicker than smaller control animals (Table 1). At 16 months the growth selected broodstock continued to maintain a significant size advantage (now 34%) and again showed considerable advancement in sexual maturation, with the majority of the select group having rapidly proceeded to the secondary female stage of sexual development (0 immature, 12 males, 35 females), while the control group was slower to develop reproductively (12 immature, 22 males, and 13 females). This data suggests that size, rather than environmental or behavioral conditions appear to be more important in determining the timing of sexual development and sex change in captive stocks of Pacific threadfin.

Table 1. Survival, growth and reproductive development of control and select parental stocks of Pacific threadfin. Individual fish (identified by PIT tags) were weighed, measured, and sexed at approximately 6, 12, and 16 months age. Reproductive maturation is represented as the ratio of reproductively immature (I), male (M), and female (F) fish in each group.

Age (mth)	Date	Survival		Weight (g)		Length (cm)		Reprod. Matur. (I:M:F)	
		control	select	control	select	control	select	control	select
6	Aug 01	100%	100%	377	516	25.6	28.9	50:0:0	50:0:0
11	Jan 02	94%	100%	686	896	30.9	34.2	37:10:0	9:41:0
16	Jun 02	94%	98%	730	980	32.4	35.9	12:22:13	0:12:35
31	Sep 03	94%	84%	947	1173	35.6	38.2	11:22:14	1:10:31

The growth selected group initiated sporadic spawning activity in May 2003 (27 months age) and the control group began sporadic spawning in September (31 months age). Typical of first spawning fish, spawning activity is highly sporadic and fertility rates are quite low (<15%) impairing hatchery stocking. Attempts to stimulate spawning using hormonal implants, and thus speed up project advancement, were relatively unsuccessful. In July 2004, at ~3½ years of age, both populations produced fertile spawns allowing hatchery stocking for the final phase of the project.

Biological performance of select lines. In July 2004, both the domesticated F1 control and growth-selected F1 broodstock populations spawned in the same month, generating fingerlings for determination of heritability of growth performance of Pacific threadfin in captivity. In addition, a second "control" group of fingerlings (F1 controls) was reared in parallel for reference. Larvae from all three lines were successfully reared using standard Pacific threadfin hatchery procedures. An unexpected finding was that both domesticated lines, and in particular the growth-selected line, had much lower rates of opercular deformities than fingerlings generated from wild-collected broodstock (Fig. 3).

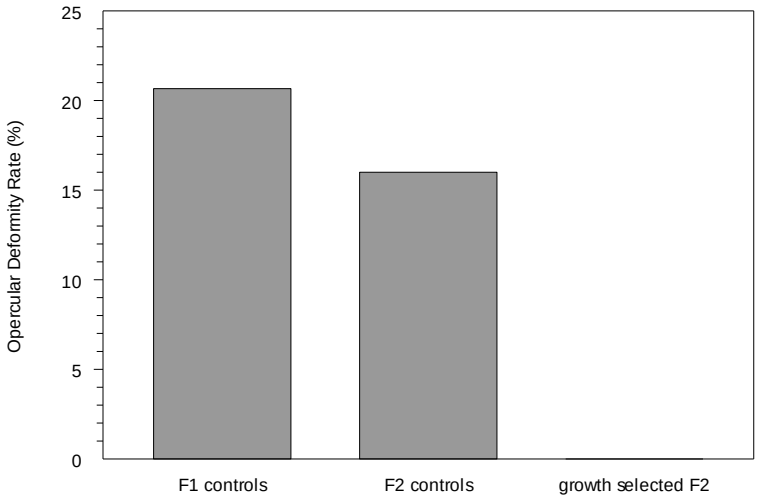

Figure 3. Opercular deformity rates of fingerlings generated from wild-collected broodstock (F1 controls), domesticated broodstock (F2 controls), and growth-selected F2 broodstock (n=25).

Lines were maintained in separate growout tanks until three months of age when most juveniles had reached sufficient size (>10cm) to survive the stressors associated with tagging. Note that juveniles less than 10cm in length, known as runts, generally do not survive the tagging process and were culled from the populations. Interestingly, about 10% of the F1 controls and F2 controls were runts while less than 1% of the growth selected F2 juveniles were runts. Although unavoidable, the culling of more runts from controls than growth selected line created an artificial bias for domesticated controls over the selects. Fish were then split into two replicated $20m^3$ tanks evenly mixed fish from each of the three treatment groups for subsequent growout of juvenile to market size (Figure 4). Note that this approach allows for more direct comparison of growth performance under identical conditions but prevents collection of feed conversion efficiency data.

Despite coming out of hatchery at slightly smaller size (0.7g) than F2 controls (1.1g), the growth selected F2 line began to exhibit better growth performance compared to the F2 control group by about four months of age and improved growth performance compared to F1 controls, generated from wild-collected broodstock, by six months of age. At the end of this study the growth selected F2 line demonstrated a 25% increase in weight compared to the non-selected F2 controls and 17% increase in weight compared to F1 controls derived from wild-collected broodstock suggesting that growth performance is highly heritable in the Pacific threadfin. It is interesting that the relative improvement in growth performance does not manifest itself during hatchery, nursery, or early growout stages suggesting the opportunity for selection at multiple stages in the production process. The poorer performance of the domesticated (F2) controls compared to F1

fingerlings derived from wild-collected broodstock is not unique to this study, and is a common finding associated with the early stages of domestication.

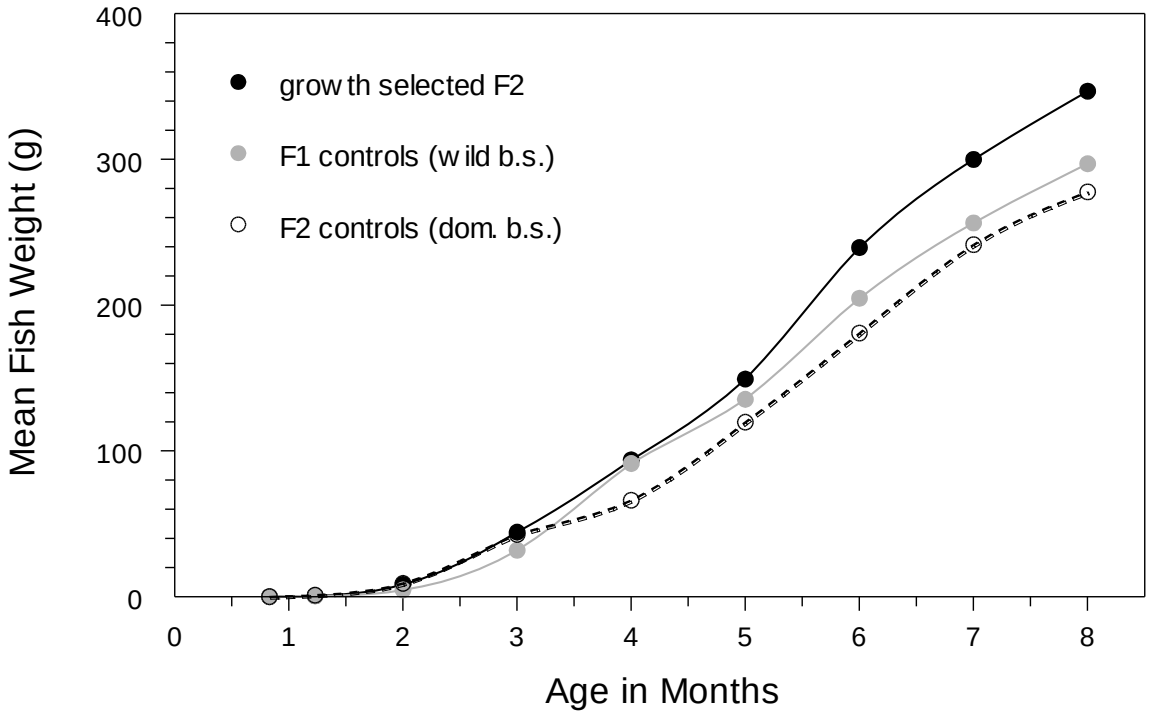

Figure 4. Growth (i.e., changes in body weight) from hatchery at day 25 until harvest of juvenile Pacific threadfin derived from growth selected F2 broodstock (closed circles), wild-F1 control broodstock (grey closed circles), and domesticated F2 control broodstock (open circles, dashed line).

Despite considerable improvement in mean and median growth performance through selection, the growth-selected line maintained considerable range in size at harvest. It appears that most of the selective growth advantage can be accounted for a greater frequency of faster growing individuals (Figure 5). It is not clear whether further rounds of selection would help reduce the number of runts (i.e., smaller fish) or whether the lower growth rate is more related to environmental factors. We also noted that six month harvest weights in the second phase of this study (181 to 240g) were considerably lower than in the phase one group from which the control and growth selected lines were derived (377g). The similar growth rates seen in the F1 controls derived from wild broodstock suggests that this phenomena is not linked to domestication, but more likely related to the higher growout densities and frequent stressing of the animals associated with the monthly handling of fish necessary to generate the required growth performance data. Fish sampled around market size at seven and eight months of age showed remarkably similar fillet ratios, with mean values ranging from 52 to 54% and clearly not significantly different between treatment growth lines.

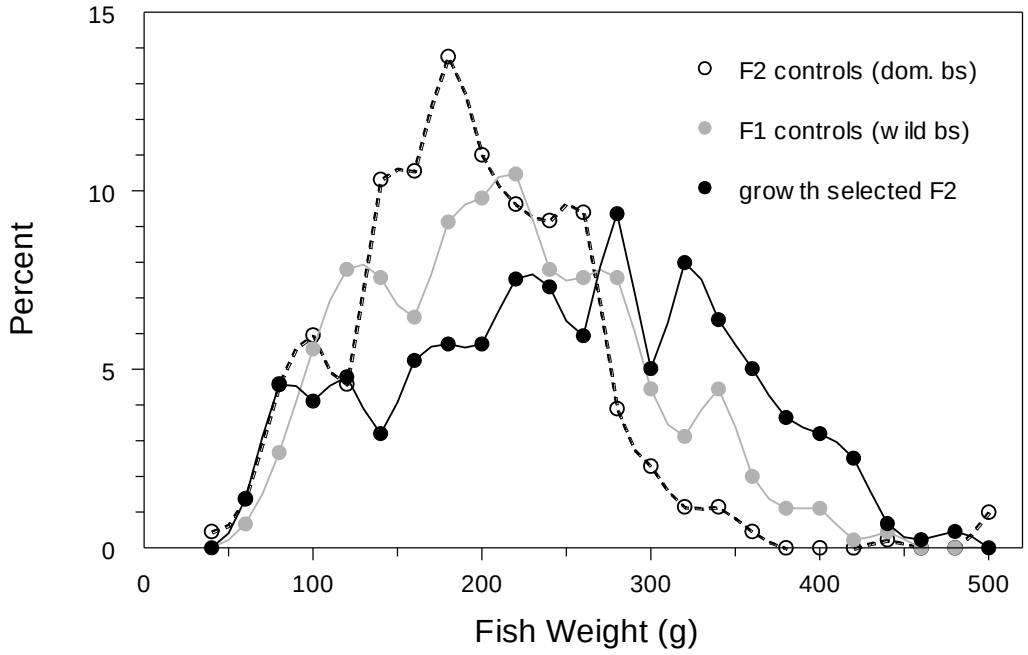

Figure 5. Relative size distribution of the three growth lines, domesticated controls (green), production controls (red) and growth-selected (yellow) of Pacific threadfin at four months growout.

Conclusion

This study demonstrates that genetic selection for growth offers a relatively simple, although long-term process to improve the relative biological performance for captive stocks of Pacific threadfin. Although these studies are clearly rudimentary in scope, they demonstrate that growth performance in this species is highly heritable. Improvements in growth performance will reduce time to market and yield overall gains in farm profitability. These benefits would be further enhanced through further rounds of selection. However, a more extensive breeding program for Pacific threadfin and other marine species in Hawaii will require long-term support in terms of both resource allocation and financial support. These efforts also indicate a continuing need for basic research on reproduction and growth processes in support of this growing industry and the application of modern breeding techniques, including molecular genetics to accelerate progress.

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